

Text-book of histology : including the microscopic technic / by Dr. Philip Stöhr.

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TEXT-BOOK
OF
HISTOLOGY

INCLUDING
THE MICROSCOPIC TECHNIC

BY
DR. PHILIPP STÖHR
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FIFTH AMERICAN FROM THE TENTH GERMAN EDITION

TRANSLATED BY DR. EMMA L. BILSTEIN
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EDITOR'S PREFACE TO THE FIFTH EDITION.

This edition has been revised in accordance with the tenth German edition, and presents much that is new, both in revision of the former text and additions throughout the entire work. Among the most important changes are—

Fifty new illustrations, a number in colors, and new color schemes of the Spleen, Lung, Kidney, and Retina.

Five new chapters upon the following subjects :

The Form of Glands, based on the investigations of Maziarski.

The Spleen, based on the researches of Weidenreich.

The Urinary Bladder, based on the researches of Lendorf.

The Seminal Passages, based on the researches of Felix.

The Development of the Hairs, based on the author's own researches.

There are, besides, many lesser alterations and additions, including the latest investigations on the Morphology of the Cell and the Structure and Chemistry of the Tissues ; also the addition to the technical part of the application of Formol and its mixtures.

The editor acknowledges gratefully the appreciation of Professor Stöhr's work shown by many teachers and students, and trusts that this new edition may yet increase its usefulness.

ALFRED SCHAPER.

BRESLAU, *August, 1903.*



EDITOR'S PREFACE TO THE FIRST EDITION.

Stöhr's text-book is well known to the histologists of all nations and held in high esteem by them. To the German medical student it has become an indispensable guide. During the ten years of its existence it has reached an extraordinary sale and passed through six revised editions. It has been translated into Italian (1887), French (1890), and Russian (1891), and has thus come into the hands of the students of these nations. These facts are sufficient to guarantee the value of the work without further recommendation. Although excellent text-books of Histology already exist in English, still the peculiarity and special superiority of Stöhr's text-book justifies, in our opinion, its translation into English for the convenience of American and English students.*

It is especially intended for the use of students, but even professional histologists and physicians will find in it much valuable information, as well as suggestions for technical purposes. The chief merit of the work lies, on the one hand, in the brevity and perspicuity of the descriptive text, elucidated by illustrations which have thus far never been excelled; and, on the other hand, in the simplicity and certainty of the methods for preparing the most important microscopical specimens. The young student is thus enabled to practice histological methods privately, at a minimum cost, in connection with his courses in the university. The preparation of almost all of the specimens enumerated in the book can be made simply by means of teasing, isolation, or cutting with the razor, but those students who have a microtome at their disposal will also find, in an Appendix, brief directions for the preparatory treatment (embedding in paraffin and celloidin) of specimens for sectioning with the microtome.

With the permission of Prof. Stöhr we have made several immaterial, but for an American edition very desirable, changes in the text, and have considered it preferable to place the technical part as a whole at the end of the book rather than in sections after the several

* In 1888 Stöhr's text-book was utilized in Kendrick's Physiology, but in such a fragmentary form and so intermingled with selections from other authors that its chief merits were entirely lost. This use of the book can not be considered as an English translation proper.

chapters. Furthermore, we have enlarged the chapter on the Uterus, in order to give detailed consideration to the various functional conditions of the organ, and added to the book an entirely new chapter on the Placenta. Eight new illustrations (Figs. 229, 230, 232, 233, 234, 236, 237, 238) were necessary for these additions.

The editor is under great obligation to the translator, Dr. Billstein, for her successful efforts in reproducing the conciseness and clearness of the German original. Further, he desires to express his gratitude to Prof. Philipp Stöhr for placing at his disposal the original electrotypes, and to Drs. Böhm and von Davidoff for the illustration of the virginal uterus (Fig. 229) from their "Lehrbuch der Histologie." He also feels deeply indebted to Prof. Charles S. Minot for kind assistance, for valuable criticism, and for permission to use two illustrations (Figs. 231 and 234) from his text-book of "Human Embryology"; and, finally, to Messrs. P. Blakiston, Son & Co., Philadelphia, for the very satisfactory reproduction of the new drawings, and for their many courtesies during the preparation of the American edition.

ALFRED SCHAPER.

HARVARD MEDICAL SCHOOL,
BOSTON, *June, 1896.*

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PART I.

GENERAL TECHNIC.

I. THE LABORATORY APPOINTMENTS.

1. INSTRUMENTS.

The Microscope.—From my own experience I can recommend the microscopes made in the optical works of Leitz in Wetzlar, Seibert in Wetzlar, and Zeiss in Jena, having repeatedly tested their excellent workmanship.*

It is not advisable for the beginner to purchase a microscope without first submitting it to an expert for examination. In order to preserve the microscope in good working condition it is necessary to protect it from dust; when in frequent use it is best to keep it under a bell-glass, in a place not exposed to sunlight. The tarnish which forms on the tube should be rubbed off with a dry piece of soft filter-paper.

* Students of the first semester are advised to refrain from the purchase of high-power oculars and immersion-systems. These should be bought shortly before entering upon bacteriologic work.

The following outfits are recommended:

Leitz.—Catalogue No. 36, 1895. Microscope No. 4 *b*. Price, 370 *M.* = \$92.00. Without homogeneous immersion and ocular IV, 265 *M.*

Seibert.—Catalogue No. 25, 1895. Microscope 3 *c*. Price, 449 *M.* = \$112.00. Without homogeneous immersion, objective 3, and ocular 0, 283.50 *M.* = \$71.00.

Zeiss.—Catalogue No. 30, 1895. Combination (p. 116) 7 *b*. Price, 602 *M.* = \$150.00. Without homogeneous immersion, 442 *M.* = \$110.00; or 8 *b*. Price, 559 *M.* = \$140.00. Without homogeneous immersion, 399 *M.* = \$100.00.

The majority of the work for this book was carried out with a Leitz microscope.

Editor's remark: Of American microscopes, those made by the Bausch & Lomb Optical Co., Rochester, N. Y., and New York City, are recommended.

For histologic work the following outfit is suitable:

Stand BB.—Oculars, 1-inch and 3-inch. Objectives, $\frac{2}{3}$ -inch and $\frac{1}{8}$ -inch. Catalogue 1895. Price, \$62.50. For cytologic and bacteriologic work a $\frac{1}{2}$ -inch oil-immersion objective (price, \$44) and an Abbé condenser and iris-diaphragm should be added. For convenience a double or triple revolver for the objectives is desirable.

Smirches on the lenses* and on the mirrors should be removed with soft leather, and if this does not answer the purpose,—as, for example, when a lens is smeared with canada-balsam,—a small piece of fine linen moistened with a drop of pure alcohol should be used. In the latter procedure great care must be exercised lest the alcohol penetrate the setting of the lenses and dissolve the balsam with which they are cemented together. Therefore the balsam should be *quickly* rubbed off with the moistened linen and the lens carefully dried. After using an immersion lens the cedar oil clinging to it should be removed by means of a patch of linen moistened with benzin; the free surface of the cover-glass of the preparation examined with the immersion lens should be cleaned in the same way. The screws of the microscope should be cleaned with benzin.

A good *razor*, flat on one side. It should always be kept sharp, and before each use should be drawn without pressure over the *strop*. The honing of it should be left to the instrument-maker. The razor should be used only in the preparation of microscopic sections.

A fine *whetstone*.

A pair of small, straight *scissors*.

A pair of easily-closing small *forceps*, with smooth or only slightly grooved points.

Four *dissecting needles* with wooden holders: two are to be heated, then slightly bent, heated again and thrust into solid paraffin, by which they are again hardened. The other two must be kept clean and sharply pointed; for delicate dissections the needles must be pointed and polished, first on the whetstone and then on the strop.

A flexible *section-lifter*, for the transfer of sections from fluids to the slide, is very useful but not absolutely necessary. A scalpel having a broad blade can be used instead.

Pins, quills, cork disks, a fine sable brush.

A *crayon*, for writing on glass. (If the glass is oily it must first be cleansed with alcohol.)

Slides, of clear glass, not more than 1 to 1.5 mm. in thickness, with the edges ground.

Cover-glasses, measuring 15 to 18 mm. in diameter, are generally large enough; the thickness may vary from 0.1 to 0.2 mm.; those with a greenish shimmer at the edges are preferable to the pure white covers, which blur in time.

* The objective lenses must not be unscrewed.

Small, wide-mouthed bottles. One dozen, capacity 30 c.c. and over, with cork stoppers.

Several glass *preparation jars* (preserve jars), with tightly-fitting covers. Height, 8 to 12 cm. ; diameter, 6 to 10 cm.

A *cylindrical graduate*, capacity 100 to 150 c.c.

A glass *funnel*, upper diameter 8 to 10 cm.

A *pipet*. Small pipets may be prepared by heating in a gas-flame a glass tube 1 cm. thick and 10 cm. long, pulling one end to a point and placing on the other a small rubber bulb.

A dozen *watch-glasses* of 5 cm. diameter.

A dozen *test-tubes*, 10 cm. long and 12 mm. wide.

Glass rods, 3 mm. thick, 15 cm. long, some drawn to a point at the end.

Old bottles that have been thoroughly cleansed will answer as receptacles for reagents. In most cases the bottles can be cleansed with water, but sometimes it is necessary to rinse them with crude hydrochloric acid or with potash lye, then with ordinary water, then with distilled water, and finally with alcohol.

Glass dishes ("Stender" dishes) 6 to 8 cm. in diameter, with ground covers, are not absolutely necessary, but very useful.* In many cases they may be replaced by saucers, food dishes for birds, etc.

A few sheets of thin, white *filter-paper*, large and small gummed labels, soft pieces of linen (old handkerchiefs), a towel, a large and a small bottle-brush.

A large earthen jar for refuse.

2. REAGENTS.†

General Rules.—Large quantities of reagents should not be kept on hand, because many decompose in a comparatively short time. Certain reagents (see below) should be procured or prepared shortly before they are to be used. Each bottle should be provided with a large label on which its contents are designated; it is advisable to write on the label not only the formula of the reagent, but also the mode of its application.

* Most of the glassware, including slides and cover-glasses, here enumerated may be obtained of W. P. Stender, Leipzig; or, in the United States, of the Bausch & Lomb Optical Co., New York.

† The reagents must be obtained from a reputable dealer. Excellent dyes and reagents may be had of Dr. Grübler, chemical and physiological laboratory, Leipzig, Bayer'sche Strasse 63. In the United States Grübler's stains and reagents are sold by Eimer & Amend, New York, and others.

All the bottles must be tightly closed with cork or well-made glass stoppers. The fluid should not reach to the lower surface of the cork.

1. *Distilled water*, 3 to 6 liters.

2. *Normal salt solution*, 0.75 per cent. (sodium chlorid, 1.5 gm., distilled water, 200 c.c.).

The cork must be provided with a glass rod reaching to the bottom of the bottle. This solution spoils easily and must be frequently prepared afresh.

3. *Alcohol*.—(a) *Ninety-five per cent. alcohol*.—About 500 c.c. should be kept on hand. The alcohol of commerce is 95 per cent., and in the majority of cases is entirely satisfactory for microscopic purposes. If it is desired to obtain alcohol free from water (absolute alcohol), drop into the bottle a few pieces of copper sulfate heated until white (15 gm. to 100 c.c. of alcohol). When these become blue they must be replaced by new pieces or be reheated. Fresh quicklime serves the same purpose, but acts more slowly.*

(b) *Ninety per cent. alcohol*.—Prepare 500 c.c. by diluting 475 c.c. of 95 per cent. alcohol with 25 c.c. of distilled water.

(c) *Eighty per cent. alcohol*.—Prepare 500 c.c. by diluting 425 c.c. of 95 per cent. alcohol with 75 c.c. of distilled water.

(d) *Seventy per cent. alcohol*.—Prepare 500 c.c. by mixing 370 c.c. of 95 per cent. alcohol with 130 c.c. of distilled water.

(e) *Fifty per cent. alcohol*.—Prepare 500 c.c. by mixing 265 c.c. of 95 per cent. alcohol with 235 c.c. of distilled water.

(f) *Thirty-three per cent. alcohol*.—(Ranvier's one-third alcohol).—This is prepared by mixing 40 c.c. of 95 per cent. alcohol with 60 c.c. of distilled water.

4. *Acetic acid*, 50 c.c.—The official is 30 per cent.

5. *Glacial acetic acid*.—This should be procured shortly before it is required. The commercial acid is 96 per cent.

6. *Nitric acid*.—A bottle holding 100 c.c. of concentrated nitric

* For the preparation of mixtures containing a smaller percentage of alcohol this equation will serve :

$$\begin{aligned} 100 : 95 &= x : \% \\ \text{e. g., } 90\% , 100 : 95 &= x : 90 \\ 95x &= 90 \cdot 100 \\ x &= \frac{9000}{95} = 94.7 \text{ or } 95. \end{aligned}$$

Therefore, to obtain 100 c.c. of 90 per cent. alcohol, 95 c.c. of 95 per cent. alcohol must be mixed with 5 c.c. of distilled water. For our purposes the errors of this ratio are too insignificant for consideration.

acid of 1.18 sp. gr. (containing 32 per cent. of acid hydroxid) should be kept in stock.

7. *Hydrochloric acid*, pure, 50 c.c.

8. *Formol*.—The aqueous 40 per cent. solution of formaldehyde occurs under two designations in commerce: (a) formol (Meister, Lucius & Brüning in Höchst am Main); (b) formalin (Chem. Fabrik auf Aktien, formerly Schering, Berlin). For microscopic purposes formalin is the less suitable.

9. *Chromic acid*.—A 10 per cent. stock solution should be prepared by dissolving 10 gm. of fresh crystalline chromic acid in 90 c.c. of distilled water. From this prepare:

(a) A 0.1 per cent. chromic-acid solution (10 c.c. of stock solution to 990 c.c. of distilled water), and—

(b) A 0.5 per cent. chromic-acid solution (50 c.c. of stock solution to 950 c.c. of distilled water).

10. *Potassium bichromate*.—This should be kept on hand in two solutions:

(a) Thirty gm. to 1000 c.c. of distilled water.

(b) Thirty-five gm. to 1000 c.c. of distilled water (for Kopsch's fluid, No. 12, and for the Golgi mixture, No. 16).

At room temperature it dissolves in from three to six days. Therefore make the solutions with warm water or place the bottles near the stove.

11. *Potassium-bichromate-acetic acid* (Tellyesnickey's fluid). To be prepared shortly before using, by adding 5 c.c. of glacial acetic acid to 100 c.c. of 3 per cent. solution of potassium bichromate (No. 10 a).

12. *Potassium-bichromate formol* (Kopsch's fluid). To be prepared shortly before using, by adding 20 c.c. of 40 per cent. formol (No. 8) to 80 c.c. of 3.5 per cent. solution of potassium bichromate (No. 10 b).

13. *Müller's fluid*.—Dissolve 30 gm. of sodium sulfate and 60 gm. of pulverized potassium bichromate in 3000 c.c. of distilled water. The solution can be made with the aid of heat, like No. 10.

14. *Müller-formol mixture* (Orth's mixture). Invariably to be prepared immediately before using, by mixing 10 c.c. of formol (No. 8) with 100 c.c. of Müller's fluid (No. 13).

15. *Zenker's fluid*.—Dissolve 25 gm. of potassium bichromate, 10 gm. of sodium sulfate, and 50 gm. of mercuric chlorid in 1000 c.c. of warm distilled water. Before using add 1 c.c. of glacial acetic acid to each 20 c.c. of the mixture.

16. *Golgi's mixture* (osmio-bichromate mixture).—This is prepared by pouring together 54 c.c. of the 3.5 per cent. solution of potassium

bichromate (10 b) and 6 c.c. of the 2 per cent. osmic-acid solution (No. 22). It should be prepared shortly before it is to be used.

17. *Cox-Golgi mixture*.—This is prepared by pouring together 40 c.c. of a 5 per cent. solution of potassium bichromate, 40 c.c. of a 5 per cent. solution of corrosive sublimate, 32 c.c. of a 5 per cent. solution of potassium chromate, and 88 c.c. of distilled water. This mixture may be kept in stock.

18. *Ten per cent. phosphomolybdic acid*.—Fifty c.c., kept in the dark.

19. *Iron solution*.—Dissolve 2.5 gm. of ferric alum— $(\text{NH}_4)_2\text{Fe}_2(\text{SO}_4)_4$ —in 100 c.c. of distilled water.

20. *Picric acid*.—Keep on hand 50 gm. of the crystals and 500 c.c. of a saturated aqueous solution, in which undissolved crystals in a stratum 2 to 3 mm. deep must always lie on the bottom of the bottle. It dissolves readily.

21. *Chromic-acetic acid*.—To 50 c.c. of the 0.5 per cent. chromic-acid solution (9 b) add 50 c.c. of distilled water and from 3 to 5 drops of glacial acetic acid.

22. *Osmic acid*.—This may be obtained from the dealer—50 c.c. of a 2 per cent. solution—shortly before it is needed. It is very expensive. It should be kept in the dark or in a dark glass bottle and if well stoppered can be preserved many months.

23. *Chromic-acetic-osmic acid* (Flemming's mixture).—Prepare a 1 per cent. chromic-acid solution (5 c.c. of the 10 per cent. solution [No. 9] to 45 c.c. of distilled water) and add 12 c.c. of 2 per cent. osmic acid and 3 c.c. of glacial acetic acid. This mixture is not injured by light and can be kept in stock.*

24. *Platinum chlorid*.—Prepare a 10 per cent. stock solution, 2 gm. dissolved in 20 c.c. of distilled water.

25. *Platinum-acetic-osmic acid mixture* (Hermann's mixture).—Pour into 60 c.c. of a 1 per cent. solution of platinum chlorid (6 c.c. of stock solution and 54 c.c. of distilled water) 8 c.c. of 2 per cent. osmic-acid solution and 4 c.c. of glacial acetic acid.

26. *Saturated sublimate salt solution*.—Put 7.5 gm. of common salt into one liter of distilled water; after solution add 125 gm. of crystalline corrosive sublimate and dissolve by the aid of heat. Filter the warm solution. On cooling, white acicular crystals form on the bottom of the bottle.

27. *Silver nitrate*.—A 1 per cent. solution (1 gm. of silver nitrate in

*Tissues fixed in old Flemming's fluid often stain badly, because the acetic acid has evaporated; 5 to 20 drops of acetic acid newly added to the solution removes this defect.

100 c.c. of distilled water) should be procured a short time before it is to be used. In a dark place or in a dark bottle it can be preserved for a long time.

28. *Gold chlorid*.—A solution of 1 gm. of gold chlorid in 100 c.c. of distilled water should be procured shortly before it is to be used. It must be kept in the dark or in a dark bottle. For gold-chlorid staining it is necessary to have No. 29.

29. *Formic acid*, 50 c.c.

30. *Concentrated potash lye* (35 per cent.), 30 c.c. The bottle must have a rubber stopper that is pierced by a glass rod. It should be procured from the druggist.

31. *Glycerol*.—One hundred c.c. of pure glycerol are to be kept in stock; also a solution of 5 c.c. of pure glycerol in 25 c.c. of distilled water. The growth of fungi, which soon takes place in this mixture, may be prevented by the addition of a small piece of camphor or thymol. The cork of the bottle should be provided with a glass rod.

32. *Xylol*.—On account of its sensitiveness in preparations incompletely dehydrated xylol is not recommended to beginners.

33. *Carbol-xylol*.—Prepare by adding 22 gm. of crystalline carbolic acid to 100 c.c. of xylol. This reagent will clear sections that are not fully dehydrated.

34. *Xylol-balsam*.—A solution of canada-balsam in xylol. The cork of the bottle should be provided with a glass rod.

35. *Cover-glass cement*.—Dilute Venetian turpentine with enough ether to make an easily flowing liquid; then filter warm (in a heated funnel) and inspissate the filtrate on a sand-bath. The proper consistency is attained when a drop transferred with a glass rod to a slide hardens at once and becomes so firm that it cannot be indented with the finger-nail. Because of the danger of fire, it is better to have the cement prepared by the druggist.*

36. *Hansen's hematoxylin*.—(a) Dissolve 1 gm. of crystallized hematoxylin in 10 c.c. of absolute alcohol and preserve it in a stoppered bottle. (b) Dissolve 20 gm. of potassium alum in 200 c.c. of distilled water, with the aid of heat and when cold filter. (c) Dissolve 1 gm. of potassium permanganate in 16 c.c. of distilled water, at room temperature. On the next day pour solutions *a* and *b* into a porcelain capsule, add 3 c.c. of solution *c* and, with constant stirring, heat the mixture to boiling and boil about one minute. Cool quickly by floating the

**Editor's remark:* In the United States an excellent fluid cover-glass cement is prepared by J. D. King, Cottage City, Mass.

porcelain capsule in cold water. When cold the mixture should be filtered; it is then ready to use. Cloudiness, or the development of fungi in the mixture, does not depreciate its effectiveness in the slightest degree. It is to be kept on hand.

37. *Delafield's hematoxylin*.—(a) Dissolve 1 gm. of crystallized hematoxylin in 6 c.c. of absolute alcohol. (b) Dissolve 15 gm. of ammonia alum in 100 c.c. of distilled water, with the aid of heat and when cold filter. Pour the two solutions together and let the mixture stand three days in a wide-open vessel exposed to the light; then filter and mix with 25 c.c. of pure glycerol and 25 c.c. of methyl-alcohol. After three days filter the mixture. It does not deteriorate with age and should be kept in stock.

38. *Weigert's hematoxylin*, for the demonstration of the medullated nerve-fibers of the brain and the spinal cord. Heat 1 gm. of crystallized hematoxylin in 10 c.c. of absolute alcohol, plus 90 c.c. of distilled water, and when cold filter. It should be prepared shortly before it is to be used. The application of this stain demands the aid of the following three fluids:

39. *Saturated solution of lithium carbonate*.—Dissolve 3 or 4 gm. of lithium carbonate in 100 c.c. of distilled water. This should be prepared the day before using.

40. *Solution of potassium permanganate (0.25 per cent.)*.—Dissolve 0.5 gm. of potassium permanganate in 200 c.c. of distilled water. This may be kept on hand.

41. *Acid mixture (Pal's mixture)*.—Dissolve 1 gm. of pure oxalic acid and 1 gm. of potassium sulfite (K_2SO_3) in 200 c.c. of distilled water. This mixture should be prepared one day before using and be kept in a well-stoppered bottle.

42. *Mallory's hematoxylin*.—Pour 10 c.c. of 10 per cent. phosphomolybdic acid into 200 c.c. of distilled water; in this dissolve (without heating) 1.75 gm. of crystallized hematoxylin and add 5 gm. of crystalline carbolic acid.

43. *Neutral carmine solution*.—Dissolve 1 gm. of the best carmine in 50 c.c. of cold distilled water to which 5 c.c. of a solution of ammonia (liquor ammonii caustici) have been added. The deep, cherry-red fluid should stand in an open vessel until it has no odor of ammonia (about three days) and then be filtered. It is to be kept in stock. The odor of this solution immediately becomes very disagreeable, but this does not depreciate its staining power.

44. *Picrocarmine*.—Pour 5 c.c. of solution of ammonia into 50 c.c. of distilled water and to this mixture add 1 gm. of the best carmine.

Stir with a glass rod. After complete solution of the carmine (in about five minutes) add 50 c.c. of a saturated solution of picric acid and let the whole stand in a wide-open vessel for two days. It is then to be filtered. Abundant fungous growth does not diminish the staining power of this excellent medium.

45. *Alum-carmine*.—Dissolve 5 gm. of alum in 100 c.c. of warm distilled water and add 2 gm. of carmine. Boil this mixture ten or twenty minutes and when cold filter; finally, to the clear, beautiful, ruby-red fluid add 2 or 3 drops of liquefied carbolic acid.

46. *Carmalum*.—To be obtained in solution of Dr. Grüber.

47. *Borax-carmine*.—Dissolve 4 gm. of borax in 100 c.c. of warm distilled water; when the solution has cooled add 3 gm. of the best carmine, stirring meanwhile, and then 100 c.c. of 70 per cent. alcohol. At the expiration of twenty-four hours the fluid should be filtered. It filters very slowly, requiring twenty-four hours or more.

Staining with borax-carmine requires after-treatment with 70 per cent. acid-alcohol, which is prepared by adding 4 or 6 drops of pure hydrochloric acid to 100 c.c. of 70 per cent. alcohol.

48. *Paracarmine*.—Dissolve 4 gm. of carminic acid (Grüber), 0.5 gm. of aluminum chlorid, and 4 gm. of calcium chlorid in 100 c.c. of 70 per cent. alcohol. This preparation keeps for a long time.

49. *Sodium carminate*.—Dissolve 2 gm. of pigment in 200 c.c. of distilled water.*

50. *Safranin*.—Dissolve 2 gm. of pigment in 60 c.c. of 50 per cent. alcohol (32 c.c. of 95 per cent. alcohol in 28 c.c. of distilled water). It is to be kept in stock.

51. *Eosin*.—Dissolve 1 gm. of pigment in 60 c.c. of 50 per cent. alcohol. This should be kept in stock.

52. *Orange*.—Dissolve 1 gm. of pigment in 60 c.c. of 50 per cent. alcohol.

53. *Congo-red*.—Dissolve 1 gm. of pigment in 100 c.c. of distilled water. From this stock-solution prepare—

(a) A $\frac{1}{30}$ per cent. solution: 3 c.c. of stock-solution in 100 c.c. of distilled water.

54. *Vesuvium*, or—

**Editor's remark*: Of the carmine stains, *alum-cochineal* should be highly recommended. Because of the certainty of its action and the simplicity of its application it is very useful in the hands of the beginner. It is prepared by boiling 60 gm. of powdered cochineal and 60 gm. of alum in 800 parts of water for about twenty minutes, filtering the decoction, and adding a small piece of camphor or thymol to prevent the growth of mold. It can be kept in stock for a long time.

55. *Methyl-violet B*, may be kept in stock in a saturated aqueous solution (1 gm. in 50 c.c. of distilled water).

56. *Methylene-blue*.—Dissolve 1 gm. in 100 c.c. of distilled water. This solution keeps well, as does the following, which is required for after-treatment.

57. *Ammonium picrate*.—Dissolve 3 gm. in 100 c.c. of distilled water.

58. *Acid fuchsin* (= rubin S).—Dissolve 1 gm. of the pigment in 100 c.c. of distilled water.

59. *Van Gieson's picrofuchsin*.—To 10 c.c. of 1 per cent. solution of acid fuchsin (No. 58) add 100 c.c. of saturated aqueous solution of picric acid (No. 20).

60. *Resorcin-fuchsin after Weigert* (modification of Pranter).—Dissolve 0.02 gm. of the dry pigment, to be obtained of Dr. Grüber, in one part by weight (not volume) of official nitric acid and 100 parts by weight of 70 per cent. alcohol.

61. *Westphal's alum-carmin dahlia*.—Dissolve 1 gm. of dahlia in 25 c.c. of absolute alcohol, add 12 c.c. of pure glycerol and 5 c.c. of glacial acetic acid, and pour into this mixture 25 c.c. of alum-carmin (No. 45, p. 25). Preserve in a well-stoppered bottle.

II. THE PREPARATION OF MICROSCOPIC SPECIMENS.

INTRODUCTION.

Very few organs of the animal body are of a structure suitable for microscopic examination without special preparation. They must possess a certain degree of transparency, which is attained either by separating the organs into their elements or by cutting them into thin sections—that is, either by *isolating* or by *sectioning*. Further, very few organs possess a consistency that, without treatment, allows of the cutting of sufficiently thin sections; they are either too soft, in which case they must be *hardened*, or too hard (calcified), in which case they must be *decalcified*. But fresh objects can be neither *hardened* nor *decalcified* without injury to their structure; both processes must be preceded by treatment which rapidly kills the structural elements and at the same time preserves their natural form. This procedure is called *fixation*. Usually, the preparation of thin sections is possible only after fixation and hardening, followed in some cases by decalcification, of the object. The sections, too, require further treatment; they may be forthwith rendered transparent by means of *clearing media* (which can be also successfully used in the examination of fresh objects), or they may be *stained* before being made transparent. The staining materials are invaluable aids in microscopic investigations. They can be applied in the examination of fresh and even of living organs. A large number of the most important facts have been discovered by means of them. Introduced into the blood-vessels, *injected*, they enable us to trace the branching and course of their finest ramifications.

§ 1. NATURE OF THE MATERIAL.

For the study of the structural elements and the simplest tissues, amphibians (frogs, salamanders) are recommended. The best is the spotted salamander,* the elements of which are very large. For the study of organs, mammals should be chosen. In many cases our

**Editor's remark:* Or the American *Amblystoma*, *Necturus*, etc.

rodents (rabbits, guinea-pigs, rats, mice), also young dogs, cats, etc., are suitable. Still no opportunity to secure human organs should be neglected. Perfectly fresh material can often be obtained at surgical clinics. Material may also be had at autopsies, if not made too long after death; with the exception of the mucous membrane of the intestinal tract, which decomposes very quickly after death, many organs can be used.

In general it is advisable to place the organs while yet warm in the fixing fluid. In order to accomplish this the following injunctions must be observed: Fill the bottles selected for the reception of the objects with the appropriate fluid and provide them with a label on which is designated the object, the fluid, the date, and in some cases the hour; then place the dissecting instruments near at hand; then kill the animal.*

§ 2. KILLING AND DISSECTING THE ANIMALS.

Kill amphibians by cutting through the vertebral column of the neck with strong scissors and destroy brain and spinal cord by means of a needle introduced through the wound into the vertebral canal and the cranial cavity. Cut the throat of mammals by a deep incision reaching as far back as the vertebral column, or pour chloroform on a cloth and press it to the nose of the animal.† Small animals, up to the size of four centimeters, and embryos may be placed entire in the fixing fluid; after about six hours the thoracic and abdominal cavities should be opened by incisions. In the dissection, if possible, an assistant should hold the extremities of the animal. Small animals can be extended on cork or wax plates and secured by strong pins thrust through the feet. The organs must be carefully removed. This is best done with scissors and forceps. Crushing or pressing the parts, or taking hold of them with the fingers, must be entirely avoided. Only the edge of the object may be grasped by the forceps. Attached foreign matter, mucus, blood, contents of the intestines, must not be scraped off with the scalpel, but should be removed by slow twirling in the respective fixing fluids [or by gently shaking the object in normal salt solution (p. 20) before placing it in the fixing medium.—ED.].

In the following methods it is not possible to avoid moistening scissors, forceps, needles, glass rods, etc., with different fluids, for example,

* To take parts from the *living* animal is an entirely needless cruelty!

† *Editor's remark*: I prefer to kill medium-sized and small animals (rabbits, guinea-pigs, cats, mice, etc.) by placing them under a sufficiently large bell-glass, together with a wad of absorbent cotton saturated with chloroform.

with acids. The instruments should be cleaned *immediately* after using by rinsing in water and drying. Above all, avoid dipping a glass rod which, for instance, may be contaminated with an acid or a dye into another fluid. Apart from the fact that thereby the reagents will be spoiled, the success of the preparation is, as a consequence, often totally frustrated. Beaker-glasses, watch-glasses, etc., are easy to clean if attended to immediately after using; but if, for example, any staining fluid is allowed to evaporate and dry on them the cleansing then becomes very tedious. Therefore the cleansing of the glasses *immediately* after using should never be neglected; in case there be no time for this, they at least should be placed in water.

All vessels used for isolating, fixing, hardening, staining, etc., must be kept closed and should not be placed in the sun.

§ 3. ISOLATION.

The process of isolation is accomplished by teasing either the fresh objects or those previously treated with dissociating fluids, which render the teasing partially or wholly unnecessary. It is a difficult task to make a well-teased preparation. Great patience and exact fulfilment of the following directions are indispensable: The needles must be sharp and perfectly clean; they should be previously pointed and polished on a moistened whetstone. The minute object, at the most 4 mm. square, should be placed in a small drop of the dissociating or mounting medium on a slide and teased on a dark background if it is colorless, on a white surface if it is dark or stained. If the object is fibrous—for example, a bundle of muscle-fibers—apply both needles at one end and separate the fasciculus along its length into two; in the same way divide one of these bundles into two, and so continue until the minute individual fibers are isolated. At times it is difficult to divide the bundle along its entire length; in this case it is often sufficient to divide it for three-fourths of its length, allowing the isolated fibers to remain attached at the one end. The uncovered preparation may be examined with the low power in order to ascertain if the dissection is fine enough.*

The following isolating fluids are recommended:

(a) FOR EPITHELIAL CELLS.

Ranvier's one-third alcohol (p. 20) is an admirable isolating

* Uncovered preparations lying in a small amount of fluid often appear indistinct, exhibit black borders, etc., errors which may be corrected by the addition of a sufficiently large drop of fluid and the application of a cover-glass.

medium. Place small pieces from 5 to 10 mm. square (*e. g.*, of the intestinal mucous membrane) in about 10 c.c. of this fluid. After four hours (in the case of stratified squamous epithelium after ten to twenty-four hours or later) take out the pieces with the forceps, *carefully* and *slowly*, and tap them *lightly* against a slide on which a drop of the same fluid has been placed. By this manipulation many isolated epithelial cells fall off; occasionally shreds are detached, which can be separated into their elements by gently stirring them with a needle. Then apply a cover-glass (p. 49) and examine. If it is desired to *stain* the object, carefully transfer the entire piece from the alcohol to about 6 c.c. of picrocarmine (p. 24). In two or four hours place the object very carefully in 5 c.c. of distilled water, and in five minutes tap it against the slide, which this time should have on it a drop of diluted glycerol (p. 23). Apply a cover-glass. The preparation can be preserved.

(b) FOR MUSCLE-FIBERS AND GLANDS.

A 35 per cent. solution of potassium hydroxid is suitable (p. 23). Small cubes from 10 to 20 mm. in diameter should be placed in 10 to 20 c.c. of this fluid. In about an hour the objects fall apart into their elements, which may then be lifted out with a needle or a pipet and examined under a cover-glass in a drop of the same lye. The action of diluted potash lye is totally different; examined in a drop of water the elements are rapidly destroyed. If the isolation is not successful, if instead a jelly-like softening occurs, the potash solution is too old. Therefore a freshly prepared solution should always be used. The preparations, even when successful, cannot be preserved.*

A mixture of potassium chlorate and nitric acid may be used. This is prepared by throwing into 20 c.c. of pure nitric acid so much potassium chlorate (about 5 gm.) that an undissolved residue remains on the bottom of the bottle. In from one to six hours, occasionally later, the object is sufficiently dissociated, and should then be transferred to distilled water, in which it should stay for one hour, but may remain for a

**Editor's remark:* According to S. H. Gage ("Proc. Amer. Soc. Micr.," 1889, p. 36), the action of the caustic potash may be at any time most satisfactorily checked by replacing it with a 60 per cent. solution of potassium acetate, or by the addition of sufficient glacial acetic acid to neutralize the caustic potash and form acetate of potash. After the action of the caustic potash is checked the elements may be preserved indefinitely *en masse* in a 60 per cent. solution of acetate of potash, or after being treated with a saturated solution of alum, in 40 per cent. alcohol or glycerol. After the last treatment the elements may even be satisfactorily stained with hematoxylin or alum-carmin.

week without injury. Then the object is placed on a slide, where, in a drop of diluted glycerol (p. 23), it can be easily dissected. If the nitric acid is well washed out the preparation can be preserved and can also be stained under the cover-glass (p. 53). Placing the unteased objects in picrocarmine (see *a*, p. 29) will not be successful, because this staining fluid makes them brittle.

(c) FOR GLAND-TUBULES.

Pure hydrochloric acid is admirable. Small pieces about 1 cm. in diameter should be placed in 10 c.c. of the acid and in from ten to twenty hours transferred to about 30 c.c. of distilled water, which must be renewed several times during twenty-four hours. The isolation is then easily accomplished by carefully spreading out the pieces with needles in a drop of diluted glycerol. The preparation can be preserved.

§ 4. FIXATION.

General Rules.—(1) For fixation a *large* quantity of the fluid should be used, exceeding the volume of the object 50 to 100 times. (2) The fluid must always be *clear*, and as soon as it becomes turbid must be replaced by fresh fluid. It often becomes turbid within an hour, or sooner, after the introduction of the object. (3) The objects to be fixed should be as *small* as possible; in general they should not exceed 1 or 2 c.c. Should it be necessary to preserve the object entire (*e. g.*, for subsequent orientation), many deep incisions should be made in it from five to ten hours after placing it in the fixation medium. The object should not lie on the bottom of the receptacle, but should be suspended within it or placed upon a thin layer of defatted cotton-wool or glass-wool.

1. *Ninety-five per cent. alcohol* is especially suitable for fixing glands, skin, blood-vessels, etc. It acts simultaneously as a hardening medium. Objects fixed in alcohol can be sectioned after twenty-four hours; * therefore it is well adapted for the rapid preparation of specimens. Special attention should be given to the following details: (1) The alcohol must be renewed in from three to four hours, even though it is not turbid. (2) The objects should not lie in contact with the glass, lest they adhere to it; † they should be either suspended on a thread in the alcohol or placed on a little wad of cotton on the bottom of the vessel.

* One should not too long delay using objects fixed in absolute alcohol, for the elements gradually deteriorate; they should be sectioned in from three to eight days. Sections of objects that have lain only twenty-four hours in absolute alcohol occasionally stain poorly.

† Such areas appear strongly compressed in the sections.

Weaker alcohol, for example, 90 per cent. alcohol, acts very differently, it shrivels the object and therefore cannot be used instead of 95 per cent. alcohol.

2. *Chromic acid* is mainly used in two aqueous solutions :

(a) As a 0.1 or a 0.5 per cent. solution (p. 21), which is especially suitable for organs that contain much loose connective tissue. This strong solution imparts a superior consistence to connective tissue, but has the disadvantage of making the staining difficult ; it is also suitable for the fixation of karyokinetic figures. The objects remain in the chromic-acid solution for from one to eight days, are then washed in running water for from three to four hours or, if this is not possible, placed for the same length of time in water renewed three or four times, then transferred to distilled water for a few minutes, and finally hardened in alcohol of gradually increased strength (§ 5) and protected from daylight (p. 35, remark *).

(b) As a 0.05 per cent. solution, which may be prepared by diluting the 0.1 per cent. solution with an equal volume of distilled water. The application is the same as that of solution *a*, except that the objects remain only twenty-four hours in solution *b*.

Chromic acid solutions penetrate slowly ; accordingly, if the tissue is submitted to the action of the medium for so brief a period as twenty-four hours, only small pieces, 5 to 10 mm. in diameter, should be preserved.

3. *Nitric acid* in a 3 per cent. solution (3 c.c. of concentrated nitric acid [p. 20] to 97 c.c. of distilled water), like the strong chromic acid solution, is an admirable medium for organs rich in connective tissue. The objects remain for from five to eight hours in this solution and without the previous use of water are transferred directly into alcohol of gradually increased strength for hardening (§ 5).

4. *Formaldehyde*, in from 8 to 10 per cent. solution (prepared by diluting 20 c.c. of commercial formol [p. 21] with 80 c.c. of distilled water) is a good medium for the fixation of cell structures ; it acts similarly to osmium solutions.* The objects should remain 48 hours or more in the formaldehyde solution and are then at once transferred to absolute alcohol, in which they are hardened for at least two days.

5. *Potassium-bichromate-acetic acid* (p. 21).—Place the objects in the liquid and after from 18 to 24 hours wash them for about three hours in (if possible running) water and then harden in gradually strengthened

* Cf. also the substitution of the osmic acid in the Golgi mixture.

alcohols (p. 35). The advantage of this reagent lies in its high power of penetration and in the rapid course of the process : fixation and hardening are completed in from 4 to 5 days. This method, that with the exception of the liver has yielded me very good results, requires more time for staining, *e. g.*, with Hansen's hematoxylin from 15 to 60 minutes, with safranin 24 hours instead of 5 minutes. Bulk staining of small pieces is easily accomplished.

6. *Potassium-bichromate-formol* (p. 21).—Place the objects in the liquid and after 24 hours transfer them to 3.5 per cent. solution of potassium bichromate ; in from 3 to 6 days wash in (if possible running) water for from 3 to 6 hours and harden in alcohols of ascending degrees of strength (p. 35).

7. *Müller's fluid*.—The objects remain for from one to six weeks* in a large volume (up to 400 c.c.) of this solution, are then washed in (if possible) running water, rinsed in distilled water, and, finally, hardened in the series of gradually ascending alcohols, under exclusion from daylight (p. 35, remark*). Who does not follow with painstaking conscientiousness the previously specified general rules for fixation will secure imperfect results, for which even otherwise experienced microscopists have held the blameless Müller's fluid responsible.

8. *Müller-formol mixture* (p. 21).—After 4 days' fixation the objects are transferred to pure Müller's fluid (p. 21) ; the subsequent treatment is the same as with Müller's fluid.

9. *Zenker's fluid*.—Metal instruments must be cleansed immediately after dipping them into this fluid. The objects should remain in it for from 10 to 24 hours, allowing about 60 c.c. of the reagent to each one-centimeter cube of tissue, should be washed in running water for the same length of time, rinsed in distilled water, and hardened in the dark in alcohols of gradually increasing strength (p. 35). For the removal of the sublimate precipitates that occur in the tissues add to the 90 per cent. alcohol enough tincture of iodine to impart to the fluid the color of port-wine. The objects remain for from eight to fourteen days in this iodine-alcohol, the color of which rapidly fades and therefore it requires the daily addition of enough of the tincture of iodine to maintain the tint.† Finally the objects are transferred to pure 90 per cent. alcohol, which is to be changed two or three times, and in this they may remain for a week or longer. (See also pp. 51 and 52.)

* Objects may be left in Müller's fluid for a longer period—up to six months ; often they can then be sectioned and stained without the alcohol hardening.

† If notwithstanding the preparations show sublimate precipitates they may be removed by placing the sections in iodine-alcohol for about ten minutes. Then rinse them in pure alcohol, transfer them to the staining fluid, etc.

The results with Zenker's fluid are good only when sectioning and staining are undertaken soon after completion of fixation and hardening. One year old Zenker preparations stain less well, even such as were embedded in paraffin. Then often only hemalum (p. 39) still gives satisfactory staining. For organs that are rich in smooth muscle-fibers Zenker's fluid is less suitable than other fixing media.

10. *Osmic acid solution* (p. 22).—In using this reagent care must be taken not to inhale the vapor, which is very irritating to mucous membranes. Fixation is accomplished either by immersing very small pieces, up to 5 mm. cubes, in the acid, which is usually employed in a one per cent. solution, of which only a small quantity—from 1 to 6 c.c.—need be used; or by exposing the moist object to the vapor of the osmic acid solution. For the latter purpose pour 1 c.c. of the 2 per cent. solution into a test-tube about 5 cm. in length and add an equal volume of distilled water; fasten the object by means of quills to the under surface of a cork stopper, with which the test-tube is then to be securely closed. In from ten to sixty minutes, according to the size of the object, it is removed from the cork and dropped into the fluid in the test-tube. In both cases the objects remain in the acid for twenty-four hours, and during this time the containers must be tightly closed and stood in the dark. Then the objects are taken out, washed for from one-half to two hours in running water, rinsed in distilled water, and hardened in gradually strengthened alcohols (§ 5).

11. *Chromic-acetic osmic acid* (Flemming's solution) (p. 22) is an excellent medium for the fixation of karyokinetic figures. Place the absolutely fresh, *still warm* pieces, from 3 to 5 mm. in diameter, in 4 c.c. of this fluid, in which they remain for from one to two days, or even longer. Then the pieces should be washed in running water for one hour, better longer, rinsed in distilled water and hardened in alcohols of gradually ascending strength (§ 5). The effect of this mixture on the nuclei is different at the periphery of the object than in the interior, where the chromatin networks are more distinct, because at the periphery the osmic acid, which renders the nuclear sap granular and the nuclear reticulum indistinct, acts in its purity.

12. *Platinum-acetic-osmic acid mixture* (p. 22) is very suitable for displaying sharply defined cell boundaries. It is used like Flemming's solution.*

*Solutions of osmic acid or mixtures containing osmic acid blacken fat; if it is desired to preserve osmicated fat the sections must not be cleared in turpentine, absolute ether, or xylol, which dissolve osmicated fat. Use chloroform (or clove oil) and mount in a solution of balsam in chloroform. Pure osmium solutions (but not osmium-containing mixtures) also blacken pigment.

13. *Sublimate salt solution*.—Place small cubes of tissue, at the most not over 4 mm. in diameter, for from one to six hours, according to bulk, in 20 c.c. of sublimate salt solution (p. 22); then transfer directly into 30 c.c. of gradually strengthened alcohols (§ 5, p. 35) for hardening. To the 70 per cent. alcohol and upward add tincture of iodine, as when using Zenker's medium (No. 9, p. 33). Avoid the use of metal instruments.

The fluids that have been used for fixation cannot be used again and should be thrown away.

§ 5. HARDENING.

Except when absolute alcohol is used, all the fixing methods necessitate a supplementary process of hardening. The best hardening medium is *alcohol in ascending degrees of strength*. Here, too, the rule is to use abundance of fluid and to change the alcohol when it becomes turbid or colored.* A stratum of defatted cottonwool, from 2 to 4 cm. deep, should cover the bottom of the receptacles used for hardening, in order to keep the water that settles there from the immediate vicinity of the object.

The exact application is as follows: After the objects have been fixed in one of the previously enumerated fluids and washed in water† they are placed, under exclusion of daylight, for from two to six hours, according to the size of the object, in 50 per cent. alcohol, then transferred for twelve hours each to 70 per cent. and 80 per cent. alcohol, and at the expiration of this time to 90 per cent. alcohol, in which after another period of from twenty-four to forty-eight hours the hardening is completed. In this alcohol the objects may remain for months before their final preparation. The 90 per cent. alcohol employed for hardening should be collected and used for burning or for hardening liver for embedding.

* Objects fixed in chromic acid or in Müller's fluid, if not subjected to prolonged washing,—and this must be avoided because of incipient decomposition,—yield substances to the alcohol which with the simultaneous action of daylight appear in the form of precipitates; on the other hand, if the object is kept in the dark no precipitates are formed and though the alcohol becomes yellow it remains clear. It is on this account that the exclusion of daylight has been recommended above; it is sufficient to place the bottles in a dark part of the room. Even the 90 per cent. alcohol must be changed once daily so long as it becomes intensely yellow.

† An exception is made in the case of objects that have been fixed in 3 per cent. nitric acid. These should be transferred directly from the fixing fluid to the 70 per cent. alcohol, which must be changed several times during the first day.

§ 6. DECALCIFICATION.

The objects to be decalcified must not be placed fresh in the decalcifying fluid; they must be previously fixed and hardened. For this purpose place small bones up to the size of a metacarp, teeth entire, and pieces from 3 to 6 cm. long sawed from the larger bones in 300 c.c. of Müller's fluid for from two to four weeks and, after previous washing, harden them in 150 c.c. of gradually strengthened alcohols (§ 5). After the bone has been in the 90 per cent. alcohol for three days or longer it is washed for twenty-four hours in running water and then transferred to the decalcifying fluid—diluted nitric acid, prepared by adding from 9 to 27 c.c. of pure nitric acid to 300 c.c. of distilled water. Large quantities, at least 300 c.c., of this fluid should be used and changed *daily* at first, later every four days, until the decalcification is completed. The process is controlled by thrusting in a needle or by making an incision with a scalpel, which should be at once carefully cleaned. Decalcified bone is flexible, soft, and easily cut. Fetal bones, heads of embryos, etc., are decalcified in weaker nitric acid (1 c.c. of pure nitric acid to 90 c.c. of distilled water) or in 500 c.c. of a saturated aqueous solution of picric acid (p. 22). The process of decalcification requires several weeks for thick bones, from three to twelve days for fetal and small bones.

So soon as the decalcification is completed the bones are placed for twenty-four hours in 5 per cent. solution of potash alum, then washed in running water for twenty-four hours, and again hardened in gradually strengthened alcohols (§ 5).

It not infrequently happens to beginners that they transfer the bone to alcohol before it is fully decalcified, and then in the attempt to section it they discover that it is not yet ready for use. In such cases the entire procedure of decalcification must be repeated. If the action of the decalcification medium is too prolonged, it eventually leads to the complete destruction of the objects.

§ 7. SECTIONING.

The razor must be *sharp*, for success in sectioning depends upon the sharpness of the knife. The blade must be moistened with alcohol; water is not suitable, because it does not adhere evenly to the surface of the blade. Therefore, at each third or fourth section dip the knife into a shallow glass dish containing 30 c.c. of 90 per cent. alcohol, which at the same time serves for the reception of the sections that are cut. The razor is to be held in a *horizontal* position, lightly grasped,

with the thumb on the side of the cutting edge, the fingers toward the back of the blade, the dorsum of the hand directed upward. The object to be sectioned must first have a smooth surface, which is made by cutting off a slice of the necessary thickness with a single movement of the razor. From this surface the sections may now be taken; they should be cut with a light, not too rapid movement, as smooth as possible, and of uniform thinness. The knife must not be pushed, but should be *drawn* through the object, and that this may be done the portion of the blade adjoining the handle should be applied to the object. Ten to twenty sections should be made; they may be transferred by means of a needle or by immersing the blade in the alcohol.* Then place the dish on a black surface and search for the best sections. The thinnest sections are not always the most useful; for many preparations—for example, for a preparation through all the coats of the stomach—thick sections are recommended. For a general view large, thick sections should be prepared; for the study of minute structures, thin sections; for the latter purpose small fragments from 1 to 2 mm. on a side are often satisfactory, or the marginal portions of thick sections.

If the object to be sectioned is too small to be held with the fingers, it should be embedded. The simplest method consists in placing the object in a cleft in a piece of hardened liver.

Ox-liver or, better, human lardaceous or amyloid liver may be used. The latter may be obtained from the pathologic laboratories. Dog's liver, to be obtained from the physiologic laboratory, is also recommended. The liver should be cut into pieces about 3 cm. high, 2 cm. broad, and 2 cm. thick, and these hardened in 90 per cent. alcohol, which must be changed within twenty-four hours; in three to five days the liver attains the necessary hardness. The embedding is then accomplished by making an incision in one of these pieces from the top half-way down and inserting the object into this cleft. If the object is too thick, furrows can be cut in the liver with a small scalpel and the object fitted into these. The object requires no further staying except, perhaps, binding with a silk thread.

As a rule I embed objects in liver; very thin sections can be made so soon as one has a certain amount of skill and this can be easily acquired in the course of a few weeks.

§ 8. STAINING.

Before using a stain it should always be filtered. A small funnel

* Very thin sections that are not to be stained or that have been stained in bulk may be transferred directly to the slide by inclining the blade and slipping or rinsing them off.

can be made by simply twice folding a piece of filter-paper 5 cm. in diameter and supporting it in a cork frame, which can be made by cutting out a piece 2 cm. square from a cork plate 5 cm. square. The frame is then mounted on four long pins. Such a funnel and frame can be used repeatedly, but only for the same fluid. The sections should not float on the surface of the staining fluid; they must be *submerged* with needles.

1. *Nuclear staining with Hansen's hematoxylin* (p. 23).—Filter from 3 to 4 c.c. of the staining fluid into a watch-glass and in it place the sections. The time in which the sections stain varies greatly. Sections fixed and hardened in alcohol stain in from one to three minutes. If Müller's fluid or potassium-bichromate-acetic acid was used for fixing, the sections must remain in the staining fluid somewhat longer, up to five minutes and more.*

From the stain the sections are transferred to a watch-glass containing distilled water, in which they are washed,—*i. e.*, gently moved about with the needle to remove the excess of dye,—and then placed in a glass containing 30 c.c. of distilled water. In this the sections must remain at least five minutes, during which their blue-red color gradually changes to a beautiful deep blue, which becomes the purer the longer (up to twenty-four hours) the sections are allowed to remain in the water. When a preparation is overstained the hematoxylin can be partially extracted by placing the sections in a watch-glass containing 5 c.c. of distilled water to which 2 or 3 drops of acetic acid have been added. In about 5 minutes the sections become lighter and are then transferred to distilled water, which must be changed several times, by means of which the color, made red by the acetic acid, becomes blue again. At first the sections have a faded blue tint; usually the differentiation occurs in about five minutes, but sometimes not for hours. When it is complete certain details can be recognized even by the unaided eye.

Beginners are recommended to leave the sections for different lengths of time—one, three, or five minutes—in the stain, in order to learn the time required to produce successful staining. The chief essential in

* Sections fixed in the strong solution of chromic acid or in Zenker's fluid, or objects not entirely free from acid, often stain very slowly, occasionally not at all. This defect can be remedied either by keeping the objects from two to three months in 90 per cent. alcohol, which must be changed two or three times during this period, or by treating the sections for from five to ten minutes with 5 c.c. of distilled water to which from 3 to 7 drops of 35 per cent. solution of potassium hydroxid have been added. The sections are then to be transferred for from one to two minutes to a watch-glass containing pure distilled water and from this into the hematoxylin. In from five to ten minutes such sections will also stain.

hematoxylin staining is *thorough washing*; if the water becomes blue, it must be replaced by fresh. The used stain should be poured back through the filter into the hematoxylin bottle. The watch-glass should be immediately cleaned.

Instead of Hansen's hematoxylin P. Mayer's *hemalum* (hæmalum pur., Grübler) may be used. It is prepared by dissolving with the aid of heat 0.5 gm. of hemalum in 25 c.c. of 90 per cent. alcohol and mixing this with a solution of 25 gm. of alum in 500 c.c. of distilled water. The application is the same as for Hansen's hematoxylin. It can also be used for bulk staining, allowing 24 hours for penetration. Large objects stained in bulk must be washed out with a 1 per cent. solution of alum.

2. *Nuclear staining with alum-carmin* (p. 25) *or with carmalum* (p. 25).—Filter from 3 to 4 c.c. of the staining fluid into a watch-glass, place the sections in it, and allow them to stain for at least five minutes. The advantage of alum-carmin lies in this, that the sections may be left in it for a longer period without becoming overstained, which is more apt to occur with hematoxylin; a disadvantage is that alum-carmin is a pure nuclear stain, while in hematoxylin staining the protoplasm too acquires color, a gray or gray-violet tone, and is thereby more easily recognized.

3. *Diffuse staining*.—For staining the protoplasm and the inter-cellular substance.

(a) *Slow staining*.—A small drop of neutral carmin solution (p. 24) is transferred by means of a glass rod to a capsule containing 20 c.c. of distilled water, on the bottom of which lies a small piece of filter-paper.* The sections remain over night in this fluid. The paler the rose color of the fluid the longer the time required for staining and the more beautiful the result will be. The beginner is always inclined to regard the pale-rose fluid as too dilute to secure good staining, until on the following day the deep pink to red sections teach him better.

This stain can be used alone only in a few cases, but is highly recommended for double-staining. The sections should be stained first with the carmin solution, then with hematoxylin.

Staining with *orange* for from 12 to 24 hours (10 c.c. of 95 per cent. alcohol to which from 2 to 4 drops of solution of orange (p. 25) have been added) yields effective pictures. Stain first with hematoxylin, then with orange or with *eosin* (2 to 4 drops of eosin (p. 25) in 10 c.c. of distilled water).

(b) *Rapid staining*.—Add 10 drops of a solution of eosin (p. 25) to 3 or 4 c.c. of distilled water. In this the sections remain for from one

* If the filter-paper is omitted the sections stain only on the one side.

to five minutes, are then washed in distilled water, and then placed in 30 c.c. of fresh distilled water (see No. 1, p. 38). The stain may be used alone or combined with hematoxylin; in the latter case the whole procedure of hematoxylin staining is to be carried out first, then that of eosin staining.

4. *Staining of the chromatin substance.*—For nuclear division. Place the objects for from five to ten minutes in a watch-glass containing 10 c.c. of distilled water and one drop of pure hydrochloric acid; wash them for one minute in distilled water and transfer them to a watch-glassful of safranin solution (p. 25), in which they should remain five minutes. The sections or membranes are then lifted out with the needle and placed in about 5 c.c. of absolute alcohol for decolorization. When the sections no longer give off much of the dye (usually in from one to two minutes), they are transferred to 5 c.c. of fresh absolute alcohol for one minute, then cleared and mounted (§ 10, 3, p. 50). If the immersion in absolute alcohol is too prolonged, it may lead to total decolorization of the preparation. Failure in staining is usually due to an insufficient amount of acetic acid in the Flemming's solution (p. 22, remark).

5. *Staining in bulk.**—Nuclear staining of the entire object before sectioning:

(a) *Borax-carmin.*—The fixed and hardened objects are placed in 30 c.c. of borax-carmin for twenty-four hours if they are small (5 mm. square), for from two to three days if they are large. From this they are transferred directly to 25 c.c. of acid-alcohol (p. 25); the used borax-carmin may be returned to the bottle. In a few minutes the acid-alcohol acquires a red color † and must be replaced by fresh, which should be again renewed in about fifteen minutes; this renewal must be repeated until the alcohol no longer becomes red. ‡ The object is then transferred to 90 per cent. alcohol, and if after twenty-four hours it is not sufficiently hardened to be sectioned, it is placed for twenty-four hours or longer in 95 per cent. alcohol.

* *Editor's remark:* It is especially for staining in bulk that *alum-cochineal* (recommended on p. 25, remark) proves very useful. It has the advantage of not overstaining, and does not need in its application a special discharging fluid. Stain the pieces for about twenty-four hours and wash them in several changes of water to remove the excess of stain and the alum; then transfer to alcohols of gradually increased strength.

† Preparations fixed in Müller's fluid often give off very little dye.

‡ This may require from one to three days; during the first day the fluid should be changed every two hours, subsequently every four hours. If you wish to be economical take a needle and gently push the object out of the area of red fluid in which it lies into an uncolored portion of the alcohol.

(b) *Paracarmine*.—This stain (p. 25) penetrates easily and in this respect is preferable to borax-carmine; pieces from 2 to 3 cm. on a side can be stained in bulk in 24 hours. The pieces are transferred from the paracarmine to 70 per cent. (not acid) alcohol, that when colored is to be changed for fresh; follow by 90 and 95 per cent. alcohol. This reagent stains not only nuclei, but also in light tone the protoplasm. Overstaining can be corrected by placing the pieces (or sections) in 40 c.c. of 70 per cent. alcohol plus 1 c.c. of glacial acetic acid. Then treat for 12 hours with 90 per cent. alcohol and from this transfer to 95 per cent. alcohol.

6. *Picrocarmine*.—Double-staining: nuclei and connective tissue red, protoplasm yellow.

Filter about 5 c.c. of the staining fluid (p. 24) into a watch-glass. The length of time in which picrocarmine acts differs greatly for individual objects and can be approximately given only in the special directions. When the staining is completed the dye is filtered back into the bottle and the object transferred for from ten to thirty minutes to 10 c.c. of distilled water. (The latter procedure is omitted in staining under the cover-glass, p. 53.) If the object, *e. g.*, a section, is to be dehydrated in absolute alcohol (p. 50), it must not be allowed to remain in this reagent longer than from one to two minutes, because the alcohol extracts the yellow stain; or the decolorization can be prevented by adding a small crystal of picric acid to the absolute alcohol.

Picrocarmine is preferably used in the examination of fresh objects. If the solution is good a very pretty stain is obtained, that is improved by subsequent treatment with acidulated glycerol, which renders it crisp and clear.

7. *Nuclear staining with anilin dyes*.—For this purpose the best anilin dyes are *vesuvin* (p. 25) and *methvl-violet B* (p. 26). Filter 5 c.c. of the staining fluid into a watch-glass; in this place the sections, which acquire a very dark color in from two to five minutes; they are then washed in distilled water and transferred to a watch-glass containing absolute alcohol, in which they give off the dye abundantly. In a few minutes, from three to five, the sections become paler, and individual parts (*e. g.*, the glands of the skin) can be detected by the unaided eye. The sections are now to be transferred to another watch-glass containing 5 c.c. of absolute alcohol, and in about two minutes they may be cleared and mounted in balsam. The result is a very beautiful permanent nuclear stain. A disadvantage lies in the necessity for using so much absolute alcohol.

8. *Safranin* (p. 25) can be similarly employed. The sections stained

for five minutes are washed for thirty seconds in a watch-glass containing 95 per cent. alcohol and then transferred to absolute alcohol, which must be replaced by fresh so soon as it becomes intensely red. In from five to fifteen minutes—the time varies according to the thickness of the sections—they are sufficiently decolorized and are then to be cleared and mounted in xylol-balsam (p. 50).

9. *Methylene-blue for staining axis-cylinders*.—This method is applicable only to perfectly fresh, "overliving" preparations. Prepare a one-fifteenth per cent. solution, by adding 1 c.c. of a 1 per cent. solution (p. 26) to 15 c.c. of distilled water. The fresh preparation is treated on the slide with a few drops of this diluted staining fluid and meanwhile covered with a watch-glass, to prevent evaporation, but not so as to make an hermetic cover, since the access of atmospheric air is necessary to the success of the staining. The reaction occurs in from one to one and a half hours; it can be rendered more certain by gently moving the preparation to and fro and by placing it in an oven at 36.5° to 37.7° C. In order to prevent the drying of the preparation during this period a drop of the diluted staining fluid or of normal salt solution should be added from time to time. Then cover with a cover-glass. The result is a beautiful blue coloration of the axis-cylinders. Other elements often are stained, the nuclei, connective-tissue fibers, etc., and with more prolonged action of the reagent also the medullary sheaths of the nerves. The preparation may be preserved as follows: replace the staining fluid with a drop of ammonium picrate solution (p. 26) according to the method given on page 53; this converts the blue color to violet; then place a drop of glycerol at the edge of the cover-glass, and it will gradually take the place of the evaporating water of the ammonia solution. After eighteen to twenty hours add another drop of glycerol and secure the cover-glass with cement (p. 49). In the course of 24 hours the preparations become thoroughly transparent and not until then do they admit of close investigation. They must not be exposed to sunlight, in which they fade; in any case they soon lose their original beauty (see further Leontowitsch, on "The Innervation of the Human Skin." Internat. Monatsschr., Bd. 18, 1901).

10. *Mucus-staining with Delafield's hematoxylin*.—Filter three drops of this stain (p. 24) into a watch-glass containing 25 c.c. of distilled water. In this dilute solution the sections (preferably of objects fixed in Flemming's mixture*) are placed and remain for two or three

* Preparations that have been fixed in Müller's and in Zenker's fluid are also suitable for mucus-staining.

hours. Usually at the end of this period the mucus (*e. g.*, in the goblet-cells) is stained an intense blue, which can be ascertained by examining with low magnification the sections as they lie in the solution. It is often necessary for the sections to remain in the solution for a longer time. Then they are washed for one minute and mounted in balsam, according to the rules given in § 10, 3, p. 50. The nuclei also stain blue. Very pretty pictures are obtained by a combination with safranin and picric acid, as in No. 11.

11. *Triple-staining* is accomplished in the following manner: The sections stained in Delafield's hematoxylin are placed for five minutes in safranin (p. 25) and then transferred to 5 c.c. of absolute alcohol, which must be changed twice within fifteen minutes. The sections are next placed for one minute in 5 c.c. of absolute alcohol to which five drops of a saturated alcoholic solution of picric acid have been added (1 gm. of picric acid to 15 c.c. of absolute alcohol), washed for thirty seconds in pure absolute alcohol, and mounted in balsam (p. 50).

Result: mucus blue; nuclei red; protoplasm and fibers yellow.

12. *Van Gieson's staining*.—Treat sections with Hansen's hematoxylin (p. 38) for 30 minutes. Place the overstained sections in:

- (a) 5 c.c. picrofuchsin (p. 26), 1–3 minutes,
- (b) 5 c.c. distilled water, 10–30 seconds,
- (c) 5 c.c. 90 per cent. alcohol, 1 minute,
- (d) 5 c.c. absolute alcohol, 2 minutes,
- (e) 5 c.c. xylol, and when thoroughly cleared,
- (f) xylol-balsam.

Result: connective tissue shining red, elastic tissue and muscle-fibers yellow, epithelium and nuclei brown.

This method should be applied to thin sections only and succeeds best after alcohol, sublimate, or nitric acid fixation, less well after fixation with solutions of chromic acid or its salts. The duration of the stain is brief. This latter disadvantage can be overcome by acidulation (placing the sections previously to (a) and subsequently to (b) for 1 minute in 5 c.c. of acid alcohol, see No. 5, p. 40).

13. *Staining of elastic fibers*.—Sections that have been fixed in any medium (preferably in alcohol) are placed in 5 c.c. of resorcin-fuchsin (p. 26) for from 8 to 24 hours, then transferred to absolute alcohol, that after one minute is to be renewed. In from 2 to 5 minutes the sections are cleared in xylol (not carbol-xylol) and mounted in balsam. These preparations, in which success is very easily attained, exhibit even the finest elastic fibers dark blue on a light ground. Sections can be fore-stained for 20 minutes in borax-carmin (p. 40), dried with filter-paper (p.

50, remark §), and placed directly in resorcin-fuchsin. The free acid of the latter provides for the differentiation. Eventually orange (p. 39, 3) may be applied as a ground stain.

14. *Staining of connective-tissue fibrils.*—By means of glass rods place thin sections of objects fixed in any medium (preferably in alcohol) in 5 c.c. of 10 per cent. phosphomolybdic acid, and after from one to ten minutes wash for a couple of seconds in distilled water; stain for from five to twenty minutes in 5 c.c. of Mallory's hematoxylin (p. 24), rinse well in distilled water and place in 10 c.c. of 50 per cent. alcohol; after another five minutes dehydrate in absolute alcohol, clear in xylol, and mount in xylol-balsam (see § 10, 3, p. 50). The connective tissue stains intensely blue. If it is desired to stain nuclei, the sections must be forestained with safranin (p. 40, 4), or with borax-carmin (p. 40, 5). Everywhere, in glands, mucous membranes, the skin, etc., I have obtained very instructive pictures.

15. *M. Heidenhain's iron-hematoxylin.*—For staining centrosomes, secretory capillaries, cement bars, and gland granules. Fix the object preferably in sublimate (p. 35), in Zenker's medium (p. 33), or in Flemming's mixture (p. 34), for granules in potassium-bichromate-formol (p. 33); embed in paraffin, cut on the microtome, and fasten the sections (which should be very thin) to the slide (see Microtome Technique). Transfer the slide with the sections from the absolute alcohol to a capsule containing 50 c.c. of the iron solution (p. 22); after from six to twelve hours remove from the mordant, rinse for a couple of seconds in distilled water, and place for from twelve to thirty-six hours in a mixture of 30 c.c. of Weigert's hematoxylin (p. 24) and 30 c.c. of distilled water.* The sections, which have become black and wholly untransparent, are now rinsed in tap-water and then returned into the iron solution for bleaching and differentiation. When this is accomplished wash them for about fifteen minutes (not more) in running water,—common water is indispensable,—stain with picrofuchsin (cf. No. 12, p. 43), and after the customary preliminary treatment mount in xylol balsam (p. 50). When the decoloration is slowly and carefully done this admirable method easily succeeds, but the exact duration of this process cannot be given; the slide must be frequently removed from the iron solution, washed with tap-water, and examined with a high-power objective, to ascertain if the differentiation is completed.

* This diluted hematoxylin can be repeatedly used and should be saved. Old Weigert's hematoxylin is preferable to the freshly prepared stain.

16. *Silver staining*.—For the exhibition of cell boundaries and the staining of cement-substance.*

The use of metallic instruments must be avoided ; glass rods should be employed and quills instead of pins.

The object is immersed for from one-half to ten minutes, according to its thickness, in from 10 to 20 c.c. of a 1 per cent. or weaker (see Special Technic) solution of silver nitrate (p. 22), which meanwhile becomes milky and turbid ; it is then removed with glass rods, washed, placed in a porcelain capsule containing 100 c.c. of distilled water, and exposed to direct sunlight. In a few minutes a faint brown coloration appears, the sign of a successful reduction. So soon as the object has become a deep red-brown (usually in from five to ten minutes) it is taken out, placed in a watch-glass containing distilled water to which a few grains of common salt have been added, and at the end of five or ten minutes transferred to 30 c.c. of 70 per cent. alcohol and stood in the *dark* ; in from three to ten hours the 70 per cent. should be replaced by 90 per cent. alcohol. The immersion in the silver solution must be done under exclusion of sunlight ; the reduction, on the other hand, must be undertaken only in sunlight.† If the sun does not shine the object, after treatment with the silver solution and washing in distilled water, is to be preserved in the dark in 30 c.c. of 70 per cent. (later 90 per cent.) alcohol, and in this exposed to sunlight at the earliest opportunity.

17. *Golgi's "black" reaction*.—For demonstration of the elements of the nervous system and the secretory passages.‡

This method unites fixing and staining. The objects must be as fresh as possible and in general their diameter should not exceed 4 mm. It is not easy to cut fresh brain or other organs into pieces of this size without bruising the delicate tissue ; therefore place larger pieces (up to 2

* The cross-striations that appear in different tissues and organs when treated with silver nitrate, particularly in nerve-fibers, blood-vessels, cartilages, etc., are artifacts ; they appear where colloid structures coagulate under the action of silver nitrate, especially when under the simultaneous influence of an acid.

† The reduction takes place in ordinary daylight, but slowly, and yields less satisfactory results.

‡ *Editor's remark* : In American laboratories a modification of Golgi's method by Cox is often used with excellent results. This modification is particularly recommended to beginners, because it is very simple and nearly always successful. In its application the following directions should be observed : Put small cubes, 2 cm. or less, of the organs of the central nervous system of adult or newborn animals of from six to ten weeks in the Cox-Golgi mixture, the formula of which is given on page 22 (No. 17), using 10 to 20 times the volume of the object treated. Change the fluid at the following intervals : after twenty-four hours ; three days ; eight days ; fifteen days ; twenty-one days ; thirty days. The objects should remain in the mixture

cm. cubes) in a small glass jar containing freshly prepared Golgi's mixture (p. 21), which is to be covered and stood in the dark (in winter it must be put in an oven having a temperature of about 25° C.). In from one to two hours the pieces can easily be cut into slices about 4 mm. in diameter. The quantity of Golgi's fluid to be used is regulated by the number of the slices, each slice requiring about 10 c.c. of the mixture. In from two to six days, less often fifteen days,* the slices are taken out, quickly washed for a couple of seconds in distilled water, gently dried with filter-paper, and placed in 0.75 per cent. silver solution (30 c.c. of the 1 per cent. solution [p. 22] plus 10 c.c. of distilled water, and for each piece 10 c.c. of this fluid).† A brown precipitate immediately envelops the pieces. They should be left in the silver solution for two days (which need not stand in the dark and *must not* be placed in the oven), and they may remain in it for six days without injury; they are then placed for from fifteen to twenty minutes (not longer) in 20 c.c. of absolute alcohol, then embedded in elder-pith (or in celloidin, see Microtome Technic) and cut into thick sections.

Each section should be at once examined, *without* a cover-glass, with the low power, in order to ascertain its usefulness; if it is good it is placed for from one to two minutes in a watch-glass containing absolute alcohol, then for a few minutes in carbol-xylol, then transferred to the slide. The xylol is removed by light pressure on the section with clean filter-paper and the preparation covered with a few drops of xylol-balsam. A cover-glass *must not* be applied, because it would prevent evaporation of the moisture in the section, which when retained destroys the Golgi preparations. Not infrequently, especially when the carbol-xylol has not been satisfactorily removed, the xylol-balsam gradually withdraws from the preparation, which in consequence appears spoiled, but may be fully restored by the application of a fresh drop of balsam. At first the preparation should be examined with the low-power objective; when the balsam has become dry the high power may be used.

until they are to be sectioned, and will keep in good condition for about ten months. Then transfer them directly into 95 per cent. alcohol for one hour; into alcohol-ether (equal parts) for a half hour; into thin celloidin solution (in alcohol-ether) for one hour. Mount on a block with thick celloidin solution (see Microtome Technic) and harden in 80 per cent. alcohol for from one to two hours. Cut at once sections from 50 to 100 μ thick; clear them in a mixture of xylol, three parts, and carbolic acid, one part, in which they may remain for weeks without injury. Mount in balsam and *cover the sections with a cover-glass*. In time the specimens thus preserved are not infrequently marred by the appearance of corrosive crystals, but the impregnation of the elements of the nervous tissue remains intact.

* See Special Technic.

† The used Golgi mixture is to be thrown away.

The results obtained by this method, when successful, are altogether admirable; single elements of the nervous system (never all), occasionally also blood-vessels, lymph-vessels, connective-tissue fibers, secretions, muscle-fibers, and epithelial cells stand out in full relief, black on a light background. But the method is subject to various accidents. Almost invariably the best sections are disfigured by black precipitates; these occur chiefly at the edges of the preparation; in order to avoid them it has been suggested that a layer of coagulated blood be applied to the fresh object. Very often the reaction fails entirely, especially when the action of the Golgi mixture was too prolonged; then the so-called "double method" may lead to success. If the first sections show nothing, the objects should be again treated with Golgi's fluid for from twenty-four to thirty-six hours and for the same length of time with the silver solution. A second failure may be occasionally crowned with success by a second repetition of the procedure. In the application of Golgi's method practice and patience are important factors.

Instead of the costly Golgi mixture (p. 21) potassium-bichromate-formol (p. 21) can be used. Put pieces of tissue of ca. 2 cm. diameter in 50 c.c. of Kopsch's fluid (do not place in the oven) and after 24 hours transfer to 3.5 per cent. bichromate solution (10 *b*, p. 21) and let them remain in this for from 3 to 6 days. Treatment with the silver solution is the same as after fixation with the osmium-bichromate mixture. Even with material 48 hours old the impregnation still succeeds.

Impregnated preparations that have been treated either with the osmium or the formol-bichromate mixture can be further fixed and stained. For this purpose transfer the sections from the alcohol to a mixture of 100 c.c. of 0.75 per cent. salt solution (p. 20) and 200 c.c. of 95 per cent. alcohol (these large quantities are indispensable), and stir them about frequently with a glass rod, for a period of from 10 to 15 minutes; next place them in a glass capsule containing about 20 c.c. of 80 per cent. alcohol and let them stand on a white background, in the light (not in sunlight), for a half day. By this means the black precipitates, that in the alcohol-salt mixture very rapidly faded to a pale yellow, become dark again. Then stain with carmalum (p. 39) or with Delafield's hematoxylin (p. 42). In staining the parietal cells (cf. Technic No. 108) use also eosin (3 *b*, p. 39). Preparations so fixed and stained can be preserved in xylol-balsam and covered with a cover-glass.

18. *Gold staining*.—For the demonstration of nerve terminations. Steel instruments must not be used; all manipulations in the gold solution are to be performed with rods of glass or wood. Put 8 c.c. of a 1

per cent. gold-chlorid solution and 2 c.c. of formic acid into a test-tube and heat the mixture to the boiling-point; let it boil up three times. Into the *cooled* mixture very small cubes of tissue (at most 5 mm. square) are placed for one hour, during which they must be kept in the dark; then they are washed in distilled water and exposed to the light in a mixture of formic acid, 10 c.c., and distilled water, 40 c.c. Sunlight is not necessary. The reduction takes place slowly, often not until after twenty-four or forty-eight hours, the exterior of the cubes meanwhile assuming a dark violet hue. When the reduction is completed place the tissue in 30 c.c. of 70 per cent. alcohol, and on the following day in an equal quantity of 90 per cent. alcohol, in which, to hinder further reduction, they must remain in the dark for at least eight days before their final preparation.

§ 9. INJECTING.

The filling of the blood- and lymph-vessels with colored masses is a special art that can only be acquired through much practice. The knowledge of the many little devices employed can scarcely be attained through didactic teaching, however painstaking and explicit. Here practical instruction is indispensable. Accordingly, since this book is intended for beginners, it seems wise to refrain from entering upon a detailed account of the technic of injecting.

He who desires to attempt injecting must have an accurately closing, smoothly working hand-syringe, provided with cannulæ of different sizes. For an injecting mass I advise Berlin blue (Grübler), 3 gm. dissolved in 600 c.c. of distilled water. It is best to begin with the injection of single organs, for example, the liver, which is preferable because it gives useful results, even though the blood-vessels are but partially filled. The injected object should be fixed for from two to four weeks in Müller's fluid (p. 33) and hardened in gradually strengthened alcohols (p. 35). The sections must not be too thin. For injecting the lymph-vessels Chinese tusche is recommended (see Lendorf, *Anatom. Hefte*, Bd. 17, p. 370).

§ 10. MOUNTING AND PRESERVING OF THE PREPARATIONS.

The finished sections and other objects prepared according to the foregoing methods, in order that they may be examined under the microscope, are finally mounted on a slide and covered with a cover-glass. The media in which the sections are mounted are: (1) *water*; or, if the section is to be cleared and preserved, (2) *glycerol*; or (3) *xylol-balsam*.

The *transfer* of the object to the *slide* is usually done in this way : a small drop of a suitable fluid is placed on the middle of the slide ; the section is then taken up on the section-lifter and with the aid of the needle slipped off onto the slide. Very thin sections are better lifted on the end of a glass rod and by rolling of the latter transferred to the slide. When the section is smoothly mounted, it is covered with a cover-glass.* The latter must be grasped by its *edges*, not by its *surfaces*. It should be taken in the left hand, one edge placed in contact with the slide, and then, supported on its under surface by a needle held in the right hand, slowly lowered upon the preparation. It is simpler to suspend a drop of the mounting medium from the under surface of the cover-glass and then to let it softly fall upon the preparation. The fluid in which the section is mounted must occupy the *entire* space between cover-glass and slide. If the amount of fluid is insufficient, which is recognized by the large air-bubbles under the cover-glass, another drop should be placed *at one edge* of the cover-glass by means of a pointed glass rod. If there is too much fluid—here the beginner strives to perpetrate impossibilities—the excess which has escaped from beneath the edges of the cover-glass should be absorbed with filter-paper. *The upper surface of the cover-glass must always be dry.* Small air-bubbles under the cover-glass may be removed by cautiously raising and lowering the cover several times with the needle (see further, p. 51).

1. The examination of the unstained and the stained sections in *water* or *normal salt solution* should never be neglected, since many structural peculiarities—for example, connective-tissue formations—stand out distinctly in these media, which under the clearing influence of glycerol or xylol-balsam almost entirely elude observation. Preparations mounted in water or salt solution cannot be preserved.

2. Preparations mounted in *glycerol* can be preserved ; in order to prevent the shifting of the cover-glass it should be secured with cover-glass cement (p. 23). The edge of the cover-glass must be *perfectly dry* ; this is an *indispensable preliminary condition*, because the cement adheres only to a dry glass surface. The drying is accomplished in this wise : remove the excess of glycerol surrounding the cover-glass with filter-paper and then with a cloth moistened in 90 per cent. alcohol and

* Examinations with *low* powers, without a cover-glass, are permissible only for the most superficial orientation : *e. g.*, to ascertain if an object has been sufficiently teased. In all other cases the cover-glass is *indispensable*. In order to convince one's self of this an uncovered section should be examined, then covered with a cover-glass and examined again. Many a good preparation that one neglects to cover appears useless. Examinations with high-power objectives without a cover-glass are in general not allowable ; they should only be attempted with certain methods, *e. g.*, Golgi's.

turned over the finger-tip carefully wipe the slide clean all around the cover-glass without disturbing the latter. Heat a glass rod and thrust it into the hard cement; * place a drop at each corner of the cover-glass and trace a continuous band from 1 to 3 mm. wide, in such a way that one edge rests on the cover-glass, the other on the slide. Finally, re-heat the rod and smooth the surface of the band of cement.†

Preparations mounted in glycerol often do not become transparent until the second or third day. Hematoxylin and other dyes soon fade in it; picrocarmine and carmine, on the contrary, are permanent.

3. The mounting of objects in *xylol-balsam* is the most popular preserving method. In comparison with glycerol it has the advantage of keeping the colors, but has one disadvantage: it clears more vigorously than diluted glycerol, and thus renders many delicate structures completely invisible.

Sections in alcohol or water cannot without further treatment be mounted in balsam; they must be *previously dehydrated*. For this purpose the sections are lifted with a needle (very thin sections with needle and section-lifter) and placed in a covered watch-glass containing 5 c.c. of 95 per cent. alcohol. In making this transfer as little as possible of the water should be allowed to adhere to the section. If a section-lifter is used, the water clinging to it should be absorbed with filter-paper; if the sections are lifted on a needle, the water can be removed by bringing the filter-paper into gentle contact with them. Thin sections remain in the 95 per cent. alcohol two minutes; thick sections, ten minutes or more.‡ Then the sections are transferred for *clearing* to a watch-glass containing 3 c.c. of carbol-xylol § or xylol, || as much as possible of the alcohol being removed with filter-paper before placing them in the clear-

* Glass rods fracture very easily in this procedure, nevertheless are preferable to metal rods, because the latter cool too quickly. The fracturing can be prevented in a measure by heating the glass rod to redness, meanwhile turning it continuously; only rods insufficiently annealed break when they are dipped into the cement.

† *Editor's remark:* King's fluid cover-glass cement (p. 23, foot-note) is to be applied with a small brush.

‡ Beginners are recommended to transfer the sections from the water to 5 c.c. of 90 per cent. alcohol, and then to place them in an equal quantity of 95 per cent. alcohol.

§ Thin sections may be transferred from the 95 per cent. alcohol directly on to the slide, the superfluous alcohol removed by means of absorbent paper, and a drop of carbol-xylol applied. At first the xylol will retreat from the section and must be led back with the needle; when the clearing is completed, which can be ascertained under the microscope with the low power, the xylol should be absorbed with filter-paper and a cover-glass with a drop of balsam applied. When examining uncovered sections lying in xylol both xylol and section often become clouded by the moisture exhaled in breathing; in this case drain off the clouded xylol and add a fresh drop.

|| On account of its greater sensitiveness to water and because it evaporates so easily the manipulation with xylol is more difficult. Many a good preparation spoils at the last moment, because the xylol has been allowed to evaporate.

ing agent. If the watch-glass is placed on a black background the effect of the oil can be watched, and it will be seen that the sections gradually become transparent. Care must be taken not to breathe into the watch-glass, or the xylol will immediately become turbid. If some areas of the section do not become transparent within two or three minutes (such areas appear white and opaque in direct light, black-brown in transmitted light), this indicates that the section is not dehydrated and it must be put back into absolute alcohol. When the clearing is completed the section is transferred to a dry slide, the superfluous xylol* absorbed by gentle pressure with a strip of smooth filter-paper,† and a cover-glass, on the under surface of which a drop of balsam is suspended, applied. If several sections are to be mounted under *one* cover, arrange them close together with a needle; then, by means of a glass rod, apply a thin, even layer of balsam to the under surface of the cover-glass and place it on the sections. Large air-bubbles are driven out by placing a small drop of balsam at the edge of the cover-glass; on the following day it will be seen that the air-bubbles have retreated from beneath the cover. Small air-bubbles disappear spontaneously and may be neglected.

It not infrequently happens to beginners to discover that the balsam becomes turbid, and finally renders the entire preparation, or parts of it, untransparent. This is due to incomplete dehydration. If the clouding is slight, which under the microscope is seen to consist of minute drops of water, a gentle warming of the slide is often sufficient to remove it. In the case of much-clouded preparations, place the whole slide in carbol-xylol for half an hour; then carefully lift off the cover-glass, place the section for two minutes in carbol-xylol, in order to dissolve off the adherent balsam, and then dehydrate in 4 c.c. of absolute alcohol, which should be changed in five minutes; clear in carbol-xylol and mount in balsam.

The balsam dries slowly, therefore the slides must not be stood on edge, but be kept in a horizontal position.

The series of processes through which a fresh object must pass until it is preserved as stained sections is a very long one. When, for example, the directions in the Special Technic require "fixation in Zenker's fluid, hardening in gradually strengthened alcohols, staining of

* The carbol-xylol in the watch-glass that has been used for clearing may be returned to the bottle.

† The double folded strip is held fast by the left hand to the left end of the slide and, lying upon the preparation, is gently stroked from left to right by the index finger of the right hand.

sections in hematoxylin and eosin, and mounting in balsam," the procedure is as follows :

1. Place the fresh object, about 1 cm. in diameter, in 60 c.c. of Zenker's fluid * for twenty-four hours.
 2. Wash in (if possible running) water for twenty-four hours.
 3. Place in 20 c.c. of distilled water for about fifteen minutes.
 4. Transfer to 50 c.c. of 50 per cent. alcohol for twenty-four hours ; from now on the object is to be kept in the dark.
 5. Transfer to 50 c.c. of 70 per cent. alcohol for twenty-four hours.
 6. Transfer to 50 c.c. of 90 per cent. alcohol and tincture of iodine for from eight to fourteen days, daily adding tincture of iodine.
 7. Transfer to pure 90 per cent. alcohol, which is to be changed two or three times.
- The object thus fixed and hardened can be sectioned at once or may remain indefinitely in the 90 per cent. alcohol, which perhaps should be once renewed.†
8. Transfer the sections from the alcohol to 5 c.c. of hematoxylin for five minutes.
 9. Transfer to 30 c.c. of distilled water for from ten minutes to two hours.
 10. Stain in 5 c.c. of diluted eosin for from one to three minutes.
 11. Wash in 5 c.c. of distilled water for two minutes.
 12. Dehydrate in 5 c.c. of absolute alcohol for five minutes.
 13. Clear in 5 c.c. of carbol-xylol for five minutes.
 14. Mount in xylol-balsam.

§ 11. EXAMINATION OF FRESH OBJECTS.

I have placed this method last because it is the most difficult and presupposes a somewhat practised eye. This practice is most readily acquired by previous examination of prepared (hardened, stained, etc.) objects ; having once clearly perceived and studied peculiarities of structure it is then not difficult to detect them in fresh objects, even though the majority of the details leave something to be desired in point of distinctness. The following instructions should be observed :

The slide and cover-glass must not be oily. They should be

* The quantities named are calculated only for this 1 cm. cube ; for several or for larger objects more fixing and more hardening fluid must be used.

† The following quantities are intended for from three to six sections ; for a larger number of sections the quantity of the absolute alcohol in particular should be increased.

cleansed with alcohol and dried with a perfectly clean cloth.* Then transfer *one* drop of a 0.75 per cent. salt solution (p. 20) to a slide, place in it a *small* piece of the object to be examined and cover it with a cover-glass. Pressure must be carefully avoided; if the structures are very delicate support the cover-glass on two strips of thin paper placed at the sides of the object. If the object requires no further treatment the cover-glass should be sealed with paraffin to prevent evaporation. Melt a small piece of paraffin on the blade of an old scalpel and let it flow, not from the tip but from the edge, on to the rim of the cover-glass; gaps that may occur in this frame of paraffin can be closed with the reheated scalpel. In most cases the influence of certain reagents (acids, alkalies, stains) is studied directly under the microscope. It is then necessary to remove a portion of the medium in which the object happens to be mounted (in the present instance the salt solution) and to replace it by another fluid. For this purpose place a drop of picrocarmine at the right edge of the cover-glass, by means of a glass rod. Should the drop not touch the edge of the cover-glass, do not incline the slide, but lead it with a needle to the appropriate position. It may now be seen that a little of the staining fluid mingles with the salt solution, but does not properly flow under the cover-glass. In order that this shall occur place at the left edge of the cover-glass a little piece of filter-paper† and immediately the picrocarmine will be seen to diffuse under the cover-glass and occupy the entire area.‡ Then remove the filter-paper and let the stain act; when the staining is completed,—this can be ascertained under the microscope,—place at the right edge of the cover-glass a drop of diluted glycerol to which, in picrocarmine staining, as much acetic acid is added as will drop from a steel needle (hence a very small drop), and again apply the filter-paper to the left edge of the cover-glass. In this way a whole series of fluids can be passed through beneath the cover-glass and their action on the tissues tested. Some of these fluids, for example, picrocarmine, must remain in contact with the objects for a very long time if they have been previously fixed with osmic acid. In this case evaporation is prevented by placing the object in a *moist-chamber*. For the construction of a moist-chamber a porcelain

* For removing the oil from new cover-glasses, heating them on a piece of sheet-iron for five minutes over the flame of a Bunsen burner is recommended.

† Cut a strip 4 cm. long and 2 cm. broad, fold it square, and place the paper tent thus formed on the slide, so that one of the 2 cm. ends, which must be *perfectly straight*, touches the left edge of the cover-glass.

‡ After the first drop has penetrated place two or three additional drops at the right edge of the cover-glass.

plate and a small bell-glass 9 cm. in diameter are required. Pour water into the plate to the depth of 2 cm. and stand in the middle a small glass dish or a cork disk supported on wooden pegs; on the latter place the slide with the preparation and cover the whole with the glass bell, the free edge of which must be submerged in the water.

§ 12. STORING OF PERMANENT PREPARATIONS.

The finished preparations should be promptly labeled. Labels of cardboard about 1.2 mm. thick, glued to the slide with fish-glue (isinglass) are preferable to those of gummed paper; the slides can then be placed one upon the other without injury to the preparations. The labels should be as large as possible (2 cm. square for slides of English form) and should bear the name of the animal, of the organ, and if possible a brief statement of the method used. Of the cases * for storing the preparations only such should be chosen in which the slides lie flat, not those in which they stand on edge.

* The best and cheapest cases are made by Th. Schröter, Leipzig, Connewitz. I recommend for box form *pattern O* (for about 300 slides), price 2 *M.* (50 cents); for tray form, *P*, with spring covers (for 10 to 20 slides according to size), price 45 Pfg. (about 12 cents). The tray form has the great advantage of allowing all the specimens to be seen at once. In the United States Schröter's boxes and trays are supplied by King & Co., New York, the Bausch & Lomb Optical Co., New York, and other dealers.

III. MANAGEMENT OF THE MICROSCOPE.

In conformity with the position taken in the introduction, an exhaustive description of the optic and mechanic parts of the microscope cannot be entered upon here. Figure 1 will recall to the reader the usual names of the several parts of the microscope.

The first requisite in the use of the microscope is perfect cleanliness of all its parts (see also p. 17). The surface of the mirrors, objectives, and oculars should not be touched with the fingers. The objective should be held with the lower end directed toward the window and the clearness of the reflected image thus tested. Foreign matter on the ocular can be detected by rotating the latter in the tube, when anything that is adherent will revolve.

After the ocular has been placed in the upper end of the draw-tube and a low-power objective screwed on the lower end of the tube or on the revolver, the field of view should be illuminated with light reflected from a suitable source by the concave mirror placed below the stage. This is accomplished by moving the mirror tentatively in all directions, with the diaphragm widely open and the front lens of the objective about 1 cm. above the level of the stage, till the eye, looking simultaneously through the eye-piece into the microscope, sees the field brightly and uniformly lighted.* The concave mirror should be used with dry lenses, except when a substage condenser is employed.

The light reflected from a white cloud or from a white window-blind illuminated by the sun is recommended; less desirable but still useful as a source of light is the blue sky. Direct sunlight must be avoided. In using artificial illumination the light should be taken from the inner surface of a white lamp-shade, not directly from the flame. A screen of green glass placed between the mirror and the source of light, or between the mirror and the object, agreeably subdues artificial

* The rays of light reflected from the mirror in this position pass perpendicularly through the object on the stage. This is called *central illumination*. For distinguishing slight differences of level between adjacent parts of an object it is of advantage to use *oblique or lateral illumination*, to obtain which the mirror is moved to the side so that the rays reflected from it strike the object obliquely. When lateral illumination is used the diaphragm and the cylinder in which it is mounted must be removed, that the opening in the stage be as large as possible.

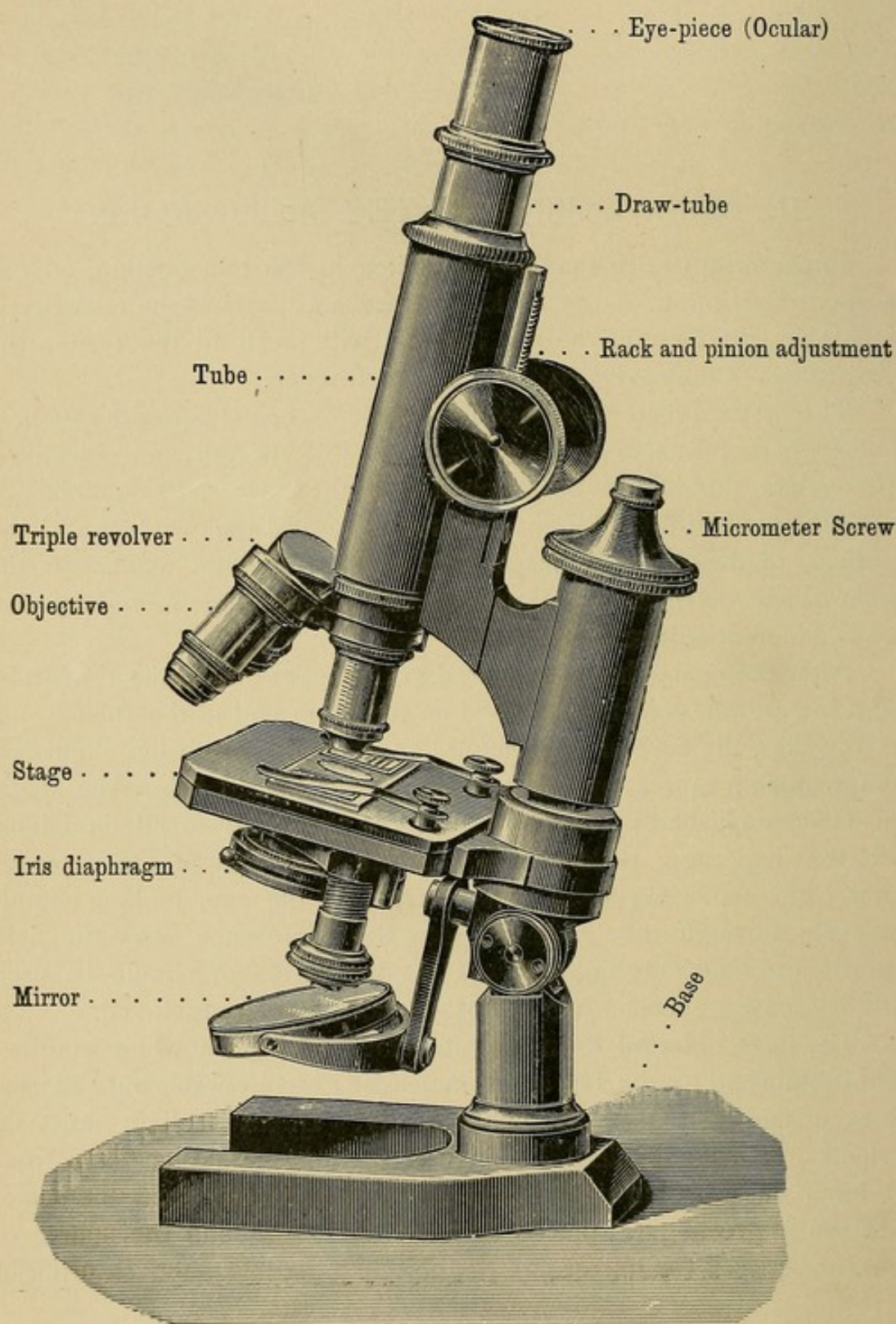


FIG. 1.—LEITZ MICROSCOPE, STAND II (one-half actual size).

light, without essentially injuring the definition of the image. It is self-evident that the microscopist should not sit in direct sunlight; the instrument should be placed about a meter from the window.

Having secured the light the examination may begin. *Always examine first with the low-power, then with the high-power objective; do not use strong oculars; they narrow and darken the field of view and render the examination much more difficult.** The low- and medium-power oculars (Leitz, Oc. I) of the usual outfit supplied with the microscope answer for the great majority of cases.

The increased magnification obtained by pulling out the draw-tube is seldom necessary. With low-power lenses a diaphragm having a large opening should be used; with high-power lenses a diaphragm having a small opening. In focusing the object the coarse adjustment by rack and pinion is used first; the objective is placed near to the object, but at a distance greater than its focal length and then, with the eye applied to the ocular, the tube is gradually lowered until the indistinct outlines of the image appear, which is then brought into distinct view by means of the fine adjustment or micrometer-screw. The left hand should hold the slide, while the right should remain at the micrometer-screw. Since only the points lying in a single plane of the object can be in focus and distinctly seen at one time, the preparation must be examined with slight raising and lowering of the tube, that is, with change of focus by gently turning the micrometer-screw. In using the microscope the habit should be formed of keeping both eyes open.

One should never neglect to examine the preparations with a hand-lens. For this purpose the oculars (*e. g.*, Leitz, Oc. III) can be used. The mounted specimen is held with the cover-glass side toward the light; the upper or back lens of the ocular is placed directly against the slide, which is examined at the lower or front lens.

SKETCHING.

An invaluable aid to study is the sketching of the microscopic object. The power of observation is made considerably keener and many details which otherwise would be completely overlooked are discovered while the sketch is in progress. The most attentive examination cannot replace the advantages which sketching affords. Even those who have little practice in drawing should nevertheless try to sketch the preparations under both low- and high-power objectives. For this purpose the drawing-paper should be on a level with the stage, the left eye applied to the microscope, the right eye directed to the paper and the pencil-point. At first this is somewhat difficult, but a little practice will soon give the necessary facility.

* The majority of the preparations from which the illustrations in this book were taken were examined and sketched with weak oculars.

MEASUREMENT.

For this purpose an ocular-micrometer and stage-micrometer are used.* The latter is laid on the stage of a microscope provided with an ocular-micrometer and the number of divisions of the ocular-micrometer corresponding to one part of the stage-micrometer is ascertained.† The dimensions of the spaces of the stage-micrometer being known the size of the object, which with a given magnification will occupy one or more of the divisions of the ocular-micrometer, is easily calculated. The following illustrations may render the manipulation intelligible.

With ocular I and draw-tube pushed in 5 divisions of the ocular-micrometer correspond with one division of the stage-micrometer. Each division of the stage-micrometer used = $\frac{1}{20}$ mm. Hence 5 divisions of the ocular-micrometer = $\frac{1}{20}$ (0.05 mm.), and 1 division of the ocular-micrometer = 0.01 mm. If then any microscopic object, *e. g.*, a striated muscle-fiber, the diameter of which is to be measured with this magnification, occupies 4 divisions the fiber is 0.04 mm. broad.

It is often difficult, especially with low magnification, to count the fine divisions of the ocular-micrometer. This can be more easily done by noting the longer lines marking every fifth or tenth division. For instance, with Leitz Objective 3, Ocular I, and the draw-tube drawn out, 40 divisions of the ocular-micrometer correspond with 5 divisions of the stage-micrometer. Therefore, 40 divisions = $\frac{5}{20}$ mm. = 0.25 mm., and one division of the ocular-micrometer with this magnification = 0.0062 mm., 2 divisions = 0.0124 mm., and so on.

With Leitz Objective 7, Ocular I, and draw-tube pushed in, 30 divisions of the ocular-micrometer correspond with one division of the stage-micrometer; 30 divisions = 0.05 mm., one division = 0.0017 mm., or 17μ .‡ Finally, with Leitz Objective 7, Ocular I, and draw-tube drawn out, 40 divisions of the ocular-micrometer = one division of the stage-micrometer. Therefore, 40 divisions = 0.05 mm., one division = 0.0012 mm., or 1.2μ .

* Some ocular-micrometers (Leitz) are made to rest upon the diaphragm inside the ocular; others (Seibert) to be inserted through a lateral opening; or, in some cases, special oculars (Zeiss) for measuring are made for the microscope. The actual size of the divisions of the ocular-micrometer need not be known. The stage-micrometer is a glass slide on which 1 mm. with 100 divisions is engraved. Instead of this a second ocular-micrometer, which usually contains a mm. with only 20 divisions, may be used. Measurements made with this are not as accurate, but the errors are so insignificant that they scarcely need consideration.

† Beginners often find it difficult to focus the lines on the stage-micrometer; faint or oblique illumination of the object makes it easier to detect them.

‡ One micron = μ = 0.001 mm.

He who has many microscopic measurements to make will find it useful to prepare a table for each magnification used, in which the equivalent values of 1 to 20 and from this in tens up to 100 scale divisions of the ocular-micrometer are given. It must be emphasized that the foregoing calculations by no means apply to all the microscopes made by Leitz. The values must be specially determined for every instrument by the foregoing method.

In conclusion the microscopist is advised to be patient, very patient; if his preparations are unsuccessful let him not search for the cause in the deficiency of the methods recommended,—I have often tested them—but in himself; he who cannot accustom himself conscientiously to follow the written instructions,* who grasps delicate objects with his fingers, who contaminates the reagents by pouring one into the other, who leaves objects in fixing fluids exposed to the sun or allows them to become dry, has not the right to expect good results from his slovenly work.

* The periods of time given for staining, dehydrating, etc., have only an approximate value. They vary within considerable limits in accordance with the thickness of the sections, the concentration of the solutions, etc. Experience will soon teach the microscopist to determine the precise period of time.

PART II.

MICROSCOPIC ANATOMY AND SPECIAL TECHNIC.

The animal body consists of cells which are derived from a single cell by repeated division. At the beginning of development the cells are of similar form, all are spherical structures, none is furnished with special characteristics that distinguish it from its companions. The cells are still *indifferent*. In the course of development the cells arrange

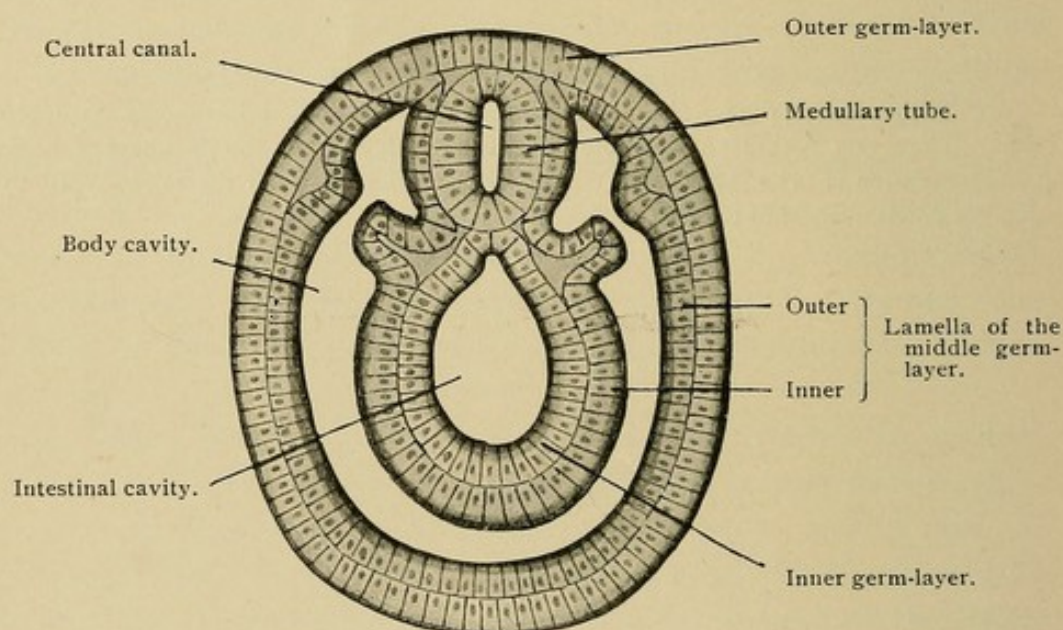


FIG. 2.—SCHEMATIC SECTION OF THE BODY OF A VERTEBRATE EMBRYO. The free side of the cells is marked by deeper shading.

themselves in the *germ-layers*; these are cell complexes, that in the lower vertebrates are for a time disposed in a simple stratum. The germ-layers thus represent an epithelium, that is, a continuous layer of cells which covers outer and inner surfaces of the body; each cell is an epithelial cell, in which a *free* side, directed toward the surface, and a *basal* side can be distinguished (Fig. 2).

In the further course of development the germ-layers become partially stratified, a not insignificant number of cells, entire or in divisions, depart from the epithelial association, whereby the cells become different

from one another, they *differentiate*. As a rule, the "differentiated" cells that have developed in a given direction are united in complexes, without definite spatial limitation, and so form a *tissue*. *A tissue, therefore, is a complex of similarly differentiated cells.* We distinguish four principal tissues: (1) the *epithelial tissue*; (2) the *supporting tissue*; (3) the *muscular tissue*; (4) the *nervous tissue*. Epithelial tissue can be developed from each of the three germ-layers. Supporting tissue is developed only from the middle germ-layer, the mesoderm; nerve tissue only from the outer germ-layer, the ectoderm; muscle tissue, in by far the greater part, is of mesodermal origin, but in isolated instances of ectodermal origin. So long as these tissues are still young they consist *only* of similar elements, only of cells; but in the process of development this condition is modified in a twofold manner. First, the cells produce special substances, which are bestowed between cells and are called *intercellular substances*. However, thereby the character of the tissue is not essentially altered. The above definition of "tissue" need be only so extended that we describe a tissue as *a complex of similarly differentiated cells and their derivatives*. More radical is the second modification, which consists in the interpenetration of a tissue of one kind by other tissues. This occurs in very different degrees. The epithelial tissues have preserved the greatest simplicity and following them the supporting tissues. But muscle and nerve in the matured state are so largely intermixed with other tissues, that even though in each the elements respectively differentiated to muscles and nerves predominate, yet one can scarcely speak of a tissue in the sense of the given definition.* Therefore the tissues are not equivalent among one another. In the lowest rank stand the epithelial and the supporting tissue; different from each other, respecting their form as well as their function, they both occur in the vegetable world and therefore we can class them as *vegetative tissues*. On a higher level, as well morphologically as physiologically, stand the muscle and nerve tissues, and being peculiar to the animal organism they are named *animal tissues*.

The fact that the tissues originate from the epithelial germ-layers does not warrant the conclusion that after complete differentiation of the chief tissues supporting, or muscle, or nerve tissue can arise from the perfected epithelial tissue. Each principal tissue then furnishes *only* its kind.

When different tissues unite in the formation of a body of definite internal structure and definite external form † they constitute an *organ*.

* For this reason the proposition has been made to take exception to the classification in tissues and to distinguish only elements and organs.

† Usually in the definition of an organ "the definite function" is included; but this does not come within the limits of a *morphologic* definition, nor is it a special peculiarity of an organ, but may be the property of a cell as well as of a tissue.

Accordingly our task resolves itself into : (1) the study of the cells and of the tissues, and (2) the study of the organs. The investigation of cells and of tissues is the object of *histology*. Histology is a division of minute anatomy, which, because of the instrument most used in its study, is called *microscopic anatomy*. The investigation of organs, also, so far as it can be done with the aid of the microscope, is the task of microscopic anatomy.

I. HISTOLOGY.

(MICROSCOPIC ANATOMY OF THE CELLS AND THE TISSUES.)

A. THE CELLS.

A cell, *cellula*, is a spatially limited structural element, which under certain conditions is able to nourish itself, to grow, and to multiply. In virtue of these properties the cell is called an *elementary organism*.

The cell of the germ-layers is a body having polar differentiation, that is, free and basal sides of the cell are typically different (p. 60). At the *free* pole the development of cuticular formations (p. 66), cilia, tactile hairs, etc., occurs, here pigment is first formed, here the discharge of secretion takes place; at the *basal* pole processes (fibrillæ, fibers) originate, by which the cell enters into association with neighboring tissues.* A line connecting the free and the basal pole is designated the chief axis of the cell.

The essential elements of a cell are the protoplasm and the nucleus, and generally, as third element, the centrosome.

1. The *protoplasm*, "cell-substance," is a soft, viscid substance of alkaline reaction, insoluble in water, highly distensible, that consists principally of albuminous substances, much water and salts, and contains a special nitrogenous proteid, the *plastin*. In the protoplasm small granules, *microsomes* (*plasmosomes* †), occur in variable quantity; when numerous they may impart to the protoplasm a dark appearance. They are irregularly distributed; namely, are absent in the superficial layer, the *exoplasm* ("cuticular stratum"), which is somewhat denser and perhaps possesses a special function. With the aid of very high magnifying powers it is seen that protoplasm possesses a structure: a framework of fibrils ("filar-

* This polar differentiation can be demonstrated in many cells of the epithelial, muscular, and nervous tissues in the developed organism; in other cells, particularly in those of the supporting tissues, insuperable difficulties still exist; the question suggests itself whether with the differentiation of these elements from the germ-layers the polar differentiation was not lost or even whether it developed at all.

† "Plasmosomes," in contradistinction to the granules of the nucleus, that then are to be named "karyosomes." Specifically developed plasmosomes, that are united in fibrils, have been named "mitochondria" (fibril-granules); they are especially developed in semen-cells.

mass," "mitom") often forming a network, which is embedded in an apparently homogeneous ground substance ("interfilar-mass," "cytolinin") chemically distinct from the filar-mass (Flemming).* A portion of the plasmosomes lie embedded in the fibrils; individual fibrils are nothing but linear arrangements of plasmosomes.

In many instances the protoplasm exhibits still other structures of different signification, as follows:

1. Canaliculi of two kinds: (a) secretory capillaries in gland-cells (p. 85); (b) delicate tubules, that communicate with the lymph-spaces exter-

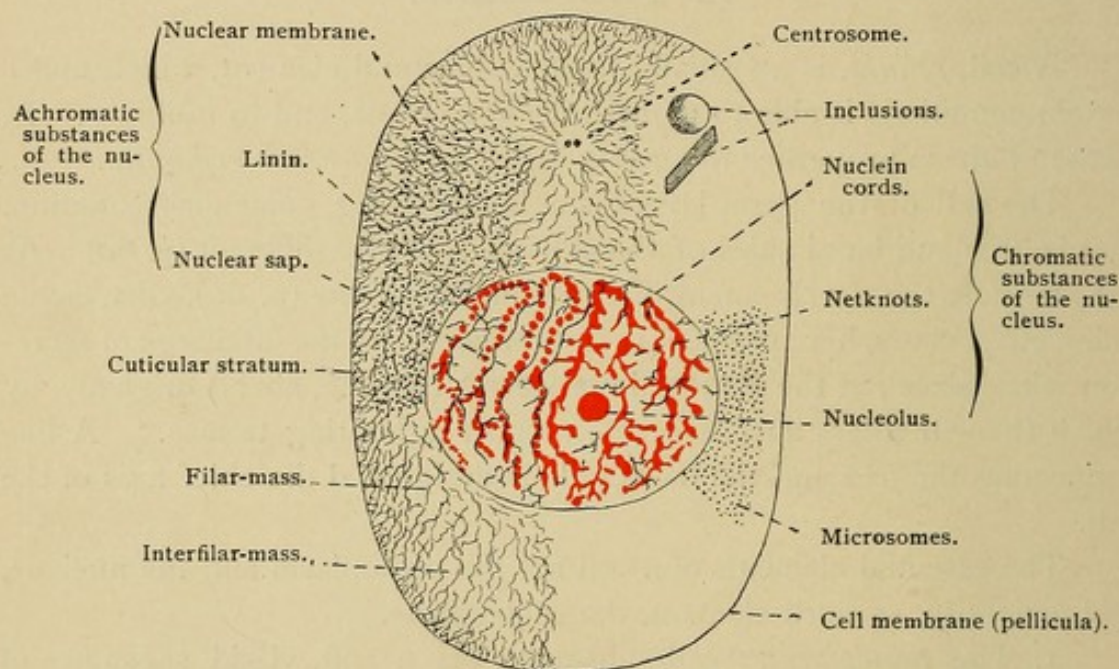


FIG. 3.—SCHEME OF A CELL. Microsomes and filar-mass only partly sketched.

nal to the cell; they were first discovered in spinal ganglion cells (Fig. 62), but may also be demonstrated in intestinal epithelial cells, in gland-cells, in egg-cells, etc. Since they provide for the nutrition of the cells they have been named "trophospongium."

2. Closed networks, that do not open at the periphery of the cell; this "apparato reticulare" has been found in nerve-cells (Fig. 63), cartilage cells, many gland-cells, and in the corneal endothelium. Their meaning is still obscure; possibly they belong to the same category as

3. Various formed cell-contents, rings, capsules, cord-fabrics and the like.

*Opinions regarding the structure of the protoplasm are by no means agreed. According to Bütschli the structure is foamy, that is, it contains small spaces, or cavities, that do not communicate with one another. A third theory holds that the protoplasm is composed of granules ("granula"); the fact that many fibrils of the filar-mass consist of granules arranged in rows has brought this "granule theory" new adherents; on the other hand, the teaching of Altmann, according to which the granules are the true elementary organisms ("bioblasts"), has been generally discarded.

2. The *nucleus* is a usually vesicular, clear, sharply defined body lying in the interior of the cell, that consists of several proteid substances, *nuclein* (chromatin), *paranuclein* (pyrenin), *linin*, *nuclear sap*, and *amphipyrenin*. By their affinity for stains nuclein and paranuclein are distinguished from the other three so-called achromatin substances, but differ chemically from each other. For example, on the addition of distilled water the structures composed of nuclein disappear, while those composed of paranuclein remain intact. In the simplest case (in spermatozoa) the nucleus is a compact mass of nuclein, to which the paranuclein is attached, but usually it consists of a network of fine linin threads and coarser nuclein cords.* The latter are of different caliber, and at isolated places are thickened to knots, the *netknots*, that must not be confused with the nucleoli. Linin and nuclein form the *nuclear network*, the interstices of which are occupied by one or more nucleoli, consisting of pyrenin, and by the nuclear sap. The nuclear membrane, not always present, consists of amphipyrenin; often a membrane is simulated by a thin superficial layer of nuclein. The nuclear network and the nucleoli are subject to momentous changes, according to the age of the cell.

Most cells contain but one nucleus; only a few have several nuclei (some wandering cells, giant cells, and others). Nonnucleated cells (horny cells of the epidermis, colored blood corpuscles of mammals) originally possess nuclei, but lose them in the course of development.

3. The *centrosome* (central corpuscle) is an exceedingly diminutive corpuscle, that consists of a homogeneous, less often honeycomb mass, the centropasm, and a very much minuter corpuscle, the centriole† (centriolum). The centrosome lies in the protoplasm, which here is differentiated to a sometimes clear, sometimes dim, encircling court ("archoplasm," "idiosome"); sometimes it lies in the neighborhood of

Pseudopodia
(p. 76, remark *). Centrosome.

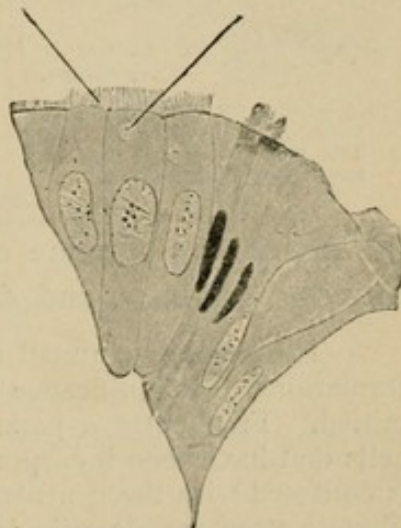


FIG. 4.—PORTION OF A SECTION OF EPITHELIUM OF THE LARGE INTESTINE OF MAN. $\times$ ca. 660.

* In particularly favorable objects it can be seen that the nuclein cords consist of rows of granules, that lie upon the linin fibrils; this relation is sketched in the left half of the schematic cell, Fig. 3.

† Our knowledge of the minute structure of the centrosome has been obtained through the study of invertebrate animals; the cells of vertebrate animals are too small for investigations of this kind. The centriole has not yet been demonstrated in any vertebrate animal cell.

the nucleus (Fig. 5), sometimes remote from this, frequently between the free surface and the nucleus * (Fig. 4). For the purpose of cell division the centrosome passes through a cycle of phenomena (p. 69) the duration of which is very variable; the phase in which a duplication of the centrosome, the "diplosome," occurs endures the longest.† For this reason the diplosome is found in the majority of resting cells, that is, cells not in the immediate process of division.‡

An unessential element of the cell is the *cell-membrane*, an independent, continuous, membranous border stratum, which is distinctly marked off from the protoplasm; it is wanting in many cells and when present is either a transformation of the peripheral zone of the protoplasm or a

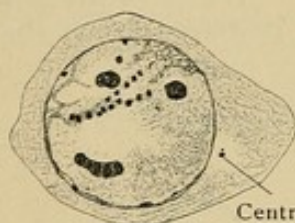


FIG. 5.—CELL OF THE BONE-MARROW OF A RABBIT. $\times 1500$. The centrosome lies near the nucleus, in a clear space.

secretory product of the latter. When the membrane surrounds the cell on *all sides*, it is named *pellicula*; when it lies only on the free surface, that is, only on *one side*, it is named *cuticula*. By *crusta* is understood the denser border zone of the cell, that without sharp demarcation gradually passes into the protoplasm beyond. Other unessential elements are the inclusions occurring in

the protoplasm of some cells, the pigment, glycogen, etc., crystalloids, secretion granules, and drops of oil, of aqueous and mucous fluids.§

The term "paranucleus" has been used to designate widely different formations, the significance of which individually is not yet everywhere determined. Frequently a paranucleus is simulated by fragments of degenerated cells that have been incorporated by living cells; in other cases the paranucleus is confused with the centrosome, with masses of secretion, or with the protoplasmic structures described on page 64.

* In many gland-cells the centrosome lies where the secretion accumulates, the discharge of which is accomplished by contraction of the protoplasmic framework lying between the masses of secretion. In the cells of the intestinal epithelium (p. 76) provided with pseudopodia the centrosome lies close beneath the point of origin of the pseudopodia; taking into account the similar behavior of the ciliated cells of the epididymis and the behavior of the centrosome of the seminal filaments (cf. The Reproductive Organs), as well as the rôle of the centrosome in mitosis (p. 69), the inference that the centrosome is the (active or passive?) center of motor processes is highly probable. In the spermatocytes of *ascaris megalocephala univalens* and in carcinoma cells the centrosome has been observed in the interior of the nucleus.

† The doubling of the centrosome is preceded by a division in two of the centriole.

‡ The diplosome phase is the most practicable for estimating the duration of the resting stage of the cell, because thereby the competency of the resting cell for the earliest possible inception of the mitotic process is indicated.

§ Such products of metabolism, when they appear in the form of small particles, may be termed "granules" and are not to be confused with the "plasmosomes" (p. 63), which represent structural elements of the cell. In practice the distinction between the two may frequently be involved in extraordinary difficulties.

Cells differ greatly in form. They may be: *spherical*, the typical form of all cells in the embryonal period, while in the adult, for example, resting leucocytes are spherical; *discoid*, *e. g.*, the colored blood corpuscles; *polyhedral*, *e. g.*, the liver-cells; *cylindrical* or *columnar*, *e. g.*, the epithelium of the small intestine; *cubical*, *e. g.*, the epithelium of the capsule of the crystalline lens; *flattened* (so-called squamous epithelium), *e. g.*, the epithelial cells of the blood-vessels; *spindle-shaped*, *e. g.*, many connective-tissue cells; *elongated into fibers*, *e. g.*, smooth muscle-fibers; *stellate*, *e. g.*, many ganglion-cells. The form of the nucleus usually corresponds to the form of the cell. It is oval in cylindric, spindle-shaped, and occasionally also stellate cells; rounded in spherical, cubical and many stellate cells. Lobulated, so-called polymorphous nuclei are found in leucocytes and in giant-cells; they are an expression of activity on the part of the cell, tending to locomotion or change in form, or to increased metabolic energy.

The size of cells varies from forms microscopically small, $4\ \mu^*$ (colored blood corpuscles), to macroscopic bodies (eggs of birds, of am-

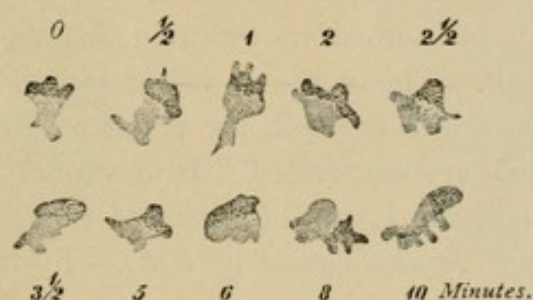


FIG. 6.—LEUCOCYTES OF A FROG. $\times 560$. Changes in form observed during ten minutes; 0, at the beginning of the observation; $\frac{1}{2}$, a half minute later, etc. Technic No. 49.

phibians). The size of the nucleus corresponds in general to that of the protoplasmic body; only mature ova, despite their great dimensions, have tiny nuclei.

In some instances the cells unite in a *syncytium*, that is, a common protoplasmic mass, in which occasionally the scattered nuclei still indicate the otherwise undefinable cell territories.

The *vital properties* of cells will be discussed here only in so far as they can be studied by direct microscopic observation; other details must be sought in textbooks of physiology. Accordingly, the phenomena of motion in cells, the reproduction of cells, and those microscopic processes which are associated with the secretory activity of cells will be considered.

* A micron, $\mu\kappa\rho\nu = \mu = 0.001\text{ mm.}$

The *phenomena of motion* occur in the form of ameboid* activity, of ciliary movement, and of contraction of certain fibers (muscle-fibers). The ameboid motion is the most important; of wide-spread occurrence, it has been observed in nearly all the cells of the animal body. In well-marked cases, *e. g.*, in leucocytes, the protoplasm of the cell projects finer or coarser processes (pseudopodia), that divide and flow together again and in this way produce the greatest variety of forms. These processes may be retracted or they may become attached somewhere and partially draw the remainder of the cell-body after them, the result of which is locomotion, or the so-called "wandering" of cells. The wandering cells play an important part in the economy of the animal body. The processes can flow around granules and cells and thus enclose them in the cell-body, an occurrence described as the *feeding* of the cell.† Cells that can transform or "digest" such inclusions are named *phagocytes*. Ameboid movements take place very slowly, in warm-blooded animals only on artificial warming of the object. For ciliary motion and the phenomena of contraction, see the chapters on epithelial and on muscle tissue, respectively.

There is another phenomenon of motion, which, however, does not occur in the living cell. This is the so-called *molecular motion*, an oscillation of minute granules in the cell, the result of molecular currents in the fluid in which they are suspended. It may often be observed in the salivary corpuscles (see the Lymph-follicles of the Tongue).

Reproduction and Multiplication of Cells.—Formerly, two kinds of cell formation were distinguished, spontaneous generation (*generatio æquivoca*) and generation by division. According to the theory of spontaneous generation, cells were supposed to originate in a suitable fluid, the *cytoblastema*. Something of this kind may formerly, in unthinkably early ages, have occurred; but now we recognize only *one* kind of cell generation, that is, reproduction by *division* of preexisting cells. "Omnis cellula e cellula."‡

In the division of a cell, first the nucleus and then the protoplasm divides into two usually equal parts. In this process a special grouping

* This movement is exhibited in its perfection by unicellular organisms named amebæ; thence the phrase "ameboid motion."

† This must not be confused with the *nutrition* of the cell, which is effected by a series of complicated chemical processes within the cell; diosmotic currents, imbibition, molecular pressure, etc.

‡ Likewise, a new nucleus can arise only by the division of an existing nucleus. The theory of spontaneous generation of nuclei, according to which nuclei originate directly from the protoplasm and independently of existing nuclei, lacks unequivocal evidence.

and rearranging of the nuclear substances (p. 65) take place according to definite laws. This mode of division is called *indirect division*, division by *mitosis*,* *karyokinesis*. Its cycle is usually divided into three phases, as follows :

(1) Prophase.

The centrosome and the nucleus approach each other and finally the former arrives in the immediate neighborhood of the nuclear membrane, surrounded by the bright halo (p. 65), in which the radiating, delicate fibrils now become more distinct; collectively these fibrils are called *astrosphere*. The centrosomes, previously duplicated in the resting state of the cell (p. 66), now move apart and instead of the *single* astrosphere enveloping the diplosomes there are two present, one for

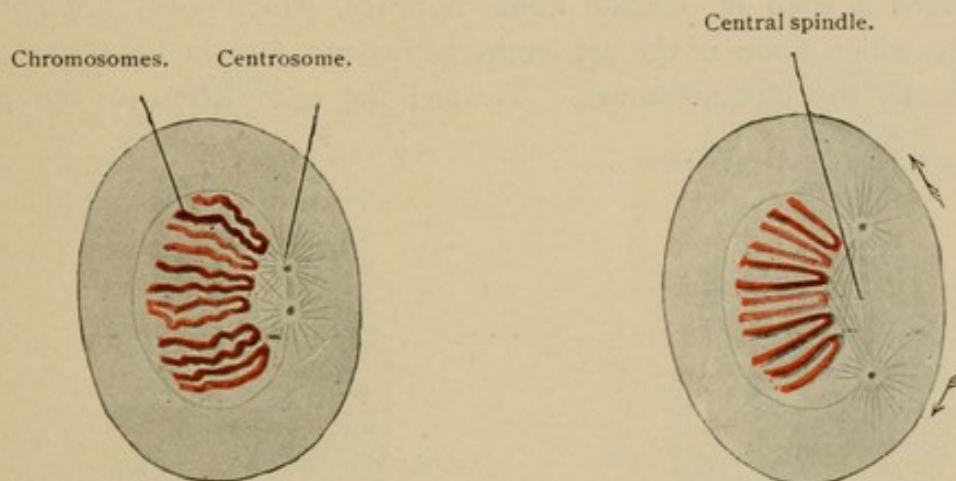


FIG. 7.—SCHEME OF THE CLOSE COIL.

FIG. 8.—SCHEME OF THE LOOSE COIL.

each centrosome (Fig. 7). Then the nucleus enlarges, the nuclear network becomes richer in chromatin, and its nuclein cords soon appear in the form of convoluted segments† (chromosomes), that are placed transversely to the long axis of the nucleus. The number of the chro-

* *μίτος*, the thread, because in this process threads are visible in the nucleus. There is a second mode of division, in which the nuclei divide simply by constriction, without the occurrence of a regular grouping of the nuclear structures. This is called *direct* or *amitotic division*. However, it is very probable that this kind of division in vertebrates, under normal conditions, has not the significance of a *physiologic* reproduction and multiplication of cells, but occurs only in cells which are degenerating—for very often the division of the protoplasm does not follow, so that only a multiplication of nuclei takes place. This frequently happens in leucocytes, also in epithelial cells—*e. g.*, in the superficial epithelial cells of the urinary bladder of young animals.

† These segments are present in many resting nuclei, but owing to the numerous lateral branches, by which they unite with their neighbors in a network, they are not easily distinguished. At the beginning of division the lateral branches are drawn in, whereby the segments become thicker and appear more distinct. In other nuclei the chromatin is disposed in a single thread, that later divides transversely into chromosomes.

mosomes is constant for each animal species, in man probably twenty-four.

The form of the chromosomes is usually that of a loop. The closed ends are directed toward the centrosomes, the *polar side* or *polar field*, the free ends toward the opposite pole of the cell. In this stage the chromosomes form a *close coil* (Fig. 7); but they soon grow thicker and less tortuous, thereby converting the close coil into the *loose coil* (Fig. 8). In the latter the curves of some of the loops can be detected pointing toward the opposite pole (*cf.* Fig. 14).

Meanwhile the two centrosomes, increasing mainly at the circumference, move apart and wander along the nuclear membrane, each to a point 90° distant from its original position. Between the retreating centrosomes a span of delicate fibers appears, which form the *central spindle*, to which fibers of the astrospheres become attached and can now be traced to the chromosomes. Toward the completion of the pro-

Polar radiation. Nuclear spindle.

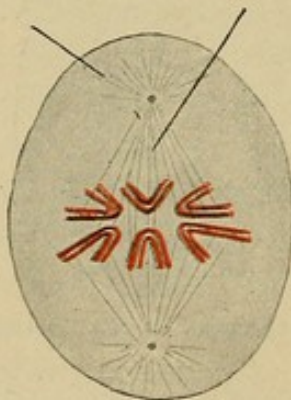


FIG. 9.—SCHEME OF THE MOTHER STAR.

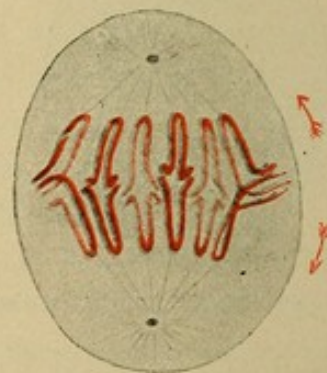


FIG. 10.—SCHEME OF METAKINESIS.

phase the nuclear membrane vanishes and the nucleolus becomes invisible.

(2) Metaphase.

The centrosomes have reached diametrically opposite points,* their fibrils, with which perhaps parts of the nuclear membrane are associated, extend to the chromosomes and now appear in the figure of a spindle, the *nuclear spindle*, at each apex of which lies a relatively very large centrosome encircled by the astrosphere, which in this stage is also known as the "polar radiation."† The chromatin loops move

* Up to this point this description of the behavior of the centrosome is not invariably applicable; for example, in *ascaris megalocephala univalens* the centrosome divides in the interior of the nucleus, which elongates and allows a centrosome to emerge at each end. With their exit the nuclear spindle is formed. In succeeding events the processes are identical.

† Remains of the central-spindle still lie in the axis of the nuclear-spindle.

to the equator of the spindle, in the future plane of division of the nucleus, and are soon arranged so that their closed ends are directed toward the axis of the spindle, their free ends toward the equator (Fig. 9). Viewed from an apex of the spindle this grouping of the segments has the appearance of a star, the *mother star* (monaster).

During the formation of the mother star, often earlier, in the first stages of the prophase, the chromatin loops divide longitudinally, so that each single loop forms two *sister loops*. This is followed by division of the nucleus exactly into halves, while by the contraction of the spindle fibers (?) the one sister loop is drawn to one pole, the other sister loop to the other pole of the nuclear spindle. This process is named *metakinesis* (Fig. 10); it is involved in a separation of the centrosomes from each other. In this stage the nuclear segments appear in the figure of

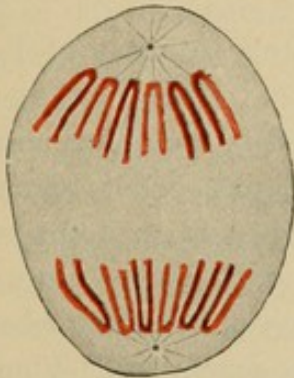


FIG. 11.—SCHEME OF THE DAUGHTER STARS.

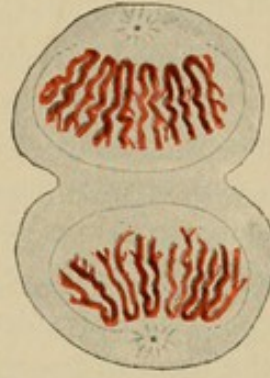


FIG. 12.—SCHEME OF CELL DIVISION.

two *daughter stars*, they form the *dyaster*. Each daughter star exhibits polarity (Fig. 11).

(3) Anaphase.

These relations are soon obliterated, inasmuch as the centrosome diminishes again, then duplicates itself, and the chromosomes thrust out lateral twigs for anastomosis with neighbor chromosomes and so reproduce the fabric of the resting nucleus. While the spindle and the greater portion of the polar radiation become invisible, and a new nuclear membrane appears, beginning opposite the "polar side," the nucleus swells by imbibition of nuclear sap, becomes spherical, and nucleoli appear; at the same time, at the equator of the cell, a division of the hitherto quiescent protoplasm begins (Fig. 12), which leads to complete separation into halves.

In rare cases, especially in those of a pathologic nature, the nucleus simultaneously divides in the mode of mitosis into more than two nuclei.

The duration of cell division, in which the progress of the indi-

vidual stages is not equally rapid, varies from a half hour (in man) * to five hours (in amphibia).

Special modifications of cell division are the so-called endogenous cell formation and budding. The *endogenous cell formation* occurs in cells that possess a firm envelop (egg-cells, cartilage cells). The process of division is precisely the same as that previously described, except that all the descendants of one cell (mother cell) arising by successive divisions (daughter and grand-daughter cells) remain inclosed in the common capsule (Fig. 44). Gemmation or *budding* indicates a kind of division in which the products are of unequal size, or in which the cell develops processes that are set free by constriction and become independent cells (see bone marrow).

The young cells always resemble in character the mother-cells. Such a case as connective-tissue cells arising from the division of an epithelial cell never occurs (*cf.* also p. 61).

The phenomena of secretion.—(See Secretory Activity of Epithelial Tissue.)

The *duration of life* in nearly all cells is brief; the old elements disintegrate, new ones appear in their places. Dying cells are characterized by decrease in the volume of both nucleus and protoplasm. The latter often seems to have an indented border or it stains more deeply, while the chromatin substance of the nucleus either diminishes or appears in the form of fragments that stain homogeneously. Vacuoles in the protoplasm or in the nucleus are signs of dying cells. Dying cells in abundance may be observed in epithelia, where formerly they were often regarded as peculiar kinds of cells (*cf.* also Fig. 26).

The *growth* of cells preeminently concerns the protoplasm and only exceptionally takes place equally in all directions, in which case the original form of the cell is retained (*e. g.*, egg-cell); as a rule, an unequal growth occurs. As a result of unequal growth the original form is altered; the cell becomes elongated, or flattened, or branched, etc. The majority of cells are soft and susceptible to change in form from mechanical influences; as, for example, the cylindric epithelial cells in the empty urinary bladder, which in the filled organ are low, flattened structures; or, the epithelial cells of the peritoneum, which by distention acquire three times their former superficies.

Secretory products of cells.—The secreted materials are either wholly removed (as most glandular secretions) or they become rigid and remain

* The disappearance of the mitotic figures in the human cadaver is not complete until after an elapse of forty-eight hours.

in contact with the cells. To the latter belong certain *intercellular substances*; many of these are a secretion of cells, others are produced by a transformation of the peripheral layers of the cell protoplasm, still others, by complete metamorphosis of the cells (some cells undergo degeneration in the development of the intercellular substance). It is very difficult to distinguish whether individual intercellular substances were formed by the one process or the other; many points in this matter are still the subject of lively controversy.

The intercellular substances occur either in small amount, as structureless, soft, perhaps fluid, *cement-substance*, between epithelial cells, connective-tissue cells, smooth muscle-fibers, etc.; or in large amounts, exceeding the mass of the cells, and then are called *ground-substance*. The ground-substances are either formless (homogeneous) or formed; in the latter case they are for the most part transformed into fibers or granules of different nature. The scanty remnants of formless substance found between the fibers or granules also are called cement-substance.

Union of cells.—Cells unite with one another either by contact (union by contiguity) or they extend into one another by means of shorter or longer processes (union by continuity). Such processes may possess delicate fibers, or fibrillæ, that extend through several cells, as for example, in stratified squamous epithelium and in the elements of nerve tissue. The independence, the possibility of territorial isolation of the individual cells is not impaired thereby. In other cases, however, cells originally distinct fuse into a common protoplasmic mass, a *syncytium*, in which then only the nuclei, often stationed at very irregular intervals, occasionally indicate the individual cell territories. The independence of the cells is thereby more or less sacrificed.

TECHNIC.

No. 1.—For the study of nuclear structure and karyokinesis amphibian larvæ are most suitable. Those most readily procured are the larvæ of the water-salamander, which in the months of June and July abound in every pool. Place *freshly caught* specimens, 3 to 4 cm. long, in about 100 c.c. of chromic-acetic acid (p. 32). After three hours place the larvæ in running water for eight hours and then in 70 per cent. alcohol. At the expiration of four hours, or later, the objects are ready for further treatment.

(a) *Nuclear structure.*—With a scalpel carefully scrape the epithelium from the skin of the abdomen, with two pairs of fine forceps strip off the thin corium, stain it for from one to three minutes in 5 c.c. of Hansen's hematoxylin (p. 38), and mount in xylol-balsam (p. 50). Between the round glands beautiful connective-tissue cells with large nuclei may be seen. The fibrillar structure of the protoplasm, the centrosome,

the astrosphere, and the finer structures of the nucleus can be recognized only by the employment of complicated methods and the highest magnification. The results obtained by ordinary methods are like those pictured in figure 13.

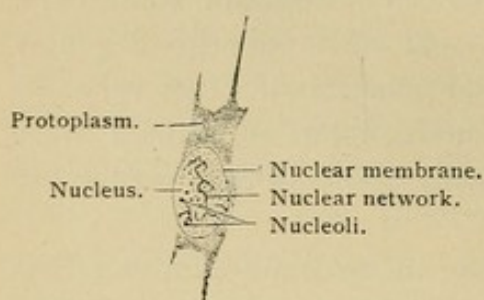


FIG. 13.—CONNECTIVE-TISSUE CELL FROM THE CORIUM OF TRITON TÆNIATUS. SURFACE VIEW. $\times 560$. Only the coarser filaments of the nuclear network can be distinctly seen; with this magnification the finer filaments appear as minute dots, the nucleoli as parts of the nuclear network.

The cross-striped muscles of the tail and the membranes of smooth muscle-fibers (the latter may be readily obtained by stripping off the muscularis of the intestine) also furnish beautiful slides.

(b) *Karyokinesis*.—With a pair of fine scissors cut round the margin of the cornea and strip off the same; stain and preserve like *a*. The preparation must be placed on the slide with the convex surface of the cornea upward; in the epithelium, even

with the low-power objective, many karyomitotic figures may be seen, which are revealed by their intense color; with stronger magnification pictures such as are represented in figure 14 can be seen. By this method

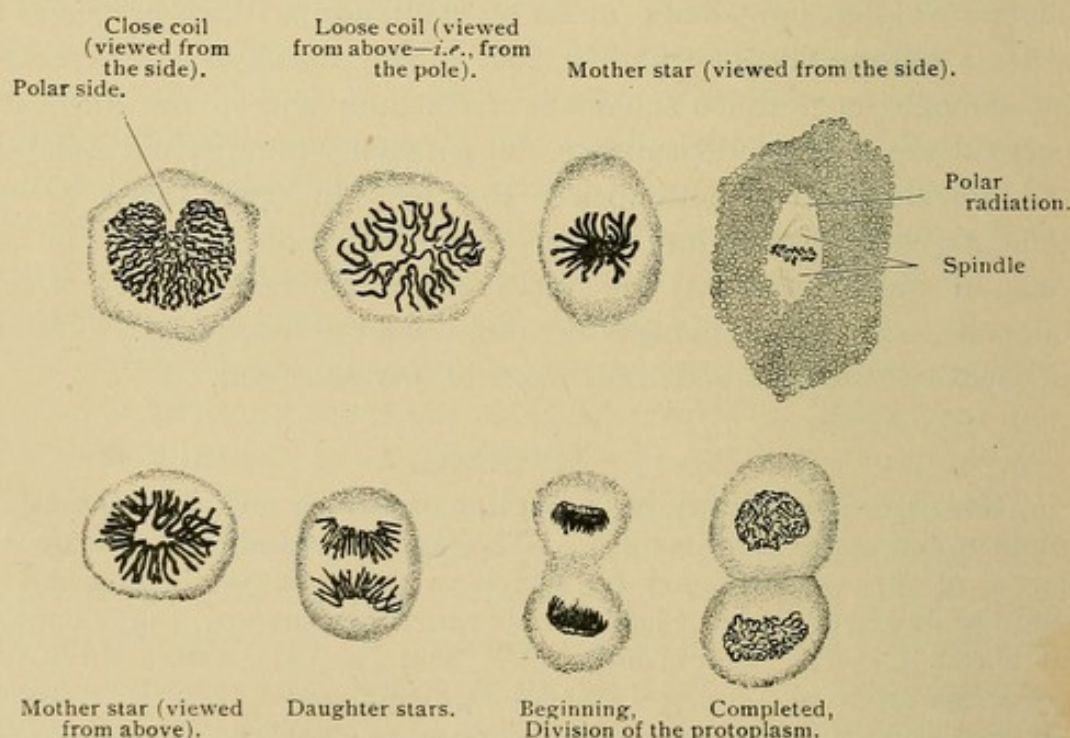


FIG. 14.—KARYOKINETIC FIGURES FROM A SURFACE PREPARATION OF THE EPITHELIUM OF THE ORAL CAVITY OF TRITON ALPESTRIS. $\times 560$.

the nuclear spindle and the polar radiation can be perceived only in especially favorable preparations—*e. g.*, eggs of siredon and of trout.

The centrosomes and first stages of spindle formation can only be seen with immersion lenses and in preparations made after technic No. 3 (p. 88).

The delicate lamellæ suspended from the convex side of the cartilaginous gill-arch, as well as the epithelium of the floor of the oral cavity, are highly suitable objects. Occasionally not a single karyokinetic figure is found. Isolated figures may sometimes be observed in preparation *a*.

B. THE TISSUES.

I. THE EPITHELIAL TISSUES.

The elements of epithelial tissue, the *epithelial cells*, are sharply defined cells consisting of protoplasm and nucleus. A cell membrane is frequently absent, often only a crusta is present (p. 66). The majority of epithelial cells are soft and plastic, yield readily to the pressure of surrounding elements, the result of which is great diversity of outline. In general two principal forms can be distinguished: the *flat* and the

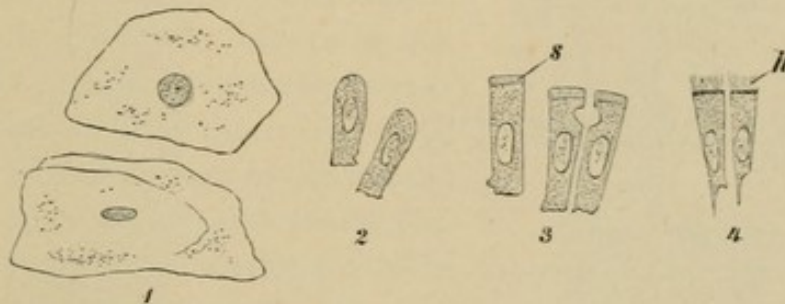


FIG. 15.—ISOLATED EPITHELIAL CELLS OF THE RABBIT. $\times 560$. 1. Squamous cells (mucous membrane of mouth). Technic No. 96. 2. Cylinder cells (corneal epithelium). 3. Cylinder cells, with cuticulae, *s* (intestinal epithelium). 4. Ciliated cells; *h*, cilia (bronchial epithelium). Technic on page 29, § 3 *a*.

cylindrical (better, prismatic). These extremes are united by numerous transitional forms.

The flat epithelial cells, *squamous epithelial cells*, *pavement epithelial cells*, rarely are symmetrical in form, excepting the pigmented epithelium of the retina, which consists of tolerably regular hexagonal cells (Fig. 16); generally the contour is very irregular.

The cylindrical epithelial cells, *cylinder cells*, seen from the side are elongated elements, the height of which considerably exceeds the breadth; seen from above they appear hexagonal; therefore they are in reality prismatic.

Cells that are as high as they are broad are called cubical epithelial cells.*

Many cylinder cells have a sometimes homogeneous, sometimes

* Such cells are frequently also called pavement cells.

striated border on their free upper surface * (Fig. 15, 3 s); it is a cuticula. Other cylinder cells are beset with delicate filamentous processes † (cilia) on their free surface, that during life are in constant active vibration to and fro in a definite direction. These are called *ciliated cells*.

The specially differentiated *sensory* or *neuro-epithelial cells* will be described in connection with the organs of special sense.

Continuous layers of epithelial cells, which cover outer and inner surfaces of the body, are called "epithelia." The epithelia are sometimes composed of a single stratum, sometimes of several strata, and accordingly the following varieties are distinguished:

1. *Simple* (one-layered) *squamous epithelium* (Fig. 16): in the pigmented layer of the retina, the alveoli of the lungs, the pericardium, the pleura, the peritoneum, the rete testis, the membranous



FIG. 16.—PIGMENTED EPITHELIUM OF THE RETINA OF MAN. Viewed from the surface. $\times 560$. Technic No. 180.



FIG. 17.—SIMPLE CYLINDER EPITHELIUM OF THE SMALL INTESTINE OF MAN. $\times 560$. c. Striated cuticular border. z. Cylinder cell. tp. Tunica propria. Technic like No. 110.



FIG. 18.—STRATIFIED SQUAMOUS EPITHELIUM OF THE LARYNX OF MAN. $\times 240$. 1. Cylinder cells. 2. Polygonal cells. 3. Squamous cells. Technic No. 128.

labyrinth, also the epithelium of the articular cavities, of the tendon sheaths, of the endocardium, the blood- and lymph-vessels.‡

* The striae are the boundaries of minute rods (Fig. 17, c) that occasionally can be distinctly seen even with medium magnification; between them processes of the protoplasm, "pseudopodia," the length of which often varies greatly, can extend toward the free surface. Such pseudopodia can be seen also on the epithelial cells of the human large intestine (Fig. 4). The so-called *brushborder* of the renal epithelium is likewise constructed of minute rods, that differ from similar formations only in their greater delicacy; whether this border belongs to the cuticular formations is questionable, for the assumed constancy of the structure is disputed (see the chapter on the kidneys).

† Very high magnifications show that each cilium is in relation with a granule, the "basal corpuscle," lying close to the free surface, which perhaps is the center of motion for the cilium.

‡ The last-named five epithelia are also called *endothelia*, their elements, *endothelial cells*. In normal anatomy these names are superfluous. It has recently been suggested to designate at least the epithelium of the blood- and lymph-vessels as endothelium.

With these is enumerated the epithelium formed of a single layer of cubical cells, occurring in the plexus chorioidei, on the inner surface of the capsule of the lens, in the thyroid gland, and in the majority of other glands.

2. *Simple cylinder epithelium* (Fig. 17): in the intestinal canal and in the ducts of many glands.

3. *Simple ciliated epithelium* (Fig. 15, 4): in the smallest bronchi, in the uterus and oviducts, in the accessory spaces of the nasal fossæ, in the central canal of the spinal cord.

4. *Stratified* (many-layered) *squamous epithelium* (Fig. 18): not all the elements of this variety are squamous cells; the lowermost stratum is composed of cylinder cells; superposed on this are several strata of very differently shaped cells, mainly irregularly polygonal, over which lie successive strata of cells that, as they approach the surface, become progressively thinner and flatter. The stratified squamous epithelium occurs in the mouth, in the pharynx, in the esophagus, on the vocal cords, on the ocular conjunctiva, in the vagina, and in the female urethra. The epidermis also consists of a stratified squamous epithelium, which is characterized by the cornification of the cells of the superficial strata, which are transformed into horny scales without nuclei. Cornified cells are also found on the hairs and nails, but in these situations they are nucleated.

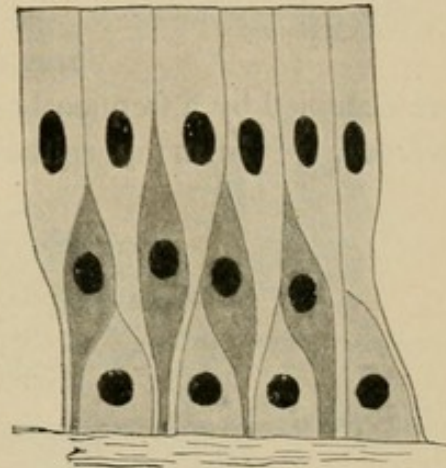


FIG. 19.—SCHEME OF A MANY-ROWED EPITHELIUM.

Stratified cylindric and stratified ciliated epithelium are also recognized, although it has been shown, particularly in sections, that this stratification is simulated by the arrangement of the nuclei of the cells at different levels in several transverse rows; the cells themselves all rest upon the connective-tissue base, but do not all extend to the free surface (Fig. 19). Such epithelium is accordingly one-layered and is distinguished from the ordinary "simple" epithelium, in which the nuclei stand in *one* row—"single row"—as *many-rowed* (multi-lineal) epithelium. Doubtless the majority of the stratified cylinder and ciliated epithelia hitherto described are merely "many-rowed." Accordingly we recognize the following:

5. *Stratified* (possibly many-rowed) *cylinder epithelium*: in man is found only on the conjunctiva palpebrarum, in the main excretory ducts

of certain glands, and in a division of the male urethra. The arrangement of the strata is similar to that of—



FIG. 20.—MANY-ROWED CILIATED EPITHELIUM. $\times 560$. From the respiratory nasal mucous membrane of man. 1. Oval cells. 2. Spindle-shaped cells. 3. Cylinder cells. 4. Connective tissue. Technic No. 200.

6. *Stratified ciliated epithelium*: only the most superficial cells are cylindric and carry cilia; in the deepest layers the elements are mainly spherical, in the middle layers spindle-shaped (Fig. 20). Stratified ciliated epithelium is said to occur in the larynx, in the upper portion of the pharynx, and in the eustachian tube; probably it is only many-rowed, like the epithelium of the nasal fossæ, the trachea, the large bronchi, and the epididymis, in which all cells actually reach to the connective tissue.

Between the epithelial cells extremely narrow clefts often occur, *intercellular spaces*, which are occupied by a frequently very scanty, soft, perhaps fluid, *intercellular substance*.* In many epithelia (in the cylinder epithelia of the mucous

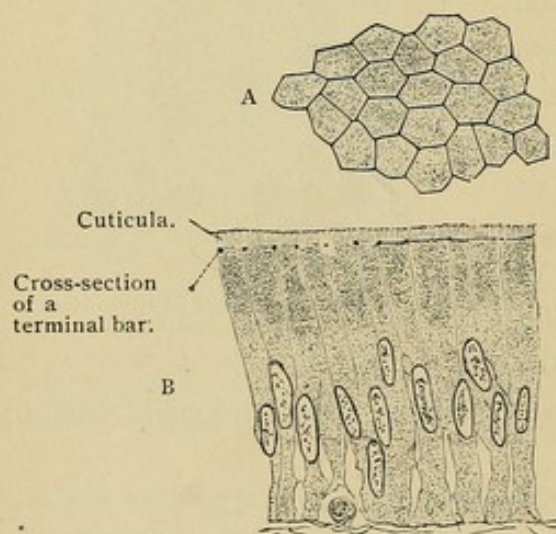


FIG. 21.—CYLINDER EPITHELIUM OF AN INTESTINAL VILLUS OF MAN. Magnified about 600 times. Network of terminal bars: A, view of free surface; B, lateral aspect; on the left the cross-sections, on the right the lateral surfaces of the terminal bars are seen. Technic No. 3.

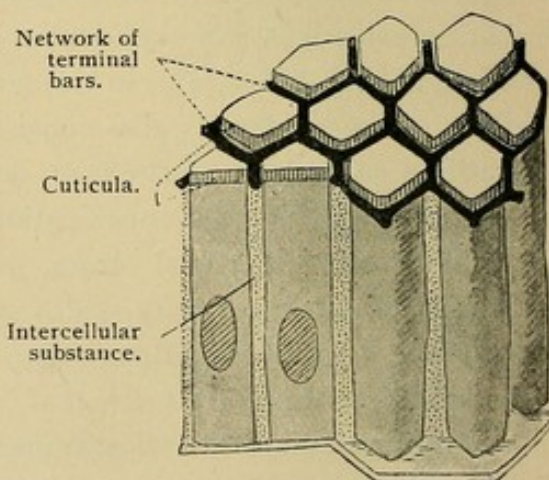


FIG. 22.—SCHEME OF THE NETWORK OF TERMINAL BARS. The two cells on the left are divided lengthwise into halves; the two on the right are drawn as complete cylinders or prisms.

membranes and in the majority of the glandular epithelia, also in the stratified epithelium of the mucous membrane of the tongue and in the transitional epithelium of the urinary organs) the intercellular spaces are

* Because in the human skin these intercellular spaces have been successfully injected through the lymph-vessels, it was believed that this substance is identical with ordinary lymph. However, this is not correct, for the intercellular substance of epithelium reacts differently; it becomes black when treated with silver nitrate.

closed toward the free surface by very delicate bars of a peculiar cement-substance; since these bars, "*terminal bars*" (*Schlussleisten*), are connected with one another they form a "*network of terminal bars*" (*Schlussleistennetz*), in the meshes of which the ends of the epithelial cells directed toward the free surface are inserted.

The union of the epithelial cells is effected in such a manner that either they present smooth surfaces of contact to one another,—namely by the intervention of intercellular substance,—or they interlock by variously shaped processes, the latter being pressure-effects. The delicate spines and thorns visible on the surfaces of many epithelial cells have been regarded as similar processes. But these are connecting bridges,* often cord- and net-like, which pierce the intercellular substance and establish an intimate union with neighbor epithelial cells. Cells provided with such thorns and ridges are called *prickle-cells*; the processes are aptly designated by the appropriate name of *intercellular bridges* (Fig. 23). They were first seen on the polygonal cells of stratified squamous epithelium,† but they also occur on the cells of simple squamous and simple cylinder epithelium,—for example, of the stomach and of the intestines,—but there they are extremely delicate and can be demonstrated only by the application of special methods. The length of the intercellular bridges and the diameter of the "*intercellular clefts*" occurring between them vary greatly in the different forms of epithelium and in the different physiologic states of the tissue.‡

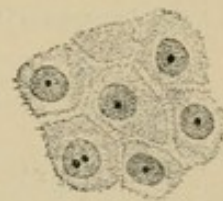


FIG. 23.—FROM A VERTICAL SECTION OF THE STRATUM GERMINATIVUM OF THE EPIDERMIS. $\times 560$. Seven squamous epithelial cells united by intercellular bridges. Technic like No. 89.

Epithelium possesses no blood-§ and lymph-vessels, but nerves are found in some situations, for example, in the epithelium of the skin and of many mucous membranes.

* These bridges contain fibrils, that can be traced in the interior of the cells (the *filarmass*, p. 63), and are the ground on which such epithelium was said to have a "*fibrillar*" structure, a description that can only lead to perplexity, because, for example, it tends to produce confusion with the fibrillar structure of connective tissue, which is something wholly different.

† The basal surfaces of the cylinder cells of stratified squamous epithelium are also provided with short processes, directed toward the subjacent connective tissue, the "*rivet-fibers*" (*Haftfasern*), that can be made visible only by means of complicated methods.

‡ In fresh, living tissues (e. g., in the tail of amphibian larvæ) the intercellular spaces can scarcely be seen, but in certain conditions dependent on disturbance of the circulation of lymph become more distinct. Then the intercellular spaces appear as tiny vacuoles in the hyaline border stratum of each epithelial cell. The thicker the stratified epithelium is, the wider are the intercellular spaces, the longer are the intercellular bridges; which on the one hand elucidates the importance of the spaces in the nutrition of the epithelium, on the other hand furnishes the explanation of the slight size of the spaces and bridges in the simple epithelia.

§ See also the Urinary Organs.

SECRETORY ACTIVITY OF EPITHELIAL TISSUE.

Many epithelial cells possess the faculty of secreting and discharging substances which are not used in the growth and repair of the tissues. Such cells are called *gland-cells*. The secreted substances are either stored in the body (secretions) or, being of no further use, removed from the body (excretions). The performance of the processes of elaboration and discharge of secretions (or excretions) is manifested by certain changes in the form and contents of the gland-cell, which

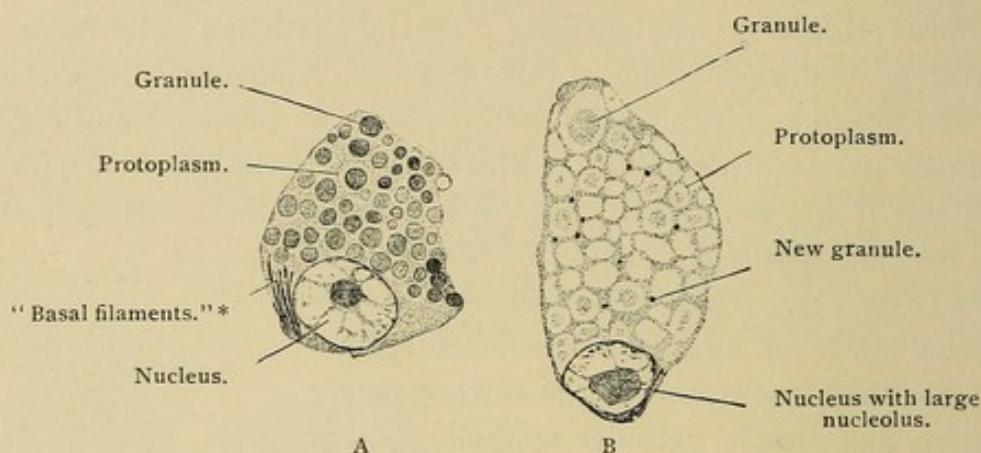


FIG. 24.—TWO SEROUS GLAND-CELLS FROM THE SUBMAXILLARY GLAND OF A GUINEA-PIG. $\times 1260$. In cell B the granules have passed into the unstainable state; new stainable granules are beginning to develop in the protoplasm. Technic No. 120.

indicate the empty and the loaded $\dagger$ condition, states of rest and activity respectively. In many cells, for example, in the serous gland-cells, the

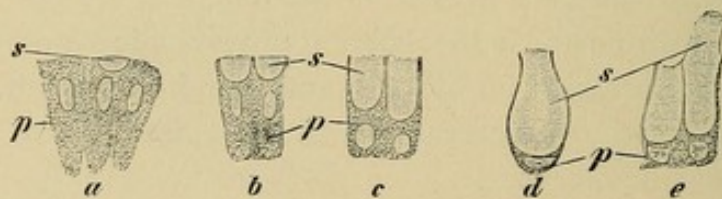


FIG. 25.—SECRETING EPITHELIAL CELLS. From a thin section of the mucous membrane of the stomach of man. $\times 560$. *p*. Protoplasm. *s*. Secretion. *a*. Two empty cells; the cell between them shows beginning mucoid metamorphosis. *c*. The cell on the right is discharging its contents; the granular protoplasm has increased and the nucleus has become round again. Technic No. 108.

empty state, barring certain phenomena of the nucleus (p. 81), is manifested by a smaller volume and a darker appearance of the cell; higher

* The basal filaments occurring in the serous gland-cells and in the chief cells of the gastric glands may be portions of the filar substance, "ergastoplasm," or possibly they too are concerned in the production of the secretion.

$\dagger$ The terms "empty" and "loaded" relate to the *finished* secretion and its impending discharge, not to the precursory stages; other authors employ instead the expressions "resting" and "active."

objectives and special methods reveal granules* that stain intensely (Fig. 24 A). These "granula" grow, lose the faculty of staining (Fig. 24 B), and become transmuted into drops of secretion; the cell thereby passes into the loaded state, which may be demonstrated also by simple methods and is indicated by an increased volume and a clearer appearance. The drops of secretion, occasionally even granules, are discharged into the lumen of the gland. In other gland-cells, for example, in many mucous gland-cells, the elaboration of secretion is likewise initially associated with granules, which, however, soon become transformed into a transparent mass, the mucus (Fig. 25, *s*) that accumulates at the "collecting center," situated on the side of the cell adjacent to the lumen or to the free surface, and is more or less sharply defined against the still unaltered protoplasm (*b*, *p*).† As the process of secretion advances, larger and larger masses of the protoplasm are converted into secretion and the nucleus and remnant of unaltered protoplasm are crowded to the base of the cell; as a consequence of this compression the oval nucleus (*a*, *b*) gradually becomes round (*c*) or even flat (*d*). The volume of the cell when filled with secretion is considerably increased. Finally, the secretion gradually escapes; simultaneously the protoplasm is regenerated and the nucleus moves upward to its original position, and restore to the cell, now diminished in size, the appearance of the empty state.

The majority of gland-cells do not degenerate in the act of secretion, but are able to repeat the process again and again. The sebaceous glands furnish an exception, for their secretion is formed by the disintegration of cells, like the goblet-cells.‡ In the latter, in one-layered epithelium, the processes of elaboration and expulsion of secretion occur simultaneously (Fig. 26); at first the secretion is produced more rapidly than it is discharged and it accumulates in the cell (2), but finally expulsion exceeds production, the cell gradually empties itself completely and dies (4). In stratified or many-rowed epithelium the formation of secretion begins in the young goblet-cells in the depths of the glands; the expulsion does not occur until the elements have matured and reached the free surface.

* These are not the microsomes (p. 63) stainable by certain methods, *e. g.* Altmann's methods.

† The collecting center by no means consists only of secretion; between the masses of the latter there is a delicate net or framework of protoplasm, that also encloses the centrosome.

‡ The testicle and the ovary afford a peculiar instance, the gland-cells of which after discharge of their secretion undergo further development.

The gland-cells lie isolated between other epithelial cells* or are united in groups and form *glandular tissue*.

THE GLANDS.†

The glands, *glandulæ*, are glandular tissue buried beneath the surface of the body, which either has the form of cylinders, *tubuli*, or pouched sacs, *alveoli*.

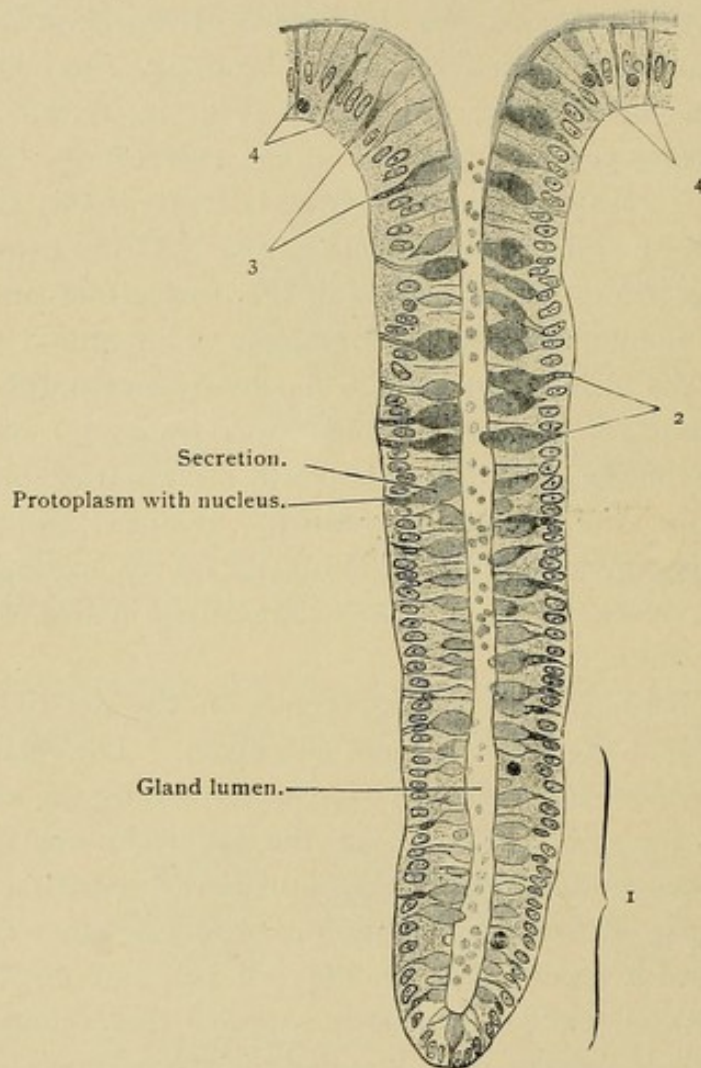


FIG. 26.—CRYPT OF LIEBERKÜHN FROM A SECTION OF THE LARGE INTESTINE OF MAN. $\times 165$. The secretion formed in the goblet-cells is deeply stained. In zone 1 the goblet-cells show the beginning of secretion; that expulsion has begun is evident from the presence of drops of secretion in the lumen of the crypt. 2. Goblet-cells with much secretion. 3. Goblet-cells containing less secretion. 4. Dying goblet-cells, some of which still contain remnants of secretion. Technic No. 112.

The structure of glands, in particular of their secreting divisions, is easy to recognize only when they present simple or but slightly branched tubuli or

* They are then called unicellular glands; they are very common among invertebrates, also occur in man as goblet-cells (see The Digestive Organs).

† The glands consist almost exclusively of epithelium; supporting tissue and blood-vessels, important as the latter are in a physiologic respect, morphologically are relatively subordinate. This furnishes the justification for describing the glands, which of course are organs, as the conclusion to the epithelial tissues.

alveoli. The majority of glands are profusely branched, twisted and coiled to a dense ball, that can scarcely be unravelled. Sections of such balls exhibit clusters of vesicles ("acini"), that may be equally as well taken for alveoli as for cross-sections of tubuli. This explains the antagonistic statements of individual authors. Hitherto the only dependable method for the exhibition of the gland lumen was either that by injection or by impregnation of the secretion after Golgi. If the lumen appeared in the shape of a branched line of uniform thickness, it was inferred that the form was tubular; if the lumen exhibited terminal or lateral evaginations the structure was said to be alveolar. Hitherto I also made my classification according to this principle. It was of course incomplete, since it did not include any consideration of the varying thickness of the wall. Finally the plate reconstruction method has led to recent success in representing the entire structure (lumen and wall) on an enlarged scale. It may be that in details corrections must still be made; however the necessity for a new classification is already evident and is given in the following.

There are two principal forms of glands, the tubular and the alveolar* glands. Between these two there is a transitional form originating from the tubular and represented by the alveolo-tubular glands. All three forms occur either individually independent or united in groups; they are accordingly classified as simple and compound glands.

A. Tubular glands.

1. *Simple tubular glands*, which have the form of either a simple or a branched tube (Fig. 27); the latter form may be named a tubular system.

2. *Compound tubular glands*, which consist of a variable large number of tubular systems (Fig. 27).

Unbranched simple tubular glands are the following: some of the fundus glands of the stomach; the majority of the coil and ceruminous glands, and the intestinal (Lieberkühn's) glands (regarding the latter see the chapter on the intestines).

Branched simple tubular glands are the following: some of the fundus glands, a few of the coil glands, and the glands of the uterus.

Compound tubular glands are the serous glands of the tongue, the serous divisions of the small glands of the respiratory apparatus (and the small glands of the oral cavity?), and the tear glands. Also the

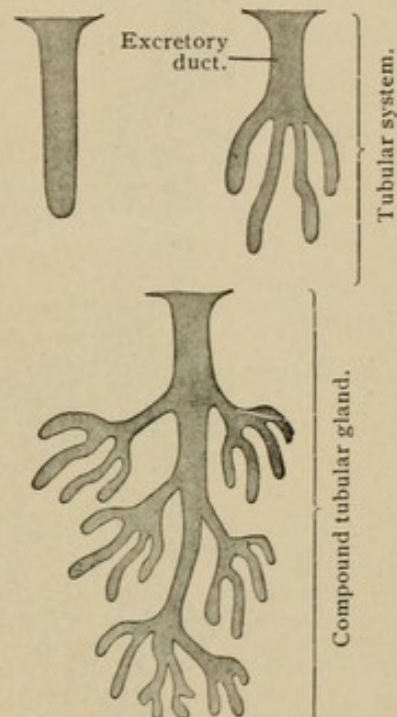


FIG. 27.—SCHEMES OF TUBULAR GLAND FORMS.

* From alveus, a pouched sac.

kidneys, the testicles, and the liver. The ramifications of the latter two glands anastomose regularly with one another and form nets; therefore the testicles and the liver are also called "reticular glands." Isolated anastomoses between glands have been observed in the fundus glands of the horse and in the serous lingual glands and the bulbourethral glands of man.

B. *Alveolo-tubular glands.*

1. *Simple alveolo-tubular glands* appear to occur only in the form of branched ducts; they form an alveolar-tubular system (Fig. 28).

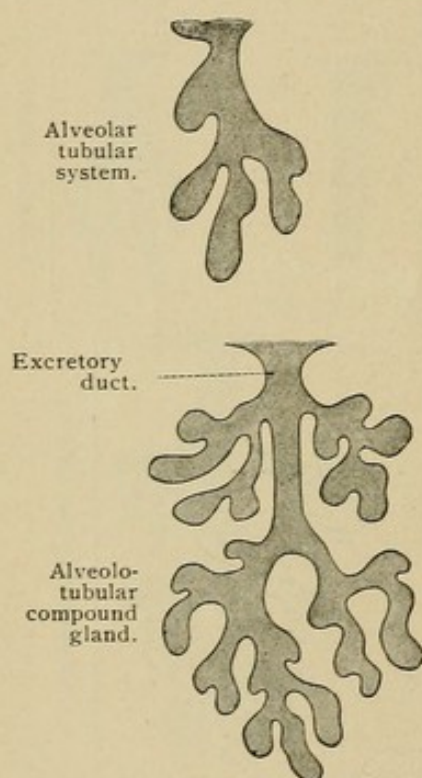


FIG. 28.—SCHEMES OF ALVEOLO-TUBULAR GLAND FORMS.

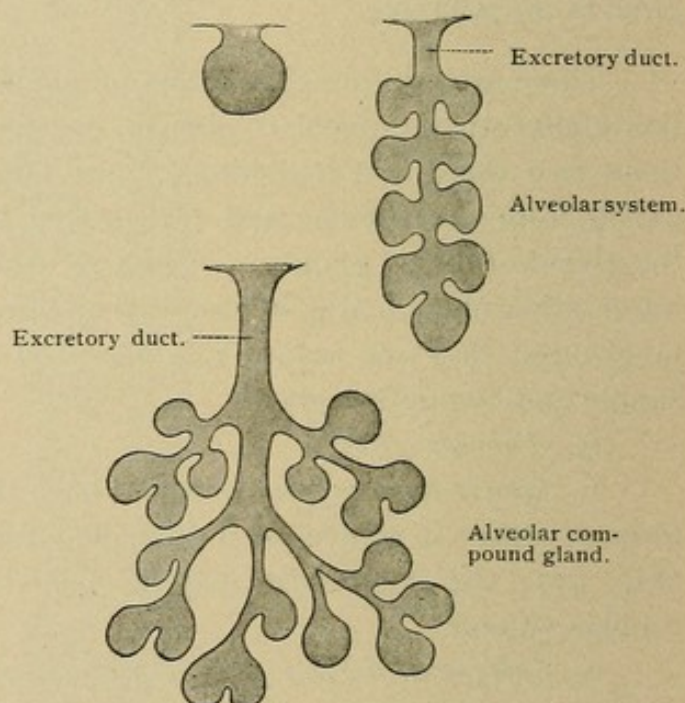


FIG. 29.—SCHEMES OF ALVEOLAR GLAND FORMS.

2. *Compound alveolo-tubular glands*, which consist of several alveolar-tubular systems (Fig. 28).

Branched simple alveolo-tubular glands are the pylorus glands, the urethral glands, and the small mucous glands of the tongue, the gums, and the esophagus.

Compound alveolo-tubular glands are the larger mucous glands, the sublingual gland, the mucous divisions of the submaxillary gland, the glands of the respiratory apparatus and the oral cavity, the duodenal glands, the bulbourethral glands (vestibulares majores?), the prostate, the lungs, and the mammary glands.

C. Alveolar glands.

1. *Simple alveolar glands* are simple or branched pouched sacs possessing an excretory duct (Fig. 29); the branched form is called an alveolar system.

2. *Compound alveolar glands*, which consist of several alveolar systems (Fig. 29).

Unbranched simple alveolar glands are the smallest sebaceous glands.

Branched simple alveolar glands are the larger sebaceous glands and the tarsal (Meibomian) glands.

Compound alveolar glands are some divisions of the parotid gland, the serous divisions of the submaxillary gland (the smallest glands of the oral cavity?), and the pancreas. In all the compound alveolar glands slender follicles, partly provided with evaginations, are to be found, so that the *entire* glands are to be annexed to the alveolo-tubular glands, with the reservation that they are distinguished from the alveolo-tubular forms by the predominance of the alveolar type.

In the majority of glands, particularly in those visible to the naked eye, a sheath is formed by the surrounding connective tissue, which sends septa into the gland and divides it into complexes of varying size, the *gland lobules*. The septa are the carriers of the larger blood-vessels and nerves. The glands may secrete throughout their entire extent, but usually only that part lying near the blind end, the *gland body*, is specialized for this purpose, while the part forming the connection with the surface serves for the conveyance of the secretion, and is called *excretory duct*.

Glands *without* excretory ducts are the *thyroid* and the *ovary*. The former has an excretory duct in the embryonal period, which disappears in the course of development; the tubuli also suffer change. The gland vesicles ("follicles") of the ovary in an embryonal period also were in connection with the superficial epithelium. These connections, which might be called excretory ducts, disappear, and the expulsion of the products formed in the ovary (the ova) takes place by the bursting of the vesicle. The ovary is a *dehiscent* gland.

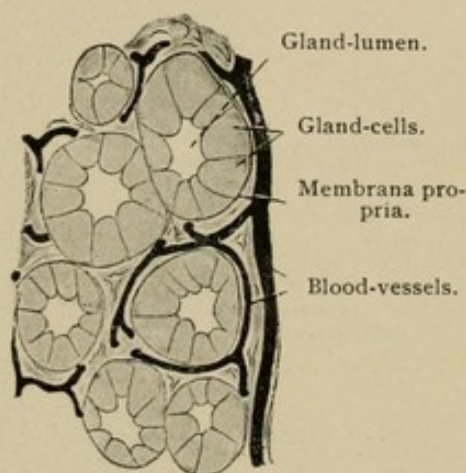


FIG. 30.—SECTION OF A MUCOUS GLAND OF THE TONGUE OF A RABBIT. Blood-vessels injected. The nuclei of the gland-cells were indistinct. $\times 180$. Technic like No. 125 b.

All gland tubuli and alveoli consist of a usually simple layer of gland-cells, which encircle the lumen of the gland and are in turn surrounded by a special modification of the connective tissue, the *membrana propria* or *basement-membrane** (see p. 94). On the outer side of the basement-membrane lie the blood-vessels (Fig. 30). Hence the gland-cells are inserted between the blood-vessels and the lumen of the gland, and on the peripheral side receive from the blood-vessels (or from the lymph clefts encircling the latter) the necessary materials for the formation of secretion and on the other (central or lumen) side discharge the elaborated product.

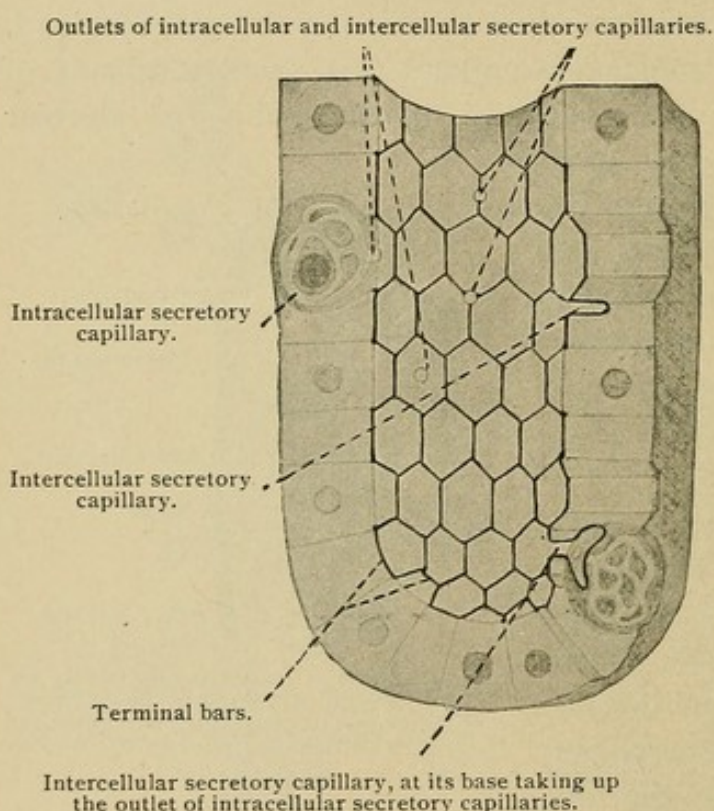


FIG. 31.—SCHEMATIC MODEL OF A HUMAN FUNDUS GLAND.

In many glands the axial (central) lumen sends off delicate lateral branches, *secretory capillaries*, that are situated sometimes between the gland-cells, "intercellular secretory capillaries," sometimes in the interior of the gland-cells, "intracellular secretory capillaries."† They can be made visible only by special methods and then appear in the form of sometimes simple, sometimes branched, or even netlike, anastomosing, tubules, that do not extend to the *membrana*

propria and the blood-vessels, but are separated from these by at least a portion of a gland-cell.

* Occasionally stellate cells occur between the *propria* and the gland-cells, which unite with one another and as "basket cells" embrace the gland tubuli. It is not yet decided whether they are epithelial or connective tissue cells or smooth muscle-fibers.

† It was for a long time very difficult to prove whether secretory capillaries were situated between or within the cells. Now we possess points of corroboration which make the distinction possible. Intercellular capillaries in cross-section are bordered by at least two terminal bars in cross-section, while in longitudinal section the terminal bars are seen running alongside the walls of the capillaries (Fig. 31). Further, intercellular capillaries are recognized by their sharp outline, which is due to a thickening of the exoplasm (p. 63). The intracellular capillaries lack this sharp outline, also any recognizable relation to terminal bars.

The intercellular secretory capillaries occur in the serous glands of the tongue, in the parotid, in the serous divisions of the submaxillary, the sublingual and related glands, in the bulbourethral glands, in the lacrimal glands, and in the pyloric glands. Intercellular and intracellular secretory capillaries occur side by side in the coil-glands, in the liver, and in the gastric glands. It is probable that the intracellular secretory capillaries are merely transient formations.

Secretory capillaries appear to be wanting in the pure mucous glands, in the mucous divisions of the mixed glands, in the intestinal, duodenal, and uterine glands, in the thyroid, the hypophysis, and the kidney.

The microscopic appearance of the gland-cells changes with their periodic functional condition. In some glands all the cells simultaneously exhibit the same functional appearance. In other glands different functional states are encountered at the same time, even within the same tubule or alveolus. The latter is the case in many mucous glands, in which the loaded cells crowd the empty cells more or less completely away from the lumen of the gland (see also the chapter on the glands of the oral cavity). The nuclei of many gland-cells also exhibit varying appearances corresponding to the changing functional state; often in empty cells the nucleus exhibits a delicate chromatin network and a conspicuous nucleolus, while in loaded cells the nucleolus is invisible and the chromatin cords appear in the form of coarse fragments.*

The finer ramifications of the excretory ducts of some glands, which are particularly conspicuous because of the form and structure of their epithelial cells, must be regarded as belonging to the gland body. These ramifications are not merely excretory ducts, but on them also devolves the function of excreting certain materials (salts); accordingly they belong to the secreting divisions of the glands. The difference in the structure of these branches renders their division into two parts

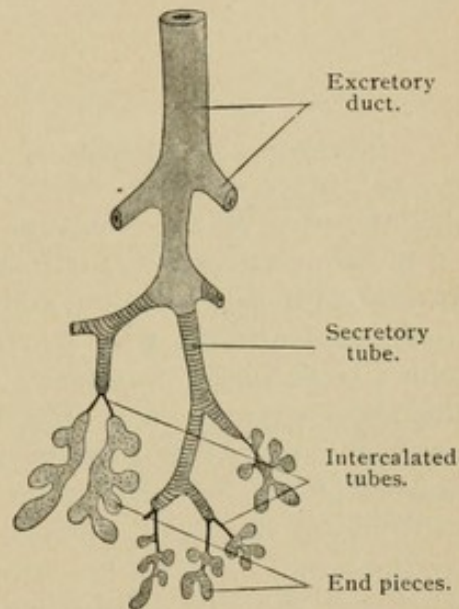


FIG. 32. Schematic drawing of the different divisions of a gland. (Submaxillary of man.)

* There is no doubt that portions of the nucleus, in the form of stainable granules, pass into the protoplasm, but whether these portions may be regarded as true secretion granules is questionable; the more so since such phenomena may be observed in other cells, for example, in the cells of the spinal ganglia.

desirable : the first portion, proceeding from the *terminal pieces*,* is narrow and lined sometimes with flat, sometimes with cubical cells ; it is called the *intercalated tubule*. The adjoining portion is wider and clothed with tall cylinder cells, the bases of which show distinct longitudinal striation, formed by rows of granules ; it is called the *secretory (salivary or mucous) tube*. The relative length of the intercalated and secretory tubes varies greatly in individual glands.

The *excretory ducts* consist of a simple or stratified cylinder epithelium and a sheath of connective tissue mingled with elastic fibers.

Accordingly in the most complicated cases the gland consists of the following divisions : (1) The excretory duct, which dividing passes into (2), the secretory tubes, which continue in (3), the intercalated portions, which lead into (4), the terminal pieces, the axial lumen of which receives (5), the secretory capillaries.

TECHNIC.

No. 2.—Living *ciliated cells* are obtained as follows : Kill a frog (p. 28), place it on its back, and with scissors cut off the lower jaw, so that the roof of the cavity of the mouth is exposed. From the mucosa of the roof cut out a small strip about 5 mm. long, place it on the slide in a drop of salt solution, and apply a cover-glass. With low magnification the beginner will scarcely perceive anything, unless currents in which large blood corpuscles are suspended lead him to the right place ; therefore examine with the high power and search the edges of the preparation. At first the movement of the cilia is so lively that the observer cannot see individual cilia ; the entire ciliated border waves ; the picture has been aptly compared to that of a corn-field swayed by the wind. After a few moments the rapidity of the movement diminishes and the cilia can be plainly seen. If the movement ceases, it can be restored by the application of a drop of concentrated potash solution (p. 23) ; the effect is transient, so that the eye of the observer must not be removed from the ocular while the fluid passes under the cover-glass. The addition of water soon suspends the movement.

No. 3.—*Terminal bars*.—Fix small pieces of intestine, from 0.5 to 1 cm. long, in Flemming's mixture (p. 34), or in sublimate-salt solution (p. 35), and harden in alcohols of gradually increased strength (p. 35) ; embed in paraffin, cut thin sections (about 10 μ) on the microtome, and fasten them to the slide (see Microtome Technic). Stain after Heidenhain's iron-hematoxylin method (p. 44), and mount in xylol-balsam. The bars can be distinguished as black streaks or dots, even with good dry systems (Fig. 21). With immersion systems the centrosomes can be seen in such preparations, but only the practiced microscopist can succeed in finding them.

* This is the designation of the blind ends of the gland ducts, which take up the secretory capillaries.

II. THE SUPPORTING TISSUES.

In the epithelial tissues the cells constitute the principal mass, but in the supporting tissues they are secondary, while the *intercellular substance* (ground substance, matrix) is conspicuously developed and variously differentiated. The preponderance of the intercellular substance, which also functionally plays the more important part, is characteristic of the group of supporting tissues. According to the nature of the intercellular substance they are divided into : (1) connective tissue ; (2) cartilage ; (3) bone.

I. CONNECTIVE TISSUE.

The matrix or intercellular substance of connective tissue is more or less soft ; the cells are few in number. Several varieties are distinguished : (a) mucous connective tissue, (b) fibrillar connective tissue, and (c) reticular connective tissue.

(a) *Mucous connective tissue* consists of round or stellate branched cells and a large quantity of undifferentiated, muciferous intercellular substance containing delicate connective-tissue bundles (see below). In the higher animals it is found only in the umbilical cord of very young embryos, but it is widely distributed in many lower animals.*

(b) *Fibrillar connective tissue* consists of abundant intercellular substance and of cells.

The intercellular substance consists of connective-tissue fibrillæ (connective-tissue fibers),† exquisitely fine filaments (0.6μ), which are united by a small quantity of homogeneous cement substance into bundles of varying thickness, the *connective-tissue bundles*. These bundles are soft, flexible, slightly extensible, are characterized by their pale contours, their longitudinal striation, their wavy course,‡ and by their chemical properties. On treatment with picric acid they separate into their fibrils, swell on the addition of dilute acids, *e. g.*, acetic acid,

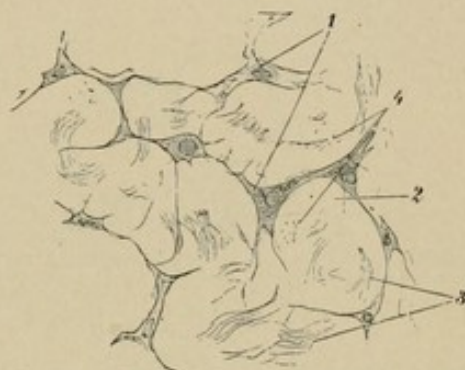


FIG. 33.—FROM A CROSS-SECTION OF THE UMBILICAL CORD OF A HUMAN EMBRYO ABOUT FOUR MONTHS OLD. $\times 240$. 1. Cells. 2. Intercellular substance. 3. Connective-tissue bundles mostly in oblique section, at 4 in true cross-section. Technic No. 4.

* Regarding the vitreous body, which some authors hold consists of mucous tissue, see the chapter on the vitreous body.

† Here *fibrillæ* and *fibers* are synonymous, while in the striated elements of muscle tissue a number of fibrillæ form a fiber.

‡ Hence the name *wavy* or *curly* connective tissue.

and become completely transparent, are destroyed by alkaline fluids, and on boiling yield *glutin*. The substance of the glutin-yielding connective tissue is called *collagen*.

According to one view the first connective-tissue fibrils originate in the *interior* of the cell; on another theory they arise *external* to the cell and in the latter case are a metamorphosis of the ground substance.*

The ground substance of fibrillar connective tissue invariably contains *elastic fibers*, but in fluctuating quantity (Fig. 35). They are characterized by their sharp, dark outlines, their strong refractive power, and—in contrast with the connective-tissue bundles—their extraordinary

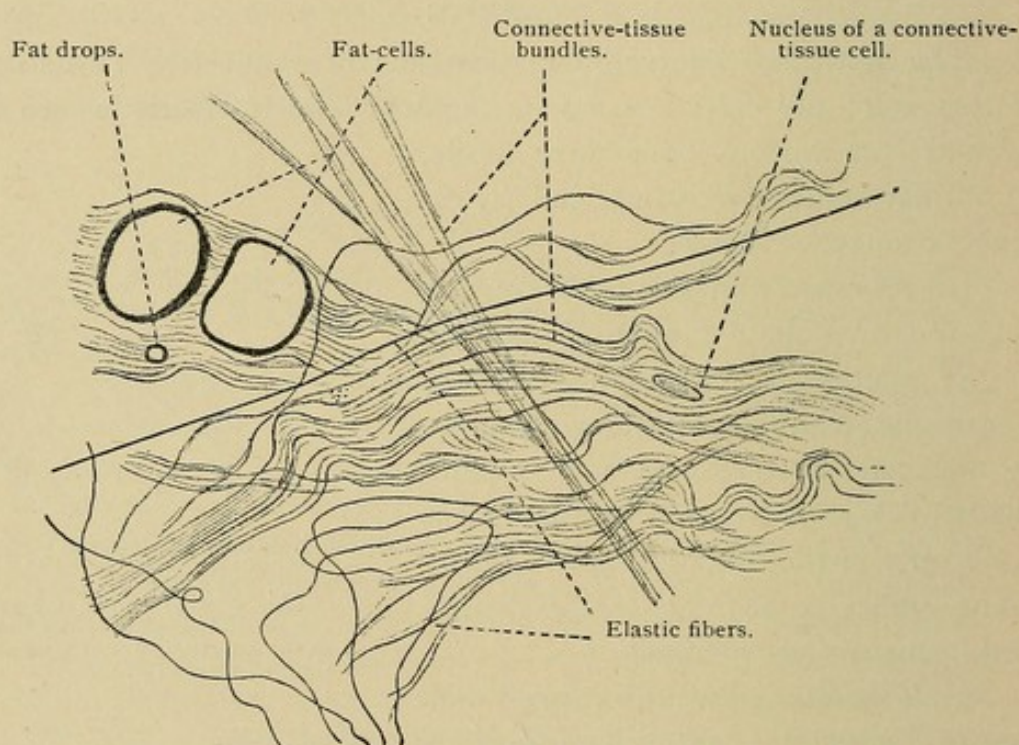


FIG. 34.—CONNECTIVE-TISSUE BUNDLES OF DIFFERENT THICKNESSES FROM THE INTERMUSCULAR CONNECTIVE TISSUE OF MAN. $\times 320$. Technic No. 5.

resistance to acids and alkalies. The substance of the elastic fibers is named *elastin*.† The elastic fibers vary from immeasurably fine to $11\ \mu$, and usually occur in the form of finer or coarser networks, the meshes of which are sometimes narrow, sometimes large.

* Flemming holds that a fibril-containing stratum is formed in the peripheral portion of the cell, which separating becomes intercellular substance and as such can produce new fibrillæ. Perhaps the different opinions are harmonized in this statement.

† There are cases, chiefly pathologic,—for example in the withered skin of the face of aged persons—in which the elastic fibers stain weakly with specific acid dyes (No. 13, p. 43) and on the other hand react strongly with basic dyes; the substance of such fibers is called *elacin*. Contrawise, degenerating fibers of glutinous connective-tissue stain strongly with the specified acid dyes; this modified substance of connective tissue has been named *collastin*.

Narrow-meshed networks composed of thick elastic fibers form the transition to elastic membranes (Fig. 36), which are either homogeneous or finely striated and are perforated with holes of different sizes (hence the name fenestrated membranes), and doubtless are produced by the fusion of broad elastic fibers.

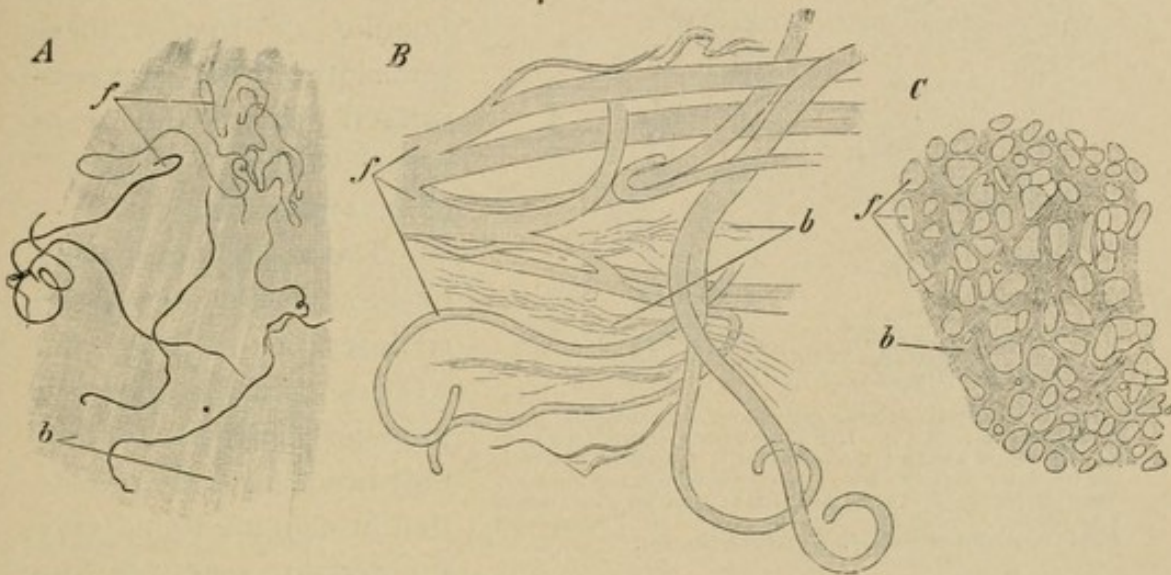


FIG. 35.—ELASTIC FIBERS. $\times 560$. *A*. Fine elastic fibers: *f*, from intermuscular connective tissue of man; *b*, connective-tissue bundles swelled by treatment with acetic acid. Technic No. 12. *B*. Very thick elastic fibers: *f*, from the ligamentum nuchae of an ox; *b*, connective-tissue bundles. Technic No. 13. *C*. From a cross-section of the ligamentum nuchae of an ox; *f*, elastic fibers; *b*, connective-tissue bundles. Technic No. 14.

When the quantity of elastic fibers predominates over the number of connective-tissue bundles, the tissue is spoken of as *elastic tissue*.

Hitherto the elastic fibers were regarded as transformations of the ground substance (perhaps of the existing connective-tissue bundles); according to recent investigations they are held to originate in the cells, in the form of the minutest depositions, that fuse into delicate nets; then the cells, it is supposed, degenerate, whereby the fibers are liberated (?). In the beginning of their development the elastic fibers are thin, but progressively with their growth increase in thickness by apposition.

The *cells* (Fig. 37 *A*) are irregularly polygonal, plump, and processless; or stellate with processes, strongly flattened and variously bent or indented. The compression and notching are explained by the adaptation of the cells to the narrow spaces occurring between the connective-

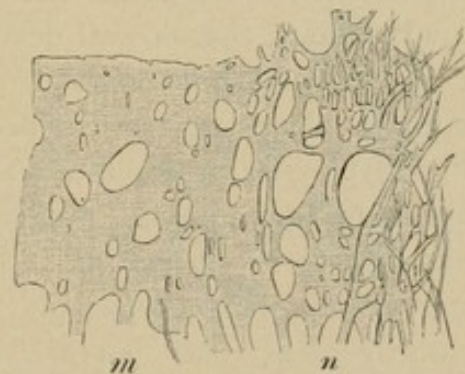


FIG. 36.—Network (*n*) of thick elastic fibers, on the left passing into a fenestrated membrane, *m*. From the endocardium of man. $\times 560$. Technic No. 15.

tissue bundles. Not infrequently the flattened cells form complete sheaths about the connective-tissue bundles.* If such a bundle be treated with acetic acid it swells and bursts the ensheathing cells, of

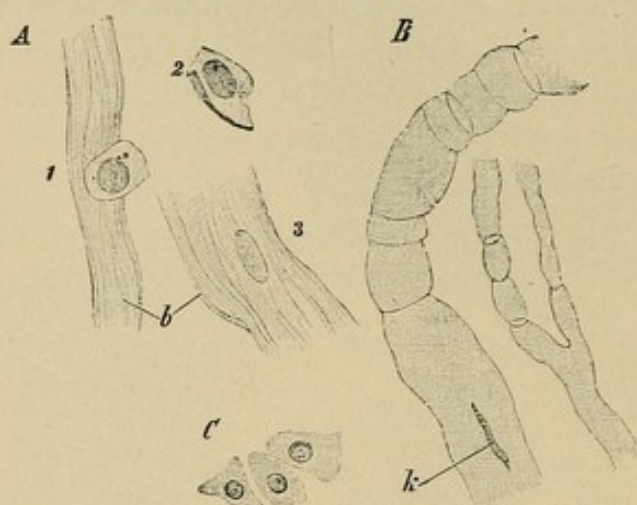


FIG. 37.—A. Connective-tissue cells from intermuscular connective tissue. $\times 560$. 1. Flat cell lying partly on a connective-tissue bundle; 2, notched cell; 3, nucleus of a cell, the protoplasm is invisible; b, connective-tissue bundles. Technic No. 6. B. Connective-tissue bundles with encircling fibers; k, nucleus. Technic No. 9. C. Plasma-cells from the eyelid of a child. Technic No. 191.

which annular or other-shaped fragments remain and constrict the swelled bundle. Formerly these remnants of cells were considered fibers and were called "encircling fibers" (Fig. 37 B).

The protoplasmic body of the connective-tissue cell encloses a nucleus and often contains pigment granules; in the latter case they become *pigment cells*,† that in man are found only in certain areas of the

skin and in the eye, but in the lower animals are very widely distributed. Connective-tissue cells may contain fat globules, that, when they are very large, coalesce and give a spherical form to the cell, which is then designated a *fat-cell* (Fig. 38). In such cells the protoplasm occupies only a narrow peripheral zone, in which lies the extremely flattened nucleus, that in well-developed, but not in atrophic, fat-cells invariably contains one or more sharply circumscribed fat droplets (Fig. 39). The protoplasmic zone often is so thin as to be invisible. Aggregations of fat-cells lead to the construction of a formation interwoven with numerous blood-vessels, lymph-vessels, and nerves, called *adipose tissue*, which plays a very important physiologic rôle in connection with metabolism.

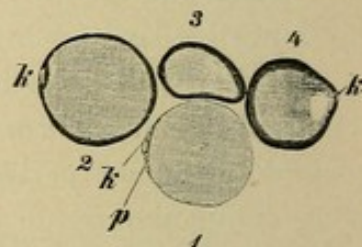


FIG. 38.—FAT-CELLS FROM THE AXILLA OF MAN. $\times 240$. 1. The equator of the cell in focus; 2, objective somewhat elevated; 3, 4, forms changed by pressure; p, traces of protoplasm in the vicinity of the flat nucleus, k, Technic No. 10.

* The form of the connective-tissue cells is not in any respect characteristic; especially when they lie together in groups their resemblance to epithelial cells often is complete. Regarding the true nature of such elements, designated by the perilous name of "epithelioid" cells, the embryonal history alone, not the form, can give the solution.

† Not every pigment cell is a connective-tissue cell; there are also pigmented epithelial cells, e. g., in the eye.

In high degrees of emaciation fat-cells are found in which all the fat except a few tiny drops has disappeared and in place of it there is a pale protoplasm mixed with a mucoid fluid; the cell is no longer spherical,

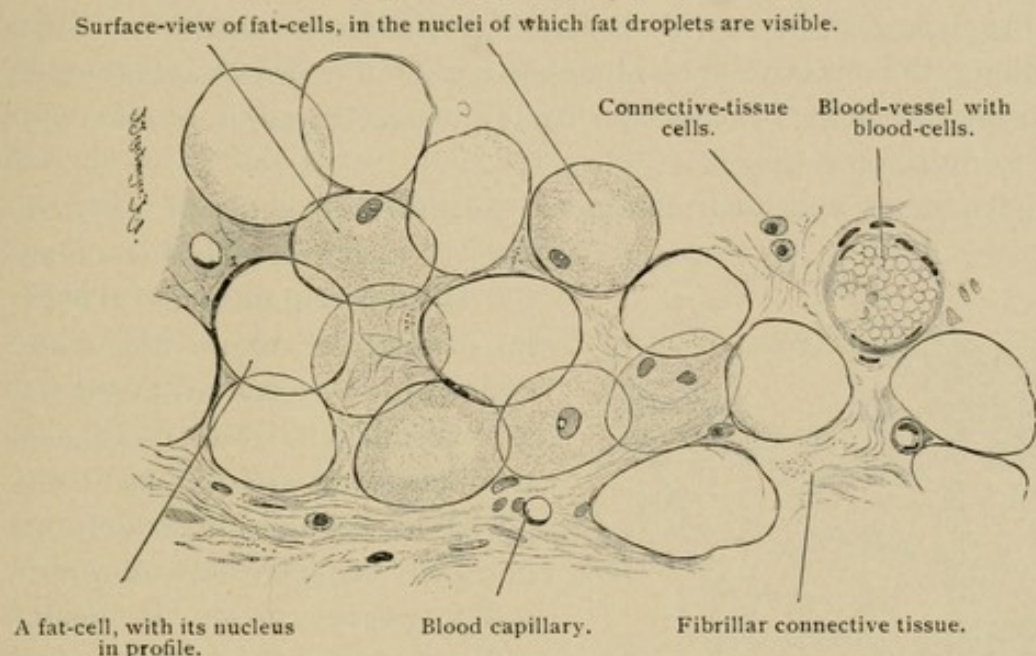


FIG. 39.—ADIPOSE TISSUE FROM THE HUMAN SCALP. $\times 240$ (about). Technic No. 11.

but has become flattened. Such cells are named *serous fat-cells* (Fig. 40). In many fat-cells spherical masses of needle-shaped crystals, the so-called *margarin crystals*, appear after death.

Finally, cells are found in connective tissue that are not connective-tissue elements, but *leucocytes* (see p. 137) that have passed out of the blood-vessels. They are described as *wandering cells*, to distinguish them from the connective-tissue cells, which are designated *fixed cells*; a classification that cannot be rigidly carried out, since in some conditions (mainly pathologic) the fixed connective-tissue cells can migrate;* therefore it is better to term the latter "histogenetic," the wandering leucocytes "hematogenetic" wandering cells.

The *plasma-cells* and *mast-cells* occurring in fibrillar connective tissue in widely varying quantity must be regarded as peculiar forms of

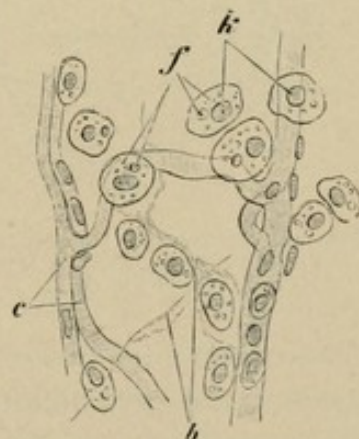


FIG. 40.—SEROUS FAT-CELLS FROM THE AXILLA OF AN EXTREMELY EMACIATED INDIVIDUAL. $\times 240$. *k*, Nucleus; *j*, oil-droplets. *c*, Blood capillaries; *b*, connective-tissue bundles. Technic No. 10.

* Under like conditions epithelial and gland-cells can wander; it is self-evident that such wandering cells cannot be included in the same category with the leucocytes.

leucocytes. The former are found principally in the vicinity of small blood-vessels and are spherical, coarsely granular, rich in protoplasm, and relatively of large size (Fig. 37); the latter contain granules that are easily stained by anilin dyes (*e. g.* dahlia), but do not stand, as their name may suggest, in any demonstrable relation to nutrition. Here also belong the clasmatocytes, branched cells the processes of which sever from the body (whence the name), dissolve, and, it is said, contribute to nutrition.

The number and distribution of the different kinds of cells are subject to considerable fluctuation.

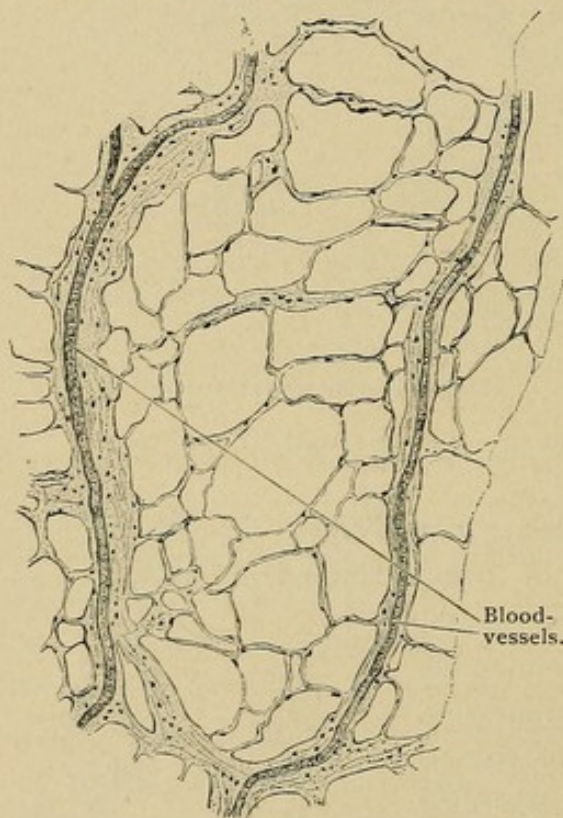


FIG. 41.—"FORMED" CONNECTIVE TISSUE. A PIECE OF THE GREATER OMENTUM OF MAN. $\times 60$. Technic No. 16.

The different elements of fibrillar connective tissue are united either without exact arrangement, as "formless" (areolar) connective tissue or are regularly disposed in definite formations, as "formed" connective tissue. *Formless connective tissue* is distinguished by its loosely united fiber-bundles interlacing in every direction; it occurs between neighboring organs and serves to connect them and fill in the interspaces. For this reason it is called "interstitial" tissue. The cells of interstitial tissue not infrequently contain fat. The *formed connective tissue* is characterized by the intimate union and regular arrangement of its bundles and comprises

the corium, the mucous membranes, the serous membranes, the periosteum, the perichondrium, the tendons, the fasciæ, the ligaments; the compact sheaths of the central nervous system, of the blood-vessels, of the eye, and of many glands.

Where fibrillar connective tissue is in immediate contact with epithelium it not infrequently happens that a structureless membrane is formed, described as *basement membrane* or *membrana propria*, also *hyaloid membrane*. It is essentially a modification of the connective tissue; possibly here and there on the epithelial side, a product of the epithelium.

(c) *Reticular connective tissue*.—The views in regard to the struc-

ture of reticular connective tissue are divided. According to an opinion formerly widely entertained it consists of a delicate network of anastomosing *stellate cells*. To this may be traced the name "cytogenous," that is, formed of cells.* There is no doubt that such networks exist in lower vertebrate animals and in embryonic stages of higher vertebrate animals, but occur only seldom in adults. In the higher vertebrates the relations are changed; here the network consists of slender bundles of fibrillar connective tissue, upon which lie flattened, nucleated cells (Fig. 42). By means of complicated methods the outlines of the cells on the fibers can be demonstrated. In fibrillar connective tissue the cells almost without exception lie upon the bundles. Finally, the fact that even in the adult fibrillar connective tissue may change into reticular tissue can be comprehended only on the assumption that the latter is a network of delicate fiber bundles. Therefore reticular connective tissue really is only a variety of the fibrillar tissue, differing somewhat from the latter chemically, in its greater resistance to reagents. The meshes of reticular connective tissue are invariably filled with densely crowded leucocytes. It occurs principally in lymph glands (better lymph nodes); for this reason it is called *adenoid tissue*, that is, tissue resembling glands.

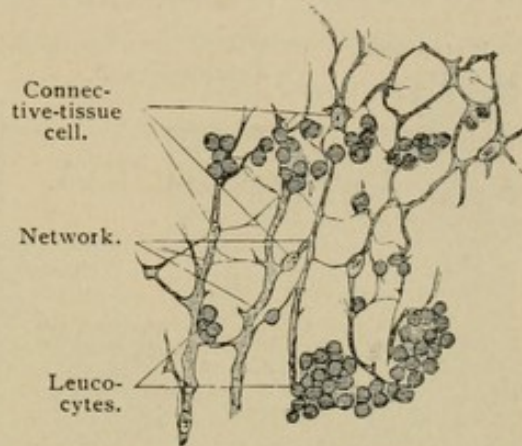


FIG. 42.—RETICULAR CONNECTIVE TISSUE. From a shaken section of a human lymph-gland. $\times 560$. Technic No. 55.

2. CARTILAGE.

Cartilage is firm, elastic, easily cut, and milk-white or yellowish in color. The *cells* present little that is characteristic in form; usually they are spherical or flattened on one side. They lie in cavities in the ground substance,† which they completely fill (Fig. 43). Not seldom the matrix immediately surrounding the cavities is specialized and forms a highly refractive, occasionally concentrically striated shell, the *cartilage capsule*. The matrix originates in excretions of the cells, through the capsules,

* Accordingly mucous tissue also might be termed cytogenous tissue.

† Whether, as in osseous tissue, the cavities are united with each other by a system of minute canals buried in the matrix is still extremely doubtful. Many such observations have been acknowledged as erroneous. The supposed canaliculi were due to shrinkage of the matrix and can be produced by treating cartilage with absolute alcohol or with ether.

which fuse into a homogeneous mass.* The parts of the capsule lying nearest to the cells are the youngest; they do not always persist, but during the process of cell-division are resorbed. Consequently the ground substance is subject to many changes. It may be free from fibrous admixture or it may be penetrated by elastic fibers or by connective-tissue bundles. Accordingly three varieties are distinguished: (a) *hyaline cartilage*, (b) *elastic cartilage*, (c) *fibrous cartilage*.

(a) *Hyaline cartilage* is of a faint bluish, pearly color. It occurs in the cartilages of the respiratory organs and of the nose, as the costal

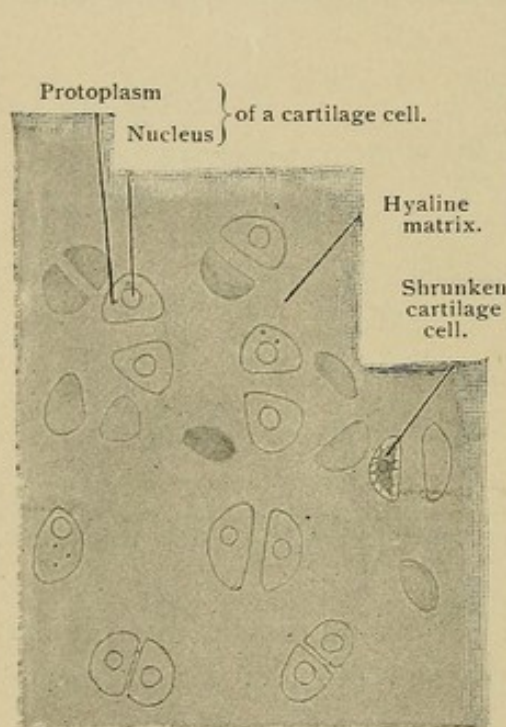
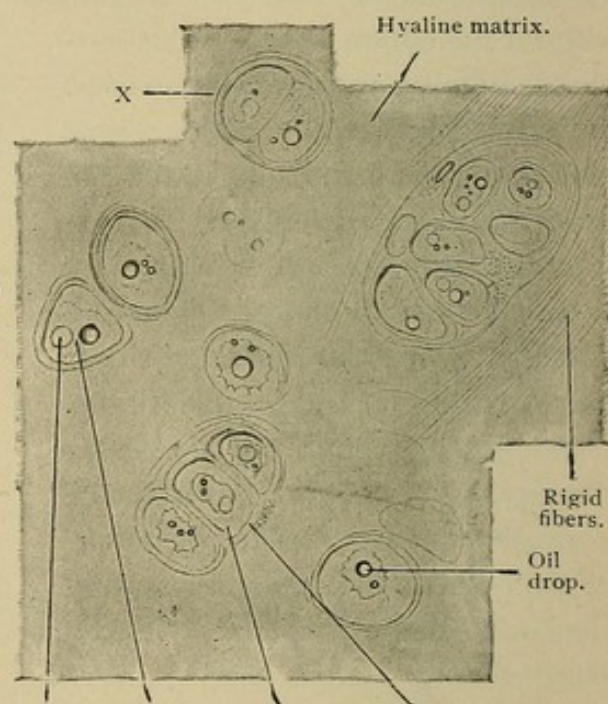


FIG. 43.—SURFACE PICTURE OF A PIECE OF THE FRESH ENSIFORM PROCESS OF A FROG. $\times 300$. Technic No. 17.



Nucleus. Protoplasm. Lacuna. Capsule.

FIG. 44.—SECTION OF A HUMAN COSTAL CARTILAGE, EXAMINED SEVERAL DAYS AFTER DEATH. The protoplasm has withdrawn from the wall of the lacuna. At X two cells lie in one capsule. $\times 300$. Technic No. 18.

The shaded cells do not lie in the focal plane and therefore shimmer indistinctly through.

and the articular cartilages, also in the synchondroses, and in the embryo in many situations where later it is replaced by bone. It is characterized by the homogeneity of its matrix, which in the ordinary methods of investigation appears amorphous throughout, but after certain manipulations, *e. g.*, artificial digestion, falls apart into bundles of fibers. Further evidence in corroboration of the fibrillar structure of the ground substance is afforded by its behavior in polarized light. It is very firm, very elastic, and on boiling yields *chondrin*.

* According to recent investigations the cartilage capsules are said to originate from the exoplasm of the cartilage cells; in this case the matrix is not a secretion, but a transformation product of the cartilage cells.

In certain cases the matrix may undergo peculiar modifications. In the laryngeal and costal cartilages it is transformed patchwise into rigid fibers, that impart an asbestos-like luster, perceptible on macroscopic inspection. In advanced age* deposition of calcareous salts may take place in the hyaline matrix, in the beginning appearing in the form of minute granules, subsequently as complete husks surrounding and enclosing the cells.

The cells of hyaline cartilage frequently occur in groups or nests, an arrangement explained by the conditions and processes of growth. Two cells may be seen within the same capsule (Fig. 44, X); they are the descendants of one cartilage cell which has undergone division by the indirect mode; in other cases a thin partition of hyaline substance may be seen between two such cells. In still other cases the septum does not develop immediately, and the process of cell-division may be repeated

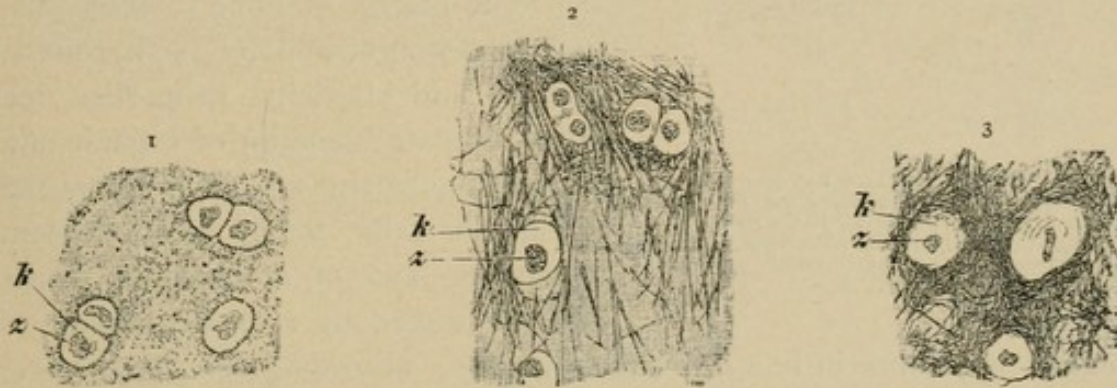


FIG. 45.—ELASTIC CARTILAGE. $\times 240$. 1. Portion of a section of the vocal process of an arytenoid cartilage of a woman thirty years old; the elastic substance is in the form of granules. 2 and 3. Portions of sections of the epiglottis of a woman sixty years old; a fine network of elastic fibers in 2, a denser network in 3. z. Cartilage-cell, nucleus invisible; k, capsule. Technic No. 19.

until groups of four, eight, and even more cells may be enclosed within one capsule (Fig. 44). Such phenomena were supposed to establish a special theory of cell-division, the so-called endogenous cell-formation (*cf.* p. 72). Not infrequently the cartilage cells in adults contain oil-globules.

(b) *Elastic cartilage* has a faint yellowish color. It occurs only in the external ear, the epiglottis, the cuneiform and corniculate cartilages, and the apex and vocal process of the arytenoid cartilages. It presents the same structural features as hyaline cartilage, but is distinguished by the networks of sometimes finer, sometimes coarser elastic fibers that penetrate the matrix. The elastic fibers do not arise directly from the cartilage-cells, but by a transformation of the matrix, and appear in the vicinity of the former as minute granules (Fig. 45, 1), that later are dis-

* In the cartilages of the larynx this may occur as early as the twentieth year.

posed in linear rows and fuse into fibers. According to an opposite view, this phenomenon is regarded as an indication of post-mortem disintegration of the elastic fibers.

(c) *Fibrous cartilage* (connective-tissue cartilage) is found in the intervertebral disks, the pubic symphysis, the head of the ulna, the articular ends of the maxilla, the sternum, the clavicle, and the ribs. The matrix contains an abundance of fibrillar connective tissue, the loose bundles of which extend in every direction (Fig. 46, *g*). The cartilage-cells are few in number, have thick capsules (*z*), and lie united in small groups or rows at wide intervals.

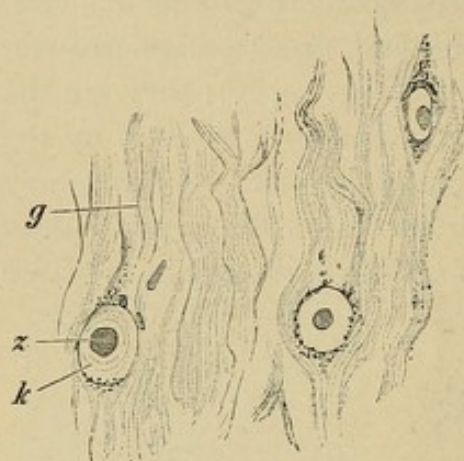


FIG. 46.—FROM A HORIZONTAL SECTION OF THE INTERVERTEBRAL DISK OF MAN. *g*, Fibrillar connective tissue; *z*, cartilage-cell (nucleus invisible); *k*, capsule surrounded by calcareous granules. $\times 240$. Technic No. 20.

3. OSSEOUS TISSUE.

The matrix of bone (osseous tissue) is distinguished by its hardness, solidity, and elasticity, properties due to an intimate blending of organic and inorganic substances.* It is composed of calcium salts, chiefly basic calcium phosphate, and of collagenous fibrils that are united by a small amount of

cement substance in fine or coarse bundles; accordingly, a *fine-fibered*, or lamellar, and *coarse-fibered*, or plexiform, bone-matrix are distinguished.† It appears homogeneous or faintly striated and contains numerous spindle-shaped spaces, 15 to 27 μ in length, the *bone lacunæ* (formerly called "bone corpuscles"), which communicate with one another through numerous branched, minute canals, the *bone canaliculi*. In this way a system of canaliculi that penetrates the entire matrix is established. Within the lacunæ, sometimes improperly called

* This union is of such a nature that either part may be removed without destroying the structure of the tissue. On treatment with acids (see Decalcifying, p. 36) the inorganic substances are withdrawn; the bone is decalcified, is rendered flexible, and is easily cut, like cartilage; therefore it is called "bone cartilage." The organic substances can be removed by cautious heating; the bone then is said to be calcined. Similarly, fossil bones are deprived of the organic substances through the prolonged action of moisture.

† The skeleton of the adult is principally formed of the fine-fibered matrix, which is characterized by distinct lamellæ (see The Organs of the Skeletal System); it contains elastic fibers. The coarse-fibered matrix occurs in the fetus in perichondral and secondary bone (see Development of Bone), and is found in the adult in sutures and at the point of insertion of tendons; it always contains, partly calcified, partly uncalcified, connective-tissue bundles, the so-called Sharpey's fibers, which also are found in the circumferential and interstitial lamellæ of fine-fibered bone.

"bone-cells," lie the nucleated *bone-cells* (Fig. 48), which have a flat-oval shape and send thin processes into the bone canaliculi. It is doubt-

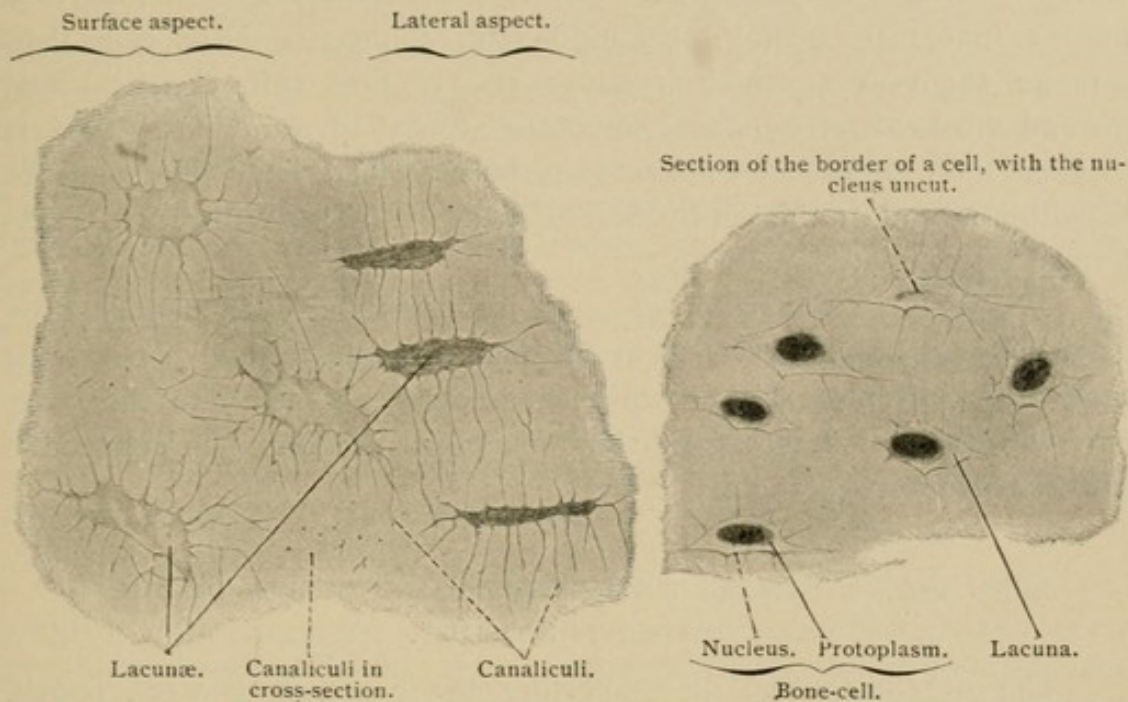


FIG. 47.—PORTION OF A GROUND SECTION OF THE INFERIOR MAXILLA OF ADULT MAN. $\times 550$. Technic No. 61.

FIG. 48.—SECTION OF THE OSSEOUS TURBIN OF ADULT MAN. $\times 550$. Technic No. 63.

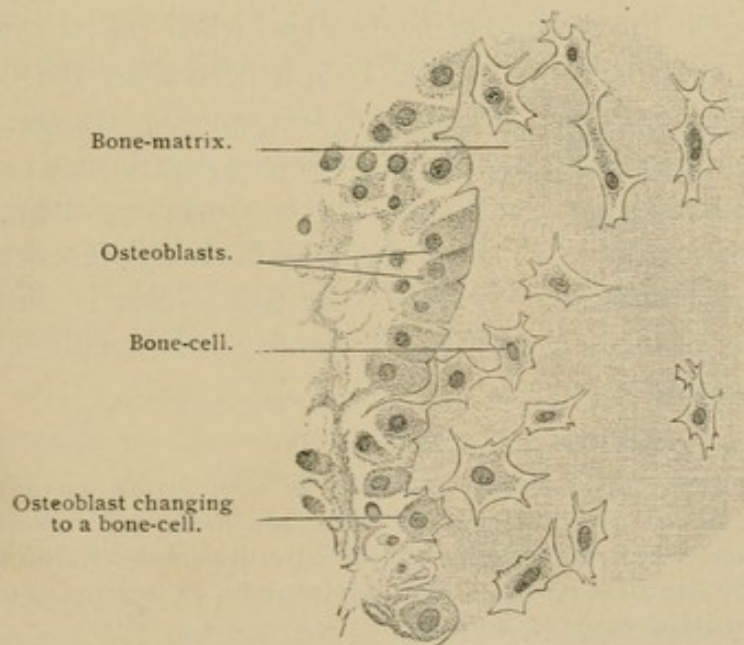


FIG. 49.—PORTION OF A CROSS-SECTION OF THE DIAPHYSIS OF THE HUMERUS OF A HUMAN EMBRYO FOUR MONTHS OLD. $\times 560$. Technic No. 67.

ful whether in the adult the bone-cells are connected by means of processes extending through the canaliculi, although such connection is readily observed in developing bone (Fig. 49).

Usually the formation of osseous tissue takes place in such a way that the ground substance of the connective tissue or of the cartilage calcifies during embryonic life. Around the trabeculæ of the calcified matrix numerous young, still indifferent, connective-tissue cells then arrange themselves, which produce the at first soft, then calcified ground substance of bone. These cells are called *osteoblasts*. At first they lie upon the osseous matrix they have formed, later they come to lie within it, and gradually, by development of processes, become transformed into stellate bone-cells.*

Dentine is a modification of bone, from which it is distinguished by its developmental history and in the fact that the formative cells, the *odontoblasts*, are not enclosed within the matrix, but penetrate the latter only with their processes. Further details will be found in connection with the structure of teeth.

THE BLOOD-VESSELS, LYMPH-VESSELS, AND NERVES OF THE SUPPORTING TISSUES.

The organs formed of supporting tissue are, in general, poorly supplied with blood-vessels,† lymph-vessels, and nerves. But supporting tissue plays a very important part as a conveying apparatus in the transference of nutritive fluids—*tissue-juice*, *lymph*—from the blood-vessels to the tissues. It moves in the ground substance and when this is soft, as in mucous and loose connective tissue, the lymph permeates the entire mass; when on the other hand the ground substance is denser, the lymph circulates in definite channels, in a *juice-canal-system* formed by the cell-spaces, the *lymph-spaces*, and the minute canals connecting them, the *lymph canaliculi* (cf. the cornea). This is the case in the more compact connective tissues‡ and in bone. Whether the tissue-juice is diffused throughout the matrix of hyaline cartilage or conveyed in definite channels is still undetermined.

* Direct transmutation of developed connective tissue or cartilage into osseous tissue does not occur. The processes collectively designated "metaplasia" are much better interpreted as signifying that indifferent formative cells of connective tissue, subject to dissimilar influences, may develop, now into bone-cells, now into cartilage-cells, or into typical tendon cells (see also the chapter on Development of Bone).

† Adipose tissue forms an exception.

‡ The lymph canaliculi occurring here stand in direct connection with the intercellular clefts of the epithelial tissues, which we must imagine as similarly permeated by the tissue juice.

TECHNIC.

No. 4.—*Mucous connective tissue*.—Place the umbilical cord of a human embryo of three or four months (or pig embryo from three to six cm. long) in 50 c.c. of Zenker's fluid (p. 33) for 24 hours; harden in 30 c.c. of gradually strengthened alcohols (p. 35). The cord will still be very soft; in order to obtain good sections it must be embedded in liver, and in cutting must be somewhat compressed with the fingers. The sections may be stained in picrocarmine (twelve hours) or in Hansen's hematoxylin (five minutes), and should be examined in a drop of distilled water (Fig. 33). In glycerol and in xylol-balsam the delicate processes of the cells and the bundles of connective tissues are invisible. In the vicinity of the blood-vessels the network of cells is less fine; therefore a field remote from the blood-vessels should be selected for study. The older the embryo, the greater is the number of connective-tissue bundles. Mount in diluted glycerol (p. 49).

No. 5.—*Fibrillar connective tissue; connective-tissue bundles*.—Prepare small strips, one or two cm. long, of intermuscular connective tissue, for example, of the thin septum between the serratus and the intercostal muscles; place a small piece on a *dry* slide and quickly spread it out with teasing needles (see "half-drying method" No. 31 *a*, p. 123), add a drop of salt solution and apply a cover-glass. The bundles of connective tissue appear wavy and pale (Fig. 34); with a little practice the sharply contoured, highly refracting elastic fibers can be distinguished and also, in favorable situations, the nuclei of the connective-tissue cells.

No. 6.—The *cells of fibrillar connective tissue* may be made visible by the addition of a drop of picrocarmine to preparation No. 5, under the cover-glass (p. 53). In most cases only the red nucleus can be perceived, especially when the cell lies wholly upon the connective-tissue bundle. In rare cases the pale yellow, variously shaped body of the cell can be seen (Fig. 37 *A*, 1 and 2).

No. 7.—*Mast-cells*.—Fix small pieces, 1 or 2 cm. square, of mucous membrane (of the mouth, pharynx, or intestine) in ninety-five per cent. alcohol (p. 31). In from three to eight days cut thin sections and stain them in 10 c.c. of alum-carmine dahlia for twenty-four hours (p. 26). Transfer them to 10 c.c. of absolute alcohol for twenty-four hours, which must be renewed once or twice during this time. Mount in xylol-balsam (p. 50). The protoplasm of the mast-cells exhibits granules stained an intense blue.

No. 8.—*Fibrillæ*.—Place a piece of tendon about 2 cm. long in 100 c.c. of saturated aqueous solution of picric acid. On the following day, with two pairs of forceps, pull the tendon apart along its length, take from the interior a bundle about 5 mm. long, and tease the same on a dry slide (*cf.* No. 31 *a*, p. 123); add a drop of distilled water, apply a

cover-glass, and examine with the high-power objective. The ultimate fibrillæ appear as exceedingly fine, pale filaments.

No. 9.—*Encircling fibers*.—With the scissors cut out a piece about one cm. square of the connective tissue within the arterial circle of Willis, wash it in a watch-glass with salt solution, with needles spread it out in a drop of the same solution on a slide, and cover. With the low power, in addition to numerous delicate blood-vessels and ordinary connective-tissue bundles, sharply contoured, refracting bundles, in distinct contrast to the remaining connective tissue, will be found, which on the use of the high power, and a diaphragm of narrow aperture, show that they likewise consist of fibrillar connective tissue. Place such a bundle in the field and treat it with a drop of acetic acid, under the cover-glass (p. 53). So soon as the acid reaches the bundle, it swells, the fibrillation vanishes and instead elongated nuclei appear. The swelling is not uniform; at irregular intervals the bundle is constricted. With dim illumination the

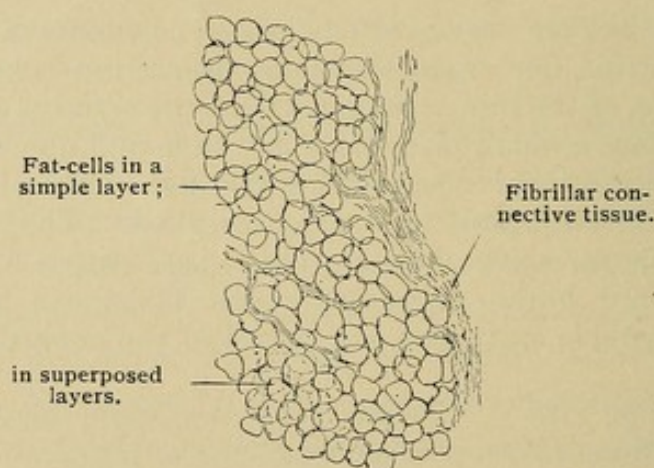


FIG. 50.—ADIPOSE TISSUE FROM A SECTION OF HUMAN SCALP. $\times 50$. Technic No. 161. Cf. Technic No. 11.

“fibers” (cell-remnants) producing the constrictions can be seen (Fig. 37 B).

No. 10.—*Fat-cells*.—Take a small piece of the reddish-yellow, gelatinous fat from the axilla of a thoroughly emaciated individual; rapidly spread out a piece the size of a split pea in the *thinnest possible* layer on a dry slide, *immediately* add a drop of salt solution and apply a cover-glass. In thin places atrophic fat-cells, like those shown in figure 40, will be seen. This preparation may be stained under the cover-glass with picrocarmine (p. 53) and preserved in diluted glycerol. Ordinary (normal) fat-cells, taken from any part of the body, are likewise to be examined in salt solution. The spherical cells should be studied with change of focus (*cf.* Fig. 38).

No. 11.—*Adipose tissue* may be seen in sections of many preparations fixed by any of the usual methods, above all of the skin (*cf.* Fig.

288). The oily contents are withdrawn by the treatment with alcohol and then the clusters of empty cell-envelopes present a picture that the beginner often finds difficulty in understanding (Fig. 50).

No. 12.—*Fine elastic fibers* may be readily obtained by treating preparation No. 5, under the cover-glass, with a few drops of acetic acid. The connective-tissue bundles swell and become transparent; the elastic fibers, on the contrary, remain unaltered and stand out sharply contoured (Fig. 35 A).

No. 13.—*Thick elastic fibers* may be obtained by teasing in a drop of salt solution a slender piece, about 5 mm. long, of the fresh ligamentum nuchæ of an ox (Fig. 35 B). The piece should not be taken from the loose, enveloping tissue, but from the tough, yellowish, fibrous portion. The preparation may be stained in picrocarmine (p. 41) and mounted in glycerol.

No. 14.—*Cross-sections of thick elastic fibers* may be obtained by drying a piece (10 cm. long and from 1 to 2 cm. thick) of the ligamentum nuchæ (it will be ready to use in four or six days) and treating it like No. 69.

No. 15.—*Fenestrated membranes*.—Take a small piece (about 5 mm. square) of the endocardium, place it in a drop of water on a slide and add, under the cover-glass, 1 or 2 drops of potash-lye. Examine the edges of the preparation (Fig. 36).

Good specimens may also be obtained from the basilar artery; place a piece of the artery cut open lengthwise in 10 c.c. of concentrated potash solution. After six hours take a small piece, about 1 cm. long, and separate the lamellæ in a drop of water on a slide; this is easily done by scraping with a scalpel. Cover and examine with the high power. The small holes in the membrane have the appearance of shining nuclei. With the low power the membrane is recognized by its dark outlines. To preserve, wash it well in 10 c.c. of water for five minutes, stain it in 3 c.c. of congo-red for from twelve to twenty hours (p. 25), and mount in xylol-balsam (p. 50).

No. 16.—*A network of connective-tissue bundles* may be obtained by spreading out a little piece of fresh human omentum in a few drops of picrocarmine. It may be preserved in diluted, nonacidulated glycerol (p. 49). Pieces of the omentum fixed in absolute alcohol and stained with hematoxylin and eosin (p. 39) may be mounted in xylol-balsam (p. 50). (Fig. 41, p. 94.)

No. 17.—*Hyaline cartilage*.—Cut off the extremely thin episternum of the frog, place it on a dry slide, cover it with a cover-glass, and examine at once with the high power. The cartilage cells completely fill the cartilage cavities (Fig. 43). For prolonged study add a drop of saline solution.

No. 18.—*Hyaline costal cartilage*.—Without any previous preparation thin sections of costal cartilage may be cut with a razor and examined in a drop of water under a cover-glass. Search for one of the glossy areas containing rigid fibers (Fig. 44). The preparation may be preserved by adding a few drops of dilute glycerol.

Fresh cartilage does not readily stain. The tissue must be first placed in Zenker's or in Müller's fluid (p. 33), then in alcohol (p. 35), and subsequently stained with Hansen's hematoxylin (p. 38). Mounted in xylol-balsam, which clears vigorously, the finer details vanish.

No. 19.—*Elastic cartilage*.—Take a piece of the arytenoid cartilage of man (better still of the ox), the elastic cartilage of the apex and the vocal process is recognized by its yellowish color. Cut a section that includes the boundary line between the elastic and the hyaline cartilage and examine it in water. Preserve like No. 18. The development of elastic fibers may often be studied in the cartilages of adults, especially in the epiglottis and in the vocal process of the arytenoid cartilage (Fig. 45, 1). See also Technic No. 128.

No. 20.—*Fibrous cartilage*.—Cut the intervertebral disks of adult man in pieces from 1 to 2 cm. square; fix in 100 c.c. of potassium-bichromate-acetic acid (p. 32) for twenty-four hours and harden in 50 c.c. of gradually strengthened alcohols (p. 35). Stain sections in Hansen's hematoxylin (p. 38) and mount in balsam (Fig. 46). Sections through the edges yield hyaline cartilage; through the central portions of the disk they exhibit large groups of cartilage-cells.

III. THE MUSCLE TISSUES.

The characteristic elements of the muscle tissues, the *muscle-fibers*, occur in two forms, named the *smooth* and the *striated*. Both are cells, the body of which is extraordinarily elongated.

1. *Smooth, nonstriated or involuntary muscle*.—The tissue of smooth muscle consists of contractile fiber-cells, spindle-shaped, cylindric, or slightly flattened elements with tapering extremities (Fig. 51). Their length in man varies from 45 to 225 μ , their width from 4 to 7 μ ; in the gravid uterus smooth muscle-fibers measuring 0.5 mm. have been found. They consist of a delicately striated protoplasm—the striations indicate that the fiber is composed of fibrillæ—and an elongated, elliptical, or rod-shaped nucleus, that is characteristic of the smooth muscle-fiber.*

The smooth muscle-fibers sometimes lie scattered in the connective tissue, sometimes are intimately united in complexes, by delicate, per-

*The diplosome lies on the longitudinal side of the somewhat eccentrically situated nucleus. Smooth muscle-fibers containing pigment have been found in the iris of fishes and amphibians, also in the human intestine and iris (dilator pupillæ).

forated connective-tissue membranes.* The development of these membranes varies greatly; while, for example, they are very tender in the musculature of the intestinal wall, so that their demonstration is possible only by certain methods, between the muscle-fibers of the ureter and yet more between those of the oviduct they are so well developed, that in specific connective-tissue staining the muscles are completely hidden. Thicker connective-tissue septa occur only at wide intervals (Fig. 52); elastic fibers are present, as well in the thicker septa as in the delicate membranes.



FIG. 51.—TWO SMOOTH MUSCLE-FIBERS FROM THE SMALL INTESTINE OF A FROG. $\times 240$. Isolated in 35 per cent. potash-lye. The nuclei have lost their characteristic form through the action of the lye. Technic No. 21 a.

The union of the complexes results either in membranes, in which their disposition is parallel, as in the muscle of the intestine, or in complicated networks, as in the urinary bladder and the uterus. The larger blood-vessels run in the stout connective-tissue septa, but the capillaries penetrate between the muscle-fibers and form longitudinal networks. The lymph-vessels follow the course of the blood-vessels and are present in conspicuous number.

For the nerves of smooth muscle, see the Peripheral Nerve-endings.

Smooth muscle-tissue occurs in the alimentary canal, in the trachea and bronchial tubes, in the gall-bladder, in the pelvis of the kidneys, in the ureters and the uri-

nary bladder, in the reproductive organs, in the blood- and lymph-vessels, in the eye, and in the skin. The contraction of smooth muscle-fiber is slow and not under the control of the will.

The musculature of the heart occupies a peculiar position. In the lower vertebrates, in frogs, for example, the cardiac muscle-fibers are spindle-shaped elements possessing elliptical nuclei, and often are more distinctly striated transversely than longitudinally (Fig. 53 A).

Fibers can also be isolated in mammals; short cylinders, which often have step-like ends (Fig. 53 B). The protoplasm is partially dif-

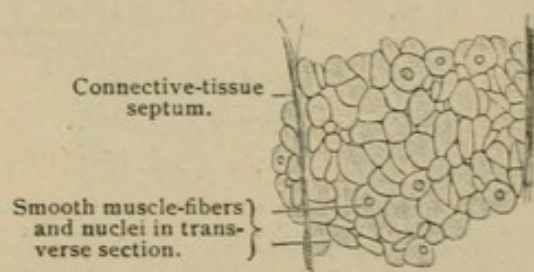


FIG. 52.—SECTION OF THE CIRCULAR MUSCLE COAT OF THE HUMAN INTESTINE. $\times 560$. The membranes are not visible here, so that the muscle-fibers appear to be in contact with one another. Technic No. 111.

* Regarding the intercellular bridges, see Technic No. 21 b.

ferentiated into cross-striated fibrils, *fibrillæ*, which not infrequently are arranged in lamellæ radially placed to the axis of the fiber (Fig. 53 *D*). The remnant of undifferentiated protoplasm, the *sarcoplasm*, relatively considerable in comparison with that of striated muscles, is found chiefly in the axial part of the fiber, from which processes radiate between the lamellæ. Owing to this longitudinal striation is often very marked. The oval nucleus is embedded in the axial part of the sarcoplasm, which very frequently contains granules of pigment or fat. A membrane equivalent to the sarcolemma of striated muscle-fibers is wanting. Characteristic of the cardiac muscle-fibers of higher animals is the union of the fibers by means of short oblique or transverse processes (Fig. 53 *B x*).

According to recent investigations these cardiac muscle-fibers are artifacts, fragments of a protoplasmic net provided with nuclei, a syncyt-

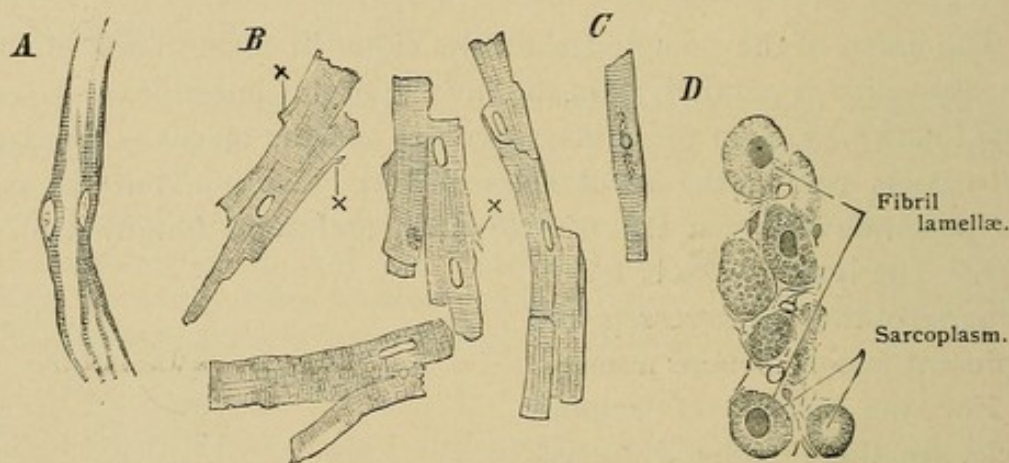


FIG. 53.—*A* and *B*, CARDIAC MUSCLE-FIBERS, isolated in potash-lye. *A*, of the frog; *B*, of the rabbit; *x*, oblique branch. $\times 240$. Technic like No. 26. *C*, from a longitudinal section, *D*, from a cross-section of a papillary muscle of man. *C* magnified 240, *D* 560 diameters. Technic No. 37.

ium, that is already present in early epochs of developmental history. The transverse lines ("cement-lines"), often distinct in longitudinal sections, the significance of which is not yet satisfactorily explained, are said to be not cell or muscle-fiber boundaries, because they are pierced by the muscle fibrillæ (*cf.* Union of cells, p. 73).

2. *Striated or voluntary muscle*.—It is only by the study of their development that the striated muscle-fibers are recognized as cells. By a colossal growth in length, by repeated division of their nuclei, as well as by peculiar differentiation of their protoplasm, they have become highly complicated structures. They have the form of long cylindrical threads, the ends of which, in the interior of the larger muscles, are rounded or pointed; at the extremities of the muscle they possess a pointed inner end and a broader end in contact with the tendon; the latter is either blunt or terminates in several stumpy, often steplike

notches. Anastomoses, divisions, and fissures occur; branched fibers are found in the muscles of the eye, the tongue, and the skin (Fig. 56, 4). They vary in length from 5.3 to 12.3 cm.,* in thickness from 10 to 100 μ . In the embryonal body no difference, or only an insignificant difference in thickness exists; after birth an unequal growth in the thickness of the muscle-fibers takes place, the intensity of which is dependent on: (1) the function of the muscle; in the adult robust muscles possess thick fibers, delicate muscles have thin fibers; (2) the nutritional condition of the individual; (3) the size of the animal, large animals possess thicker fibers than smaller ones. Hence the difference in caliber may be of a threefold nature.

Under the microscope each cross-striated muscle-fiber exhibits alternate broad *dim* and narrower *clear* transverse stripes. The substance

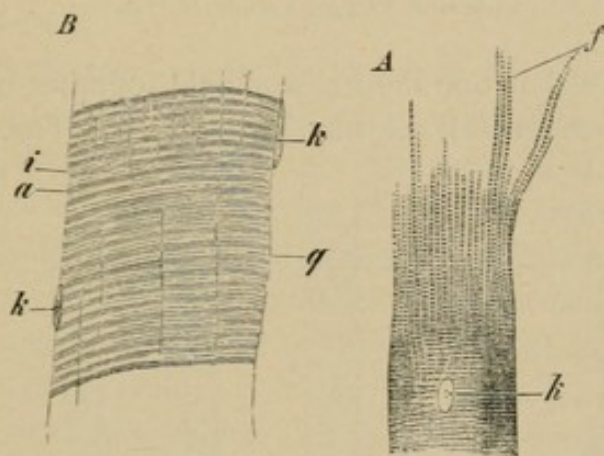


FIG. 54.—B. PORTION OF A MUSCLE-FIBER OF MAN; *a*, anisotropic, *i*, isotropic band; *q*, intermediate disk; *k*, nucleus. $\times 560$. Technic No. 22 *b*. A. MUSCLE-FIBER OF A FROG; *f*, fibrillae; *k*, nucleus. $\times 240$. Technic No. 25.

of the dim stripes is doubly refracting or *anisotropic*, that of the clear stripes singly refracting or *isotropic*.† Besides the cross-marking, a more or less distinct longitudinal striation may be observed. Treatment with certain reagents (*e. g.* solution of chromic acid) renders this striation more evident and even causes the muscle-fiber to fall apart longitudinally in delicate, likewise cross-striped, filaments, which are called *fibrillæ*. These

* It is probable that there are fibers having greater length, but their isolation entire is very difficult to accomplish.

† High amplifications show that each transverse disk is transversely divided; invariably in the isotropic (clear) zone a dim line occurs, the *intermediate disk* (Fig. 54, *q*), and above and below this a dark band, the *accessory disk*. In the anisotropic (dim) band a clear stripe, the *median disk*, has been observed. Owing to their extreme variation and their instability, these disks are of subordinate significance.

fibrillæ are the contractile structural elements of the muscle-fiber.* They are grouped into longitudinal bundles, the *muscle-columns*, in which they

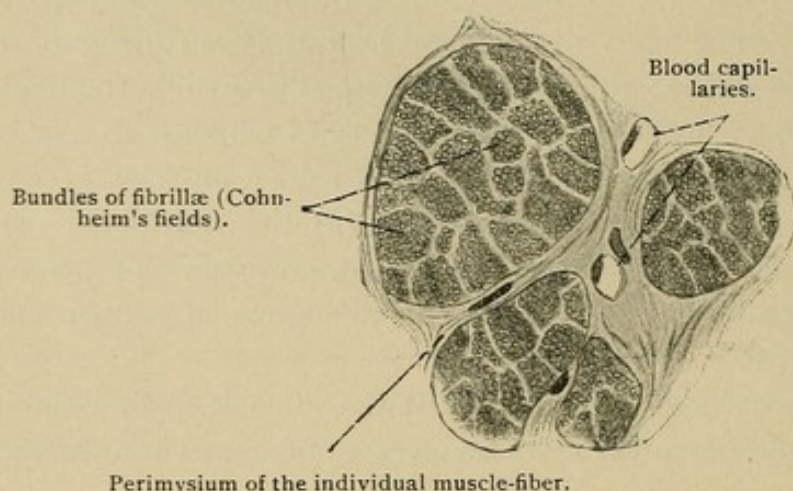


FIG. 55.—FROM A CROSS-SECTION OF THE ARYTENOID MUSCLE OF MAN. Four muscle-fibers are represented. $\times 590$. Technic No. 128.

are arranged parallel to one another and held together by the *sarcoplasm*, which also unites them with neighboring bundles. The disposition of the

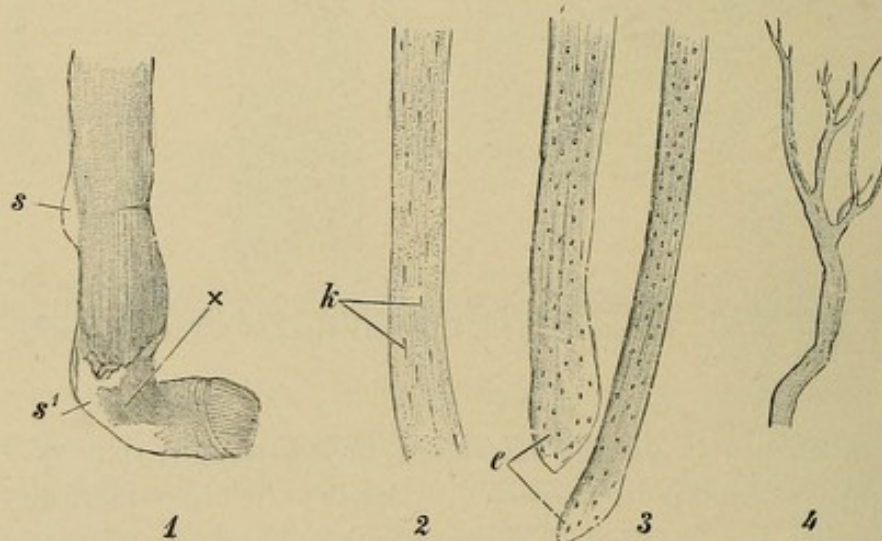


FIG. 56.—PORTIONS OF ISOLATED STRIATED MUSCLE-FIBERS OF A FROG. $\times 50$. 1. After treatment with water: *s*, *s'*, sarcolemma; at *x* the muscle-substance is torn, the cross-striation not apparent, the longitudinal striation distinct. Technic No. 23. 2. After treatment with acetic acid: *k*, nuclei; the fine stippling represents the interstitial granules. Technic No. 24. 3. After the action of concentrated potash solution: *e*, rounded ends; the numerous nuclei are swollen and vesicular in appearance. With this amplification the cross-striation in 2 and 3 is invisible. Technic No. 26. 4. Branched muscle-fiber from the tongue of a frog. Technic No. 27.

sarcoplasm is best seen in cross-section; high amplification is required. It presents the appearance of a clear network, within the meshes of which

* The muscle-fibers of some animals, after treatment with certain reagents, cleave transversely into disks. Fibrillæ and disks may be further separated into smaller, polygonal, anisotropic particles, that were called *sarcous elements*. Certain authors interpreted the disks, others the sarcous elements, as the true structural units.

are the muscle-columns in section, known as *Cohnheim's fields* (Fig. 55). The sarcoplasm contains the *interstitial granules*, consisting partly of fat and probably also of lecithin, and the *nuclei*. The latter are oval bodies placed parallel to the long axis of the muscle-fiber; in mammals, bony fishes, and some birds they are chiefly situated upon the surface of the muscle-fiber, beneath the sarcolemma; in other vertebrates they lie also in the interior of the fiber.* The number of the nuclei is from 3 to 12 times greater in thin than in thick muscle-fibers.

Each muscle-fiber is snugly enclosed in a structureless sheath, the *sarcolemma*, which represents the cell-membrane. Therefore the fiber of striated muscle consists of fibrillæ, sarcoplasm, nuclei, and sarcolemma.

The striated fibers are found in the muscles of the trunk and the extremities, of the eye and the ear, also in the tongue, the pharynx, the upper half of the esophagus, the larynx, the diaphragm, the genital organs, and the rectum.

In some animals, the rabbit, for example, two varieties of striated muscles are distinguished, the *red* (*e. g.* the semitendinosus, the soleus) and the *white* or *pale* (*e. g.* the adductor magnus); and correspondingly, two varieties of muscle-fibers: (1) dim fibers, *rich in protoplasm*, or sarcoplasm, showing less regular cross-striation, more distinct longitudinal striation, possessing in general a smaller diameter (for example, those forming the red soleus of the rabbit); (2) pale fibers, *poor in protoplasm*, more distinctly cross-striated and having in general a greater diameter. The latter represent the more highly differentiated muscle-fibers. While in certain animals the two varieties of fibers occur separately, each in particular muscles, in others—also in man—they are found intermingled in the same muscle. As a rule, the more functionally active muscles, the cardiac, ocular, masticatory, and respiratory, contain the greater number of fibers rich in protoplasm. The muscles with many pale fibers contract more rapidly, but are sooner fatigued.

The contraction of the striated fibers, compared with that of smooth muscle-fibers, is rapid and is under the control of the will. The striated fibers are united into bundles by fibrillar connective tissue, which serves also to convey the numerous ramifications of the blood-vessels and nerves supplying the muscle tissue. The lymph-vessels are few in number.

* In man, also, nuclei occur in the interior of the muscle-fiber and especially well developed in the neighborhood of the insertion of the tendon, thin and stunted in the rest of the muscle substance. The nuclei at the ends of the muscle-fibers, often numerous and arising by amitotic division, indicate that these are the places where the growth in the length of the fibers occurs.

TECHNIC.

No. 21.—*Smooth muscle-fibers (a) isolated*.—These are best obtained by treating a little piece of the stomach or intestine of a frog just killed with 20 c.c. of potash lye. Cover the glass. After from 30 to 60 minutes (in a cold room somewhat later), the intestine falls to pieces on being slightly stirred with a glass rod. If the action fails the lye is not strong enough (see p. 30). Transfer a drop containing some of the sediment to a slide (the fibers cannot be examined in water or glycerol, for the lye thus diluted will immediately destroy them); carefully apply a cover-glass and examine with the high power (Fig. 51).

After treating small pieces of intestine with 100 c.c. of Müller's fluid (p. 21) for from 8 to 14 days smooth muscle-fibers can be isolated by teasing, but successful preparations are difficult to obtain from the intestine of man and also of the frog, easier on the other hand from that of the horse (take the lower portion of the duodenum) and also of the rat.

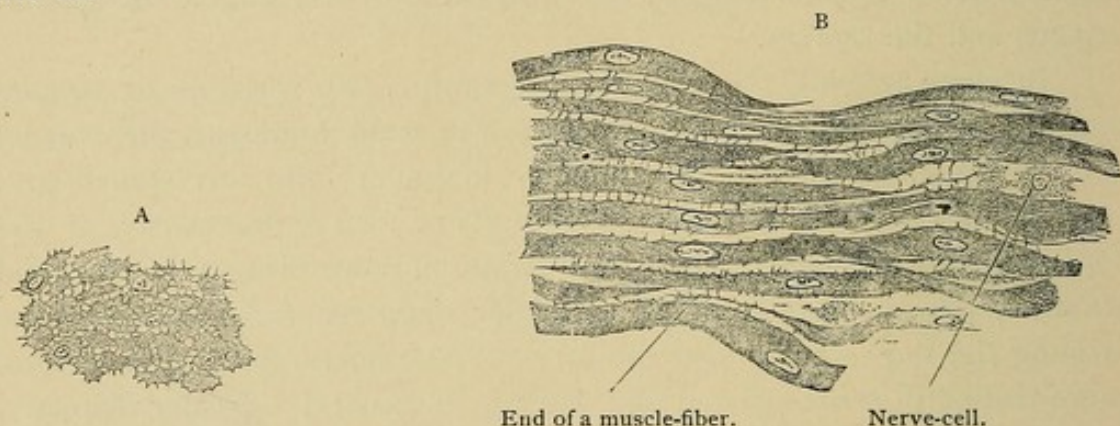


FIG. 57.—APPARENT INTERCELLULAR BRIDGES OF SMOOTH MUSCLE-FIBERS. A. Transverse section of the intestine of a rabbit. B. Longitudinal section of the intestine of a guinea-pig. $\times 420$.

(b) *Cross-sections of bundles of smooth muscle-fibers*.—Many fixation fluids cause shrinking of the fibers, which gives rise to deceptive pictures; such are the *intercellular bridges*. Pictures like that of Fig. 57 can often be obtained in very thin sections of tissue (pieces from 1 to 2 cm. long of the small intestine of a guinea-pig or rabbit *just killed*) fixed in 100 c.c. of Zenker's fluid (p. 33) and hardened in gradually strengthened alcohols (p. 35).

The twisted nuclei of smooth muscle-fibers also belong in the category of artifacts.

The demonstration of the connective-tissue membrane enveloping each individual muscle-fiber succeeds only by special staining (van Gieson, p. 43); the sections must not be thinner than 10μ .

No. 22.—*Striated muscle-fibers (a) of the frog*.—With the scissors placed flat and parallel to the course of the fibers, cut a piece about 1 cm. long from the adductor muscle of a recently killed frog. Take a fragment from the inner surface of this piece and tease it in a small drop

of salt solution (see Isolation, p. 29), add a second larger drop of the same liquid and, *without pressing*, cover the preparation with a cover-glass. With low magnification (50 diameters) the cylindrical form, the difference in thickness, occasionally also the cross-striation of the isolated fibers can be seen (Fig. 56). With higher magnification (240 diameters) the cross-striation is distinctly seen and occasionally pale nuclei and refracting granules. The presence of numerous granules within the muscle-fibers is probably an indication of active metabolic processes. Where the muscle-fibers are cut across, the muscle-substance not infrequently protrudes from the sarcolemma.

(b) *Of man*.—I have found beautiful striated fibers in muscles taken from the human cadaver injected with carbolic acid. To preserve, stain under the cover-glass with picrocarmine (p. 53) for about five minutes, then displace the staining fluid with diluted glycerol.

No. 23.—*The sarcolemma*.—Treat preparation No. 22 *a* with a couple of drops of ordinary water. In from two to five minutes it will be seen, with the low power (50 diameters), that the sarcolemma is raised from the muscle-substance in the form of transparent vesicles; at some places, where the torn muscle-substance has retracted, the sheath appears as a delicate line spanning the interval (Fig. 56, 1, *s s'*).

No. 24.—*Muscle nuclei*.—Prepare muscle-fibers after No. 22 *a*; treat them with a drop of acetic acid (p. 53). The shrunken but sharply outlined nuclei, with the lower power, have the appearance of dark, spindle-shaped streaks (Fig. 56, 3).

No. 25.—*Fibrillæ*.—Place a fresh muscle of a frog in 20 c.c. of 0.1 per cent. chromic-acid solution (p. 21). In about twenty-four hours the tissue can be teased in a drop of water and fibers will be found, the ends of which have separated into fibrillæ (Fig. 54, *A*). If it is desired to make a permanent preparation, place the muscle in water for one hour, then in 20 c.c. of 33 per cent. alcohol, ten or twenty hours; tease at once or preserve in 70 per cent. alcohol until wanted and then isolate (p. 29). If the chromic acid be removed by allowing the tissue to remain in alcohol (frequently renewed) for several weeks, the teased preparation may then be stained with picrocarmine in the moist chamber and this replaced by glycerol (p. 53). Beautiful fibrillæ can also be obtained by teasing the muscles of larval salamanders that have been fixed according to technic No. 1 and stained in bulk in borax-carmine (p. 40). Pieces of such muscle are transferred from absolute alcohol to carbol-xylol and teased on a slide in a drop of the latter. Examine with the low-power, without a cover-glass, and when individual fibrillæ are visible, remove the excess of carbol-xylol with filter paper and mount in xylol-balsam.

No. 26.—*Ends of muscle-fibers*.—Place the fresh gastrocnemius muscle of the frog in 20 c.c. of concentrated potash-lye, and treat further like No. 21 *a*. With low magnification one sees the ends of the muscle-fibers and numerous swollen, shining nuclei (Fig. 56, 3).

No. 27. — *Branched muscle-fibers*. — Remove the tongue from a recently killed frog (it is attached in front to the lower jaw, is free behind) and place it in 20 c.c. of pure nitric acid, to which about 5 gm. of potassium chlorate have been added (some undissolved chlorate must remain in the bottom of the vessel). In a few hours, with glass rods, carefully transfer the tongue to 30 c.c. of distilled water, which must be frequently changed. In this the tissue may remain a week, though it can be used at the end of twenty-four hours. For this purpose put it in a test-tube half filled with water and shake it several minutes; the tongue will fall to pieces. Turn the contents of the test-tube into a capsule and in an hour or later place a little of the sediment that has been deposited in the meanwhile in a drop of water on a slide. The tissue may be further isolated with the teasing needles, but in most cases this is superfluous. Examine with the low power. Stain under the cover-glass with picrocarmine (p. 53). Mount in dilute glycerol (p. 50). (Fig. 56, 4.)

IV. THE NERVE TISSUES.

The elements of the nerve tissues, in an early embryonal stage, are without exception cells having a spherical form, the so-called *neuroblasts*. In the course of development they become pyriform, the narrow end grows out as a long, thin process, often extending the length of a meter, and terminates in a free, branched end; it is named the *nerve-process*. From the body of the cell, now termed a *nerve-cell*, other processes may arise, which, however, are short and divide dichotomously; they are called *dendrites*. Delicate lateral branches, the *collateral fibers*, may grow from the nerve-process. The nerve-cell and the nerve-process together form a cellular unit, the *neuron* (neurodendron). The dendrites and collaterals are to be regarded as secondary processes of the neuron.

The nerve-process may remain naked throughout its course, or it may receive different sheaths; these are the *neurilemma*, or sheath of Schwann, and the *medullary sheath*.* Both clothe the nerve-process only in a portion of its length. There are stretches in which the nerve-process is entirely without covering, is naked (Fig. 58, *a*); stretches in which it is enveloped only by the neurilemma (Fig. 58, *b*) or only by the medullary sheath (Fig. 58, *c*), and, finally, stretches in which both sheaths are present (Fig. 58, *d*); in this case the medullary sheath is always the innermost envelope, lies directly upon the cylindrical nerve-process, and is itself ensheathed by the neurilemma. The nerve-process always occupies the longitudinal axis; hence the name *axis-cylinder*.

* The neurilemma is of connective tissue origin, the *myelin* forming the medullary sheath (p. 121), according to recent research, originates in the blood and is conducted to the nerve-processes by connective-tissue cells,—in the central nervous system by glia-cells.

Owing to the often great length of the nerve-process, it is not possible to investigate the neuron as a whole. As a rule, it is seen only in fragments, either the nerve-cell or the nerve-process. This explains the former division of the elements of the nerve tissues into *nerve-cells* and *nerve-fibers*, the latter being the nerve-processes with their sheaths. There are no independent nerve-fibers, each so-called fiber is a process of a nerve-cell; if the connection between the fiber and the cell is broken, the fiber dies cellulifugalward from the point of solution of continuity. For practical reasons the old classification of nerve-cells and nerve-fibers is retained.

A. NERVE-CELLS.

Nerve-cells (ganglion-cells) are found in the ganglia, in the organs of special sense, along the course of cerebrospinal, as well as sympathetic nerves, but principally in the central nervous system. They differ greatly in size (4 to 135 μ and more) and in form. There are spherical and spindle-shaped nerve-cells and irregularly-stellate forms are very common; the latter are those in which the protoplasm sends off several processes. Nerve-cells having two processes are termed *bipolar*, those having several processes *multipolar* ganglion-cells (Fig. 59). There are also *unipolar* nerve-cells; these occur in the sympathetic nerve of amphibians and universally in the olfactory mucous membrane (Fig. 348). They actually possess but a single process. The nerve-cells of the spinal ganglia, on the other

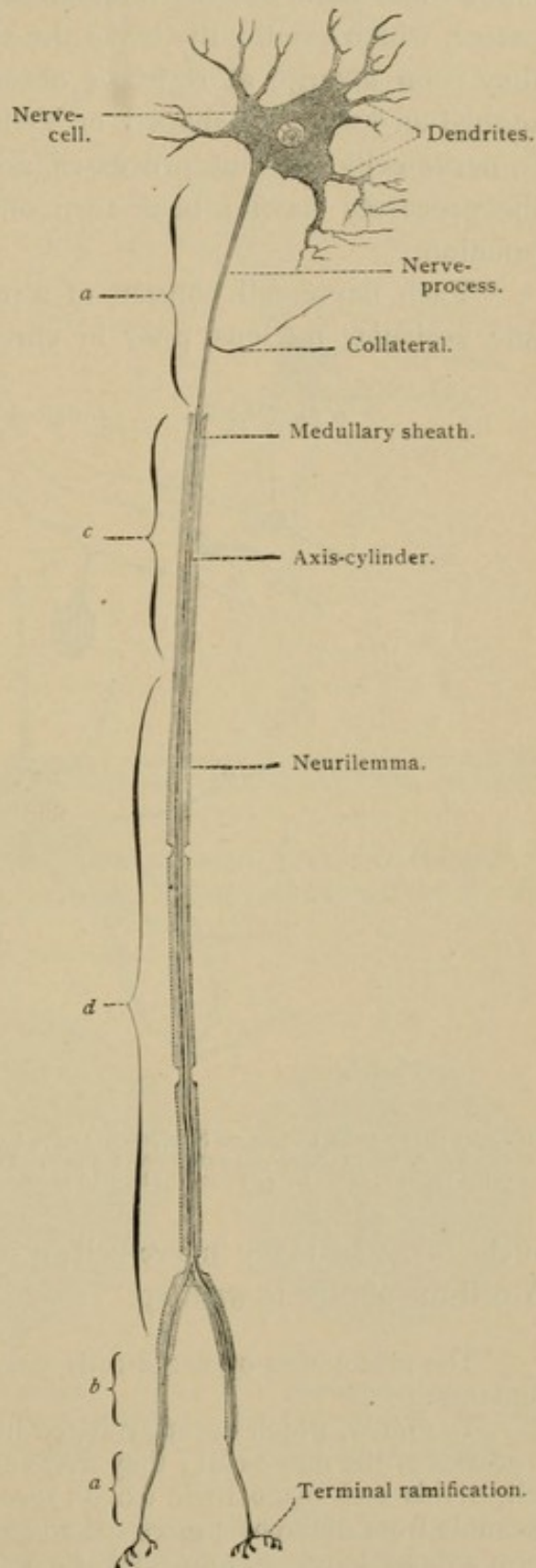


FIG. 58.—DIAGRAM OF A NEURON.

hand, are only apparently unipolar; in developmental epochs they were bipolar and then became unipolar by the gradual approach of the processes, which eventually leave the cell by a common stalk, from which they soon diverge at right or obtuse angle. These are the cells described as having T-shaped or Y-shaped processes. Apolar cells, that is, nerve-cells without processes, are either immature forms or artifacts, the processes having been torn off in the manipulation required for isolation.

Each nerve-cell consists of a protoplasm and of a quite characteristic vesicular nucleus, poor in chromatin and enclosing a conspicuous

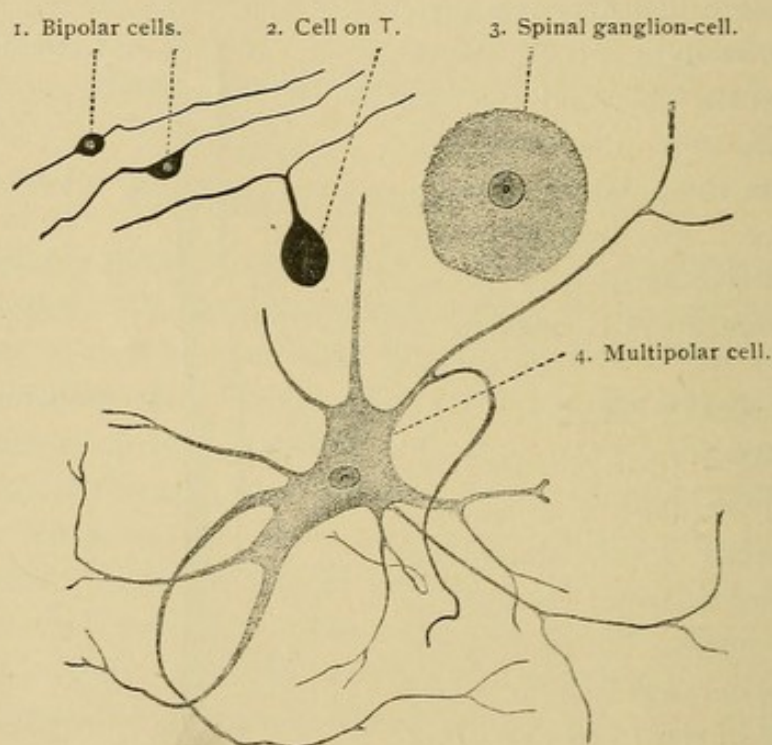


FIG. 59.—DIFFERENT FORMS OF NERVE-CELLS. $\times 236$. 1. From the spinal ganglion of a six-day embryo chick. 2. From the spinal ganglion of a calf. Technic No. 86. 3. Of man; the nerve-process torn off. Technic No. 28. 4. From the human spinal cord. Technic No. 29.

nucleolus. In many nerve-cells a centrosome has been demonstrated. A cell-membrane is wanting.

The protoplasm of nerve-cells possesses a very complicated structure. It contains:

1. *Fibrils*, which occur arranged in bundles, as well in the body as in the processes of the nerve-cell; they may enter from one process, simply pierce the cell, divide and make their exit in several processes; or, reversed, they may assemble from different processes and pass out in a *single* process, or they may form a dense tangle—more rarely an actual network—in the body of the cell. These fibrils are to be regarded as the conducting nervous elements, but whether they are the only ones is doubtful, for it is well known that conduction can also be effected without demonstrable nerve-fibrils, simply by the protoplasm.

2. *Delicate canaliculi*, the trophospongium (Fig. 62), and the apparatus reticulare (Fig. 63), see page 64. In the lower vertebrates canaliculi containing blood-cells have been found.

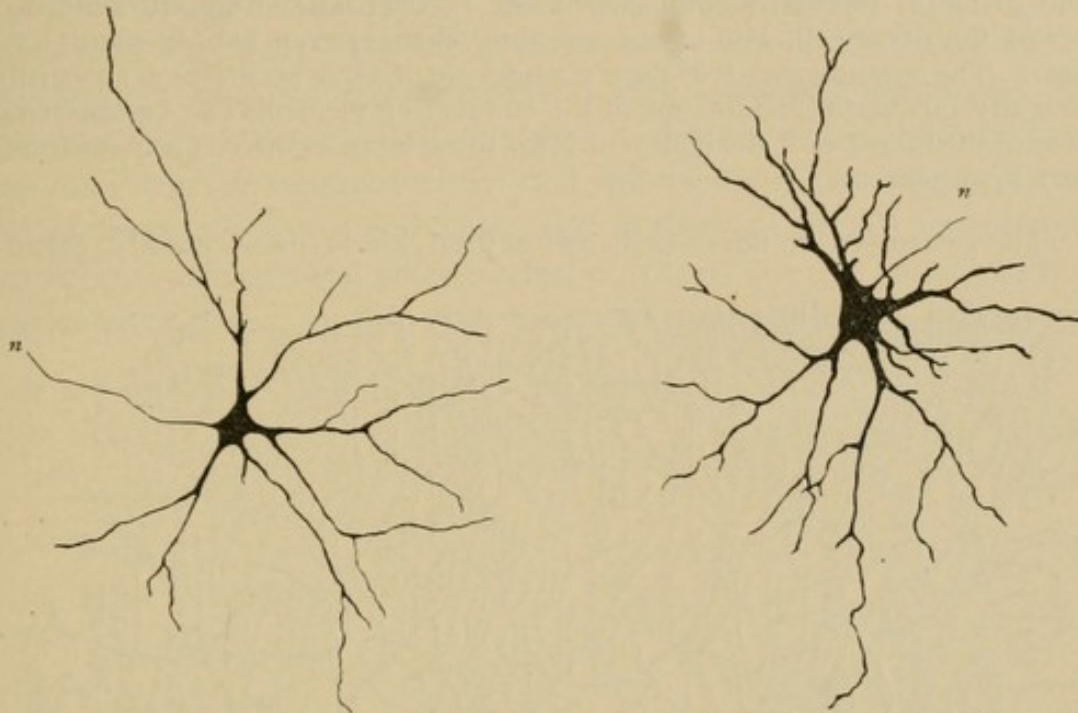


FIG. 60.—TWO FORMS OF MULTIPOLAR NERVE-CELLS FROM THE VENTRAL HORN OF THE SPINAL CORD OF A NEWBORN RABBIT, SHOWING THE RICHLY BRANCHED PROTOPLASMIC PROCESSES. *n*. Nerve-process. $\times 60$. Technic No. 76. (Schaper.)

3. *Granule groups*, which consist partly of pigment, partly of substances that can be made visible only by special methods. These substances, the *Nissl's*

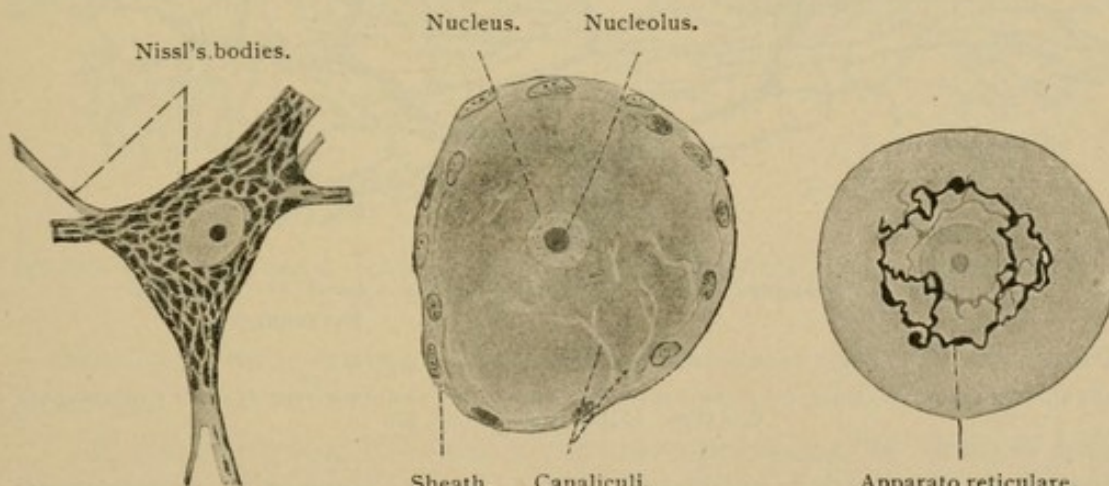


FIG. 61.—GANGLION-CELL OF THE SPINAL CORD OF A CHILD. $\times 430$. Technic No. 30.

FIG. 62.—SPINAL GANGLION-CELL OF AN ADULT CAT. $\times 430$. Technic No. 30.

FIG. 63.—SPINAL GANGLION-CELL OF A NEWBORN KITTEN. Copy after Golgi.

bodies ("tigroid"), that do not belong to all nerve-cells, are very differently shaped,* are sometimes spherical, sometimes polyhedral aggregations of gran-

* This varied structure of the protoplasm makes it intelligible that, despite the numerous interlacing ramifications of the nerve-cells and their processes, the excitation is not a diffuse but an orderly one, running in definite pathways.

ules, sometimes spindles and bands, and fill the spaces between the strands of fibrils, the canaliculi of the trophospongium. They occur also in the dendrites (see below), but extremely seldom in the nerve-process. The bodies of Nissl are in so far of especial significance, since in over-fatigue and in abnormal states of the nerve-cell, also in old age, they change, even almost wholly disappear. The circumstance that these changes are of early occurrence, appearing before any functional disturbance of the conducting elements can be observed, indicates the function of the bodies of Nissl to be more nutritive (perhaps formative) than nervous.

The processes of nerve-cells are of two kinds, most readily distin-

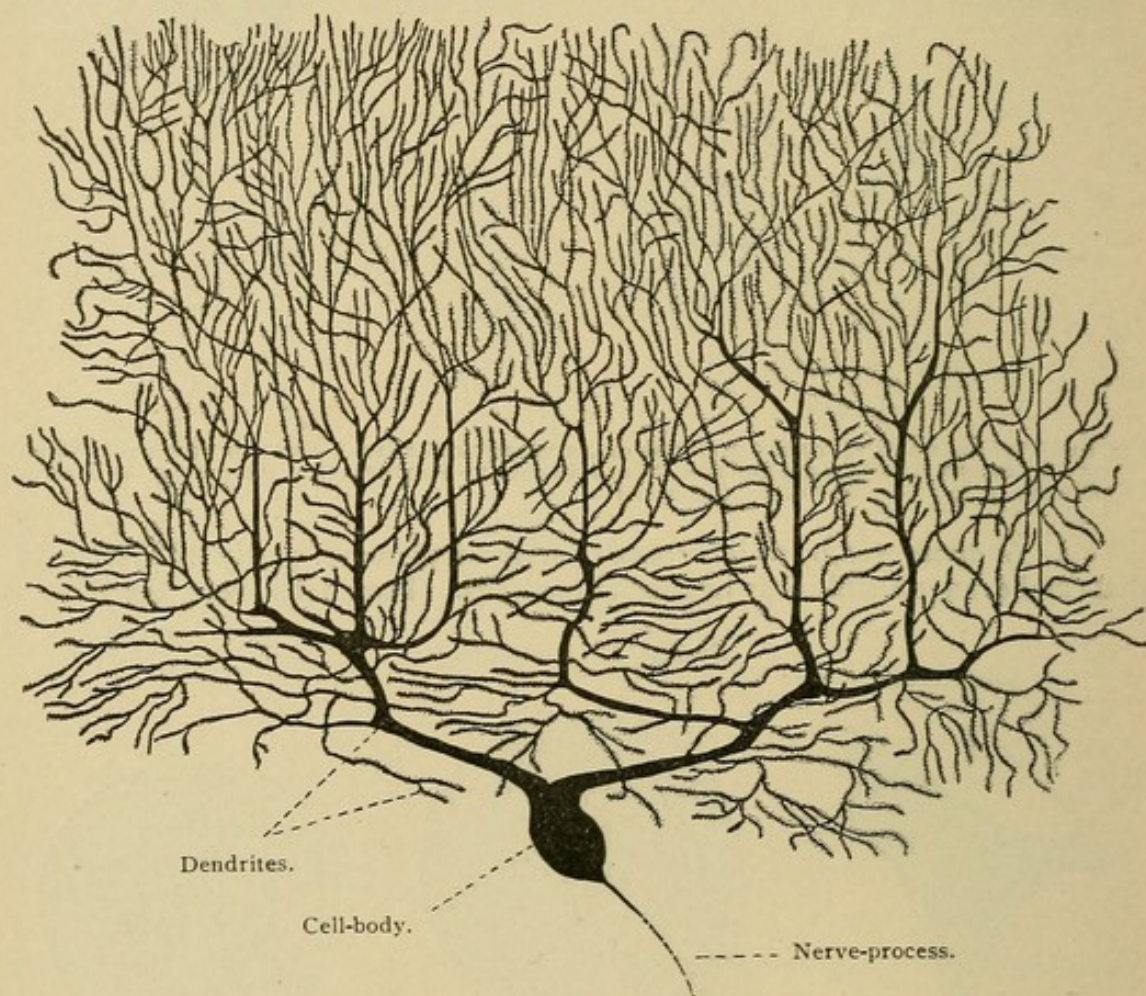


FIG. 64.—NERVE-CELL (CELL OF PURKINJE) FROM A SECTION THROUGH THE HUMAN CEREBELLAR CORTEX. $\times 180$. Technic No. 80.

guished in multipolar nerve-cells: 1. One process, the *nerve-process* (axis-cylinder, axon, Fig. 64), the only one of its kind;* it is the

* It is said there are cells with *several nerve-processes* (e. g., Cajal's cells in the cerebral cortex). In bipolar ganglion-cells, both processes of which become axis-cylinders of medullated nerve-fibers (spinal ganglion-cells of lower vertebrates and of embryos), the central process going to the central nervous system corresponds to the nerve-process, the peripheral process to a dendrite. This conception is supported by the observation that in the bipolar cells of the cochlear nerve the peripheral process develops precisely like a dendrite and only later assumes the character of a nerve-fiber.

first outgrowth of the embryonal spherical nerve-cell and is characterized by its hyaline, smooth-bordered appearance; it conducts from the cell—cellulifugal. 2. Many processes, the *dendrites* (protoplasmic processes, Fig. 64); they are a later outgrowth of the nerve-cell, are thicker, granular or finely striated, and often beset with varicosities; they conduct toward the cell—cellulipetal. The dendrites divide repeatedly and so can form an extraordinarily rich arborization, the finest twigs of which terminate in free ends (Fig. 64); in this way the cell-body acquires an enormous superficial enlargement, which, on the one hand, exalts the sustentative power, on the other hand, the susceptibility to nerve stimuli

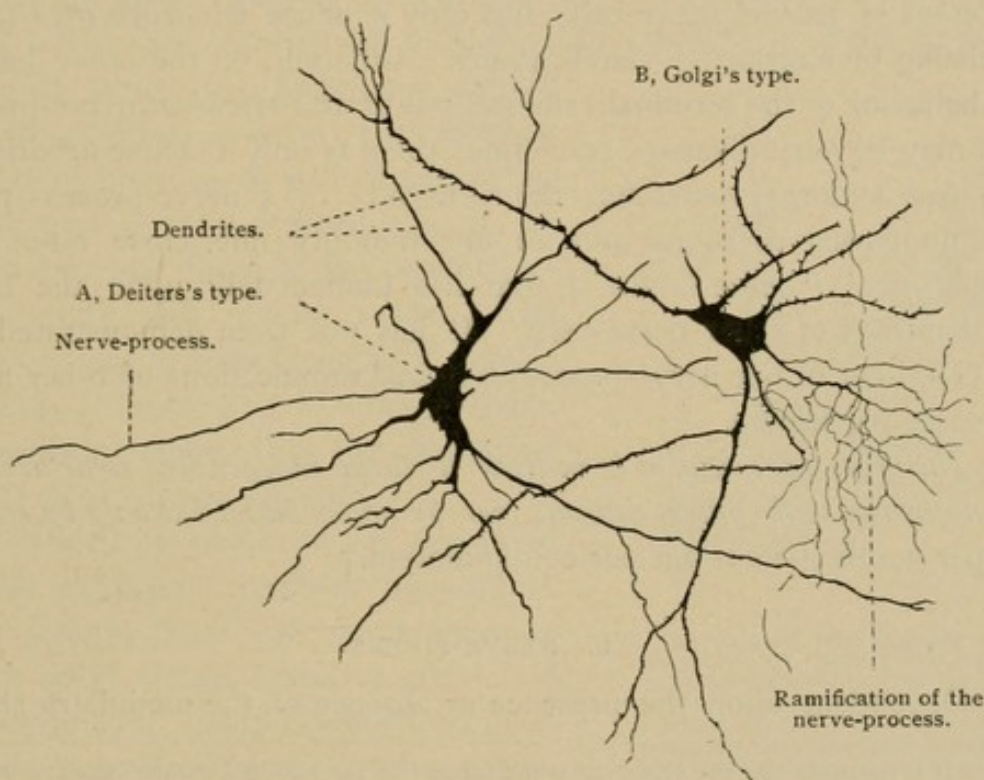


FIG. 65.—TWO NERVE-CELLS. $\times 200$. A, From a section of the spinal cord of a human embryo six months old. B, From a section of the brain of a cat. Technics No. 76 and No. 79.

—these latter being transmitted by adjacent terminal ramifications of nerve-processes.

According to the behavior of the nerve-processes two types of ganglion-cells are distinguished.

1. In *cells of the first type* (Deiters's type) the nerve-process becomes the axis-cylinder of a medullated nerve-fiber, which after running a long course, often measuring many centimeters, always terminates in an extremely delicate ramification; these cells are described as nerve-cells with a long nerve-process (Fig. 65).

During its course such a nerve-process gives off a number of delicate, branching, lateral twigs ("collaterals," "paraxons"); by no means rarely

there also occurs a division of the nerve-process in two equal nerve-processes (see The Spinal Cord, "plurifunicular cells," p. 192, remark *).

2. In *cells of the second type* (Golgi's type) the nerve-process, by continual division, resolves in a nervous ramification in the vicinity of the cell; these cells are called nerve-cells with a short nerve-process (Fig. 65).

Whether the nerve-processes belong to the one or the other type, their terminal ramifications are free, in the respect that they never directly pass into the terminal ramifications of other nerve-processes or dendrites. Accordingly there is no *nervous reticulum* formed of the processes of *several* nerve-cells, but only a dense felt-work (*neuripilem*) consisting of interlacing ramifications. Different, on the other hand, is the behavior of the terminal ramifications of an *individual* nerve-process. This may be very diverse; sometimes there is only a coarse arborization with free endings, sometimes the extremity of a nerve-process passes with numerous divisions into an uncommonly fine, close reticulum* ("Golgi-net," "nerve-lattice"), that lies immediately upon the bodies and dendrites of other nerve-cells. It has not been demonstrated that in a Golgi-net of one nerve-process terminal ramifications of other nerve-processes participate.

Therefore each neuron is in itself a closed, independent structure, that communicates with other neurons, not by anastomoses but only by contact, not per continuitatem but per contiguitatem.†

B. NERVE-FIBERS.

Dependent upon the presence or absence of the medullary sheath,

* It is probable that the Golgi-net is not formed of the terminal ramifications themselves, but consists of a peculiar substance in which the fibrils of the ramifications run; the proof is still required that the fibrils form a true net. Each nerve-cell of the central nervous system is provided with a reticulum of Golgi; a single nerve-cell may be embraced by several variously fashioned terminal ramifications of different nerves.

† Whether this statement is invariably valid is doubtful. In the retina and in the electric organ of the torpedo true nets formed of processes of several nerve-cells have been described; von Thanhofer demonstrated to me preparations that show distinct, delicate anastomoses between cells of the spinal cord. Such a connection does not affect the neuron theory any more than the intercellular bridges do the cell theory. A few authors describe the Golgi-net as "diffuse" and thereby endeavor to rehabilitate the old, long-abandoned hypothesis that the nerve-fibers arise from a general nerve-plexus. Therewith the nerve-cells are pushed aside as secondary elements, having no significance in nervous functions. This view, which is in glaring opposition to the neuron theory, is still lacking in sound anatomic, as well as experimental, evidence. When in the following nervous networks and plexuses are spoken of it is to be thus understood, that from nerve-fiber *bundles* a few nerve-fibers branch off to join other bundles. In this transfer of fibers a direct transition of one nerve-fiber into another, as a rule, does not occur.

nerve-fibers are divided into *medullated* and *nonmedullated*. Each division is susceptible of a subdivision dependent on the presence or absence of the neurilemma.

1. Nonmedullated Nerve-fibers.

(a) *Without a Neurilemma.*

These fibers consist of the axis-cylinder (nerve-process) alone ; they are therefore described as "naked" and are found in the olfactory nerve, where, grouped in bundles, they are held together by connective tissue. Similar are many fibers of the sympathetic nerve, the so-called *Remak's fibers*, which are transparent, cylindrical or bandlike in form, from 3 to 7 μ wide, about 2 μ thick, and exhibit faint longitudinal striation ; they likewise consist of bundles of naked axis-cylinders* and a very delicate sheath, on which here and there lie flat connective-tissue cells with oblong nuclei. The sheath is said to correspond to the endoneurium or, according to some authors, to the neurilemma. (See the chapter on the Central Nervous System.)

While the fibers so far described exhibit the same structure throughout their length, there are nerve-fibers of which only certain divisions are naked axis-cylinders. Such divisions occur as peripheral endings of the nerves of special sense and of sensory as well as motor nerves ; also the first division of the nerve-process proceeding from the nerve-cell is a naked axis-cylinder (*cf.* Fig. 58).

(b) *With a Neurilemma.*

Fibers consisting in their entire length of an axis-cylinder and a neurilemma are found in many invertebrates and in cyclostoma. They occur as divisions in the course of the cerebrospinal nerve-fibers (Fig. 58, *b*).

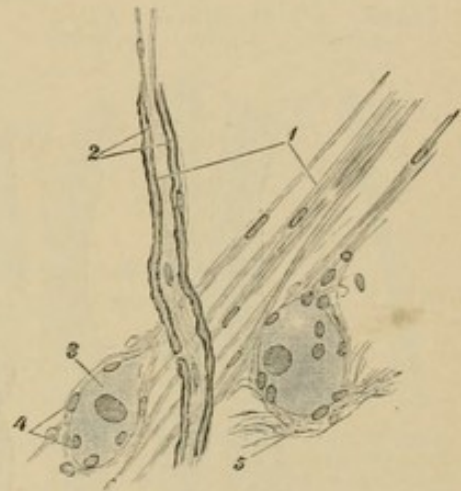


FIG. 66.—TEASED PREPARATION OF THE SYMPATHETIC NERVE OF A RABBIT. 1. Nonmedullated, 2. thin medullated nerve-fibers ; 3, ganglion-cell ; the large nucleus has lost its characteristic appearance in consequence of the treatment with osmic acid ; 4, nuclei of connective-tissue capsule ; 5, fine connective-tissue fibers. $\times 240$. Technic No. 36.

* By Remak's fibers some authors understand, not bundles of naked axis-cylinders, but individual axis-cylinder processes of sympathetic ganglion-cells.

2. Medullated Nerve-fibers.

(a) *Without a Neurilemma.*

Among these are no fibers that possess the medullary sheath in their entire length; this always clothes only one division of the axis-cylinder. Fibers consisting of axis-cylinder and medullary sheath alone (Fig. 58, c) occur only in the central nervous system.

(b) *With a Neurilemma.*

Medullated fibers possessing a neurilemma are found in the trunks and branches of the cerebrospinal nerves, also in the sympathetic nerve, and vary in thickness from 1 to 20 μ .

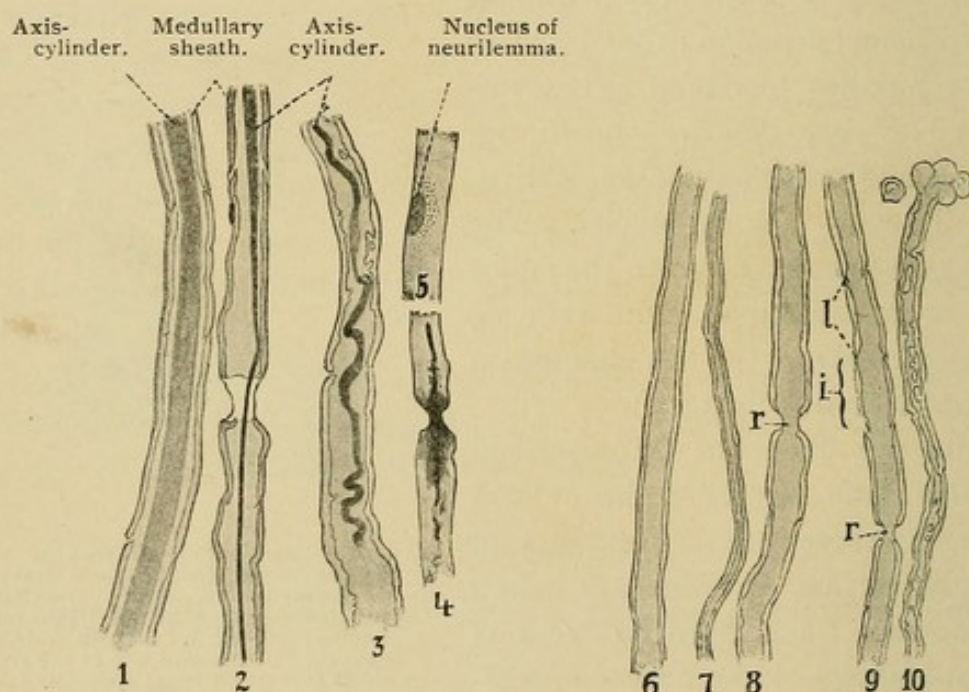


FIG. 67.—MEDULLATED NERVE-FIBERS FROM THE SCIATIC NERVE OF A FROG. $\times 280$. 1, Normal, 2, shrunk, 3, twisted axis-cylinder; 4, node of Ranvier; 5, neurilemma with nucleus. Technic No. 33. 6, 7, 8, and 9, Fresh medullated nerve-fibers; 10, post-mortem distortion of medullary substance; *r*, annular constriction; *l*, notches of Lanterman; *i*, cylindro-conical segment. Technic No. 31 a.

The thickness of the nerve-fiber bears no relation to its motor or sensory nature, but appears to be determined by its length: the longer its course the thicker is the fiber. Division of the medullated fibers occurs (1) throughout the central nervous system, principally where the collateral fibers diverge at right angles into the white substance, and (2) in the peripheral nervous system shortly before their ultimate distribution (Fig. 58).

The medullated nerve-fibers have a brief lease of life. They degenerate by a gradual breaking down of the medullary substance and axis-cylinder into a granular mass containing numerous nuclei; in this

mass both parts are regenerated, the axis-cylinder probably by outgrowth of the nerve-process of the nerve-cell.

Regarding their *finer structure* and peculiar properties, the three constituent parts of nerve-fibers comport themselves in the following manner :

1. The *axis-cylinder*, the most essential part of every nerve-fiber, exhibits a delicate longitudinal striation, which is the optical expression of its fibrillar structure. Each fibrilla represents a special conducting path and is cemented to neighboring fibrillæ by a small amount of highly aqueous, finely granular or homogeneous interstitial substance, the *neuroplasm* (axoplasm).

2. The *medullary sheath* consists of a semi-fluid, highly refracting, fatty substance, the *myelin*, which imparts to fresh medullated fibers the appearance of entirely homogeneous, glistening, opaque cylindrical threads, the structure of which can only be perceived by the help of reagents. Often it is seen that the medullary sheath is not continuous, but is divided at slightly irregular intervals by oblique incisions, the *Lantermann's notches*, into the "cylindro-conical" segments, which seem to be united to each other by cement-substance* (Fig. 67, 9). On treatment with various reagents the totally homogeneous myelin of living nerve-fibers undergoes partial transformation as it dies; at first the nerve-fiber exhibits a double contour,† later the myelin rolls up into peculiar spherical masses (Fig. 67, 10).

At certain places marked by ring-like constrictions the medullary sheath is wanting, so that the axis-cylinder and the neurilemma come into contact. These annular constrictions are termed *nodes of Ranvier* (Fig. 67, *r*); they are the localities where the nutritive fluid can approach the axis-cylinder. In the vicinity of the nodes the axis-cylinder is often provided with a biconical enlargement (Fig. 69), due to a local accumulation of neuroplasm. The treatment with silver

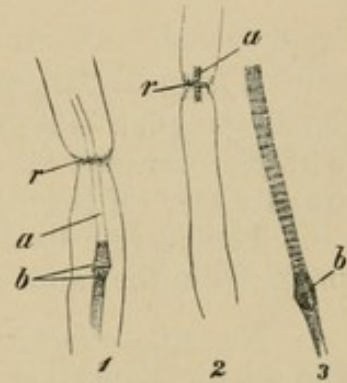


FIG. 68.—MEDULLATED NERVE-FIBERS OF A FROG, TREATED WITH SILVER NITRATE SOLUTION. $\times 560$. 1. At *r*, node of Ranvier; *a*, axis-cylinder, of which only a small extent is silvered; *b*, biconical swelling displaced downward owing to manipulation. 2. Axis-cylinder with the silvered portion *in situ*, at *a*. 3. Axis-cylinder with cross-markings. The myelin is not visible by this method. Technic No. 35.

* Many authors regard these notches as artifacts; they appear very quickly, even in fibers just removed from the animal.

† Thence the old designation: "double-contoured or dark-bordered nerve-fiber." However it is questionable whether the double contour lines are a coagulation product, for they have been seen also in the living animal. In this case the two contours would correspond to the outer and inner boundaries of the sheath.

nitrate solution reveals the neuroplasm at the nodes (Fig. 68, *r*), as well as a very distinct transverse marking on the adjacent portions of the axis-cylinder.* Each peripheral medullated nerve-fiber is provided with nodes, at intervals of from 0.08 mm. in thin to 1 mm. in thick fibers, dividing it into "interannular" segments.

3. The *neurilemma*, or sheath of Schwann, is a delicate, structureless membrane, against the inner surface of which lie elliptical nuclei surrounded by a minimal amount of protoplasm (Fig. 67, 5).

The union of the two elements of the nerve tissues in the peripheral nervous system is secured by means of connective tissue, which contains the ramifications of the blood-vessels; in the central nervous system they are united, not only by connective tissue, but by the *neuroglia* (p. 201).

TECHNIC.

No. 28.—*Ganglion-cells, fresh*.—Tease a small piece of the Gasserian ganglion in a drop of salt solution and stain under the cover-glass with picrocarmine for two minutes (p. 53). The processes of the cells usually tear off (Fig. 59, 3).

The nerve-cells of the cerebral and the cerebellar cortex may be prepared in the same way; the processes likewise are easily lost.

No. 29.—*Multipolar ganglion-cells of the spinal cord*.—Remove with the scissors as well as possible the white substance of a fresh spinal cord and place the gray residue, divided into pieces from 1 to 2 cm. long, in highly diluted chromic acid solution (5 c.c. of the 0.05 per cent. solution, page 21, to 45 c.c. of distilled water). After from 1½ hours to 8 days (the time varies according to the outside temperature) the spinal cord is macerated to a soft mass, that is to be carefully transferred into 20 c.c. of undiluted neutral carmine solution (p. 24). The pieces remain in the stain for from 10 to 20 hours, then are placed in 50 c.c. of distilled water, in order to wash out some of the color, and after 5 minutes the tissue is ready to spread in a thin film on a dry slide. With a little practice the ganglion-cells can be distinguished by their brilliantly stained red nuclei; of the bodies and processes nothing is visible to the unaided eye. Let the film dry thoroughly and then cover it over with a cover-glass on the under side of which a drop of xylol-balsam is suspended (Fig. 59, 4).

No. 30.—*Nissl's bodies*.—For the exhibition of Nissl's bodies fix and harden in absolute alcohol (p. 31) a piece 1 cm. long of the spinal cord freed from the pia (the lumbar enlargement is best, owing to its large cells) and embed it in paraffin (see Microtome Technic). Microtome sections are placed in 5 c.c. of xylol, then transferred to an equal quantity of absolute alcohol and after a minute to 70 per cent. alcohol. From this the sections are put into 5 c.c. of a 2 per cent. aqueous solution of

* These striæ are artifacts; for their significance, see p. 45, remark *.

fuchsin, that is heated over a flame until bubbles rise. Then the somewhat crinkled sections are lifted with a needle into a capsule containing a mixture of 9 c.c. of absolute alcohol and 1 c.c. of anilin oil, in which the decolorization occurs. In about 10 minutes renew the alcohol-anilin mixture. After 5 minutes transfer the sections to absolute alcohol and after 1 minute to xylol. Mount in xylol-balsam. The preparations keep for a long time. (Fig. 61.)

The *canaliculi* of the ganglion-cells can be shown by fixing in sublimate-picric acid (equal parts of No. 20, p. 22, and No. 26, p. 22). Further treatment as with Zenker's fluid (p. 32). Stain *thin* microtome sections with Heidenhain's iron-hematoxylin (p. 44) and examine with an immersion lens.

No. 31.—*Fresh medullated nerve-fibers*.—Expose the sciatic nerve of a frog just killed. With delicate scissors cut it at the level of the popliteal space and about 1 cm. higher. Isolate in a drop of salt solution.

No. 31 *a*.—Better still, tease on a dry slide by the "half-drying" method. Hold the lower end of the nerve with one needle, with another needle separate the nerve bundles along half the length of the nerve; a thin shining membrane will span the interval between the separated bundles. Add a drop of salt solution and apply a cover-glass. The membrane contains numerous isolated nerve-fibers. The manipulation must be done very rapidly (in about fifteen seconds), so that the nerve-fibers do not become dry (Fig. 67, 6, 7, 8, 9).

No. 32.—*Alterations of the medullary sheath*.—Treat No. 31 *a* with water, place a drop at the edge of the cover-glass and let it flow under. In a few minutes the formation of the myelin drops begins (Fig. 67, 10).

No. 33.—*The axis-cylinder*.—Tease dry (like No. 31 *a*) and stain with methylene blue (p. 42); the nodes of Ranvier stain first, and often so deeply that the axis-cylinder cannot be distinguished there (Fig. 67, 4). Frequently the axis-cylinder shrinks and becomes displaced within the medullary sheath, or it becomes convoluted (Fig. 67, 2, 3). On the addition of glycerol the medullary substance can no longer be distinctly recognized, but the nuclei of the neurilemma are often rendered plainly visible (Fig. 67, 5). If it is desired to preserve in xylol-balsam drain off the solution of ammonium picrate after about 18 hours, cautiously rinse with water, and let the preparation dry in the dark. Then mount straightway in balsam.

No. 34.—*Exhibition of the axis-cylinder with chromic acid*.—Expose the sciatic nerve of a rabbit recently killed, *being careful not to touch it*; place a match-stick parallel to the long axis of the nerve and secure it by means of ligatures at the upper and lower ends; cut the nerve on the farther side of each ligature and place it, with the wood, in 100 c.c. of a 0.1 per cent. chromic-acid solution (p. 21).

In about twenty-four hours cut the ligatures and tease a piece of the nerve, from 0.5 to 1 cm. long, separating it into bundles, not fibers.

Put the bundles back into the chromic-acid solution; after twenty-four hours transfer them to 50 c.c. of distilled water, and after two or three hours to 30 c.c. of gradually strengthened alcohols to harden (p. 35). It is advantageous to leave the bundles for a long time, one to eight

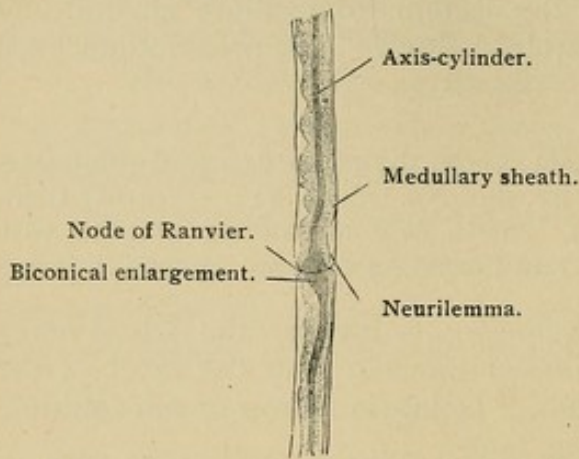


FIG. 69.—NERVE-FIBER OF A RABBIT. $\times 560$.

weeks, in 90 per cent. alcohol, as they are then more readily stained. After the hardening the bundles are to be teased in a drop of picrocarmine, placed in the moist chamber, and after the staining is completed (which according to the length of time the tissue was allowed to harden in the alcohol requires from one-half to three days), preserved in acidulated glycerol (p. 53). The nodes of Ranvier are not so distinct as in fresh and in osmium preparations, but appear as delicate transverse lines (Fig. 69). The somewhat shrunken axis-cylinder and the nuclei are stained a fine red.* Not seldom the axis-cylinder is displaced, so that the biconical swelling is not at the node, but above or below it.

No. 35.—*Nodes of Ranvier and axis-cylinders.*—Preliminary: Add 10 c.c. of a 1 per cent. solution of silver nitrate to 20 c.c. of distilled water. Kill a frog, open the abdomen by a crucial incision, turn out the viscera, and expose the nerves descending on each side of the vertebral column. Wash out the abdominal cavity with distilled water and pour over the nerves about one-third of the silver solution. After two minutes carefully cut out the delicate nerves, put them for a half-hour in the remainder of the silver solution, and *place them in the dark*. Then transfer them to 10 c.c. of distilled water, in which they may remain for from one to twenty-four hours. If a nerve is now examined in a drop of water, with the low power, the delicate sheaths composed of flat cells (see the cerebrospinal nerves) and numerous pigment cells will be seen; frequently a blood-vessel lies along the nerve. Now tease the nerve, cover it and place a small drop of dilute glycerol at the edge of the cover-glass. On examination with the high power little will be seen of the nodes and axis-cylinder, but if the preparation be exposed for several hours to daylight (or a few minutes to sunlight) the reaction takes place and the parts become silvered. The biconical swelling on the axis-cylinder often becomes displaced in teasing and is not always readily found by the beginner. With a little practice pictures like that shown in Fig. 68 are easily obtained.

* The intensity of the color depends on the quality of the picrocarmine, which unfortunately varies greatly; it can be improved by placing the glass containing the teased bundles and the stain on the embedding oven.

No. 36.—*Nonmedullated nerve-fibers*.—Tease a portion of the pneumogastric nerve of a rabbit on a dry slide (No. 31 *a*), and add a few drops of a 0.5 per cent. osmic acid solution; in five or ten minutes the medullated nerve-fibers become blackened (which may be ascertained by examination with the low power). Remove the osmic-acid solution and add a few drops of distilled water, which should be renewed in five minutes. In five minutes more remove the water, add a few drops of picocarmine, apply a cover-glass, and place in the moist chamber for from twenty-four to forty-eight hours; then displace the picocarmine with acidulated glycerol (p. 53). The tissue may be teased again after the staining is completed, which is now more easily done because the elements are more distinctly seen. With high magnification the medullated nerve-fibers appear blue-black, the nonmedullated pale gray and finely striated longitudinally. The sympathetic nerve treated in the same way exhibits more numerous nonmedullated nerve-fibers. But this nerve is somewhat more difficult to find. Cut through the greater cornu of the hyoid bone, through the hypoglossal nerve, and push them aside; behind the pneumogastric nerve lies the sympathetic, which is recognized by its three or four mm. in size, ellipsoidal, yellowish, transparent superior cervical ganglion. If the piece of the nerve lying close under the ganglion be teased, ganglion-cells the majority of which contain two nuclei* will be obtained (Fig. 66); it is difficult to isolate the cells so that their processes can be distinctly seen.

* Accidentally in figure 66 only the less usual uninucleated ganglion-cell is represented.

II. MICROSCOPIC ANATOMY OF THE ORGANS.

I. ORGANS OF THE CIRCULATORY SYSTEM.

I. THE BLOOD-VESSEL SYSTEM.

The blood-vessels are composed of connective tissue, elastic fibers, and smooth muscle-fibers, which, mingled in very different relations, are arranged in strata. In general, a uniform disposition of the elements prevails in each stratum; longitudinal in the inner and the outer, circular in the middle stratum. An exception to this occurs in the complicated structure of the heart and in the simple structure of the capillaries.

THE HEART.

The wall of the heart consists of three membranes: (1) the endocardium; (2) the powerfully developed muscular layer, the myocardium; (3) the epicardium (visceral layer of the pericardium).

(1) The *endocardium* is a connective-tissue membrane, which contains smooth muscle-fibers and numerous elastic fibers. The latter are less well developed in the ventricles than in the auricles, where they form close-meshed networks or are blended in fenestrated membranes (Fig. 36). The free surface, directed toward the cavity of the heart, is clothed with a simple layer of irregularly polygonal epithelial (endothelial) cells.

(2) The *myocardium* consists of a long-meshed network of muscle-fibers (for their structure see p. 105) which are enveloped in a delicate perimysium; the course of the strands of muscle is very intricate. The musculature of the auricles is entirely separate from that of the ventricles. In the auricles an outer transverse layer common to both and an inner longitudinal layer independent in each (more particularly in the pectinate muscle of the right auricle) can be distinguished. In addition numerous small bundles pursue independent courses in other directions. The musculature of the ventricles is much more irregular; the bundles extend in every direction, often describing a figure-of-eight in their course.

Within the territory of the auricles * the perimysium contains many

* The muscle membrane of the auricular appendages, on the other hand, is poor in elastic fibers.

elastic fibers, which multiply in old age and which are connected with those of the endocardium and the epicardium; within the territory of the ventricles the perimysium contains no elastic elements, except those belonging to the adventitia of the myocardial blood-vessels. Between the auricles and ventricles lie firm tendon bands intermingled with elastic fibers, the *annuli fibrosi*, of which the right is stronger than the left. Similar but less developed tendons lie at the arterial orifices of the ventricles. Numerous ends of muscle-fibers or of the muscle net are inserted in these tendons.

Lateral union.

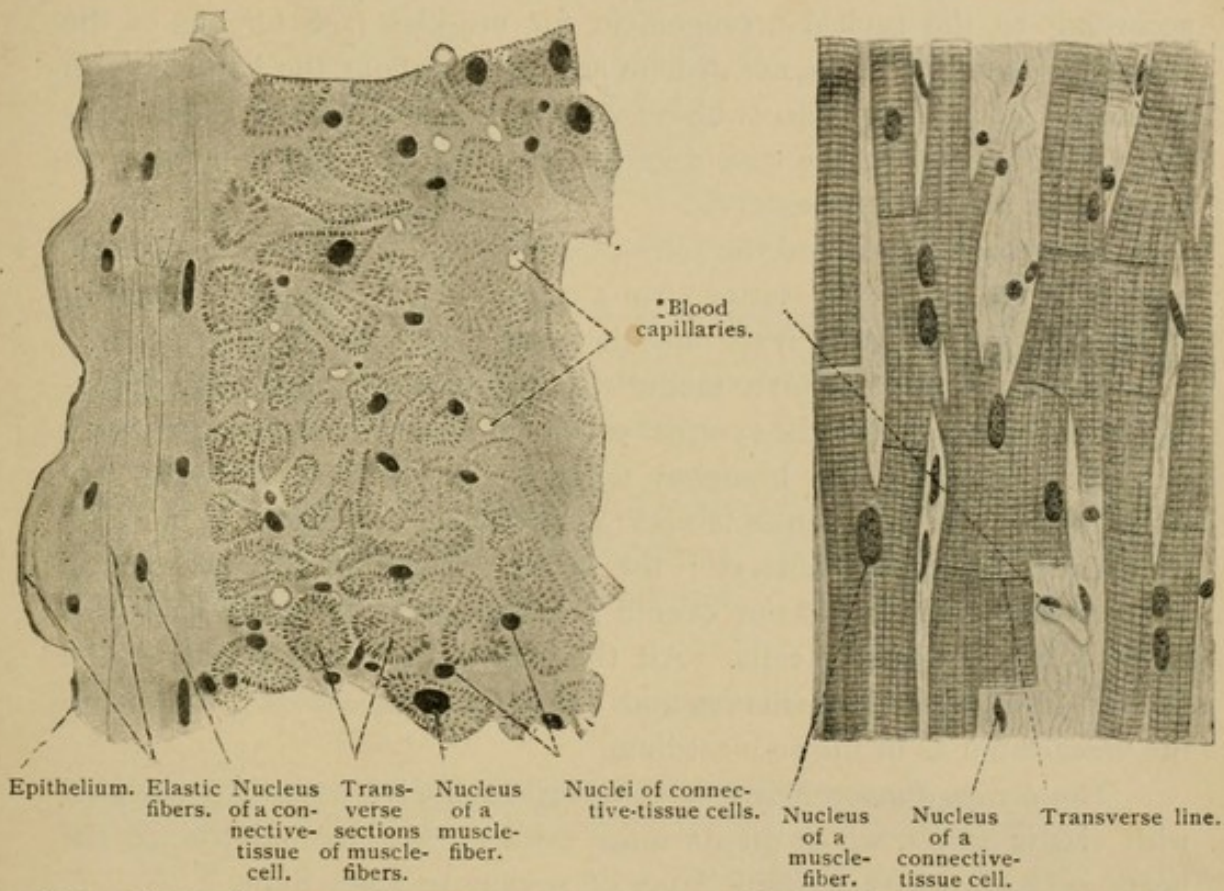


FIG. 70.—FROM A TRANSVERSE SECTION OF A PAPILLARY MUSCLE OF THE HUMAN HEART. $\times 360$. Technic No. 37.

FIG. 71.—FROM A LONGITUDINAL SECTION OF A PAPILLARY MUSCLE OF THE HUMAN HEART. $\times 360$. Technic No. 37.

(3) The *epicardium* is a connective-tissue membrane penetrated by elastic fibers and fat-cells, which on the outer surface is covered with a single stratum of squamous epithelium. The elastic fibers of the auricular epicardium pass over into the adventitia of the large veins; those of the ventricular epicardium are lost in the conus arteriosus and do not continue over into the aorta and the pulmonary artery.

The *valves* of the heart are composed of fibrous connective tissue, which is connected with that of the *annuli fibrosi*, and their surfaces are clothed by the endocardium. They contain muscle-fibers, but only in

the attached margin, and elastic fibers, which are especially abundant in the nodules of the free edges of the semilunar valves.

In many mammals (seldom in man, very fine in sheep) *Purkinje's fibers* are found in the heart wall, usually close under the endocardium; they are strings of clear cells. Their border layers contain cross-striped fibrillæ extending continuously through from cell to cell. Their nuclei multiply partly by mitosis, partly by amitosis; in the latter case cell division does not occur. These cells must be regarded as developmental forms of true cardiac muscle-fibers, since they gradually are transformed into such.

The numerous *blood-vessels* of the heart run in the musculature according to the typical arrangement for muscles (see Organs of the Muscular System). The epicardium and endocardium, the latter only in its deeper strata, also possess blood-vessels. The semilunar valves contain no blood-vessels, the cuspid valves have them only at their base, so far as their musculature extends.

The *lymph-vessels* and the juice-canals (see p. 100) occur in colossal number in the heart; the latter form a comprehensive system embracing all the free spaces between the muscle bundles and the blood-vessels.

The many *nerves*, partly medullated and partly nonmedullated, arising from the vagus and the sympathetic, form networks enclosing numerous ganglion-cells; the branches springing from these networks are partly motor (on every muscle-fiber a nerve-fiber terminates in a small eminence) and partly sensory; the latter end in terminal plexuses of different size, that spread out over a granular plate furnished with stellate (connective-tissue?) cells. All the terminal plexuses appear to be derived from medullated nerves and occur in great number, as well in the epicardium as in the endocardium.

The *pericardium* consists of compact connective tissue intermingled with elastic fibers, which on its inner surface, that directed toward the heart, is clothed with a simple layer of squamous epithelium.

THE ARTERIES.

The walls of the arteries consist of three coats: 1, the tunica intima; 2, the tunica media; 3, the tunica externa (adventitia). The elements of the tunica media are transversely disposed, those of the two other tunics chiefly longitudinally. The structure and thickness of these coats vary with the size of the artery. This makes their classification as small, medium, and large arteries desirable.

The *small arteries* are the precapillary arteries, the arterial vessels shortly before their transition into capillaries. The intima consists of elongated, spindle-shaped epithelial cells and a structureless elastic mem-

brane, the so-called *internal elastic membrane* (elastica interna), that in somewhat larger arteries assumes the character of a fenestrated membrane. The *media* is formed of a single layer (in somewhat larger arteries of several layers) of circularly disposed smooth muscle-fibers. The

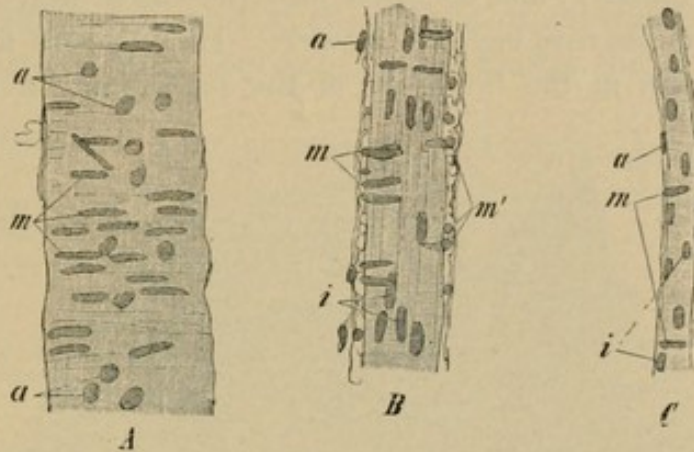


FIG. 72.—SMALL ARTERIES OF MAN. *i*, Nuclei of intima, the outlines of the cells are invisible; *m*, nuclei of circularly disposed muscle-fibers of media; *a*, nuclei of externa. *A*, artery with the surface in focus. *B*, artery with the lumen in focus; at *m'* the nuclei of the muscle-fibers of the media are seen in optical cross-section. *C*, small artery shortly before transition into capillaries; the media consists of a few isolated muscle-fibers. $\times 240$. Technic No. 39.

externa is composed of fine-fibered, longitudinally disposed bundles of connective tissue and delicate elastic fibers. It blends insensibly with the connective tissue supporting the arteries.

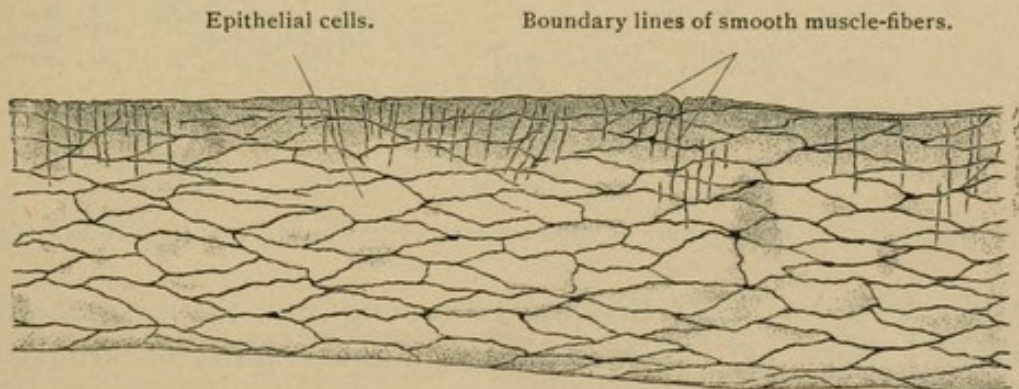


FIG. 73.—EPITHELIUM OF THE MESENTERIC ARTERY OF A RABBIT. Surface view. $\times 260$. Technic No. 40.

The *arteries of medium size* comprise all the (remaining) arteries of the body with the exception of the aorta and the pulmonary artery. The *intima* of these vessels has increased in thickness owing to the interposition between the epithelial cells and the internal elastic membrane of networks of delicate elastic fibers and a striped connective substance enclos-

ing flattened cells.* The media no longer consists only of circularly disposed smooth muscle-fibers,† that here are arranged in several superposed layers, but also contains wide-meshed nets of elastic fibers. The proportion of the two tissues in the individual arteries differs widely: in the celiac, femoral, and radial arteries the muscle tissue preponderates; in the carotid, axillary, and common iliac the elastic tissue is in excess. The *externa* has likewise become stouter. Thick elastic fibers occur in especial profusion at the boundary of the media and in many arteries

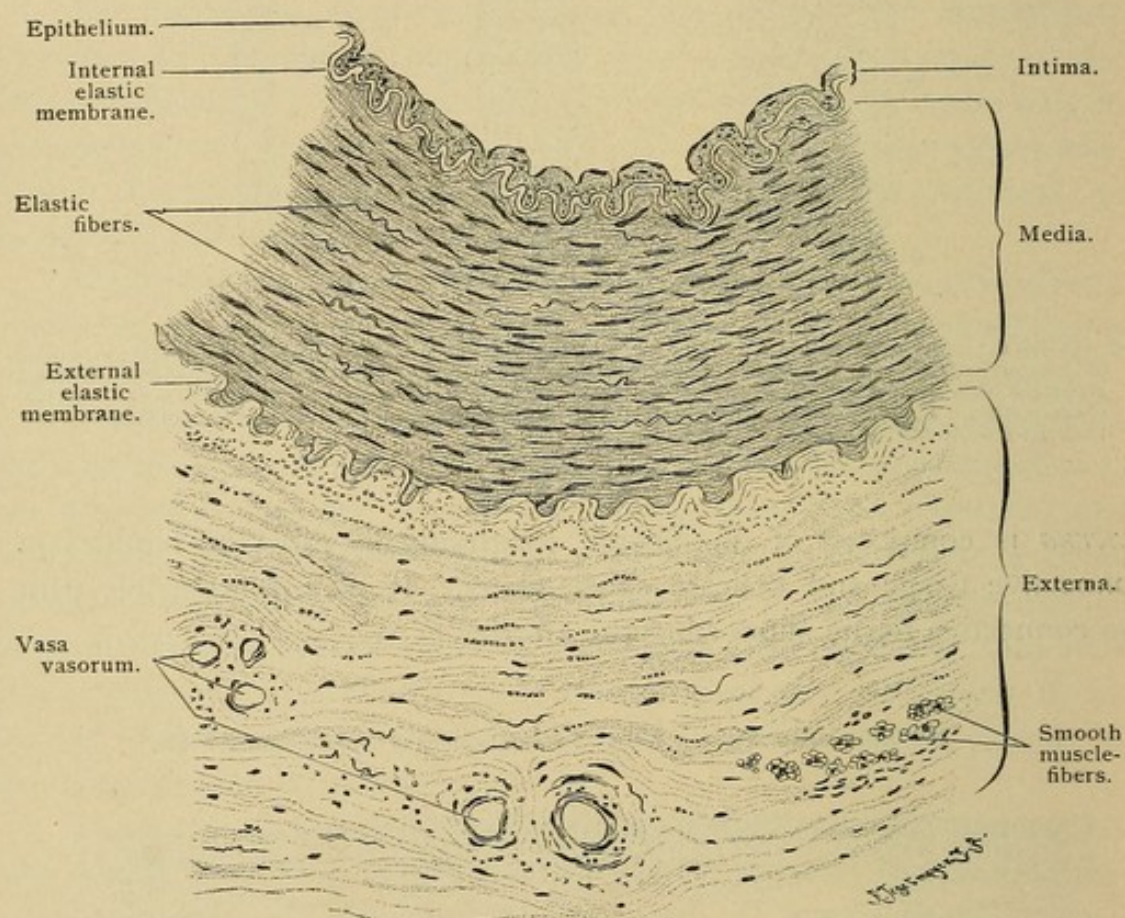


FIG. 74.—PORTION OF A CROSS-SECTION OF THE BRACHIAL ARTERY OF MAN. $\times 100$. Technic No. 37.

form an independent layer that has been designated the *external elastic membrane*‡ (Fig. 74). New elements in the externa of arteries of

* This subepithelial layer is absent in the larger branches of the abdominal aorta, in the external iliac, and in the uterine arteries of young individuals; in the larger arteries of the brain the internal elastic membrane is about three times as thick as in other arteries of like size and is characterized by longitudinal bars.

† At the inner boundary of the media longitudinally disposed muscle-fibers occur; they are especially well-developed in the subclavian artery.

‡ In the arteries of the brain the longitudinally disposed elastic fibers of the externa are very slightly developed; on the other hand, the internal elastic membrane is very well developed.

medium size are smooth muscle-fibers, that are arranged in single, longitudinally disposed bundles, never in a continuous layer.

In the *large arteries* (aorta and pulmonary artery) the epithelial cells of the *intima* are shorter and more polyhedral in outline than in medium-sized vessels; immediately beneath are the subepithelial layers, of striped connective substance, that occur in the larger medium arteries and that here also enclose flattened, stellate, or spherical cells, as well as elastic fiber-nets. These fiber-nets are the thicker the nearer they lie to

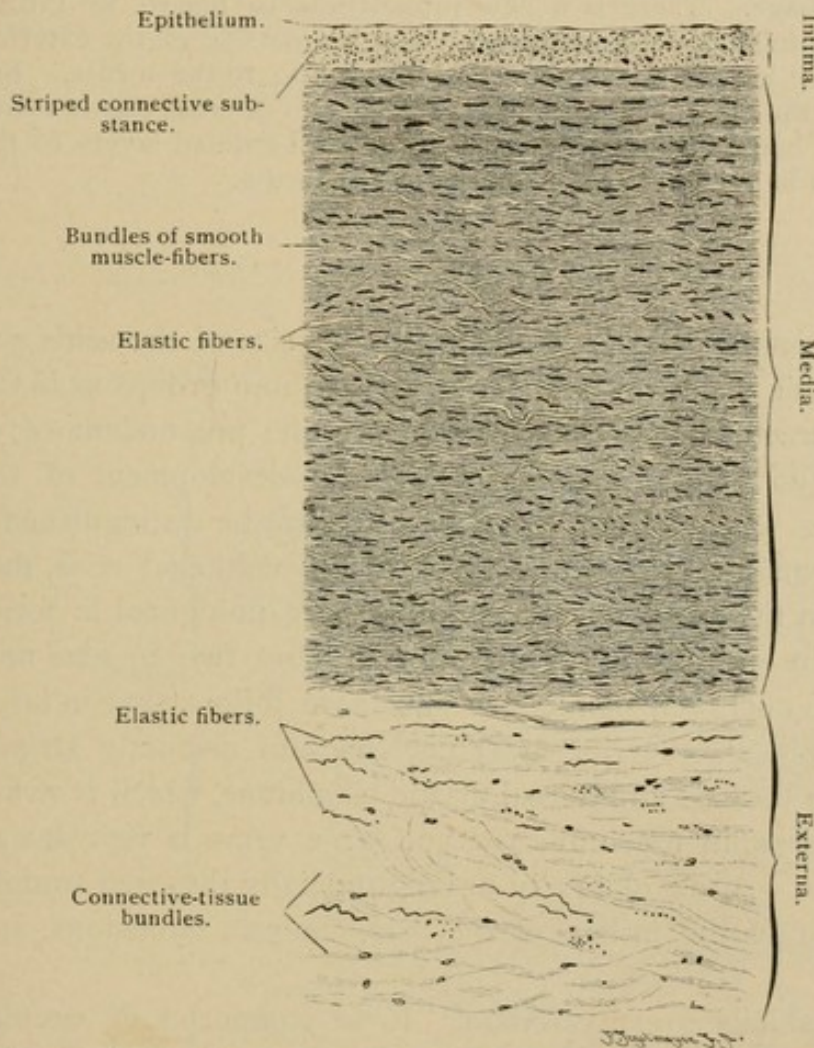


FIG. 75.—FROM A CROSS-SECTION OF THE THORACIC AORTA OF MAN. $\times 100$. Technic No. 37.

the tunica media and finally pass into a fenestrated membrane, which corresponds to the internal elastic membrane of small and medium-size arteries. The *media* of large arteries is characterized by the preponderance of richly developed elastic tissue over the muscular elements. Instead of thin fiber networks close networks of thick elastic fibers or fenestrated membranes * occur, which regularly alternate with strata of

* The elastic membranes are already present in the larger middle-sized arteries; they are especially well marked in the carotids, which closely approach the large arteries in structure.

smooth muscle-fibers. The elastic elements, like the muscle-fibers, pursue a circular course; all the elastic elements of the media are united by fibers and membranes that obliquely penetrate the muscle strata.

The *externa* of large arteries presents no essential peculiarities and differs but slightly from that of medium-sized arteries. It does not possess the external elastic membrane. Smooth muscle-fibers only occur in the externa of the large arteries of the lower animals.

The foregoing classification of the strata of the wall of the artery corresponds to present usage. There is a new proposition to regard as intima simply the epithelial tube alone, as externa all that lies outside of the external elastic membrane, the latter to be reckoned as belonging to the media. Between these two lies the media, of which the external and internal elastic membranes represent the border-lamellæ. The subepithelial striated layers of the larger arteries are to be reckoned as belonging to the media.

THE VEINS.

There is no definite proportion between the size of the veins and the thickness of their walls, no basis for a division into groups as in the arteries. The characteristic of the veins lies in the preponderance of the connective-tissue sheaths and in the slighter development of the muscular elements. As in the arteries three coats can be distinguished.*

The *intima* consists of a single layer of flat epithelial cells, that are fusiform only in the smallest veins, in others are polygonal in form. In veins of medium size, having a diameter of from two to nine millimeters, layers of nucleated connective substance follow, that in large veins (femoral, popliteal, superior cava) develop into distinctly striped layers. Following these is an internal elastic membrane, which is structureless in small veins, in medium-sized and large veins is represented by elastic networks. A few obliquely or longitudinally disposed smooth muscle-fibers occur in the intima of the iliac, femoral, saphenous, and mesenteric veins.

The *media* exhibits great variation. It is composed of circular muscle-fibers, elastic networks, and fibrillar connective tissue, and is best developed in the veins of the lower extremities (especially in the popliteal), less in the veins of the upper extremities, still less in the large veins of the abdominal cavity; finally, it is absent in a large number of veins (in those of the pia and dura, of the bones, of the retina, in the superior cava, and also in the veins proceeding from the capillaries,

* Owing to the meager development of the media some histologists have recognized only two coats, the tunica intima and the tunica externa, the layers usually regarded as tunica media being included in the latter.

the precapillary vessels). Instead only obliquely and transversely placed connective-tissue bundles occur.

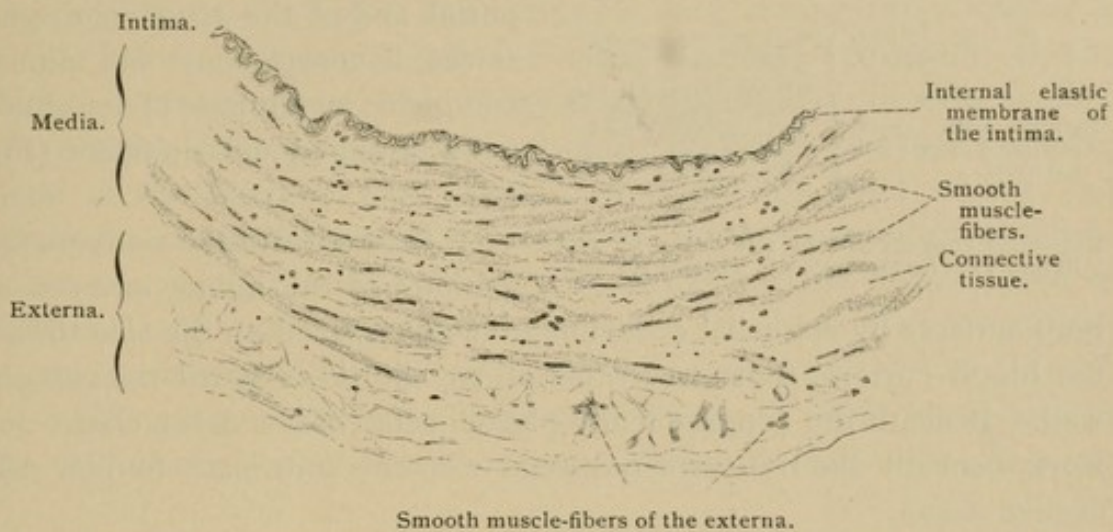


FIG. 76.—PORTION OF CROSS-SECTION OF A VEIN OF A HUMAN EXTREMITY. $\times 100$. Technic No. 37.

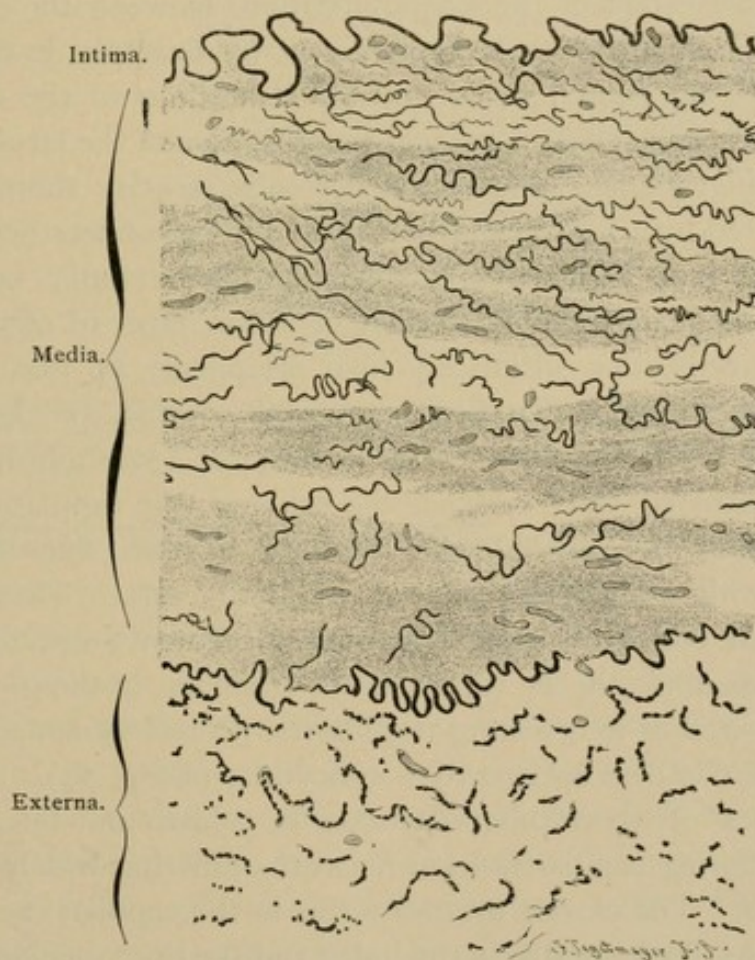


FIG. 77.—CROSS-SECTION OF A VEIN OF A HUMAN EXTREMITY. $\times 420$. The elastic elements are stained. Technic No. 38.

The usually well-developed externa consists of intercrossing bundles of connective tissue, of elastic fibers, and of longitudinally disposed

smooth muscle-fibers, that are much more richly developed in the veins than in the arteries. The externa of certain veins (*e. g.*, of the trunk of the

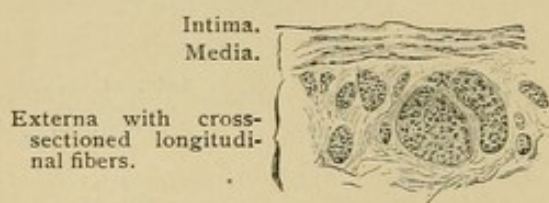


FIG. 78.—CROSS-SECTION OF THE WALL OF THE RENAL VEIN OF MAN. $\times 50$. Technic No. 37.

portal and of the renal vein) possesses a conspicuous and almost complete membrane of longitudinally arranged muscle-fibers (Fig. 78).

The *valves* of the veins are formations of the intima covered on both surfaces by epithelial cells, longitudinally placed on the side toward the blood current, transversely placed on the side toward the vascular wall. Beneath the longitudinally placed cells lies a dense elastic network, beneath the transversely placed elements a delicate fibrillar connective tissue.

The *valves* of the veins are formations of the intima covered on

THE CAPILLARIES.

The capillaries establish the communication between the arteries and the veins. There are a few exceptions, as, for example, in the corpora cavernosa of the genital organs. The transition of the arteries into the capillaries is effected by a gradual simplification of the structure of the vessel-wall (Fig. 72 *c*). The media becomes steadily thinner and finally is represented by a few circularly disposed muscle-fibers occurring at wide intervals, that ultimately disappear. The externa becomes correspondingly attenuated until it consists of a thin layer of connective tissue containing cells, that ultimately also disappears, so that at last the only part of the vessel wall that remains is the intima, the layers of which are likewise reduced until nothing is left but a stratum of plate-like, nucleated epithelial cells. Hence the walls of the capillaries consist of a simple layer of epithelial cells, the form of which may be most aptly compared with a steel pen pointed at both ends. These cells are united at their edges by a small amount of cement substance. In a few places, for example, in the capillaries of the liver, in the glomeruli of the kidney, as well as in growing capillaries, no cell boundaries can be exhibited; there is here apparently a syncytium (see p. 73).

The capillaries divide without decrease in caliber and by anastomosis with neighboring capillaries form networks differing widely in the size of the meshes. The closest meshes occur in the capillary networks of secretory organs, for example, in the lungs and the liver; wide-meshed networks, for example, in the muscles, the serous membranes, the special-sense organs. The reverse obtains in regard to the caliber of the capillaries; the widest capillaries are found in the liver, the narrowest in the retina and in the muscles.

Development of capillaries.—Only the developmental processes in post-embryonic epochs will be considered here. A conical protoplasmic mass appears on the wall of an existing capillary, resting with a broad base on the latter and terminating in a slender, tapering, free end.* In the further course of development this pointed free end unites with another approaching off-shoot, that has arisen in the same way from another point of the capillary wall. These formations are solid at first, but gradually become hollow by the extension of the lumen of the capillary, and subsequently the walls of the new vessels become differentiated to epithelial cells. The development of new capillaries is always consummated in connection with existing capillaries (*cf.* technic No. 42, p. 153).

All medium and large blood-vessels possess small blood-vessels, the vasa vasorum, that provide for the nutrition of their walls; they run

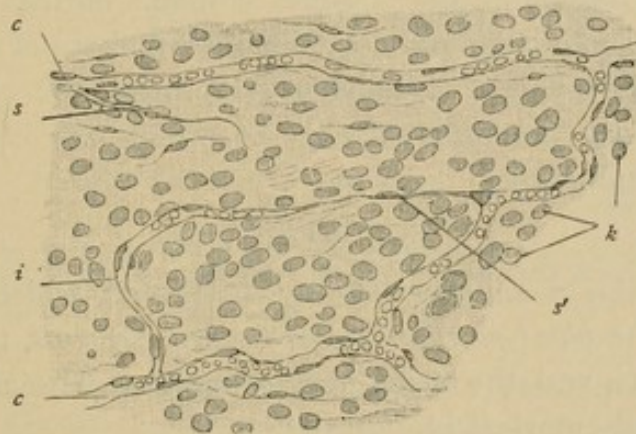


FIG. 79.—SURFACE VIEW OF A PORTION OF THE GREATER OMENTUM OF A SEVEN-DAY-OLD RABBIT. *c*, Blood capillaries, still containing blood corpuscles; *s*, capillary sprout tapering to a free solid point; *i*, young capillary, the greater part of which is hollow, at *s'* still solid; *k*, nuclei of peritoneal epithelium. $\times 240$. Technic No. 42.

almost exclusively in the externa (Fig. 74). The intima always is without blood-vessels.

On the walls of all blood-vessels, with the exception of the vessels of the substance of the brain and the spinal cord, nonmedullated and medullated *nerves* have been found, which form a plexus in the tunica media of the arteries and the veins. From this nonmedullated fibers arise, some of which supply the smooth muscle-fibers and some of which form terminal plexuses lying in the externa and in the interna and agreeing in all points with those of the heart (p. 128). The capillaries are accompanied by encircling networks of nonmedullated nerve-fibers.

* Such blind capillary sprouts may be hollowed out at an early period; corpuscles that happen to flow into them degenerate, because they are excluded from the circulation and the interchange of gases, and fall into minute fragments, that have been erroneously interpreted as hematoblasts; they have no connection with the true hematoblasts (p. 140).

Many blood-vessels are encircled by lymph-vessels, which occasionally are so wide that they form spaces completely enclosing the former, the "adventitial lymph spaces."

The walls of the blood-vessels permit not only the escape of fluid but also of corpuscular elements, *e. g.* blood-cells; this is especially the case in the thin-walled veins and the capillaries, in which the exit takes place *between* the epithelial cells. The intercellular spaces thus arising close again; permanent openings, "stomata," are not present.

The *glomus caroticum* ("carotid gland") is no gland, but consists essentially of blood-vessels. The capillaries arising from the division of the one supplying artery differ greatly in width and are surrounded by numerous chromaffine cells (see sympathetic ganglia) united in spherical groups, the so-called secondary nodules. The many veins collect at the periphery of the gland, that besides contains fibrillar connective tissue, isolated ganglion-cells, and conspicuous numbers of medullated and nonmedullated nerve-fibers. Similar in structure is the coccygeal gland (*glomus coccygeum*), the blood-vessels of which are characterized by hemispherical evaginations.

THE BLOOD.

The blood* is a slightly clammy, red liquid, which consists of a fluid substance, the *blood-plasma*, and of *formed elements*, the blood-cells, the blood-platelets, and the elementary granules. The cells are of two kinds, colored and colorless blood-cells.

The *colored blood-cells* (red blood corpuscles, erythrocytes, Fig. 80) are soft, flexible, highly elastic structures and possess a smooth, slippery surface. In man and in other mammals they usually have the form of a flat, circular disk,† slightly concave on each surface, and therefore resemble biconcave lenses. Exceptions occur in the llama and the camel, in which the colored blood-cells are oval disks. Their average diameter in man is $7.5\ \mu$, their thickness $1.6\ \mu$. The colored blood corpuscles of domesticated mammals all are smaller; the largest are those of the guinea-pig ($7.48\ \mu$) and the dog ($7.3\ \mu$). The colored blood-cells consist of a *stroma* (protoplasm) which contains spaces filled with the blood coloring substance, the *hemoglobin*. The hemoglobin imparts to the blood-cells the yellow or yellowish-green color.‡ A nucleus and

* The elements of the blood do not form a tissue, but represent a loose union of elementary parts, without definite arrangement of the same, an aggregation of cells.

† In addition there occur in human blood *spherical* colored blood corpuscles; they are smaller ($5\ \mu$) and few in number.

‡ Only when very *many* blood-cells lie one over the other do they appear red.

an actual cell-membrane are wanting (*cf.* remark *, p. 140). The colored blood corpuscles of fishes, amphibians, reptiles, and birds are distinguished from those of mammals by their oval, biconvex form, their generally greater size ($22\ \mu$ long by $15\ \mu$ broad in the frog), as well as by the presence of a round or oval nucleus; in other respects they exhibit the same properties as those of mammals.

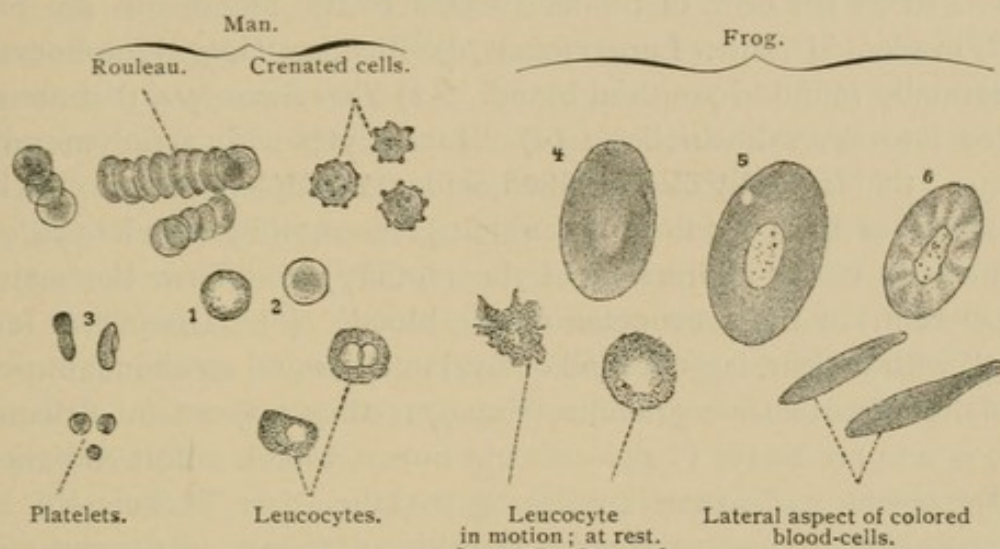


FIG. 80.—BLOOD-CELLS. $\times 600$. 1, Colored blood-cell seen in close focus, 2, in distant focus. 3, Lateral aspect of colored blood-cells. 4, Colored blood-cell, quite fresh, nucleus indistinct; 5, the same cell a few minutes later, with the nucleus plainly visible; 6, the same cell after treatment with water. Technics Nos. 43, 46, 47.

The *white or colorless blood-cells* (leucocytes) occur not only in the blood but also in the lymph-vessel system, where they are termed "lymph or chyle corpuscles." They are also found outside of the vascular systems; in bone-marrow, as "marrow-cells," and further in abundance in adenoid tissue (see p. 95), scattered in fibrillar connective tissue, and finally between epithelial and gland-cells, whither they have wandered by their power of ameboid movement; * therefore they are also called "wandering cells" (*cf.* pp. 68 and 93).

In all cases the colorless blood-cells consist of a clammy protoplasm and a nucleus, and are without a cell-membrane. A definite form cannot be described, because during life they are usually engaged in ameboid activity. In a state of rest they are spherical (Fig. 80).



FIG. 81.—COLORLESS BLOOD-CELLS OF MAN. a, Lymphocyte. b, Leucocyte with polymorphous nucleus. c, Leucocyte with neutrophile granules. $\times 600$. Technic No. 45.

* In the mucous membranes leucocytes in varying large numbers wander through the epithelium to the free surface and there degenerate. In vertebrates this wandering bears no relation to nutrient processes; on the other hand, the leucocytes play a great rôle in preventing the injurious action of microbes or of other poisonous substances.

The size and properties of the nucleus and the protoplasm have led to the following classification :

(1) *Lymphocytes*, the smallest of which measure from 4 to 7.5 μ . The quantity of their protoplasm is so small that it can scarcely be perceived by the usual methods ; it forms only a thin envelope for the relatively large round nucleus (Fig. 81*a*). They exhibit little motility, form from 22 to 25 per cent. of the leucocytes of the blood, and are found chiefly in adenoid tissue. Larger forms, described as "large lymphocytes," are normally found in youthful blood. (2) *True leucocytes*, that are subdivided into several varieties. (a) "Leucocytes with a polymorphous nucleus," that is deeply cleft or lobed, seldom multiple.* These cells have a diameter of from 7.5 to 10 μ , exhibit great motility (the lobulation of the nucleus is the expression of the motility), and form the majority (72 per cent.) of the leucocytes of the blood. (b) "Uninuclear leucocytes," with a clear, large, round or oval nucleus and an abundant protoplasm free from coarse granules (Fig. 97); they may attain a diameter of 20 μ and are scarce (1 per cent.) in human blood. Both varieties (*a* and *b*) possess a dense neutrophile granulation (see Technic No. 45 *c*) and are united to each other by transitional forms. Leucocytes with polymorphous nuclei that have wandered out of the blood-vessels into the tissues may become transformed into small uninuclear leucocytes. (c) "Leucocytes with coarse granules," from 8 to 14 μ in size, with a round or polymorphous nucleus, and distinguished by the possession of large quantities of granules, which react very differently to stains. Oxyphile (eosinophile) or basophile (= mast-cells) leucocytes are distinguished, according as the granules imbibe acid or basic stains.† The granules probably are the optical expression of metabolic processes and of phases of progressive development ; those of the oxyphile cells are perhaps albuminous substances taken up from without (see further Technic No. 45, *Further treatment*).

The determination of the proportionate number of, as well as the ratio between, the colored and colorless blood-cells is coupled with considerable difficulty and only approximately correct estimates can be given. In man one cubic millimeter of blood contains about 5,000,000 colored cells. The white cells are present in the blood in much

* Multiplicity of nuclei is often merely apparent, the delicate connecting filaments of the deeply cleft nucleus being overlooked ; the term "poly- (better multi-) nuclear" often used to describe leucocytes with polymorphous nuclei is totally inaccurate and should be discarded.

† Ehrlich, who made this classification, proceeds therein from other standpoints than the chemist ; acid dyes, *e. g.*, are those in which the coloring principle is a molecular combination existing chemically as an acid.

smaller number; there is about one in from 300 to 500 colored blood-cells, therefore about 10,000 colorless cells in one cubic millimeter of blood.

The *blood-platelets* (thrombocytes) are very unstable, colorless, round or oval disks having a diameter of from 2 to 4 μ (Fig. 80); they are capable of ameboid movement and contain a body the nuclear nature of which is, however, questionable. At times they are present in the blood in large numbers.* Their origin is obscure; the hypothesis of constriction from erythrocytes or leucocytes supported by some is disputed by others, likewise is it still undecided whether their rôle in the process of coagulation of the blood is direct or indirect.

The *elementary granules* are chiefly particles of fat transferred from the chyle to the blood. They are easily observed in the blood of the lower mammals and in herbivorous animals but are not normally present in the blood of man. Small refractive granules not of a fatty nature, that occur in variable quantity in all human blood, have been named *hematokonia* (blood dust).

After death or as a result of changes within the vessel-wall the blood coagulates under the influence of two substances, fibrinoplastin and fibrinogen, which pass into solution and unite in the plasma. The product of this union is *fibrin*. The coagulated blood separates into two parts, the *clot* and the *serum*. The clot is red and consists of all the colored and the majority of the colorless blood-cells and the fibrin, which microscopically appears as a felt of fine fibers; chemically the fibers resemble the fibers of glutinous connective tissue. The serum that collects above the clot is colorless and contains a few colorless blood-cells.

The coloring substance contained in the colored corpuscles, the *hemoglobin*, possesses the property of crystallizing under certain conditions and in nearly all vertebrates the crystals belong to the rhombic system. Their form in the different animals varies greatly; in man it is usually prismatic. Hemoglobin is readily decomposed. One of the decomposition products is *hematin*, which yields *hematoidin* and *hemin*. Crystals of hematoidin, which occur within the body in old extravasated blood, for example, in the corpus luteum, are rhombic prisms of orange-red color. The hemin crystals, when well developed, are rhombic tablets or bars of mahogany brown color; often they are very irregular in form (Fig.

* In 1 c.c. of human blood there are said to be 245,000 blood-platelets, a number that probably is below the truth, since in the method of estimating some blood-platelets always adhere to the walls of the pipet.

82, 1). In a forensic respect they are of great importance (*cf.* Technic No. 50).

Development of colored blood-cells.—From the earliest period of embryonic development and during the whole of life nucleated colored blood-cells, the *hematoblasts* (erythroblasts), are found in certain localities (see bone-marrow). Their number fluctuates and runs parallel with the energy of the blood-forming processes. By indirect division they give rise to the nonnucleated colored blood-cells, that at first contain a nucleus, but by a process of internal degeneration (not by extrusion) subsequently lose it.* As centers for the formation of blood in embryonal periods the liver and the lymph-glands, later the spleen, in the adult exclusively the bone-marrow, must be indicated.

Development of colorless blood-cells.—It is conjectured that the colorless blood-cells arise from elements having their origin in the anlage of the embryonal blood and blood-vessels and are transported with (not in) † the blood-vessels to the most diverse localities, to the anlages of the



FIG. 82.—1. Hemin crystals of man; whetstone forms on the right. 2. Crystals of common salt. 3. Hematoidin crystals of man. 1 to 3 magnified 560 times. 4. Hemoglobin crystals of the dog, magnified 100 times; a, crystal separating into fibers. Technic No. 50.

lymph-glands and lymph-nodules, where they multiply by mitosis and are carried back to the blood stream through the lymph-vessels. In the embryonal period the mother-cells of the leucocytes can produce not only colorless but also colored blood-cells.

In adult man the lymphocytes are said to be formed by the lymph-glands and lymph-nodules, the leucocytes with polymorphous nuclei in the bone-marrow (hence the name "myelocytes"), statements that are supported by clinical experience, not by anatomic investigations; the latter strongly indicate that the lymphocytes are juvenile forms from which the other varieties of leucocytes develop.

* Whether the forms demonstrated in nonnucleated elements fixed in osmium solution are transformation products, rudiments of nuclei, is questionable, since it has been shown that similar bodies occur in the nucleated blood-cells.

† This may have been the case in the earliest stages; later the lymph-glands and nodules arise from leucocytes that have migrated from the blood-vessels.

2. THE LYMPH-VESSEL SYSTEM.

THE LYMPH-VESSELS.

The wall of the larger lymph-vessels (from 0.2 to 0.8 mm. and upward), like that of the blood-vessels, is composed of three coats. The intima consists of epithelial cells and a network of delicate elastic fibers with elongated meshes. The media is formed of circularly disposed smooth muscle-fibers and a few elastic fibers. The externa consists of longitudinally arranged bundles of connective tissue, elastic fibers, and bundles of smooth muscle-fibers, likewise disposed in a longitudinal direction. The wall of the smaller lymph-vessels and of the lymph capillaries is composed exclusively of extremely delicate epithelial cells, that often have sinuous contours. The lymph capillaries are wider than the blood capillaries, frequently are beset with constrictions and dilatations, and where they branch are often considerably expanded; the networks they form are more irregular.

The question of the origin of the lymph-vessels is not yet satisfactorily decided; while some authors are of the opinion that the lymph capillaries form a closed system, according to another widely entertained view the lymph capillaries are open toward the periphery and in direct connection with the system of intercommunicating cell-spaces of connective-tissue (juice-canal-system,* p. 100).

According to the first theory the nutritive fluids (tissue juices) passed through the walls of the blood capillaries that are not used in the nutrition of the tissues penetrate the closed lymph capillaries by endosmosis; according to the second view the tissue juices flow directly from the tissues into the patent orifices of the lymph capillaries.

It is said that the lymph-vessels of the pleura and of the peritoneum are in open communication with their respective cavities through small openings, the *stomata*, between the epithelial cells, which in the pleura are found at the

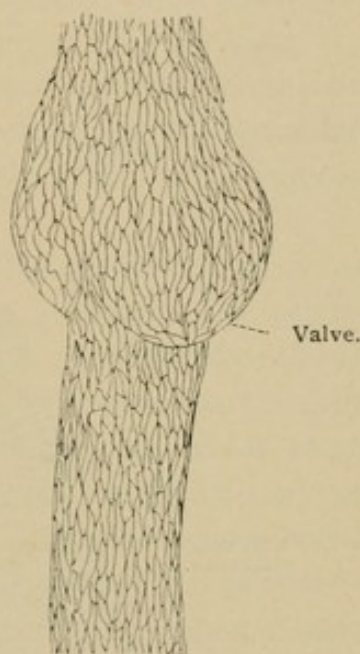


FIG. 83.—LYMPH-VESSEL OF THE MESENTERY OF A RABBIT, showing the boundaries of the epithelial cells. $\times 50$. Technic No. 40.

*The juice canaliculi are designated *lymph canaliculi* in contradistinction to lymph-vessels provided with cellular walls; other authors make lymph canaliculi equivalent to lymph-vessels plus the tissue-juice canal system.

intercostal spaces and in the peritoneum on the central tendon of the diaphragm. However, it is a question whether the stomata described in mammals are not artifacts. Stomata are unnecessary in the transfer of fluids and corpuscular elements from the peritoneal cavity into the lymph-vessels, because thin-walled lymph-vessels lie immediately beneath the peritoneal epithelium.

THE LYMPH-GLANDS.

The lymph-glands (lympho-glandulæ, lymph-nodes) are macroscopic bodies intercalated in the course of the lymph-vessels. Usually they

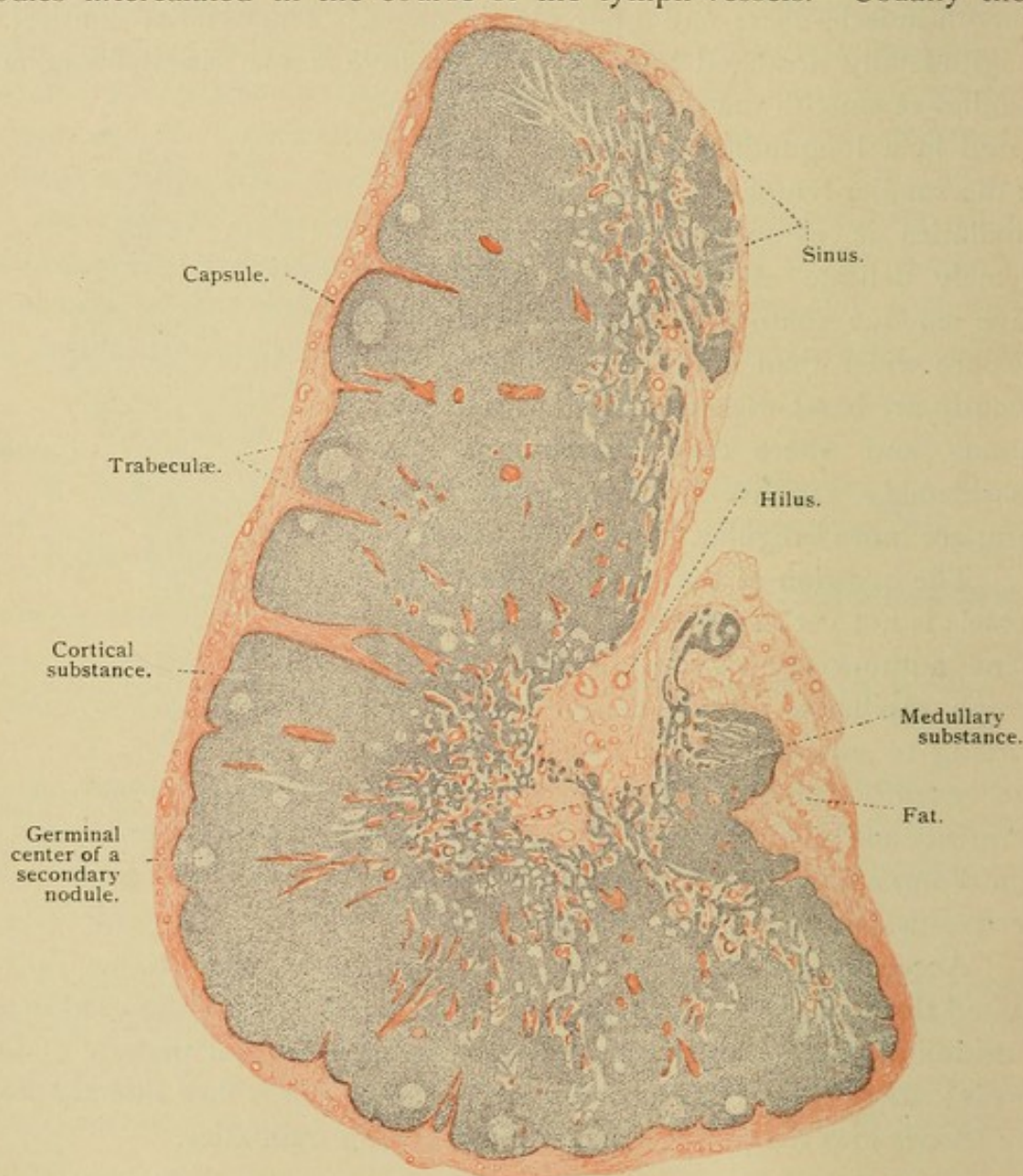


FIG. 84.—LONGITUDINAL SECTION OF A HUMAN CERVICAL LYMPH-GLAND. $\times 12$. Technic No. 53.

are rounded oval or flat, kidney-shaped structures and differ greatly in size. On one side there is often a scar-like depression, the *hilus*, at which the efferent lymph-vessels emerge.* Their construction becomes intelli-

* The afferent lymph-vessels penetrate the gland at various points.

gible if we proceed from the following conception: In certain localities from three to six lymph-vessels divide repeatedly into anastomosing branches, which soon reunite into the same or a lesser number of usually narrower lymph-vessels. In this way a kind of rete mirabile* is formed. The dividing lymph-vessels are called *afferent* vessels (*vasa afferentia*), the reuniting, *efferent* vessels (*vasa efferentia*). Within the meshes of this reticulum lie some spherical and some elliptical bodies, that consist of adenoid tissue. The spherical bodies, the *secondary nodules* (follicles, *ampullæ*), occupy the periphery, the elliptical bodies, the *medullary cords*, the center of the lymph-gland. The lymph-gland is enveloped in fibrous connective tissue, the *capsule*, which sends processes, the *trabeculæ*, into the interior of the organ (Figs. 84 and 85). Fine extensions from the trabeculæ, in the form of reticular connective tissue, pierce the walls of the lymph-vessels, penetrate the secondary nodules and the medullary cords, and form a support for the numerous leucocytes found there.

Accordingly the lymph-gland consists of a *cortical* and a *medullary substance*, the relative proportions of which vary greatly. The cortical substance contains the secondary nodules, which continue centralward directly into the medullary cords (Figs. 84 and 85). The secondary nodules and the medullary cords are surrounded by the continuations of the afferent lymph-vessels.† The latter here are greatly expanded and are termed *lymph sinuses*; they are pierced by the connective-tissue reticulum. The secondary nodules and the medullary cords are composed of *adenoid* tissue, that is, of reticular connective tissue the meshes of which are crowded with leucocytes. In many of the secondary nodules there is at times a light, spherical spot, the *germinal center*, in which karyokinetic figures are always to be found.‡ The secondary nodules are stations for the formation of leucocytes, which pass into the lymph-sinuses and thence into the *vasa efferentia*.

The *capsule* consists of fibrous connective tissue and elastic fibers, in a variable quantity increasing with age, also smooth muscle-fibers, which in the large lymph-glands of the ox are united in large strands.

* *Retia mirabilia* were first described in connection with the blood-vessels. They consist of a vascular plexus, which *suddenly* interrupts the course of the vascular stem. They occur in the course of both arteries and veins, and accordingly there are arterial and venous *retia mirabilia*. The glomeruli of the kidneys are exquisite examples of such arterial vascular networks (*cf.* Fig. 247): a small arterial stem divides into capillary twigs, which in turn reunite to a small arterial stem, which then ramifies in the usual way.

† The lymph-vessels never penetrate the interior of the secondary nodules.

‡ Multiplication of cells also occurs in the medullary cords, but in much slighter degree than in the secondary nodules.

The *trabeculae* have the same structure ; they pass between the secondary nodes and the medullary cords, but do not come into contact with them, being separated from them by the lymph-sinus. The wall of the lymph-sinus is formed of only a simple layer of plate-like cells ; similar cells clothe the surface of the secondary nodules and the medullary cords, and also the surface of the *trabeculae* and of the connective-tissue reticulum (*cf.* p. 94).

The structure of the lymph-glands here described is difficult to recognize, owing to sundry complications. These complications consist in : (1) the frequent merging of neighboring secondary nodules with each other ; (2) the union of the medullary cords in the form of a coarse net-

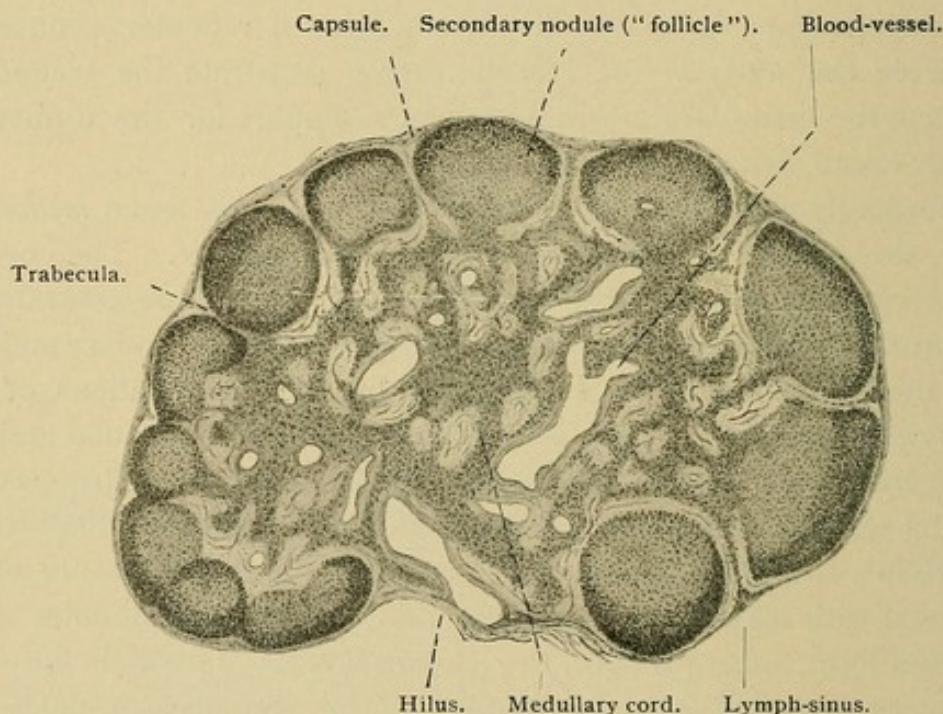


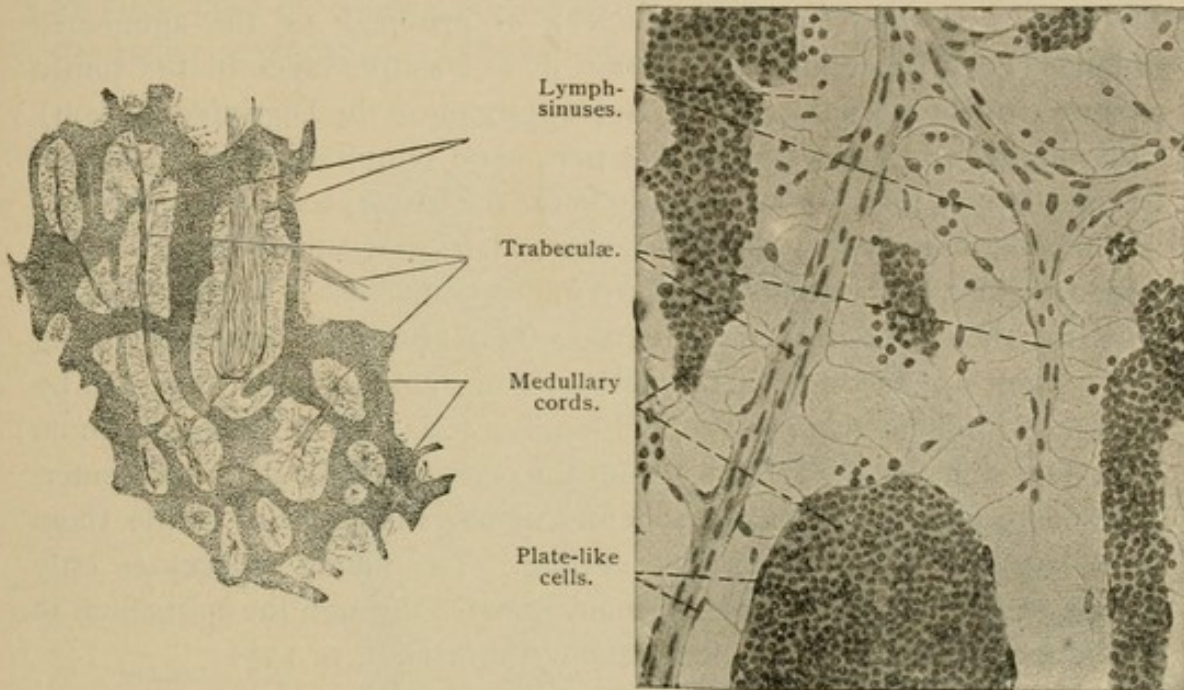
FIG. 85.—SECTION OF A LYMPH-GLAND OF A RABBIT. $\times 28$. (Schaper.) Technic No. 53.

work ; (3) the similar network formed by the *trabeculae* ; (4) the interlacing of the networks formed by the medullary cords and the *trabeculae* (Fig. 86) ; (5) the presence of leucocytes in the lymph-sinus, which must first be removed by special methods.* In this manner the secondary nodules, the medullary cords, and the leucocytes in the lymph-sinus form a soft mass, that has been named the *pulp* or *parenchyma* of the lymph-gland.

The majority of the *blood-vessels* enter at the hilus, the others at various points on the surface of the gland. The latter are delicate vessels and divide in the capsule and in the large *trabeculae*, in the axis

* *Editor's remark* : In preparations of lymph-glands it is necessary to dislodge the leucocytes to bring the lymph-sinus into view (see Technic No. 53).

of which they run. The large artery entering at the hilus divides into a number of branches, that here are surrounded by richly developed connective tissue. The branches are principally distributed to the adenoid tissue, only a few entering the trabeculæ; they pass through the lymph-sinuses, into the medullary cords, then into the secondary nodules,* and in both situations break up into richly developed capillary networks which

FIG. 86. $\times 50$.FIG. 87. $\times 240$.

FROM A VERTICAL SECTION OF A LYMPH GLAND OF AN OX. The medullary substance. In the upper half the medullary cords and trabeculæ are cut through longitudinally, in the lower half transversely. Both form a continuous network. In the lymph-sinus the delicate fibers of the reticular connective tissue, still containing a few leucocytes, can be seen. Drawn with change of focus. Technic No. 55.

supply the oxygen needed in the formation of the leucocytes. The veins emerge at the hilus.

The few *nerves* of the lymph-glands are partly medullated, partly nonmedullated bundles of fibers, that chiefly form richly branched plexuses about the blood-vessels; nerves have been found in the capsule and in the trabeculæ, but not in the nodules.

THE PERIPHERAL LYMPH NODULES.

(NODULI LYMPHATICI.)

Reticular connective tissue enclosing leucocytes is not confined to the lymph-glands; it occurs in great diffusion in many mucous membranes

* The arteries entering into the axis of the secondary nodules break up in slender non-anastomosing capillaries, which pass into a meshwork of venous capillaries lying at the border of the nodule, from which larger veins arise. The conditions here are quite like those in the spleen.

and in different degrees of development, sometimes as *diffuse*, sometimes as *definitely circumscribed* infiltrations of leucocytes. These formations are not included in the lymphatic system. But more highly developed structures, nodules with germinal centers, closely resembling the secondary nodules of the lymph-glands, are also found in the mucous membranes; these are named *peripheral lymph nodules* and are included in the lymphatic system. They occur in many mucous membranes isolated, as the *solitary nodules* (solitary follicles), or grouped, as the agminated nodules (Peyer's patches), and always lie in a simple layer in the tunica propria close beneath the epithelium (see Organs of the Digestive System). The number and distribution of the peripheral lymph nodules are subject to considerable fluctuation, not only in the different species of animals, but in different individuals; since their mass also varies and frequent transitions to circumscribed and to diffuse infiltrations exist it is highly probable that they are temporary structures that arise and disappear during life. They are distinguished from the true lymph-glands above all by their less intimate relation to the lymph-vessels, which do not form an encircling sinus for the follicle.* But the possession of a germinal center, a brooding place for young leucocytes, appears in so far to entitle them to a place in the lymph vascular system. The young leucocytes only in part enter the lymph-vessels; many wander through the epithelium to the surface of the mucous membrane (*cf.* remark *, p. 137).

THE LYMPH.

The lymph is a colorless fluid in which leucocytes (*cf.* white blood-cells, p. 137) and granules are suspended. The latter are immeasurably small, consist of fat, and are principally found in the lymph (or chyle) vessels of the intestine; frequently they are present in colossal quantity and then they impart the white color to the chyle. In other lymph-vessels the fatty granules occur sparingly. In the lymph-glands many leucocytes are found in which the envelope of protoplasm surrounding the nucleus is so thin that its presence can only be demonstrated with high magnifications.

THE SPLEEN.

The *spleen* is an organ closely allied to the blood-lymph-glands† and consists of a capsule, of trabeculæ, and of the pulp.

* The only exception exists in the rabbit, in which the sinus occurs in the agminated nodules; on the other hand, in the solitary nodules of this animal the sinus is likewise wanting.

† In some mammals, *e. g.* in the pig, lymph-glands of a dark red color, similar to that of the spleen, occur along the thoracic aorta; they are characterized by the absence of efferent and afferent lymph-vessels and in being penetrated only by blood-vessels, which take up the leucocytes formed in the gland. Such "blood-lymph glands" also occur in man, in the tissue between the vessels of the kidney.

The *capsule* is firmly united by growth to the peritoneum which covers it and consists of tough fibrous connective tissue, a few smooth muscle-fibers, and dense nets of elastic fibers, the quantity of which increases with age. From the capsule numerous, mostly cord-shaped processes, the *trabeculae*, pass into the interior of the spleen and form a continuous network; they likewise consist of connective tissue, of elastic fibers, and in man of a few, in animals (*e. g.* the dog and the cat) of an abundance of smooth muscle-fibers. The thicker trabeculae contain the larger ramifications of the blood-vessels. The meshes of the trabec-

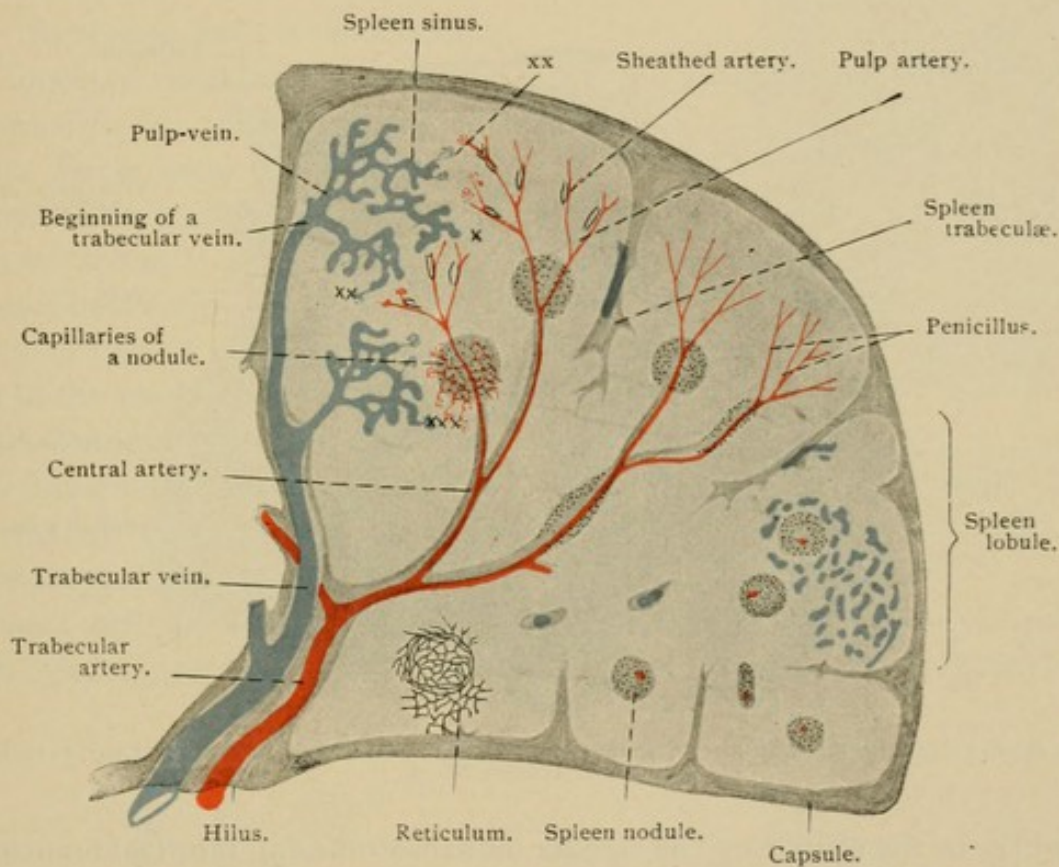


FIG. 88.—SCHEME OF THE HUMAN SPLEEN. x, opening of the arterial capillaries in the spleen sinus. xx, interruption of the closed blood course at the ends of the arterial capillaries, at the margin of the nodule, xxx. (For the sake of distinctness the spleen sinus is sketched too far removed from the margin of the nodule.)

ular network are filled with the *pulp*, a red, soft mass consisting of adenoid tissue and the smaller blood-vessels, the minute structure of which will be considered after the description of the arrangement of the blood-vessels.

The arteries entering at the hilus divide into branches, which further on, together with the veins, are enclosed in the trabeculae (Fig. 88). Then the arteries separate from the veins and the tunic derived from the trabeculae, the "adventitial sheath," as well as the tunica externa become loosened by infiltration with numerous leucocytes. These masses of leu-

cocytes may accompany the artery in its entire course as a continuous layer (*e. g.* in the guinea-pig) or may be confined to a few localities (*e. g.* in man, the cat, etc.). In the latter case the leucocytes form spherical masses of from 0.2 to 0.7 mm. in size, the *spleen nodules* (Malpighian corpuscles), or slender spindles.

The spleen nodules are usually situated in the forks of the smaller arteries, in such a manner that the artery pierces the middle or the edge of the nodule. For this reason these arteries are called *central arteries*; they send off capillaries which are well developed in the nodules, but only

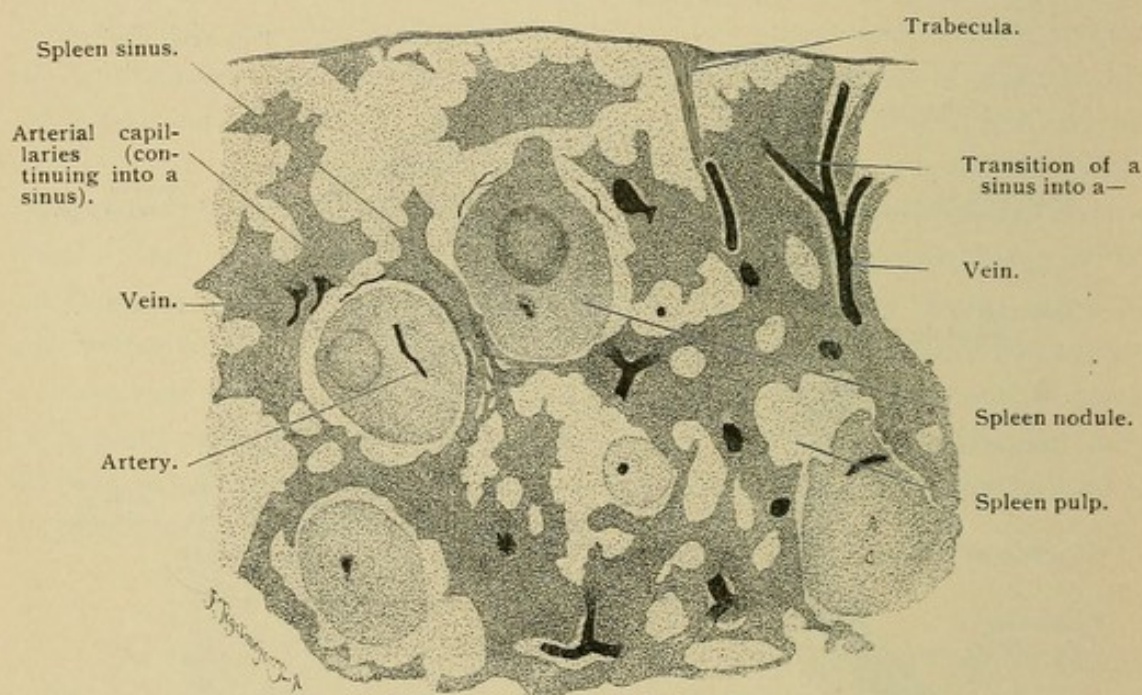


FIG. 89.—SECTION THROUGH THE INJECTED SPLEEN OF A CAT. The spleen-sinus is wider in the cat than in man. Technic No. 59.

slightly in the spindles. The slender, nonanastomosing terminal branches of the arteries,* the so-called *pulp arteries*, shortly before their transition into capillaries are provided with relatively thick walls and are called *sheathed arteries* ("ellipsoids"); the arterial capillaries arising from them empty at narrow angles into wide spaces (from 12 to 40 μ), the spleen sinuses,† which by means of wide pulp veins are connected with the large veins running in the trabeculae.

According to the foregoing description the blood-vessel system of the spleen is closed on all sides; but recent researches support, with much ingenuity, the theory advanced long since, that the path of the blood is inter-

* In injected and macerated spleens the pulp can be washed out, and then the slender terminal branches of the arteries can be seen lying together in a leash or pencil (penicillus).

† Synonyms: "ampullae," "venous capillaries," "intermediate lacunae."

rupted. This interruption occurs at the edge of the nodule and at many (not all) terminals of arterial capillaries, by the breaking up of the capillary wall. The blood then passes into the reticulum of the pulp and is transferred from here through delicate tubules into the spleen sinuses. This satisfactorily explains the fact that free erythrocytes occur in the pulp. On this theory the path of the blood in the spleen must be regarded as partly closed and as partly interrupted or "open."

By *spleen pulp* is understood the mass of vascular ramifications external to the trabeculæ and the tissue lying between the ramifications. The pulp, also designated "parenchyma"* and "red pulp," forms a network of cords which similarly to that of the lymph-glands, lies in the meshes of the trabecular net. The pulp-cords are occasionally connected with the nodules and consist of very delicate reticular connective tissue (p. 94) and numerous cellular elements. The latter are in part leucocytes, in part somewhat larger multinucleated cells, also cells containing erythrocytes

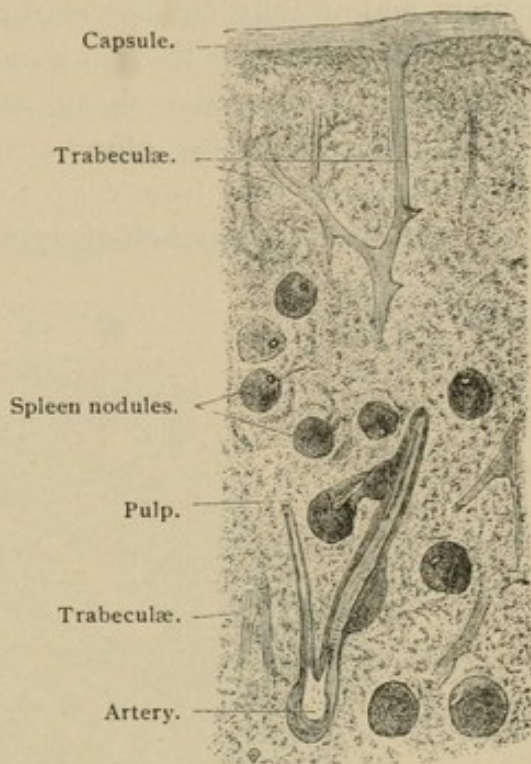


FIG. 90.—FROM A CROSS-SECTION OF A HUMAN SPLEEN, showing well-developed spleen nodules, the majority of which are pierced eccentrically by an artery. The right branch of the artery has a spindle-shaped accumulation of leucocytes. $\times 10$. Technic No. 57.

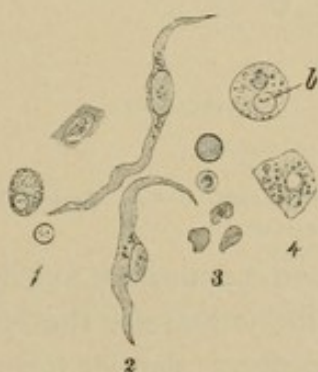


FIG. 91.—ELEMENTS OF THE HUMAN SPLEEN. $\times 560$. 1. Colorless blood-cells. 2. Epithelial cells. 3. Erythrocytes. 4. Cells containing granules; the upper one enclosing also an erythrocyte, 5. Technic No. 56.



FIG. 92.—RETICULAR CONNECTIVE TISSUE OF THE HUMAN SPLEEN. $\times 560$. Sketched from the edge of a shaken preparation. Technic No. 58.

(Fig. 91) and free erythrocytes. A granular pigment is also found in

* By the name "parenchyma" (that poured in between), earlier authors designated the masses of tissue lying between the blood-vessels in the most widely different organs. It is still customary to speak of the parenchyma of the liver, the lungs, etc.

the pulp. The nodules agree in minute structure with the secondary nodules of the lymph-glands; occasionally they even contain germinal centers and usually delicate elastic fibers. The nodules and spindles of the spleen belong to the temporary lymphatic structures; continually some undergo regressive change and new ones develop.

The portion of the blood-vessels designated sheathed or ellipsoid artery measures only from 0.15 to 0.25 mm. and has a peculiar structure; the vascular epithelium is surrounded by a thick layer of longi-

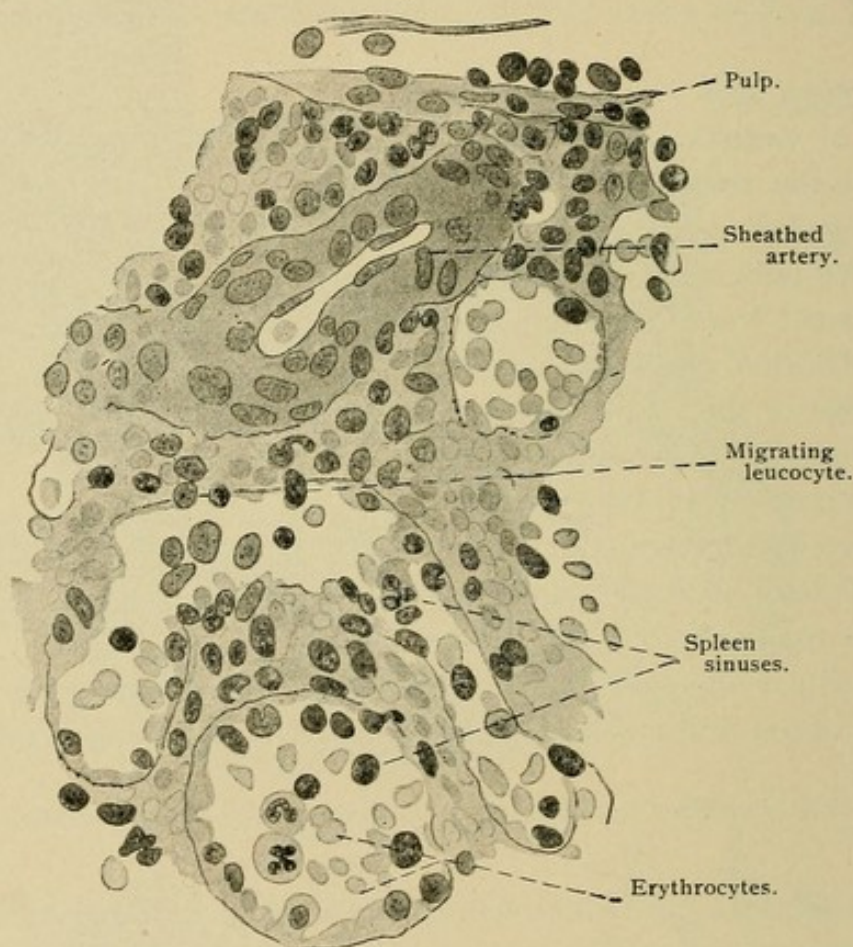


FIG. 93.—PORTION OF A THIN SECTION THROUGH A HUMAN SPLEEN. $\times 600$. Technic No. 57.

tudinally disposed fibers resembling the striped connective substance of the medium-sized arteries * (p. 129). Quite individual are the epithelial cells of the spleen sinuses, the so-called *spleen fibers*, slender forms (Fig. 91, 2), probably capable of contraction, with very prominent nuclei that protrude toward the lumen; they rest upon a thin membrane and are not in contact with one another at their edges. Here numerous leuco-

* The constant diameter (from 6 to 8 μ) of the sheathed arteries suggests that they serve to regulate the arterial blood stream, by preventing a too precipitate flooding of the sinuses and the parenchyma. They are very strongly developed in animals, *e. g.* in the porcupine, the dog, and the pig.

cytes may be seen wandering through the wall of the sinus (Fig. 93). The walls of the sinuses, like those of the veins that follow, are kept open by ring-like strands, that are not elastic in nature, but resemble reticular connective tissue. The larger veins, wholly or partially enclosed in the trabeculæ, possess no proper wall except their epithelium. The spleen venous blood is rich in leucocytes (70 times richer than

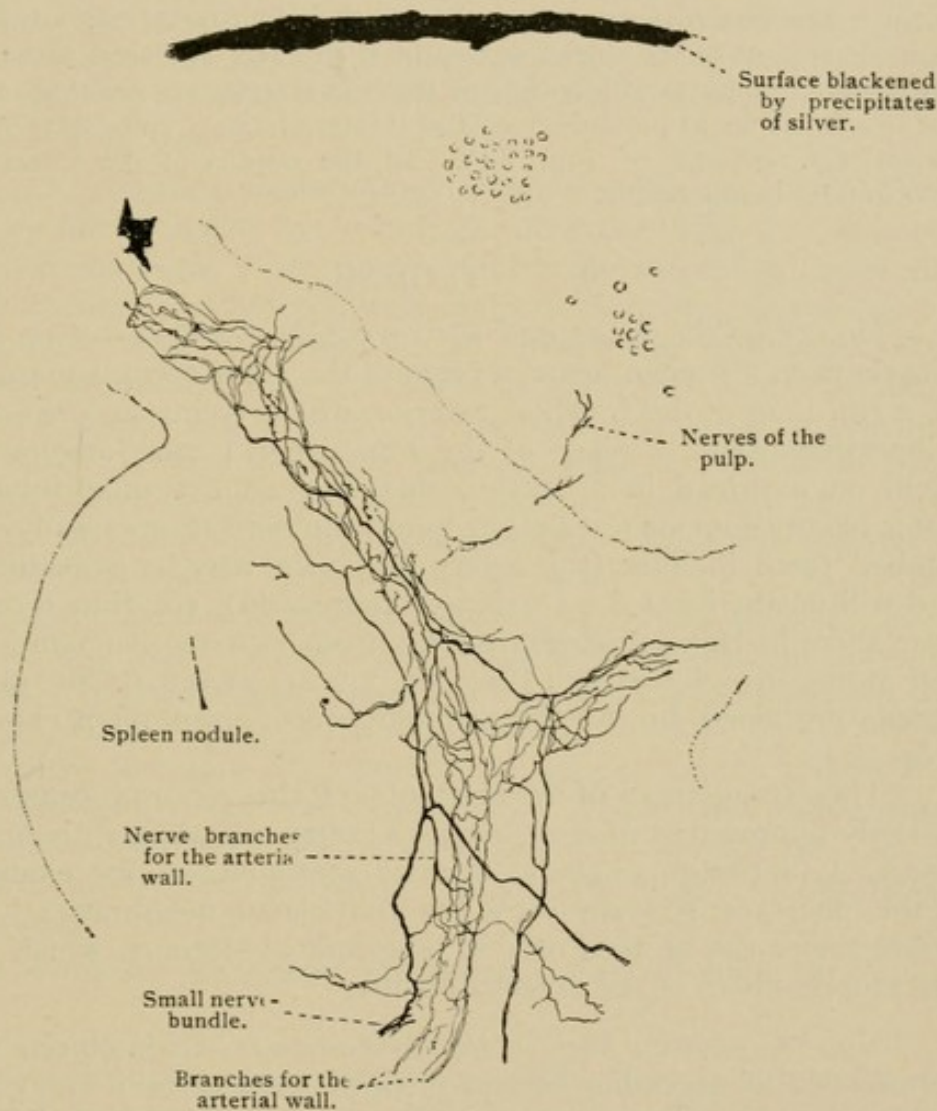


FIG. 94.—SECTION OF THE SPLEEN OF A MOUSE. $\times 85$. The boundary between the spleen pulp and the artery, the sheath of which is infiltrated in its entire length with leucocytes, is indicated by a dotted line. Technic No. 60.

spleen arterial blood) that have come through the patent beginnings of the veins, as well as through the walls of the sinuses.

The *lymph-vessels* are profuse on the surface of the spleen of animals, but in man are only slightly developed. Deep lymph-vessels, running in the interior of the spleen, are wanting.

The *nerves* consist of a few medullated fibers and many naked axis-cylinders. They enter the spleen with the arteries and ramify with

them. During their course they send branches to the musculature of the arteries (Fig. 94) and to the trabeculæ. Plexuses of nonmedullated nerve-fibers are also found in the spleen pulp; they are partly sensory in nature and probably arise from the ramifications of the medullated fibers just mentioned.

The uninjured external surface of the spleen frequently shows boundary marks of spheric lobules; the attempt to find a division into lobules in sections through the human spleen cannot be definitely carried out; though always near the surface of the spleen trabeculæ with their enclosed veins can be regarded as boundaries of lobules and that the arteries are situated in the axis of the lobule, as far as possible from the "interlobular" trabecular veins, can be shown (*cf.* scheme of Fig. 88). In the depths of the spleen a division into lobules is impossible.

TECHNIC.

No. 37.—*The heart and the large blood-vessels.*—Cut a papillary muscle from a human heart, a piece of the aorta 2 cm. square, a piece 1 or 2 cm. long of the brachial artery with its veins and the enveloping connective tissue, a piece of the renal vein 1 cm. long, and suspend them on a thread in a bottle containing 40 c.c. of absolute alcohol. After twenty-four or forty-eight hours the objects are ready to section. Embed them in liver (the artery and vein may be embedded together and will not be injured by strong compression), cut thin cross-sections, stain them in Hansen's hematoxylin, from two to five minutes (p. 38), and mount in xylol-balsam (Fig. 70, 71, 74, 75, 78). The elastic fibers remain unstained, but can be distinctly recognized, often only with high powers.

The arrangement of the elements of the externa cannot be satisfactorily appreciated in cross-sections; often all appear to be circularly disposed (a portion have a circular arrangement, for example, those of the innermost stratum of the external elastic membrane). The exact arrangement can be seen only in longitudinal sections, which also show the muscle-fibers of the externa plainly.

No. 38.—*Elastic fibers of the blood-vessels.*—Stain objects fixed in absolute alcohol according to No. 37, with borax carmine and with resorcin fuchsin (p. 43) and mount in xylol-balsam. Result: similar to figure 77, that preparation having been stained with the less efficient orcein.

No. 39.—*Small blood-vessels and capillaries.*—From the base of a human brain slowly strip off pieces of the pia from 1 to 3 cm. long (in this way delicate blood vessels that penetrate the brain vertically are withdrawn), shake them in distilled water to free them from adherent fragments of brain tissue, and place them in 60 c.c. of Zenker's fluid (p. 33) for one hour; wash them for one hour in running water, and harden them in about 40 c.c. of gradually strengthened alcohols (p. 35). Examine one of these pieces in a watch-glass on a black background and it will be seen that small vessels are isolated.

(a) With a fine scissors cut off small twigs with their ramifications, stain them for from two to five minutes in Hansen's hematoxylin (p. 38) and mount in balsam * (Fig. 72).

(b) From the larger twigs of the cerebral blood-vessels cut pieces about 5 mm. long, slit them open lengthwise, stain them in Hansen's hematoxylin, and place them on a slide with the externa side down. Mount in balsam. By changing the focus the three coats of the vessel and their general arrangement can be seen.

Capillaries can be found on examining fresh brain tissue. They are recognized by their parallel outlines and the oval nuclei of their epithelial cells; they are also found in other preparations, for example in technic No. 9, p. 102.

No. 40.—*Epithelium (endothelium) of the blood-vessels*.—Decapitate a rabbit, open the abdomen by a crucial cut made with the scissors; insert a cork frame about 2 cm. square under the mesentery, span the membrane smoothly and fasten it with quills or hedgehog spines, taking care to touch it as little as possible. Cut around the frame and place the stretched membrane with the frame in 20 or 30 c.c. of 1 per cent. silver-nitrate solution. In about thirty seconds the solution becomes turbid and milky; remove the frame, carefully wash the membrane with distilled water, place the whole in a white capsule containing 100 c.c. of distilled water and expose it to direct sunlight. In a few minutes a brown coloration appears. Now transfer the whole to 50 c.c. of 70 per cent. alcohol (the membrane must be submerged in the alcohol); in a half-hour cut out small pieces 5 or 10 mm. long and mount them in xylol-balsam. In the absence of sunlight take the preparation from the silver solution, wash it, place it for about twenty hours in 30 c.c. of 70 per cent. alcohol, then in a like quantity of 90 per cent. alcohol, and expose it to sunlight on the first opportunity. It must not be forgotten that the whole blood-vessel and not a section of it is present, so that in order to obtain a view such as that in Fig. 73 the surface of the vessel must be in focus.

No. 41.—*Elastic fenestrated membranes*.—See Technic No. 15, p. 103.

No. 42.—*Development of capillaries*.—Chloroform a seven-day-old rabbit, fasten it with pins on a cork plate, open the abdomen by a crucial incision, quickly remove the spleen, stomach, and attached greater omentum and place these parts in 80 c.c. of a saturated aqueous solution of picric acid (p. 22). In this solution the omentum, otherwise difficult to separate, spreads out easily. After one hour cut it off, transfer it to 60 c.c. of distilled water, and divide it with the scissors into pieces about 1 cm. square. Place such a piece on a dry slide, remove the water with filter-paper, and with needles spread it out as smooth as possible, which is

* Frequently the blood-vessels are filled with blood-cells, which make an exact study of the vascular wall more difficult; this obstacle can be removed by placing the fresh blood-vessels in distilled water for an hour. In this way the cells are decolorized (cf. technic No. 43).

the more easily done the less moisture there is present. Put one or two drops of Hansen's hematoxylin on the preparation. In from one to five minutes drain off the hematoxylin and place the slide with the preparation in a flat dish containing distilled water; the membrane will soon float from the slide and will remain smooth, and in five minutes should be transferred to a watch-glass containing eosin (p. 39), in which it should remain three minutes. It should then be washed for one minute in distilled water and placed on a slide; the water should be absorbed with filter-paper, any wrinkles smoothed out with needles, and a cover-glass with a drop of dilute glycerol suspended from its lower surface applied. The preparation may be mounted in balsam instead of glycerol (that is, dehydrated in 95 per cent. alcohol, cleared in carbol-xylol, and then mounted in xylol-balsam), but the finer structural details are apt to be lost. The colored blood corpuscles are stained a bright red by the eosin (Fig. 79).

In spreading the membrane on the slide delicate young capillaries may be easily torn from the older capillaries and then simulate "isolated cells containing blood corpuscles"; such artifacts, also atrophying capillaries, have been described as "vasoformative cells."

No. 43.—*Colored blood-cells of man.*—Carefully cleanse a slide and a small cover-glass (finally with alcohol). With a needle cleansed shortly before by heating prick the finger-tip at one side; lightly touch the first drop of blood that escapes with the cover-glass and *at once* put it on a small drop of 0.75 per cent. salt solution previously placed on the slide. With the high power many colored cells adhering to one another by their broad surfaces, forming the so-called rouleaux (Fig. 80), can be seen, as well as isolated colored and colorless blood-cells. The distortion of many of the colored cells is due to evaporation, in consequence of which they are beset with minute spines, are *crenated*. If a drop of water be placed at the edge of the cover-glass, the cells soon become decolorized and the water acquires a yellowish tinge; at the same time the cells become spherical, have the appearance of pale circles, "shadows," and finally disappear. In studying this process of decoloration the student is advised to concentrate his attention upon a *single* cell.

No. 44.—*Permanent preparation of colored blood-cells.*—By means of a sable pencil spread living blood, fresh from the finger, in the thinnest possible film on a carefully cleaned slide and let it dry in the air. Cover the dry preparation with a dry cover-glass and seal the edges with cement (p. 50). Among many misshapen forms a few blood-cells, well-preserved in form and size, may be found.

No. 45.—*Permanent preparations of colored and colorless blood-cells* are made by Ehrlich's dry method. This method accurately carried out, after some practice, yields good results, but with unskilful manipulation many caricatures arise and mislead the inexperienced. The employment of this method for purposes of investigation and discovery requires great skill and great caution in judgment.

Preliminary manipulations.—For each preparation two *thin* cover-glasses are required, they should not be over 0.1 mm. thick and should be cleaned by placing them for a few minutes in dilute hydrochloric acid, then in distilled water, and finally in alcohol. It is best to take cover-glasses that have never been used. Prepare a mixture of equal parts of absolute alcohol and ether (about 5 c.c. of each). Cleanse the tip of the finger first with soap and water, then with a tuft of clean cotton-wool moistened with the alcohol-ether mixture. With a clean needle (not previously used for anatomic purposes) prick the pad of the finger, made slightly hyperemic by compression; take up a cover-glass with the forceps (not with the fingers), press it lightly upon the escaping drop of blood and place it on the second cover-glass, with one edge projecting slightly. The drop of blood will spread out in a thin film between the two glasses, which are then *slipped* apart by means of two forceps. By this manipulation the influence of the insensible perspiration on the blood-cells is prevented, which otherwise would shrink or lose their hemoglobin.

Exposed to the air the blood on the cover-glasses dries in a few minutes; they are then to be placed in the alcohol-ether mixture for fixation. In from one-quarter to two hours they should be removed, again dried in the air, when they are ready for further treatment, which may be applied immediately or later, since the fixed preparations can be preserved for a long time.

Further treatment. (a) *Oxyphile (eosinophile, a) granules.*—Place the cover-glass preparations for twenty-four hours in about 4 c.c. of distilled water to which about 10 drops of eosin solution have been added. Rinse one minute in distilled water and stain for from one to five minutes in a watch-glass with Hansen's hematoxylin (p. 38). Transfer to distilled water; remove in five minutes and let the preparations dry in air under a bell-glass. Mount the dry preparation, without further treatment, in a drop of xylol-balsam. The colored blood-cells and the oxyphile granules of the colorless blood-cells are stained bright red; the nuclei are blue. The oxyphile granules occur sparingly (2 to 4 per cent.) in the leucocytes of normal blood, of lymph, and of the tissues. They are numerous in the bone-marrow of the rabbit. A magnification of 400 diameters is sufficient to find them.

(b) *Basophile (mast-cell) granules.*—Stain the dry cover-glass preparation after the method given in No. 7, p. 101, dry, and mount in balsam. These granules are rare in normal blood (0.5 per cent. at most).

(c) *Neutrophile (ε-) granules.*—(1) Dissolve 1 gm. of orange-yellow extra in 50 c.c. of distilled water; (2) 1 gm. of acid-fuchsin extra in 50 c.c. of distilled water; (3) 1 gm. of crystalline methyl-green in 50 c.c. of distilled water, and let the three solutions settle. Then mix 11 c.c. of solution (1) with 10 c.c. of solution (2) and add 20 c.c. of distilled water and 10 c.c. of absolute alcohol; to this mixture add a mixture of 13 c.c. of solution (3), 10 c.c. of distilled water, and 3 c.c. of absolute alcohol. The whole is then allowed to stand for one or two weeks. In this "triacid solution" the dry cover-glass preparation

should be placed for fifteen minutes, then washed, dried, and mounted in balsam. The neutrophile granules, which are found in leucocytes with lobulated nuclei of normal and other blood, are of a violet color and are easily seen with the usual dry high-power lenses; the oxyphile granules and the colored blood-cells are of a yellow-brown to chocolate-brown color, the nuclei a bright blue-green, though their outlines are not so distinct as in the hematoxylin preparations.

No. 46.—*Blood-platelets*.—Mix about 5 drops of an aqueous solution of methyl-violet (p. 26) with about 5 c.c. of salt solution (p. 20). **Filter** the mixture and place a drop of it on the tip of the finger; prick the finger through the drop; the escaping blood mixes with the methyl-violet; take up a drop with the cover-glass and examine with the high power. The platelets are stained an intense blue of a peculiar luster, are disk-shaped (Fig. 80), and should not be confused with the white blood-cells likewise stained blue. They are numerically variable elements, occurring in large numbers in the blood of one individual, while in the blood of another they are only to be found singly here and there. Care must be taken not to confuse them with foreign particles, which may occur even in the filtered staining solution.

No. 47.—*Colored blood-cells of the frog*.—Prepare the slide and treat the blood, taken from the recently killed animal, after No. 43.

No. 48.—*For forensic purposes*.—Since it is usually dried blood that is to be examined, dissolve small particles of dried blood in 35 per cent. potash solution on a slide; blood-stained pieces of linen may be teased in a drop of the same solution. Although the colored blood-cells of native mammalian animals are smaller than those of man, it is nevertheless impossible from the size of the blood-cell to determine its source. On the other hand, it is easy to distinguish the disk-shaped cells of mammals from the oval elements of other vertebrates.

No. 49.—*Colorless blood-cells (leucocytes) in motion*.—*Preliminary manipulations*: Carefully cleanse a slide and cover-glass with alcohol. Kill a frog, grasp it by its hind legs, dry its back somewhat with a cloth, and with fine scissors make an incision 1 cm. long parallel to and close beside the vertebral column. Introduce a capillary pipet into the wound (with the tip directed forward) and suck the tip full. A small drop is sufficient; blow it on to the slide, cover it quickly, and seal the edges with melted paraffin (p. 53). Such a preparation shows colored and colorless blood-cells; at first the nuclei of the former are indistinct. The nuclei of living colorless blood-cells are in general invisible. For the study of ameboid movement select leucocytes the protoplasm of which is partly granular and which are not spherical. The movements are slow; of this one can best convince one's self by studying a single leucocyte and making sketches of it at intervals of from one to two minutes. Study with the high power (Fig. 6).

No. 50.—*Blood-crystals*.—(a) *Hemin crystals* are easily obtained. Cut a small strip about 3 mm. wide from a piece of linen previously

saturated with blood and dried and place it with a pinhead-sized crystal of common salt on a clean slide; add a large drop of glacial acetic acid and with a glass rod stir the linen and salt for about one minute or until the acid acquires a brownish tinge. Then heat the slide over the flame until the acetic acid boils. Quickly remove the linen and examine the dry brown places on the slide with the high power (from 240 diameters up). Occasionally the *brown crystals* can be seen without the cover-glass and without a mounting medium, lying next to numerous fragments of white salt-crystals (Fig. 82, 1). For preservation add a large drop of balsam and apply a cover-glass. The hemin crystals differ greatly in form and size. Well-developed crystals lying singly or crosswise over one another, or arranged in stellate groups, whetstone forms and minute particles that scarcely exhibit crystallization are obtained from the same drop of blood. The demonstration of the hemin crystals is of great importance in a forensic respect. While it is easy to obtain the crystals in large stains on wearing apparel, it is difficult when the stains are small, especially on rusty iron, to prove that they are from blood. The instruments and reagents employed in such investigations must be absolutely free from contamination.

(b) *Hematoidin crystals* are obtained by teasing old blood extravasations; they can be recognized macroscopically by their reddish-brown color, for example, in the corpus luteum, in cerebral hemorrhages (Fig. 82, 3).

(c) *Hemoglobin crystals* are obtained by transferring 5 c.c. of the blood of a dog to a test-tube, adding a couple of drops of ether, and shaking vigorously until the blood becomes lake-colored. Then spread a few drops on a slide and let the preparation dry in the cold. When crystallization has occurred add a drop of glycerol and apply a cover-glass. The large crystals often exhibit a tendency to cleave lengthwise (Fig. 82, 4 a).

No. 51.—*Lymph-vessels*.—For the study of the *walls* of the larger lymph-vessels, select the vessels opening into the inguinal glands, that are large enough to be taken out with forceps and scalpel. Prepare like the large blood-vessels, No. 37, or after No. 39 b.

No. 52.—For the exhibition of the more *delicate lymph-vessels*, of their course and their distribution, the method of interstitial injection is often employed. The needle of a hypodermic syringe filled with Berlin blue is thrust haphazard into the tissue; this is a crude method, the results of which are of very doubtful value. Even though here and there actual lymph-vessels may thus be filled, in most cases the injection-mass is simply driven forcibly into the interfascicular clefts of the connective tissue. The value of any opinion with regard to "radicles of lymph-vessels" and to "lymph-spaces" thus exhibited is self-evident.

No. 53.—*Lymph-glands*.—For a general view the mesenteric glands of kittens are suitable. For fixation and hardening place them in 30 c.c. of absolute alcohol; in three days thin sections can be readily made and should be taken so that they pass through the hilus, which is

easily recognized macroscopically by an external depression. Longitudinal sections passing through the poles of the glands are best, though transverse sections are also useful. Stain six or eight sections in Hansen's hematoxylin for from two to three minutes, then in eosin for one minute (No. 3 (*b*), p. 39), transfer them to a test-tube half filled with distilled water and shake them for from three to five minutes. Pour the shaken sections into a flat dish; the cortex and medulla can be macroscopically distinguished by the uniformly blue color of the former and the variegated appearance of the latter. Mount in xylol-balsam. The trabeculae are but slightly developed. The fragments of adipose tissue adhering to the glands must not be taken for reticular tissue. High magnification is of no advantage, the sharp outlines disappear and the picture loses in distinctness.

No. 54.—*Lymph-glands of mature animals and of man* are difficult to understand, because the entire cortex is transformed into a continuous mass irregularly sprinkled with germinal centers. Still, thin sections of small glands fixed in Zenker's fluid (p. 33) and hardened in gradually strengthened alcohol (p. 35), stained after the method of van Gieson (p. 43), give satisfactory pictures (Fig. 84). The lymph sinuses cannot be brought distinctly to view by shaking the sections and the germinal centers are apt to fall out and leave round spaces macroscopically recognizable.

No. 55.—The mesenteric follicles of the ox are well adapted for the representation of the network of *medullary cords* and *trabeculae*. Place pieces 2 cm. long in 200 c.c. of concentrated aqueous picric-acid solution and after twenty-four hours, with a sharp knife moistened with water, try to cut thin sections. This is not so easily done as after alcohol fixation, but slightly thicker sections can be used. Place the sections for one hour in 100 c.c. of distilled water, which must be changed frequently, stain with Hansen's hematoxylin and with eosin (No. 3 (*b*), p. 39), and shake them (see No. 53). Mount in xylol-balsam (p. 50). The trabeculae are red, the medullary cords blue; with low magnification the picture is like Fig. 86; with high magnification the reticular connective tissue of the lymph sinuses can be seen; the majority of the leucocytes occupying the meshes become loosened by the treatment with picric acid and are lost in the shaking (Fig. 87).

No. 56.—*Elements of the spleen*.—Make an incision through a fresh spleen; with a scalpel obliquely applied scrape the cut surface and examine a little of the red mass adhering to the blade in a drop of salt solution. Use the high power. Often, especially in animals, only colored and colorless blood corpuscles are found; some of the latter contain minute granules. In human spleens, in addition to the numerous colored blood-cells altered in form, epithelial cells of the lymph sinuses are always found; the latter were formerly called "spleen-fibers" (Fig. 91, 2, 3). In many human spleens multinucleated cells and cells containing erythrocytes are often sought in vain (Fig. 91, 4).

No. 57.—*The spleen*.—Without cutting it, fix the entire spleen in Müller's fluid (p. 33), using one liter for a human, 200 to 300 c.c. for a cat's spleen. After two weeks for the cat's, five weeks for the human spleen, wash for from one to two hours in running water, cut out pieces 2 cm. square and harden them in 60 c.c. of gradually strengthened alcohols (p. 35). The spleen nodules can be seen on the cut surface with the unaided eye. Sections not too thin are to be stained in Hansen's hematoxylin and mounted in balsam. If it is desired to differentiate the trabeculæ, after staining in hematoxylin place the sections for a half minute in eosin * (No. 3 (b), p. 39). In successful preparations the pulp cords and the spleen nodules are blue, the trabeculæ rosy, the vessels distended with blood corpuscles brown. The sections are most satisfactory when examined with a very low power (Fig. 90); with the high power the outlines are often indistinct. Fixation in Zenker's fluid (p. 33) is recommended for thin sections, with staining after van Gieson (p. 43).

No. 58.—*Reticular connective tissue of the spleen*.—Shake a *thin* section fixed and stained according to No. 57 for about five minutes in a test-tube half filled with distilled water. Mount in glycerol. The leucocytes are difficult to dislodge; the narrow-meshed network can be seen only at the edges of the preparation (Fig. 92).

No. 59.—*Blood-vessels of the spleen* are incidentally exhibited by injecting the stomach and intestine (compare with No. 116).

No. 60.—*Nerves of the spleen*.—For this purpose the spleen of the mouse is best suited. Halve it and apply Golgi's method for the demonstration of the elements of the nervous system (p. 45). It is sometimes sufficient to place the object in the osmio-bichromate mixture (in a warm oven) for three days and for the same length of time in the silver solution; often a repetition of the whole process once or twice yields good results.

II. ORGANS OF THE SKELETAL SYSTEM.

The skeletal system mainly consists of a large number of firm bodies, the bones, which are joined together by special structures and in their entirety form the skeleton.

In the embryo the greater part of the skeleton consists of cartilage, which in the course of development is supplanted by bone and with the exception of a few remnants disappears; such remnants are the costal cartilages and the cartilages of the joints, which cover the articular surfaces of many bones. Skeletal cartilages are also found in the respiratory passages and in the organs of special sense.

* Longer staining makes the erythrocytes bright red, the trabeculæ dark red, and the easy distinction between them is lost.

THE BONES.

On sawing through a fresh bone at once it will be seen that its texture is not everywhere alike, but that the osseous tissue appears in two forms: the one, a very dense, firm, apparently structureless substance, constitutes the principal portion of the periphery and is termed *compact bone* (*substantia compacta*); the other, toward the axial cavity, appears as an irregular reticulum of thin osseous lamellæ and slender trabeculæ, and is called *spongy bone* (*substantia spongiosa*). The interstices of the spongy bone, as well as the central marrow-cavity, are filled with a soft mass, the *bone-marrow*; the surface of the bone is enveloped in a fibrous

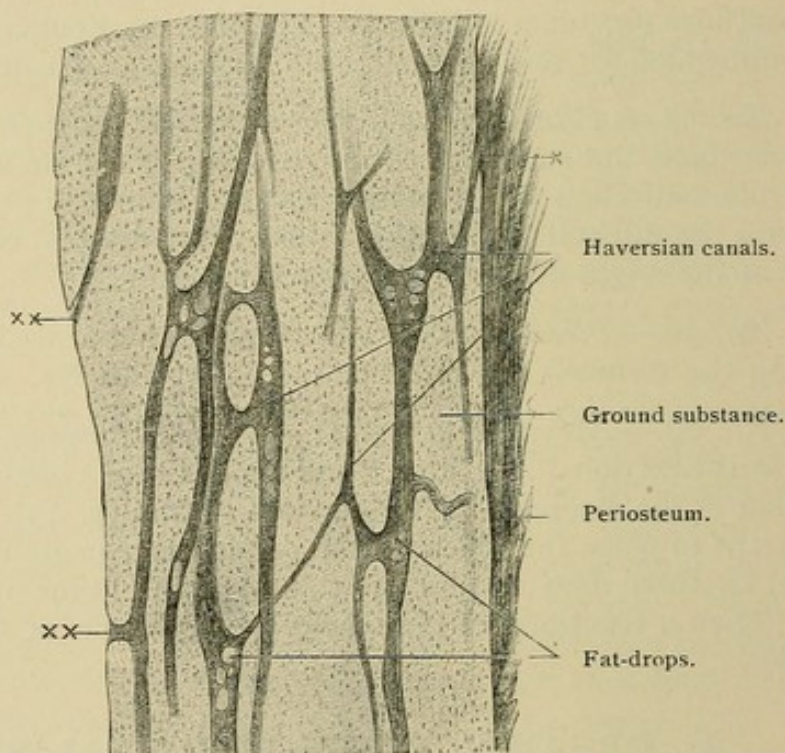


FIG. 95.—FROM A LONGITUDINAL SECTION OF A HUMAN METACARPUS. $\times 30$. Fat-drops are seen in the Haversian canals. At x Haversian canals open on the outer, and at xx on the inner surface of the bone. Technic No. 63.

membrane, the *periosteum*. The proportion between the compact and the spongy substance is somewhat different in the *short* bones, which consist chiefly of the latter, the compact substance being confined to a narrow zone at the periphery. *Flat* bones have a sometimes thicker, sometimes thinner cortex of compact substance, while the interior is filled with spongy substance. In the epiphyses of the long bones, as in the short bones, the spongy substance preponderates.

The *spongy substance* consists entirely of osseous tissue (p. 98); the *compact substance*, on the other hand, contains besides the bone canaliculi and lacunæ a second system of *larger* canals, from 22 to 110 μ wide, which divide dichotomously and form a wide-meshed network. These

canals contain the blood-vessels and are named *haversian canals*. In the long bones, in the ribs, in the clavicle, and in the inferior maxilla their course is parallel to the long axis of the bone; in short bones they run mainly in *one* direction, for example, vertically in the vertebræ; in the flat bones their course is parallel to the surface, not infrequently in lines that radiate from a point, as in the tuberosity of the parietal bone. The haversian canals open on the outer surface of the bone (Fig. 95, x), as well as on the inner surface (Fig. 95, xx) directed toward the substantia spongiosa.

The ground-substance of compact bone is arranged in lamellæ, that is, the osseous fibrillæ (p. 98) are joined in bundles and these placed side by side form thin plates or lamellæ. According to the disposition of these plates three lamellar systems can be distinguished: an annular

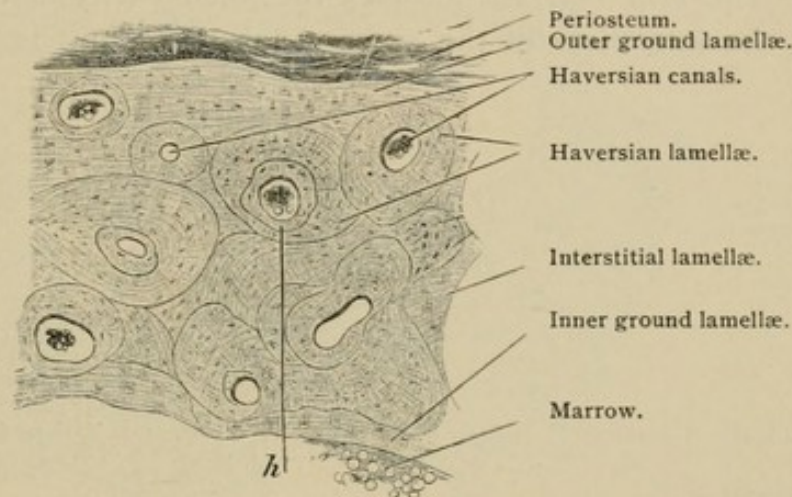


FIG. 96.—FROM A CROSS-SECTION OF A METACARP OF MAN. $\times 50$. The haversian canals contain a little marrow (fat-cells). Resorption line at *h*. Technic No. 63.

system encircling the haversian canals, which in cross-section exhibits from eight to fifteen lamellæ concentrically arranged around an haversian canal; these lamellæ are called *haversian* or *special lamellæ* (Fig. 96). Transverse sections show that the haversian systems are in contact in portions of their circumference, elsewhere are kept apart by strata of osseous lamellæ running in a different direction. These more irregularly disposed lamellæ between the haversian systems are named *intercalated* or *interstitial lamellæ*; they are connected with the third, superficial lamellar system, the *general* or *ground lamellæ*, in which the osseous strata encircle the outer surface of the bone and are called *outer ground lamellæ*; occasionally similar circular lamellæ are found on the inner free surface and are named *inner ground lamellæ*. The general lamellæ contain an extremely variable number of canals for vessels, which unlike the haversian canals are not the centers of annular systems of lamellæ;

they are called Volkmann's canals and the contained vessels, the "perforating vessels." The latter freely connect with the vessels of the haversian canals; the transition of Volkmann's canals into the haversian canals is a very gradual one.

The bone lacunæ in the compact substance have quite definite positions. In the haversian lamellar systems their long axis is parallel to the long axis of the haversian canals and they are bent in the direction of the surface, so that cut transversely they appear concentrically curved to the cross-section of the haversian canal. In the interstitial lamellæ the lacunæ are placed irregularly, in the ground lamellæ so that their surfaces run parallel to the surfaces of the lamellæ. The bone canaliculi open into the haversian canals and on the free outer and inner surfaces of the bone.

The *bone marrow* occupies the axial cavity of the tubular bones, fills the interstices of the spongy substance, and is also found in the

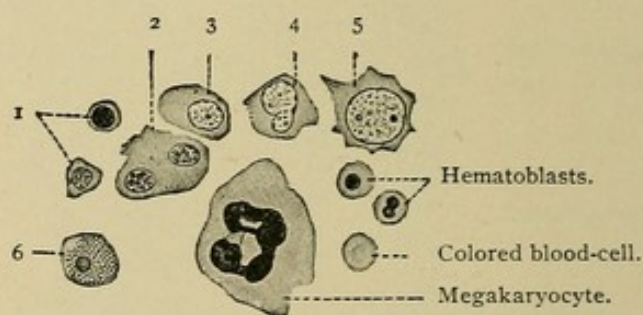


FIG. 97.—ELEMENTS OF HUMAN BONE MARROW. $\times 600$. 1-5. Various forms of marrow cells. 6. Eosinophile cell. Technic No. 64 b.

larger haversian canals. It is of a red or a yellow color and therefore two varieties are distinguished, the *red marrow* and the *yellow marrow*. The red marrow is found in the flat bones, in the vertebræ, in the base of the skull, in the sternum, in the ribs, and in all young bones (also in the long bones of small animals); the yellow marrow occurs in the short and long bones of the extremities. In old and in sick persons the marrow is mucoid and reddish-yellow and is then called *gelatinous* bone-marrow; it is characterized simply by its poverty in fat.

The elements of *red marrow* are connective tissue, marrow cells, giant cells, and hematoblasts. The scanty *connective tissue* consists of bundles of fibrillæ, connective-tissue cells, and fat-cells. In the large marrow cavities the bundles of fibrillæ are denser and form a lining membrane, the *endosteum*, while in the marrow spaces of the spongy substance they are almost entirely wanting. Elastic elements are absent. The *marrow cells* are leucocytes and predominantly uninuclear neutrophile (so-called myelocytes) and polymorphous-nuclear varieties (Fig.

81 *b, c*); also eosinophile leucocytes, mast-cells, and, in considerably lesser quantity, nongranular uninuclear cells. The giant cells are huge, extremely irregularly shaped structures, of which two varieties are distinguished: (*a*) *megakaryocytes* (Fig. 97), cells with *one* huge nucleus, varying greatly in shape; it is round, or lobed, or flat and ring-like (Fig. 109, 2 *r*) or it forms a network; (*b*) *ostoclasts* (p. 177), cells containing *several* small nuclei (Fig. 104 and 108); they invariably lie in the neighborhood of the bone, or of the cartilage, while the megakaryocytes lie in the interior of the marrow.

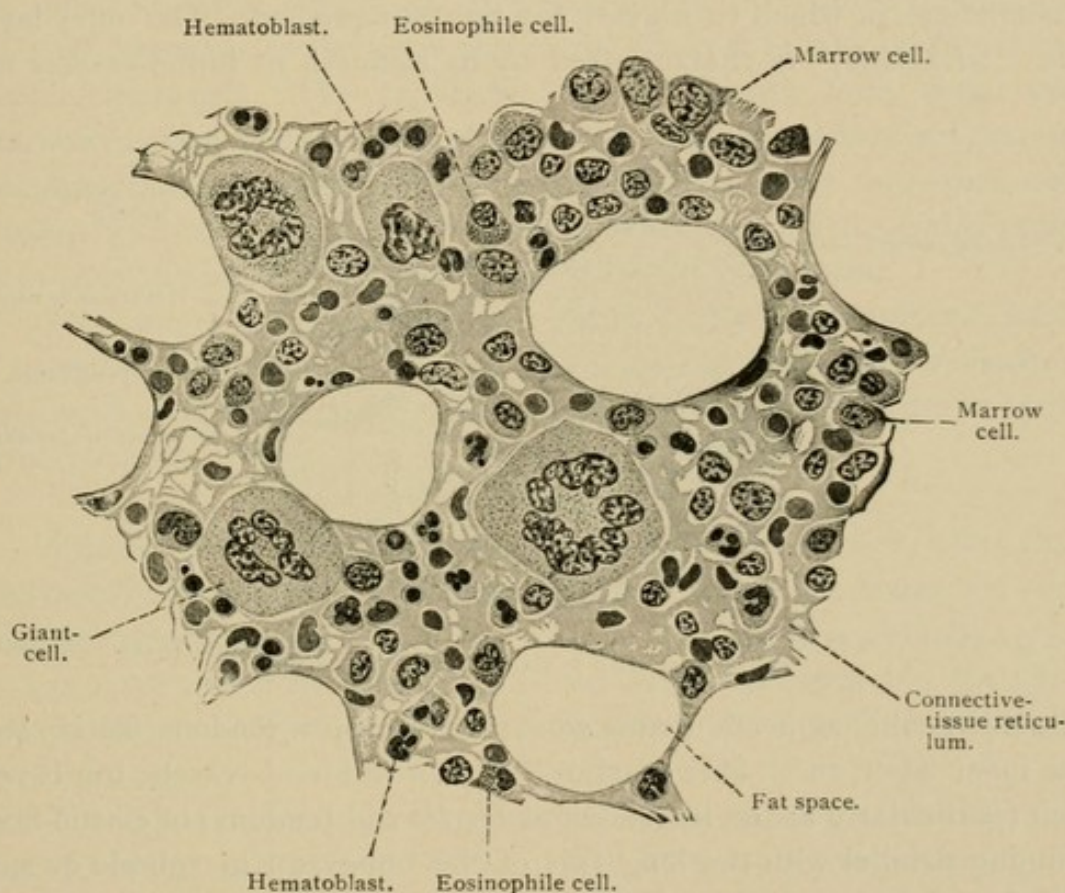


FIG. 98.—SECTION OF THE BONE MARROW OF A RABBIT, SHOWING THE DELICATE CONNECTIVE-TISSUE RETICULUM CONTAINING THE DIFFERENT ELEMENTS OF THE MARROW. $\times 400$. (Schaper.)

Many giant cells are structural anomalies related to leucocytes, being enlarged and modified forms of the latter; the osteoclasts, according to recent investigations, are said to originate in the walls of the blood capillaries, by proliferation of the protoplasm and multiplication of the nuclei of the epithelial cells and subsequent constriction from the maternal basis. By constriction of the nucleus into several parts a uninuclear giant cell may become a multinuclear cell (Fig. 109, 3 *r*); or with the nuclear particles corresponding portions of the protoplasm may be separated by constriction, resulting in uninuclear cells (*cf.* budding, p. 72). The supposition that these processes of division are the phenomena of a reversed series of processes, the merging of several cells into one, has very little probability, since the process of budding has been observed in living cells. Trophospongium canals have been observed in the giant cells (p. 64).

The *hematoblasts* are nucleated cells with yellow colored protoplasm, resembling that of the erythrocytes. They are the mother cells of the erythrocytes (Fig. 97 and 98). Yellowish pigment corpuscles occurring in various cells are regarded as remains of degenerated red blood-cells.

The *yellow marrow* consists of much fat and of connective tissue. Marrow cells and hematoblasts are found only in the yellow marrow of the head of the humerus and of the femur.

The *periosteum* is a compact membrane consisting of connective-tissue fibers, in which two layers can be distinguished. The outer layer, the "adventitia," is characterized by its richness in blood-vessels and

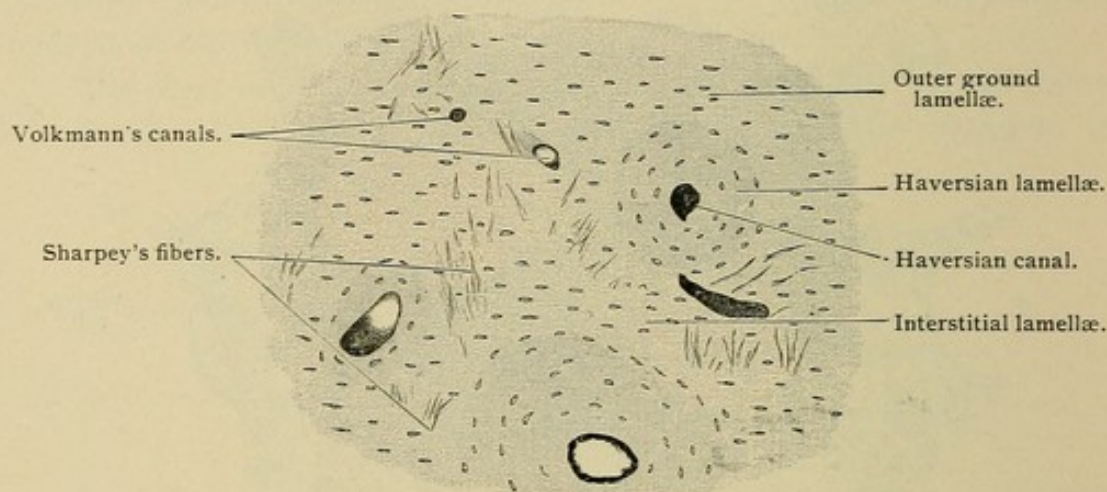


FIG. 99.—FROM A CROSS-SECTION OF THE FEMUR OF ADULT MAN. $\times 80$. Technic No. 62. The lamellæ can be recognized by the position of the lacunæ.

establishes the connection with adjacent structures, tendons, fasciæ, etc.; the inner layer, the "fibro-elastica," is poor in blood-vessels, but is very rich (particularly at the insertions of fasciæ and tendons) in elastic fibers running parallel with the long axis of the bone and in spheric or spindle-shaped connective-tissue cells. Here and there on the inner surface a layer of cubical cells* is found, that are of importance in the development of the bone. The periosteum is sometimes firmly, sometimes loosely attached to the bone; the attachment is effected by the blood-vessels passing to and from the bone and by Sharpey's fibers (p. 98, remark †), which pierce the outer ground and the adjacent interstitial lamellæ and extend in all directions (Fig. 99). In the tubular bones elastic elements of the fibro-elastica of the periosteum penetrate the bone in company with many Sharpey's fibers and without regard to the

* The vitality of the cells of the fibro-elastica is very great. The periosteum of a corpse kept at 15° C., which is transplanted 168 hours after the death of the organism is said to be still capable of producing cartilage and osseous tissue.

lamellar structure of the bone run in the more superficial strata. There are also elastic fibers that penetrate independently of Sharpey's fibers. In the bones of the vertex of the skull elastic elements are wanting.

The *blood-vessels* of the bone, the marrow, and the periosteum are in the closest connection with one another, and also with surrounding structures. Small branches (not capillaries) of the numerous arterial and venous vessels of the periosteum everywhere enter the haversian and Volkmann's canals and on the inner surface of the bone are in communication with the blood-vessels of the marrow. The latter is supplied by the nutrient artery, which on its way through the compact substance gives off branches to the same and in the marrow breaks up into a rich vascular network. The capillaries of the marrow form wide, very thin-walled,* valveless veins; of the larger, likewise valveless, veins one accompanies the nutrient artery, while the others make numerous connections with the veins of the compact substance. True *lymph-vessels* occur only in the most superficial layers of the periosteum.

The *nerves* are numerous and consist partly of medullated, partly of nonmedullated fibers. They enter the haversian canals and the bone marrow, also the periosteum, where occasionally they terminate in lamellar corpuscles.

THE ARTICULATIONS OF BONES.

Two forms of articulation are recognized: *synarthroses*, joints characterized by immobility; *diarthroses*, joints in which the bones are movable, one upon the other.

In *synarthroses* the bones are joined either by ligaments, the union constituting a *syndesmosis*; or by the intervention of cartilage, forming a *synchondrosis*.

The ligaments are partly *fibrous bands*, possessing a structure like that of tendon, partly *elastic bands*. The latter are distinguished by the possession of numerous robust elastic fibers, which are never arranged in bundles or lamellæ, but are always separated by loose connective tissue (*cf.* Fig. 35 C). The ligamentum nuchæ, the ligamenta subflava, and the ligamentum stylohyoideum are elastic ligaments.

The *sutures* also belong to the syndesmoses; they are short fibrous ligaments that extend from one serrated osseous edge to the other.

The cartilage in synchondroses is rarely only of the hyaline variety, but usually is in part fibro-cartilage and (especially at the borders in contact with the bone) in part hyaline, in which the cell-capsules are frequently calcified.

* These delicate walls were formerly overlooked, whence arose the teaching that blood-spaces without walls exist in bone marrow.

The intervertebral ligaments, which likewise belong to the synchondroses, possess in their center a soft, gelatinous substance, the *nucleus pulposus*, that contains large groups of cartilage cells; it is the remains of the notochord, the embryonic precursor of the vertebral column. At the periphery of the intervertebral ligaments there is a narrow tendinous zone.

In *diarthroses* the parts entering into a joint are the articular ends of the bones, the capsular ligament, the marginal fibro-cartilages (*labra glenoidalia*), and the interarticular cartilages (*menisci*).

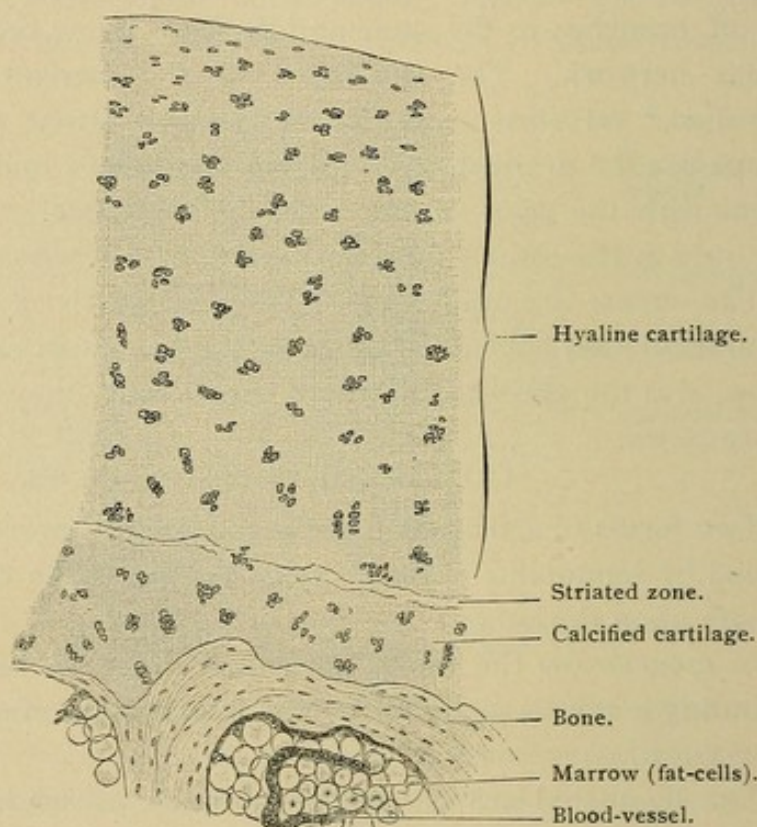


FIG. 100.—VERTICAL SECTION THROUGH THE HEAD OF A METACARPUS OF ADULT MAN. $\times 50$.
Technic No. 65.

The *articular ends of the bones* are covered by a stratum of hyaline cartilage from 0.2 to 5 mm. thick, thinning toward the edges. In the superficial parts the cartilage cells are flattened and placed parallel to the surface; those in the median parts are rounded* and are often united in groups; in the deepest portions the groups of cells are partly arranged in longitudinal rows, vertical to the surface of the bone; following, but separated by a narrow striated belt, is a small zone of calcified cartilage

* The cells of the articular cartilages have been described as having processes which extend into the adjacent cartilaginous matrix. The flattened cartilage cells of the deeper portions are said to possess lobulated nuclei.

interposed between and connecting the hyaline cartilage and the osseous tissue (Fig. 100).

Not all the articular cartilages exhibit the structure just described; the cartilages of the costo-vertebral, the sterno-clavicular, the acromio-clavicular, and the maxillary articulations, and the head of the ulna are not hyaline, but fibro-cartilage; the distal articular surface of the radius is covered with dense fibrous tissue.

The *glenoid ligaments* and the *interarticular cartilages* do not exhibit the characteristic cartilage matrix; they consist of a compact fibrous connective tissue and partly of spherical cells. To the same category belong the so-called sesamoid cartilages. The tendon sheath of the cuboid bone, however, contains genuine cartilage.

In the adult nerves and blood-vessels are wanting in the articular cartilages, also in the interarticular cartilages and the glenoid ligaments.

The *joint capsules* consist of an external fibrous layer, *stratum fibrosum*, varying greatly in thickness and possessing a structure like that of the ligaments above described, and of an internal membrane, the *stratum synoviale*, the free inner surface of which is smooth and glossy. The outer layer of the latter is composed of loose elastic fibers and fibrillar connective tissue, here and there containing fat-cells; within this is a thin lamella of parallel connective-tissue bundles, in which, toward the joint cavity, there are small spherical or stellate cells, 11 to 17 μ in size, containing a large nucleus. These cells are sometimes few in number,—at points subjected to pressure—sometimes very abundant and form distinct epithelial (endothelial) layers, covering the inner surface with a three or four-fold stratum.

The synovial membrane (*stratum synoviale*) often forms folds containing fat and projecting into the synovial cavity and on its free surface bears the *synovial villi* (Fig. 101), variously shaped processes, mostly of microscopic size, which are particularly closely set on the edges of the joint-surfaces and bestow upon the synovial membrane a reddish, velvety appearance. They consist of connective tissue and are covered with a single or double layer of epithelial cells.

The larger *blood-vessels* of the synovial membrane lie in the loose



FIG. 101.—SYNOVIAL VILLI WITH BLOOD-VESSELS FROM A HUMAN KNEE-JOINT. $\times 50$. The epithelium has fallen from the apex of the left villus, exposing the connective tissue. Technic No. 66.

connective-tissue layer; from here capillaries extend through the inner thin connective-tissue stratum and penetrate within the villi. Some of the villi are nonvascular. The lymph-vessels lie close under the epithelium.

The *nerves* run in the loose connective-tissue stratum and in part terminate in lamellar corpuscles (see End-bulbs).

The *synovia* contains more or less profoundly altered cells, fragments of cells, and oil globules, all products of a physiologic process of waste of the surfaces of the synovial membrane and the articular cartilage; also, albumin, mucus, and salts; these solid constituents amount only to six per cent., the remainder consists of water.

THE CARTILAGES.

The *costal cartilages* are of the hyaline variety; the matrix exhibits the peculiarities previously described (p. 96), the cells frequently contain fat. Their surface is enveloped by a compact fibrous membrane, the *perichondrium*, which consists of interlacing connective-tissue bundles and elastic fibers.

The *articular cartilages* are covered by the perichondrium only on their lateral surfaces, not on their contiguous surfaces. Where the cartilage and the perichondrium are in contact there is a gradual transition of the one tissue into the other and consequently the attachment between the two is very firm.

The perichondrium carries the nerves and the blood-vessels; the latter also run within growing cartilage, in buried canals. In the adult cartilage is non-vascular; the nutrition of the tissue depends upon diffusion from the surface. In advanced life the costal cartilages often contain blood-vessels because of beginning ossification.

The *cartilages of the special-sense organs and of the respiratory organs* will be described in the respective chapters.

THE DEVELOPMENT OF THE BONES.

The bones are relatively late structures to appear. The development of the muscles, nerves, vessels, brain, spinal cord, etc., is already well advanced in an embryonal period when not a trace of bone is present. At that time the skeleton of the body is formed of hyaline cartilage. With the exception of certain parts of the cranium and nearly all the bones of the face, the future osseous skeleton is represented in cartilage. For example, in the upper extremity the humerus, radius, ulna, carpus, and skeletal parts of the hand consist of cartilaginous pieces that are not hollow like the bones by which they are subsequently

replaced, but are solid throughout. The osseous skeleton then gradually appears in the place of the cartilaginous skeleton. All the osseous parts that in the embryo were preceded by cartilage are called *primary* or endochondral bone; the other bones, which have no cartilaginous precursors, are named *secondary* or connective-tissue bone.

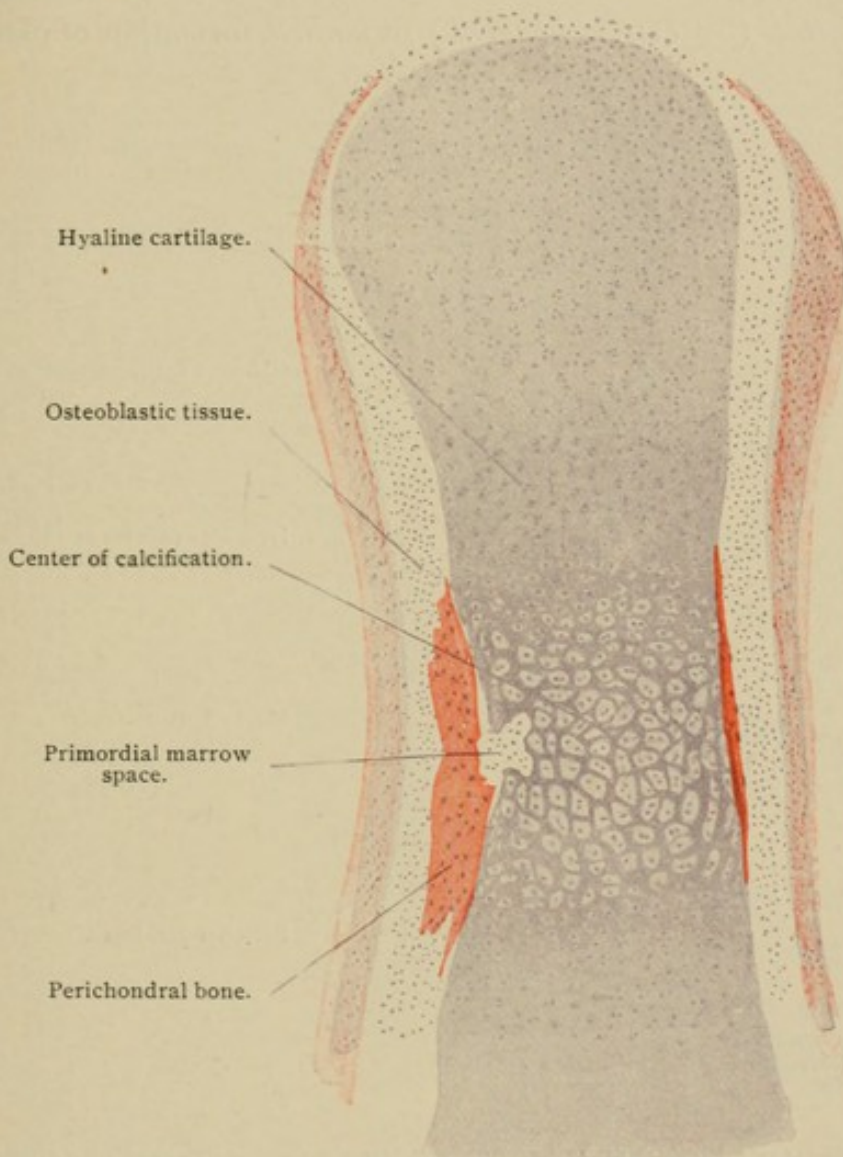


FIG. 102.—FROM A DORSO-PALMAR LONGITUDINAL SECTION OF A PHALANX OF THE LITTLE FINGER OF A HUMAN FETUS SIX MONTHS OLD. $\times 60$. At the center of calcification the lacunæ are enlarged and contain several cells; above the cartilage cells stand in groups. Each group has arisen through the repeated division of *one* cartilage cell. Technic No. 67.

The *primary bones* include all the bones of the trunk and the extremities, the greater part of the base of the cranium (the occipital bone with the exception of the upper portion of the tabular part, the sphenoid bone with the exception of the internal pterygoid plate, the temporal bone and the ossicles of the ear, the ethmoid bone, the inferior turbinal), and the hyoid bone.

The *secondary bones* include the bones forming the sides and roof of the cranium and nearly all the bones of the face.

DEVELOPMENT OF PRIMARY BONE.

Here two processes of bone-formation are to be considered: (1) *endochondral formation*, formation of osseous tissue *within* the cartilage present; (2) *periosteal* (better *perichondral*) *formation*, formation of osse-

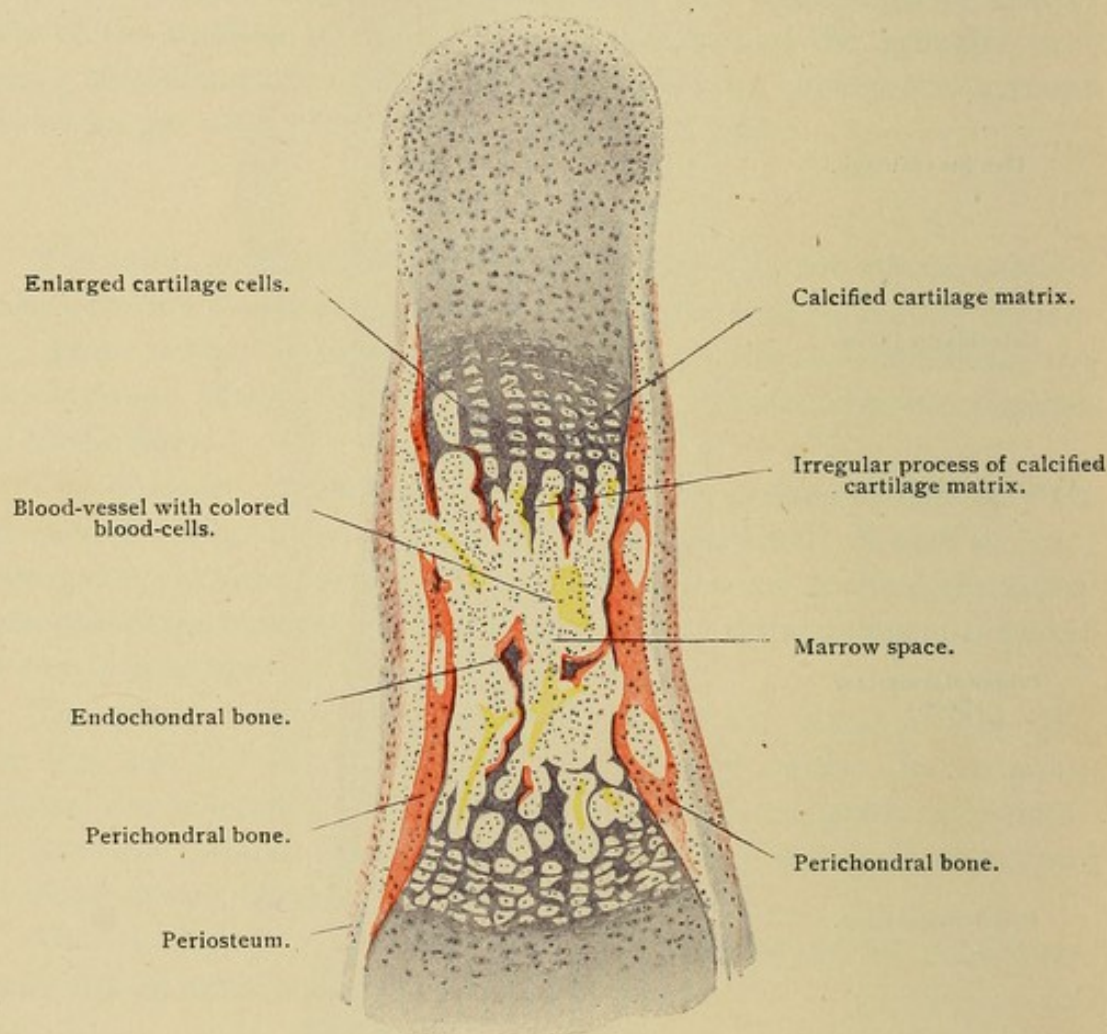


FIG. 103.—FROM A DORSO-PALMAR LONGITUDINAL SECTION OF A MIDDLE-FINGER PHALANX OF A HUMAN FETUS FOUR MONTHS OLD. $\times 60$. Technic No. 67.

ous tissue immediately surrounding, therefore upon, the cartilage. The phylogenetically older perichondral ossification usually begins earlier, but for didactic reasons will be described subsequently to the process of endochondral formation.

1. *Endochondral ossification*.—The first indications of this process consist in changes at certain places within the cartilage; the cells enlarge and divide, so that several lie in one lacuna; then a deposition of lime salts takes place within the matrix, in consequence of which it becomes

finely granular and dull; it calcifies. Such places can soon be recognized by the unaided eye, and are called *centers of ossification* (better, centers

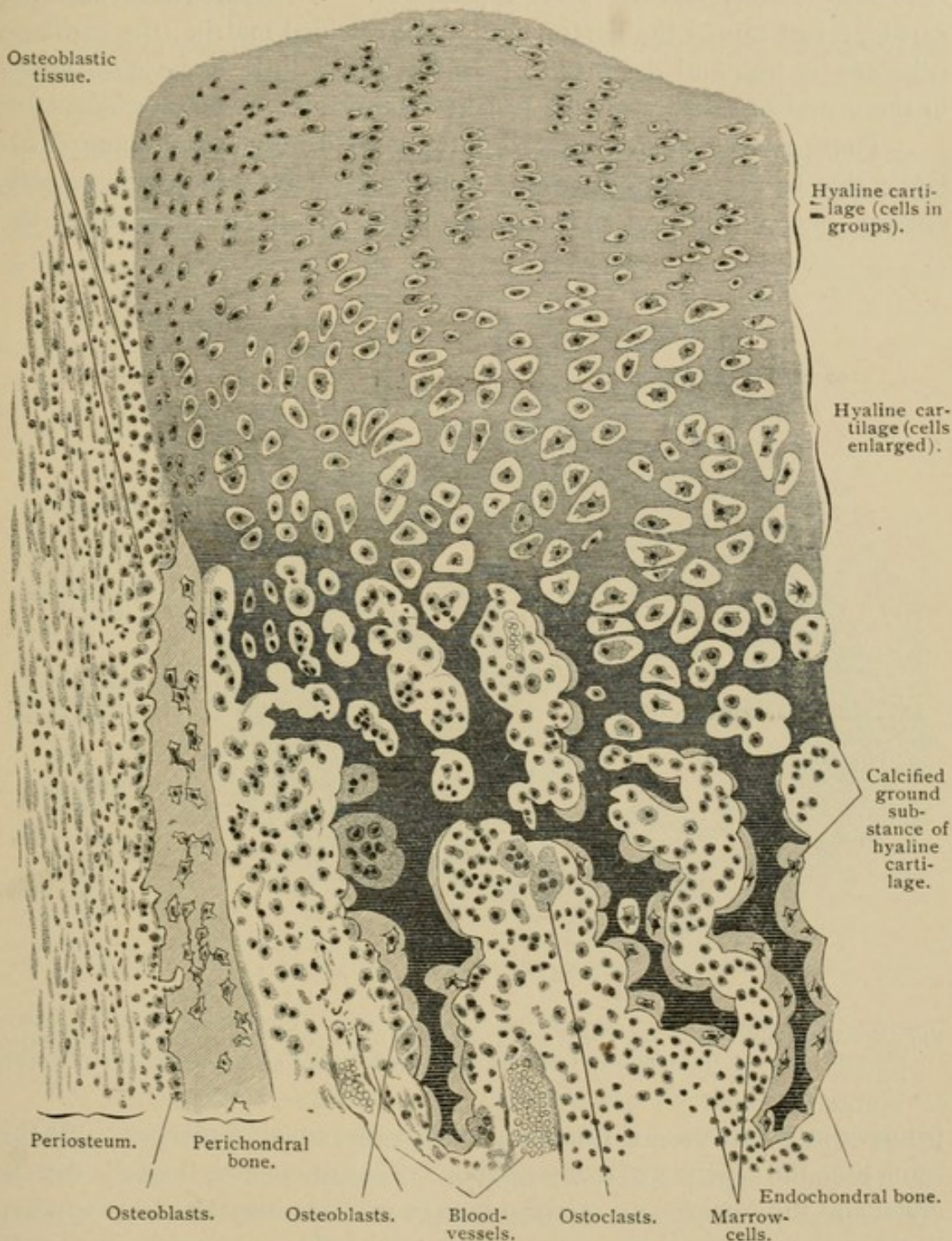


FIG. 104.—FROM A LONGITUDINAL SECTION OF THE PHALANX OF THE FIRST FINGER OF A HUMAN FETUS OF FOUR MONTHS. $\times 220$. In the endochondral bone irregular lacunæ with bone cells are seen. Technic No. 67.

of calcification, Fig. 102). The portions of the cartilage most remote from the center of calcification continue to grow in thickness and length, while at the center growth ceases and consequently the cartilage at this

point appears constricted (Fig. 102). Meanwhile, on the surface of the center of calcification a tissue rich in blood-vessels and young cells, the *osteoblastic tissue*, has made its appearance. This penetrates into the cartilage and causes the destruction of the calcified matrix; the cartilage cells are set free and degenerate. In this way a little excavation arises in the center of calcification; it is called the *primary marrow cavity*.

These processes are repeated in the immediately surrounding cartilage; that is, the cartilage ground substance calcifies, the cartilage cells enlarge, new portions of the cartilage break down, and as a result the

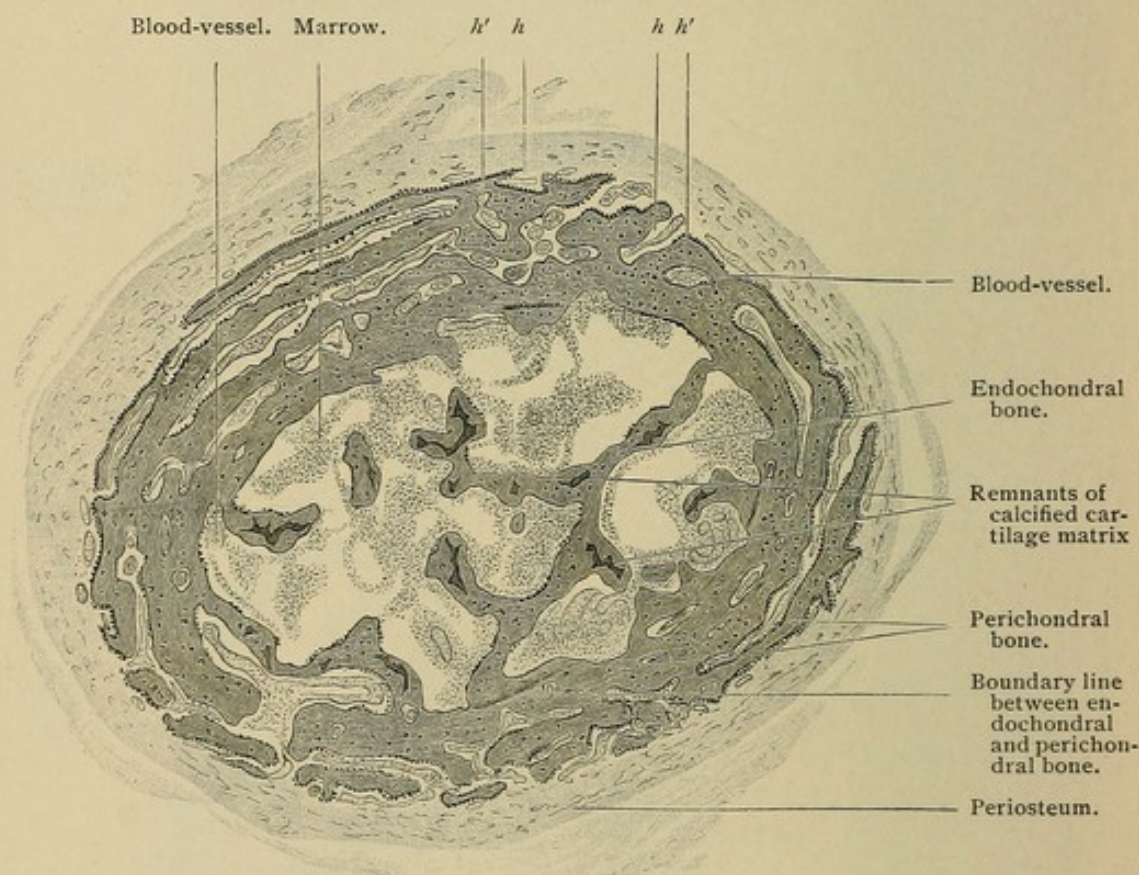


FIG. 105.—CROSS-SECTION OF THE UPPER HALF OF THE DIAPHYSIS OF THE HUMERUS OF A HUMAN EMBRYO OF FOUR MONTHS. *h*, Developing haversian spaces; *h'*, blood-vessel. $\times 35$. Technic No. 67.

primary marrow-space is gradually and continuously enlarged. At the same time the capsules of many cartilage cells are opened, the cells degenerate, and the intervening calcified matrix projects into the marrow-space in the form of irregular processes (Fig. 103). The marrow cavity is now a bay-like space, filled with blood-vessels and with primary bone-marrow, that is, with anastomosing, branched connective-tissue cells. Some of these cells, the *osteoblasts*, grow rich in protoplasm and apply themselves in the manner of a one-layered epithelium to the walls of the marrow cavity and there produce bone (*cf.* p. 100). Meanwhile, leucocytes appear

in ever increasing numbers and finally form the chief mass of the cellular elements of bone-marrow, therewith converting the primary marrow into the red marrow.

Some of the branched connective-tissue cells retain their form and later together with a fine-fibered connective tissue constitute the supporting framework of the bone-marrow. Others of these cells become fat-cells.

Through the activity of the osteoblasts the marrow cavity is soon clothed with a thin stratum of bone gradually increasing in thickness; the irregular processes of calcified ground substance are completely enveloped in young bone. Thus step by step the former solid piece of

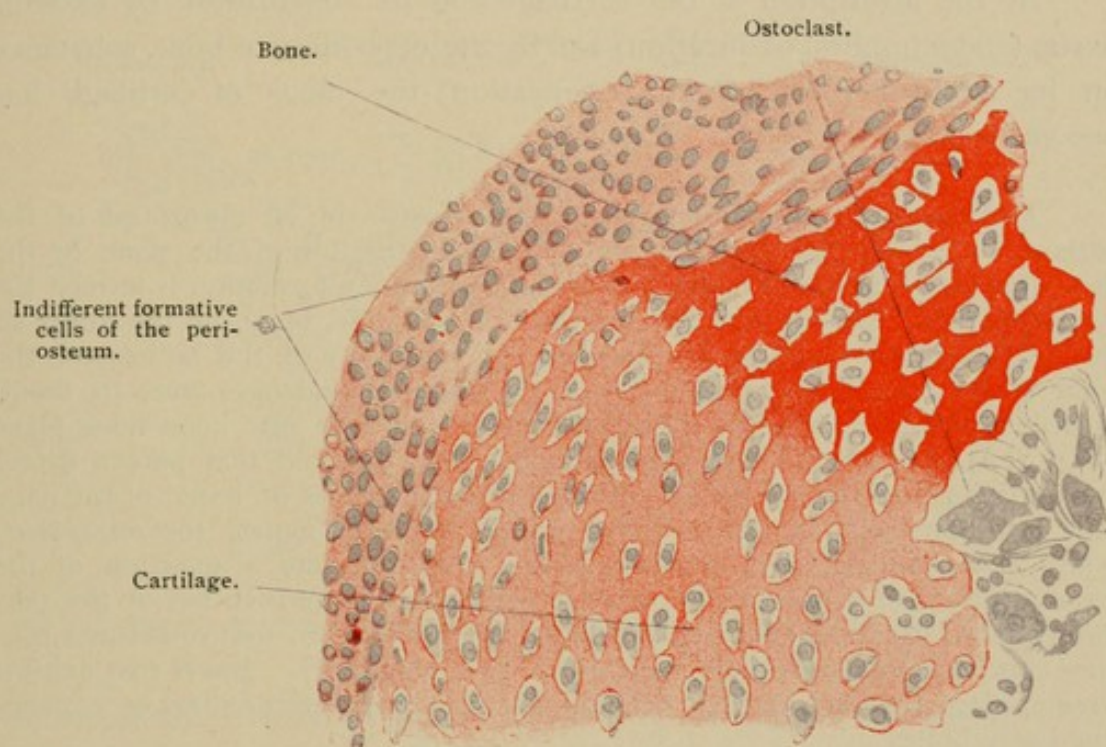


FIG. 106.—FROM A SECTION OF THE HARD PALATE OF A NEWBORN KITTEN. $\times 240$. Technic No. 67.

cartilage is transformed into spongy bone, the trabeculae of which still contain residues of calcified cartilage matrix (Fig. 105).

2. *Perichondral ossification.* This process of bone formation is likewise performed by the osteoblasts derived from the osteoblastic tissue at the surface of the center of calcification (Fig. 102). Through the activity of the osteoblasts * strata of plexiform osseous tissue are periodically formed on the surface of the cartilage (Fig. 103); these osseous masses are distinguished from the endochondral bone by the absence of remnants of calcified cartilaginous matrix, because the perichondral

* In the inner strata of the perichondral osseous cortex the osteoblasts are almost entirely absent; also in the region of the endochondral osseous trabeculae the number of osteoblasts is smaller, which doubtless is associated with the future process of resorption.

bone is formed at the circumference and not in the interior of the cartilage. The formation of the first haversian canals may be studied in the perichondral bone (Fig. 105). The latter is not formed in a continuous layer of uniform thickness, but at frequent intervals depressions may be observed containing blood-vessels surrounded by osteoblasts (Fig. 105, *h, h*); at first the depressions are mere furrows open toward the periphery, but with the progressive development of the perichondral osseous strata they are closed in (*h'*), and then represent vascular canals, the haversian canals. The osteoblasts enclosed within the canals produce new osseous strata, the future haversian lamellæ (Fig. 108).

By the absorption of the cartilage and its substitution by osseous tissue (endochondral ossification) and by the deposition of bone substance on its exterior (perichondral ossification) the piece of cartilage has become a bone.

The essence of the foregoing processes consists in an absorption of the parts of the primordial skeleton and in a reconstruction of the same by the development of bone substance. This mode of bone formation is termed the *neoplastic type*. On the articular fossa of the temporal bone, on the suture of the palate, on the inferior maxilla, on the tuberosity of the radius, on the spine of the scapula, and on the tips of the terminal phalanges areas are found in which apparently a direct transformation of cartilage into bone takes place (Fig. 106). From this the conclusion has been deduced that here a direct metamorphosis of the matrix of cartilage into the matrix of bone, of cartilage cells into bone-cells, occurs and the process has been named the *metaplastic type*. The conclusion is unwarrantable; it is not here a question of the metamorphosis of a developed cartilage cell into a bone-cell, but of the performances of indifferent formative cells of the periosteum, that sometimes produce cartilage, sometimes bone (see also p. 100, remark *). Bones that exhibit a metaplastic type are in their original anlage either perichondral or connective-tissue bones.

DEVELOPMENT OF SECONDARY OR CONNECTIVE-TISSUE BONE.

Here the foundation on which the formation of bone occurs is not cartilage, but connective tissue. Isolated bundles of connective tissue calcify; on these osteoblasts (Fig. 107) derived from embryonal cells arrange themselves and produce bone in the manner previously described. Or small groups of osteoblasts can secrete calcified substance, that becomes the starting point for the development of osseous trabeculæ. For the comprehension of connective-tissue bone it is necessary to bear in mind that it is surrounded on *all sides* by connective tissue; when osseous tissue is in contact on one side with cartilage, without the interposition of connective tissue, the resulting formation is not connective-tissue bone, but perichondral bone.

THE GROWTH OF BONES.

1. *Bones preformed in cartilage.*

(a) In *tubular bones* ossification in the diaphysis begins much earlier than in the epiphyses (in the humerus the center of ossification in the diaphysis appears in the eighth fetal week, in the epiphyses in the first year of life); blood-vessels grow into the calcified cartilage, which at first is transformed only by endochondral, later also by perichondral ossification into bone. The articular surfaces of the bone remain *permanently* cartilaginous; a *temporary* narrow zone of cartilage between diaphysis and epiphysis, the *epiphyseal cartilage*, persists until the growth of the bone

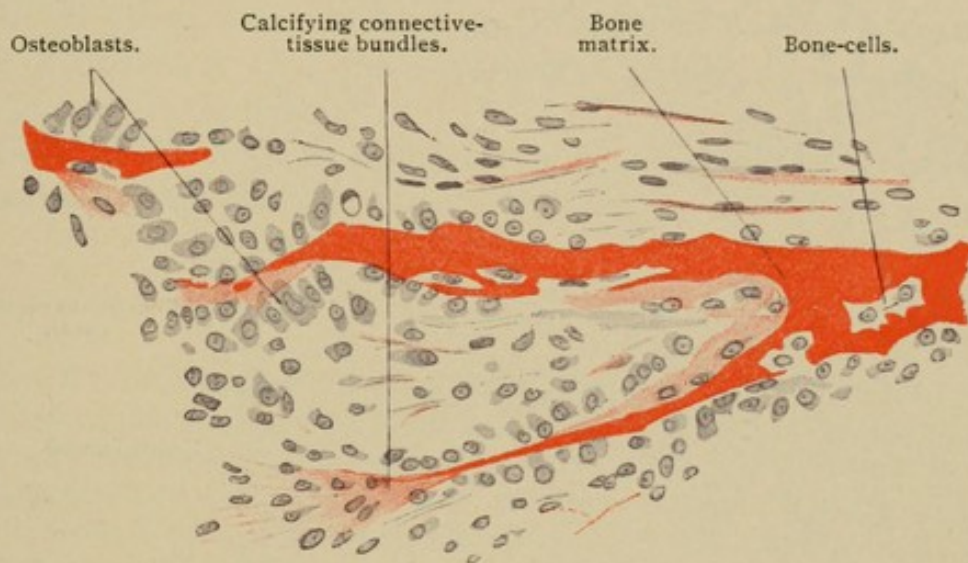


FIG. 107.—FROM A SECTION OF THE INFERIOR MAXILLA OF A HUMAN FETUS FOUR MONTHS OLD.
X 240. Technic No. 67.

is completed. Here an active growth of cartilage occurs that, by extension of the primary marrow cavities of the diaphysis and the epiphyses, is continually being supplanted by bone. In this way the bone grows in length. Increase in thickness takes place by the constant "apposition" of new periosteal strata of bone.*

(b) In *short bones* ossification takes place, as in the epiphyses, at first by endochondral ossification; after the absorption of the last superficial remnant of cartilage a perichondral osseous cortex is formed.

(c) In *flat bones* ossification is first perichondral, then endochondral.

2. *Connective-tissue bones.*

These grow in superficies and in thickness by the formation of new

* "Interstitial" growth, dependent on increase of the ground substance between the bone lacunæ, occurs only, in very slight degree, in the youngest bone substance. The bone-cells are much more numerous here than in later stages.

osseous masses at their edges and on their surfaces respectively. As a consequence of the abundant deposition of bone-substance on the surfaces, the outer and inner tables of compact bone are formed, which enclose between them spongy bone; the latter in this situation is termed diploë. The osseous masses at first possess a coarse-fibered, later (from about the first year of life) a fine-fibered matrix (p. 98).

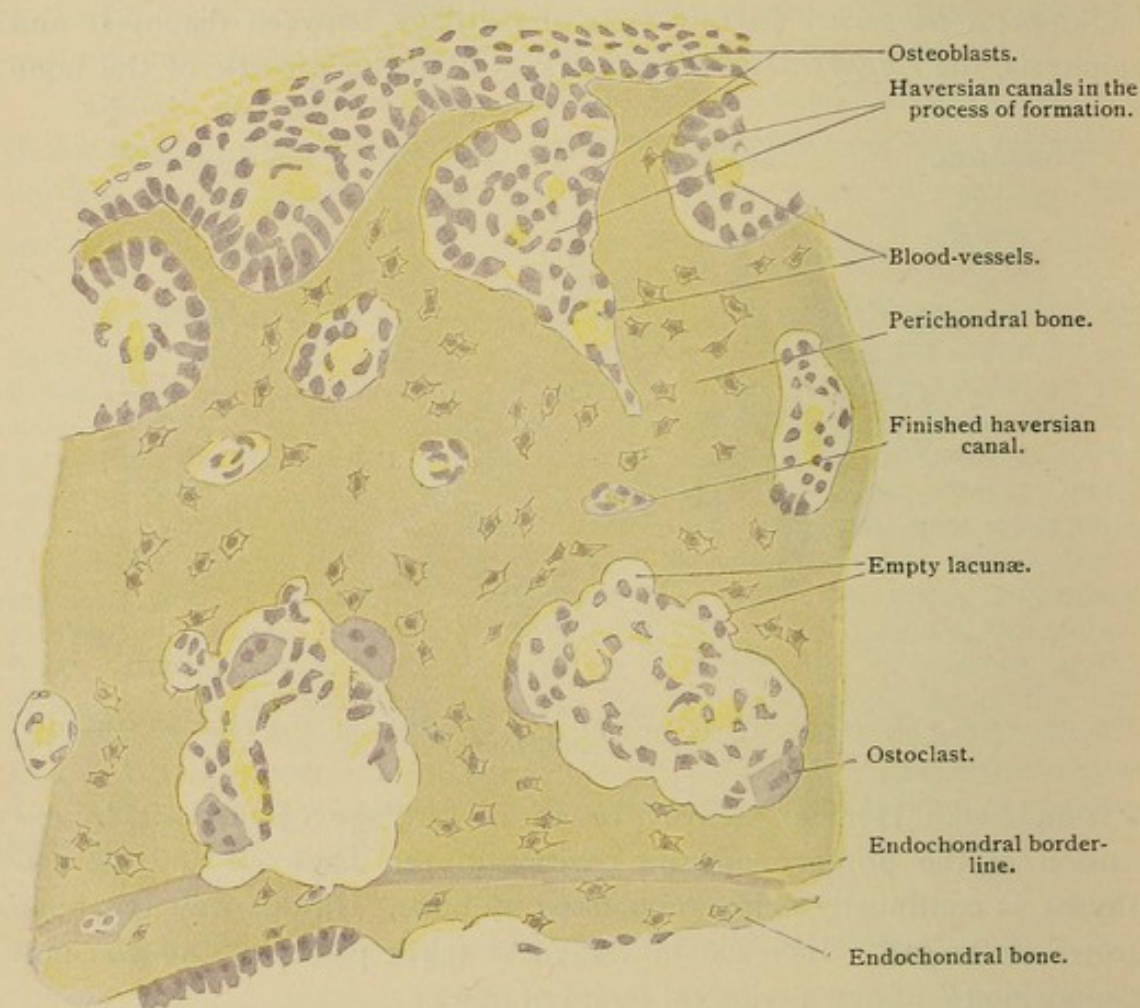


FIG. 108.—PORTION OF A CROSS-SECTION OF A TUBULAR BONE OF A NEWBORN KITTEN. Technic No. 67.

THE RESORPTION OF BONES.

Simultaneously with the first anlage of osseous tissue a counter process, resorption, becomes noticeable, by which the calcified cartilage matrix, as well as many parts of the recently formed bone, is dissolved. Resorption occurs in slight measure in flat and in short bones and on the surface of all bones until their typical shape is developed, but in extremest degree in tubular bones, in the formation of the marrow

cavity.* In this process not only entire masses of endochondral bone † are lost, but also conspicuous quantities of perichondral bone, losses that are always covered by deposition from outside of strata of new perichondral bone. Also in the interior of the substantia compacta irregular cavities are seen, the so-called *haversian spaces*, that have arisen by solution of the inner haversian lamellæ, which may become partially filled again by deposition of new osseous masses (Fig. 96, *h*). The substantia spongiosa of the bone of adults arises by resorption from the marrow cavity and from the inner surface of the haversian canals, which leads to the gradual reduction of the bone to small strips, the trabeculæ and lamellæ of the spongy substance.

In all places where the resorption of bone occurs the osteoclasts (bone destroyers, p. 163) are seen, lying in pit-like depressions (*Howship's lacunæ*) in the bone.

Even in the fully developed skeleton the processes of apposition and resorption still continue in a few localities.

TECHNIC.

No. 61.—*Ground sections of dried bone.*—The bone must not be dried before maceration, but must be placed fresh for several months in water, which should be frequently changed. Then it is dried and a piece held between two pieces of cork or folds of cloth is clamped in a vice and a section 1 or 2 mm. thick, transverse or longitudinal, is cut with a compass-saw. Glue the section with sealing-wax to the under surface of a cork-stopper (the sealing-wax should encircle the section), dip the whole for a moment in water and then file it, first with a coarse, then with a fine file, until it is perfectly smooth; the file must be frequently dipped in water, in order to wash off the adherent particles of bone and to prevent the heating of the sealing-wax by friction.

The section of bone should then be loosened by heating the sealing-wax and the smooth side stuck fast to the stopper. It must now be filed until it is so thin that the sealing-wax can be seen through it. The whole should at once be placed in 90 per cent. alcohol, in which within a few minutes the section becomes loosened from the cork. Then moisten a coarse whetstone with water, rub it with a second whetstone until the surface is covered with a little grinding-paste; lay the section in the paste, place on it a smooth cork (one without cracks), and with a *circular* motion grind it on both sides; it is not necessary to glue the section to the cork. The section when sufficiently

* For example, a femur of a three-year old child contains scarcely any of the osseous tissue present at birth.

† The osseous labyrinth of the ear forms an exception; it still contains remnants of calcified cartilage even in extreme old age.

thin is transparent; this is to be ascertained by drying it between pieces of filter-paper and examining with the low power. It should then be ground on a fine whetstone, in the same manner as on the coarse, and when both sides are smooth it should be dried with filter-paper and polished. Nail a piece of wash-leather smoothly on a board, sprinkle it with chalk, and with the tip of the finger rub the section to and fro on the leather. In this way the previously dull section acquires shining surfaces. The adherent powder may be removed by rubbing the section on fresh wash-leather. The finished section is to be placed dry on a slide and the cover-glass is secured by means of cement (p. 50).

Examine first with the low, then with the high power. The bone lacunæ and bone canaliculi are filled with air and with the customary illumination of the object from below appear black (Fig. 47). If the section is thick it may be impossible to examine it with the high power, since then the objective cannot be brought near enough to the preparation.

No. 62.—*Sharpey's fibers*.—Prepare a cross-section of the middle of the shaft of a tubular bone, preferably of a young individual, according to the method given in No. 61. Place the finished dry section for from two to five minutes in 4 c.c. of xylol and then mount in xylol-balsam. The fibers, invisible in the sections produced by other methods (No. 61 and No. 63), can be plainly seen, even with the lower power (Fig. 99).

No. 63.—*Haversian canals and bone lamellæ*.—Select the metacarpal bone of an adult; after four weeks' fixation in Müller's fluid and hardening in alcohol, decalcify in from 3 to 9 per cent. nitric acid (p. 36), harden again, and cut transverse and longitudinal sections. The compact structure of larger bones (the femur, for example) requires too much time (several weeks) for decalcification. The periosteum should be allowed to remain on the bone. For longitudinal views of haversian canals very thick sections (0.5 mm. or more) must be made. Mount in dilute glycerol (Fig. 95). Nor are very thin sections necessary for transverse cuts of the canals and lamellar systems; the lamellæ are best seen if the section be examined in a drop of distilled water and the mirror turned so that the object is only half illuminated; in this way, too, the striæ produced by the bone canaliculi, running vertically to the lamellæ, are best seen (Fig. 96). Mount in dilute glycerol, which, however, makes the lamellar systems partially indistinct. Not every part of the bone exhibits all the lamellar systems; the outer and also the inner ground lamellæ are frequently wanting. In sections taken near the epiphyses the transition of the compact substance into the trabeculæ of the spongy bone can be seen. The bone lacunæ and bone canaliculi are much less distinct in moist preparations than in dried ground sections, because the contained air has been displaced by the mounting medium. (Compare Fig. 47 with Fig. 48.)

Not infrequently the concentric lamellæ of the haversian systems

are found to be interrupted by an irregular line, the *resorption line*. Up to this line the osseous tissue previously formed has been again resorbed (p. 176). All that which lies within the line is newly deposited bone-substance. These formations are partially filled haversian spaces (Fig. 96, *h*).

No. 64.—*Red bone-marrow*.—(a) Compress the halved vertebra or the rib of a calf in a vice or with tongs; with a pipet take up a small drop of the liquid thus expressed, transfer it to a slide and, without the addition of any other fluid, apply a small cover-glass or, better, a fragment of a cover-glass. Examined with the high power red blood-cells, hematoblasts, marrow cells of different sizes, and giant cells will be seen, but not always their nuclei (Fig. 109, 1). Add a drop of picrocarmine (p. 53); the nuclei become red in from one to two minutes, but are still pale (Fig. 109, 2). If the picrocarmine is displaced by salt solution and this by dilute acidulated glycerol, the nuclei acquire a deep

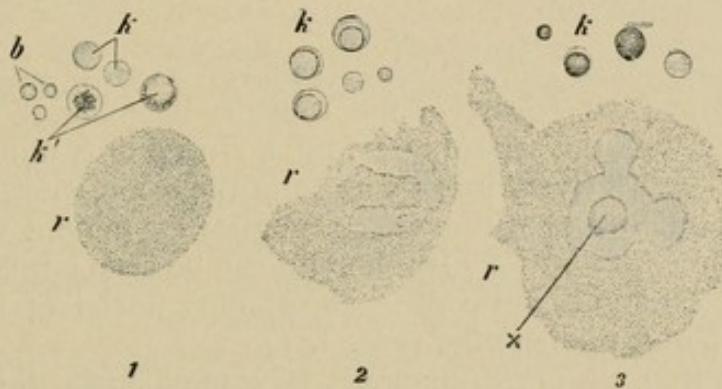


FIG. 109.—ISOLATED ELEMENTS OF FRESH BONE-MARROW FROM THE VERTEBRA OF A CALF. $\times 560$. 1. In salt solution. 2. Stained with picrocarmine. 3. After treatment with acidulated glycerol. *k*, Marrow cells; *k'*, two marrow cells containing masses of pigment-granules, the cell on the right seen from the side, the cell on the left seen from the surface; *b*, nonnucleated colored blood corpuscles; *r*, megakaryocytes; in the one on the right the nucleus is dividing by constriction, at two lateral points and at $\times$ on the surface.

color and sharp contours (Fig. 109, 3). Occasionally giant-cells are sought in vain. Human ribs are often usable.

(b) For permanent preparations proceed as follows: With a *thin* cover-glass take up a drop of the marrow expressed from a rib and make two cover-glass preparations as directed in No. 45, for dry cover-glass preparations of blood, after Ehrlich. Since the marrow does not diffuse as readily as blood between the two cover-glasses, make slight pressure upon them before slipping them apart, by means of forceps. They should not be allowed to dry, but should be placed *at once* in a concentrated aqueous solution of corrosive sublimate (5 gm. in 100 c.c. of distilled water). At the end of ten minutes transfer them to 200 c.c. of distilled water, which is to be changed in about five minutes. In another ten minutes place them in 5 c.c. of diluted eosin (p. 39, 3 *b*) for from one to five minutes, then wash for a moment in distilled water and transfer them to 5 c.c. of filtered Hansen's hematoxylin; after a minute or two place them for five minutes in distilled water; remove the water by

means of filter-paper placed at the edge of the cover-glass and place them in 95 per cent. alcohol (not longer than one minute, lest the eosin be extracted), then in carbol-xylol for three minutes. With a cloth carefully remove the oil from the film-free surface of the cover-glass, place a drop of xylol-balsam on the surface containing the film of marrow, and invert the cover-glass on a slide. The colored blood-cells, which are very often distorted, and the protoplasm of the hematoblasts are stained a brilliant rose color, the protoplasm of the remaining cells gray-violet; all the nuclei are blue. Cells containing oxyphile (eosinophile) granules are often found (Fig. 97). Cells with neutrophile and basophile granules are exhibited by treating bone-marrow according to technic No. 45.

No. 65.—*Articular cartilage*.—Select the head of the metacarpal bone of an adult and treat it according to the method given in No. 63. Cut longitudinal sections and mount them in dilute glycerol (Fig. 100). The parallel streaks often present in the hyaline cartilage are produced by the razor. The granules of the calcified cartilage disappear in the process of decalcification.

No. 66.—*Synovial villi*.—From a cadaver, as fresh as possible, cut out a piece about 4 cm. square of the capsular ligament at the *border* of the patella, and with the scissors take off a strip 2 or 3 mm. broad from the reddish, glossy, velvety inner surface of the same, moisten it with a drop of salt solution, and without a cover-glass examine it with the low power. At the edges of the strip the villi can be seen; their blood-vessels often still contain blood-cells. The shining nuclei of the epithelial cells lie close beside one another (Fig. 101).

If it is desired, the preparation may be stained under the cover-glass with picrocarmine and mounted in diluted glycerol (p. 53), but much of the original beauty is lost.

No. 67.—*Development of bone*.—Human embryos four or five months old, embryos of the sheep, pig, or cow, from 10 to 14 cm. long (measured from the tip of the snout to the root of the tail), are suitable. The latter are readily obtained at the slaughter-house; the entire uterus should be ordered. Place parts of the human embryos, the animal embryos in toto (2 or 3 in 1 liter), in Zenker's fluid for forty-eight hours. Wash in running water for forty-eight hours and harden in from 200 to 400 c.c. of gradually strengthened alcohols (p. 35). After the embryos have lain one week or longer in 90 per cent. alcohol containing tincture of iodine (p. 33), cut off the head, the extremities close to the rump,* and decalcify them in 200 c.c. of distilled water to which 2 or 4 c.c. of pure nitric acid have been added. In from two to five days, during which the decalcification medium must be changed about three times, the extremities are to be taken out (the head is probably not yet decalcified, and must remain in two per cent. nitric acid for several days more), treated with potassium alum (p. 36) then washed from one to six hours

* Pieces of the vertebral column and the ribs likewise yield instructive pictures.

in running water, and again hardened in gradually strengthened alcohols. After they have lain five days in 90 per cent. alcohol, cut the extremities into pieces 1 cm. long, which, should they still be too soft, may be placed for one or two days in 30 c.c. of absolute alcohol.

For sections showing the *first processes* in the development of bone (Fig. 102-104), embed in liver the phalanges and metacarpal bones (the latter are very long in the animals mentioned), and make longitudinal (sagittal) sections, from the flexor to the extensor surface; to be useful the sections must be taken in the axis of the extremities, those taken from the margin exhibit pictures that are unintelligible.

For *more advanced stages* make chiefly transverse sections of the humerus and femur. Sections through the diaphysis show more perichondral, sections through the epiphyses more endochondral bone.

The most beautiful *osteoblasts* are obtained in cross-sections of the inferior maxilla, which are also valuable as preparations showing the development of teeth.

For still *later stages* the skeleton of newborn animals is useful; their phalanges show tolerably early stages in the process, their carpal bones the first stages. The decalcification requires somewhat more time (up to eight days).

For *connective-tissue bone* select the parietal and frontal bones of embryos and cut horizontal sections.

All the sections are to be stained in 4 c.c. of Hansen's hematoxylin (p. 38) for from two to ten minutes, transferred to 10 c.c. of distilled water for ten minutes, then stained in 4 c.c. of eosin for one minute (p. 39), washed for two minutes in 5 c.c. of distilled water, and mounted in xylol-balsam (p. 50).

If the staining is successful, the cartilage (especially the calcified portion) is blue, the bone red. Occasionally the cartilage does not stain a bright blue; then, instead of using the usual hematoxylin solution, place the sections in 5 c.c. of distilled water plus 5 drops of filtered hematoxylin solution. In from six to fourteen hours the cartilage will be blue. The eosin staining of bone often is not uniform; the youngest portions of the bone, the margins of the osseous trabeculæ, for example, are often the most brilliantly stained.

III. ORGANS OF THE MUSCULAR SYSTEM.

The muscular system is composed of a large number of contractile organs, the *muscles*, which consist of cross-striated muscle tissue and are joined to the skeleton, the skin, the viscera, etc., by the intervention of special connective-tissue formations, the *tendons*, and by accessory apparatus of similar structure, the *fasciæ*, *tendon-sheaths*, and *bursæ*.

Each *muscle* is composed of striated muscle-fibers (p. 106) that as a rule are united in such a manner that they lie lengthwise, side by side

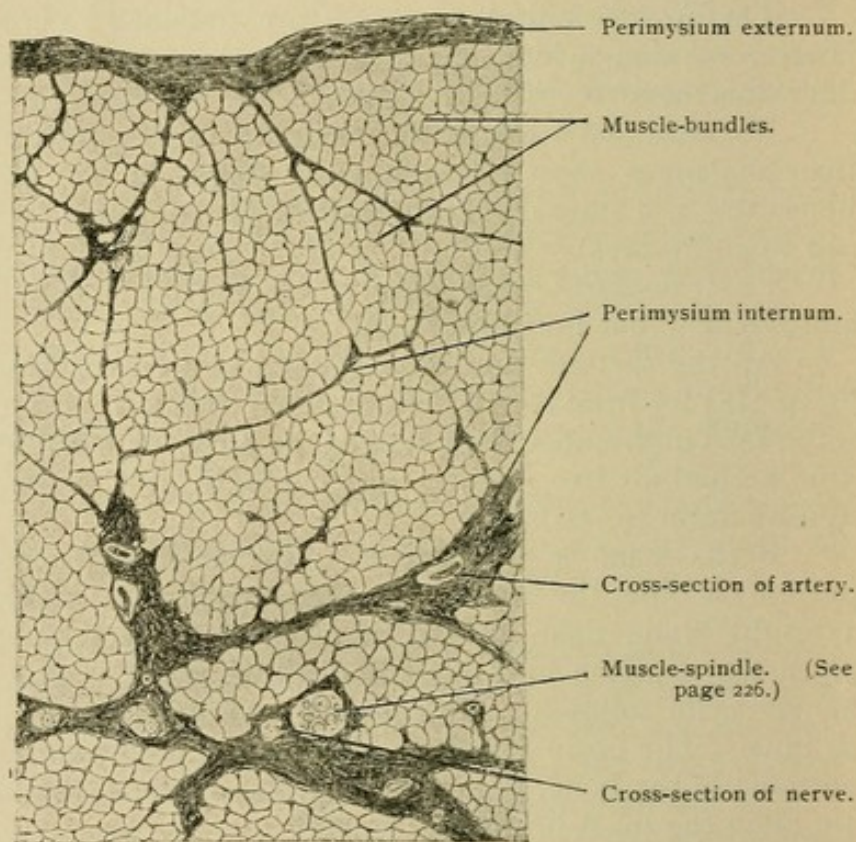


FIG. 110.—FROM A CROSS-SECTION OF THE OMO-HYOID MUSCLE OF MAN. $\times 60$. Technic No. 68.

and behind one another, and are held together by loose connective tissue, the *perimysium*. Transverse interlacing is rare, but occurs, for example, in the tongue. Neighboring muscle-fibers never are in direct contact by their sarcolemmæ, but each individual fiber is enveloped in a delicate connective-tissue sheath, the perimysium of the single muscle-fiber, which is joined to neighbor sheaths (Fig. 111).

A somewhat thicker connective-tissue sheath, the perimysium internum, encloses a large, widely varying number of fibers and in this way a muscle-bundle is formed (Fig. 110). A collection of muscle-bun-

dles * forms a muscle, the surface of which is covered by a still thicker connective-tissue sheath, the perimysium externum. The several sheaths are connected with one another.

The perimysium is composed of fibrillar connective tissue and fine elastic fibers, † occasionally contains fat-cells, and conveys the nerves, blood-vessels, and lymph-vessels. The perimysium of the individual muscle-fiber contains only capillaries and terminal branches of nerves.

The post-embryonal increase in the thickness of the muscles depends less on the multiplication by division, than on the growth in thickness of the already existing muscle-fibers.

The *tendons* are characterized by the parallel course of their fibers, by their firm union, and by their poverty in elastic fibers. They are composed of dense-fibered connective-tissue bundles, the "tendon-

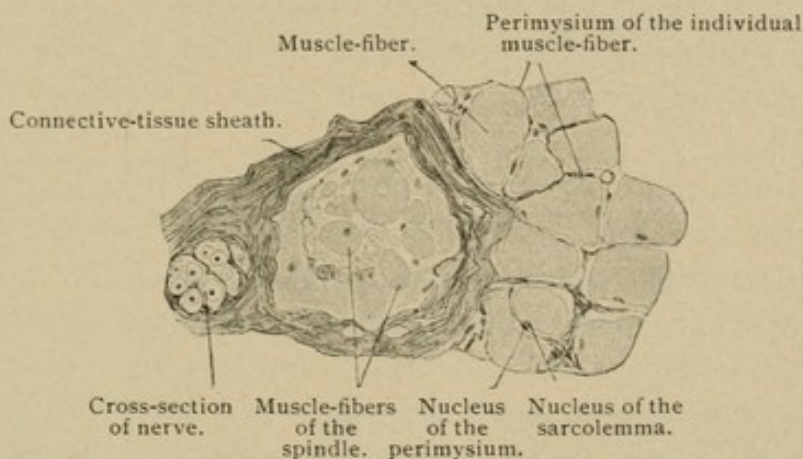


FIG. III.—PORTION OF THE SECTION OF FIGURE 110. $\times 240$.

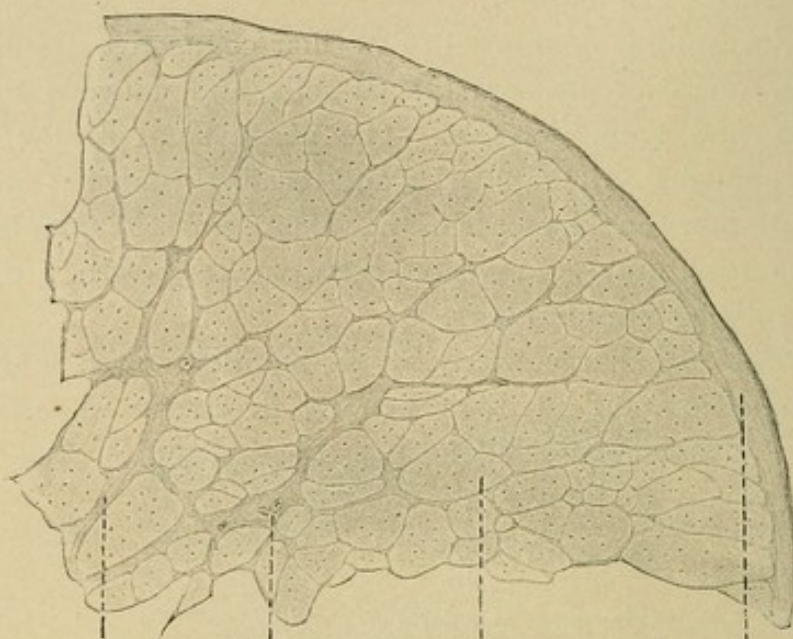
bundles," which are held together by loose connective tissue. Each of these (so-called secondary) tendon-bundles consists of a number of parallel fibrillæ running a perfectly straight course and united by a small amount of cement-substance in smaller (so-called primary) bundles. Between the primary bundles lie the cellular elements of the tendon; they are spindle-shaped or stellate, or four-sided, flat connective-tissue cells, placed behind one another in rows; they are bent like concave tiles and partially encircle the primary bundles; they unite by means of processes with neighbor cells.

Elastic fibers in large quantity are found only in the loose connec-

* The grouping of the primary bundles in secondary bundles, of which a certain number form tertiary bundles, that finally unite to form a muscle, is an arbitrary classification and in many preparations cannot be recognized.

† In the perimysium externum elastic fibers are present in great abundance; the muscles of the extremities are poor, the diaphragm is rich in elastic fibers.

tive tissue ; in the dense tendon-bundles they are very scarce and occur in the form of a fine, wide-meshed network.



Loose connective tissue. Blood-vessel. Tendon bundle. Loose connective tissue.
FIG. 112.—FROM A CROSS-SECTION OF A TENDON OF ADULT MAN. $\times 40$. The dark dots in the tendon bundles are connective-tissue cells. Technic No. 69.

The union of the muscles with tendons and fibrous membranes (periosteum, fascia) is effected by an extension of the perimysium of the

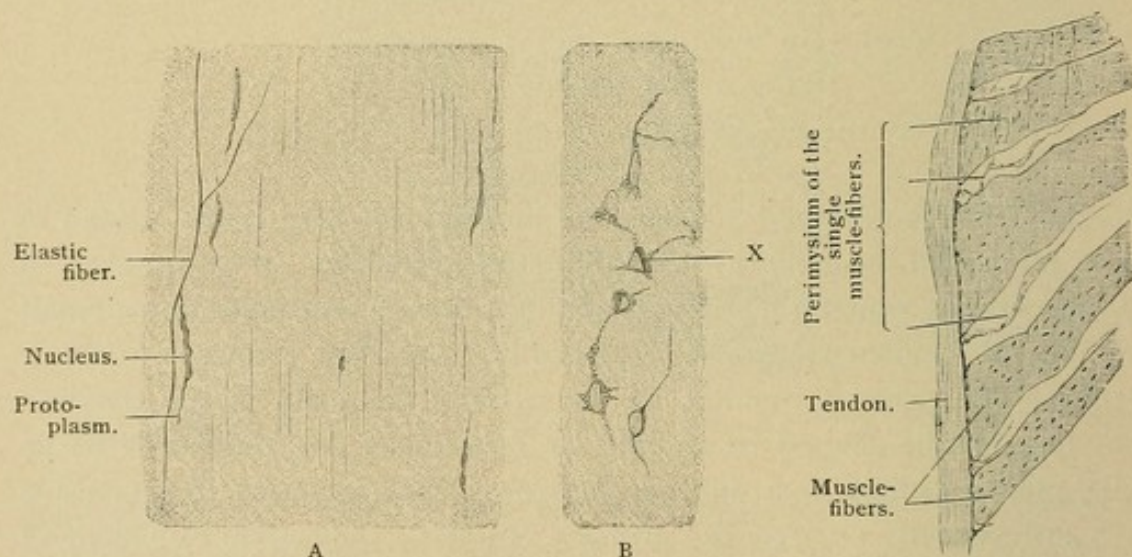


FIG. 113.—TENDONS FROM A RAT'S TAIL. $\times 240$. A. Tendon-cells viewed in profile; B. from the surface. At X the nucleus is bent so that it is seen partly in profile (the shaded portion) and partly from the surface (the light portion). Technic No. 71.

FIG. 114.—FROM A SAGITTAL LONGITUDINAL SECTION OF THE GASTROCNEMIUS OF A FROG. $\times 50$. The uppermost mark indicates the perimysium seen from the surface (as transverse lines). Technic No. 72.

individual muscle-fiber into these structures and the blending of the tissues ; the sarcolemma takes no part in this, but closely investing the

muscle-fiber terminates as a closed sac with pointed or obliquely blunted ends (Fig. 114). The radiating cross-striped muscle-fibers in the skin attach themselves to the connective tissue of the corium by pointed or forked ends.

The *fasciæ* in part exhibit the same structure as the tendons and in part they are connective-tissue membranes richly provided with elastic fibers. The latter is the case only when they form sheaths for the muscles and do not furnish surfaces for the attachment of the muscle-fibers.

The *tendon-sheaths* and the *bursæ* consist of a layer of varying thickness of connective tissue with elastic fibers, the inner surface of which is covered *patchwise* by an "endothelium"; that is, by a usually simple layer of connective-tissue cells.

Where the endothelium is wanting the connective tissue is dense and rich in rounded elements resembling cartilage cells. The majority of the tendon-sheaths have small vascular processes exactly like the synovial villi.

The *blood-vessels* of the striated muscles are very numerous and evenly distributed; the capillaries are among the most delicate in the human body and form networks characterized by elongated rectangular meshes, lying immediately upon the fibers. The veins are provided with valves even in their smallest branches. The *lymph-vessels* are few in number and follow the ramifications of the smaller blood-vessels.

For the *nerves*, partly sensory and partly motor, of cross-striped muscle, as well as for the muscle-spindles, see the *Peripheral Nerve-endings*, p. 220.

The *blood-vessels of the tendons* and the thinner *fasciæ* are very scarce and are found only in the loose connective tissue surrounding the tendon-bundles; on the other hand, the *tendon-sheaths* and the *bursæ* have a rich vascular supply. *Lymph-vessels* are found only on the surface of the tendons.

The medullated *nerves* of tendons terminate in part in a close plexus of nonmedullated nerve-fibers and in part pass into spindle-shaped expansions of the tendon, the so-called *tendon-spindles* (see the *Peripheral Nerve-endings*). End-bulbs and lamellar corpuscles are found in the perimysia, tendons, fasciæ, and tendon-sheaths.

TECHNIC.

No. 68.—*Bundles of striped muscle*.—Select a muscle in which the fibers have a parallel disposition (for example, the adductor of the rabbit) and with a sharp razor make a deep incision transverse to the course

of the fibers and 2 or 3 cm. below make a second incision; connect these by longitudinal incisions and, *without traction*, carefully remove the piece thus mapped out. For fixation place it in 100 c.c. of 0.1 per cent. chromic acid (p. 32). After two weeks wash it for 2 or 3 hours in running water and harden in 50 c.c. of gradually strengthened alcohols (p. 35). Cut cross-sections and examine them unstained in diluted glycerol. The muscle-fibers differ greatly in thickness; the very smallest are sections through the ends of the fibers. Although the muscle-fibers are cylindrical and therefore in section should appear circular, they have an irregularly polygonal outline due to mutual pressure. The perimysium of the individual fiber is better seen with the high power (240 diameters), while Cohnheim's fields (Fig. 55) can only be seen in transverse microtome sections. *Muscle-spindles* are easily found in transverse sections of the human omo-hyoid muscle.

No. 69.—*Tendons*.—Cut from a tendon a piece 5 or 10 cm. long, and let it dry in the air (but not in the sun). Thin tendons (*e. g.*, that of the flexor digitorum pedis) at room-temperature are sufficiently dry in twenty-four hours. Thicker tendons require several days. With the scalpel (not the razor) make a smooth transverse surface and then cut thin shavings from the tendon, supporting it on the thumb of the right hand and with the remaining fingers grasping the scalpel (the manipulation is the same as in sharpening a pencil). Throw the shavings into a capsule containing distilled water and in two minutes examine in a drop of the same medium (Fig. 112). To preserve, stain in 3 c.c. of picrocarmine for five minutes and mount in dilute glycerol. Very frequently a streak is seen extending across the entire section; this is produced by the knife.

Place another section, unstained, in a drop of water on a slide; treat it under the cover-glass with a drop of acetic acid; the edges of the section soon exhibit swollen convoluted bands (acetic acid reaction of connective tissue, p. 89).

No. 70.—For the study of the *minute structure of tendon, its cells and their processes*, place a thin tendon, as fresh as possible (*e. g.* that of the palmaris longus muscle), in pieces 3 cm. long in 100 c.c. of 0.5 per cent. chromic acid (p. 21) for at least four weeks. The chromic acid should be changed several times during this period. Then wash the tissue in running water one or two hours and harden it in about 40 c.c. of gradually strengthened alcohols (p. 35). The sections should be cut with a very sharp razor; often the tendon is so brittle that it falls to pieces in cutting. The sections need not be very thin. Mount them unstained in diluted glycerol. Examined with the low power and direct light (with the mirror muffled) they yield beautiful pictures, better than the preparations made like technic No. 69.

No. 71.—*Tendon-cells*.—From the tail of a rat or a mouse cut pieces of tendon 0.5 to 1 cm. long and place them in 5 c.c. of alumcarmine. The following day (or later) transfer the swollen pieces to a

dry slide and rapidly tease them (p. 29). It is not necessary to separate the tendon into very small bundles, but care should be taken that the bundles lie straight. Then cover the preparation with a drop of distilled water and a cover-glass. With the low power the rows of cells appear for the most part as dark streaks; these are the cell-nuclei seen in profile. In surface views the nuclei appear dull red. The body of the cells, the protoplasm, can only be seen with the high power; viewed laterally, it appears as a sharp, dark streak (Fig. 113 A); from the surface, paler and delicate (Fig. 113 B). Not infrequently the cells are indented, so that they are visible partly from the edge and partly from the surface. Occasionally the connective-tissue fibers can be distinguished as delicate parallel lines; the fine elastic fibers with their sharp contours are always distinct. The focus should be changed by means of the micrometer-screw and the different planes of the section examined. If the cells are not distinct add a drop of acetic acid (p. 53). To preserve, displace the water with diluted glycerol (p. 23).

No. 72.—*Muscle and tendon*.—Remove the skin from the hind leg of a frog just killed and with scissors cut off the leg above the knee-joint, just above the origin of the gastrocnemius. Fix it in 50 c.c. of Zenker's fluid (p. 33) and harden in gradually strengthened alcohols (p. 35). Cut off the muscle with a piece of the tendo-Achillis and stain it in bulk in borax-carmin (p. 40). Then harden again in 90 per cent. alcohol. Cut sagittal longitudinal sections, placing the edge of the razor on the tendon situated on the posterior surface of the muscle. Mount in xylol-balsam (Fig. 114).

IV. ORGANS OF THE NERVOUS SYSTEM.

1. THE CENTRAL NERVOUS SYSTEM.*

THE SPINAL CORD.

Topography.—The spinal cord consists of a white and a gray substance, distinguishable by the unaided eye. The arrangement and relation of these two substances are best recognized in cross-sections of the spinal cord.

The *white substance* encircles the gray substance and is partially divided by a deep anterior cleft, the *anterior median fissure*, and a poste-

* I shall confine myself here to a brief account of the topography and of the histology of the spinal cord and the brain. An exhaustive presentation of the architecture of the central nervous system, the paths of the nerve-fibers, and the complicated structures in connection with the "nuclei" of the cranial nerves in the oblongata would exceed the limits of this "Histology." The student is referred to special text-books, of which Edinger's "Vorlesungen über den Bau der nervösen Centralorgane," Barker's "Nervous System and its Constituent Neurones," and Van Gehuchten's "Anatomie du système nerveux de l'homme" are recommended.

rior *septum* (formerly called the posterior median fissure) into a right and a left half. Each half is subdivided by the furrows marking the exit of the anterior and the posterior roots of the spinal nerves into a large *lateral column*, an *anterior column*, and a *posterior column*. In the lower cervical and the upper thoracic region of the spinal cord two parts can be distinguished in each posterior column, of which the median is named

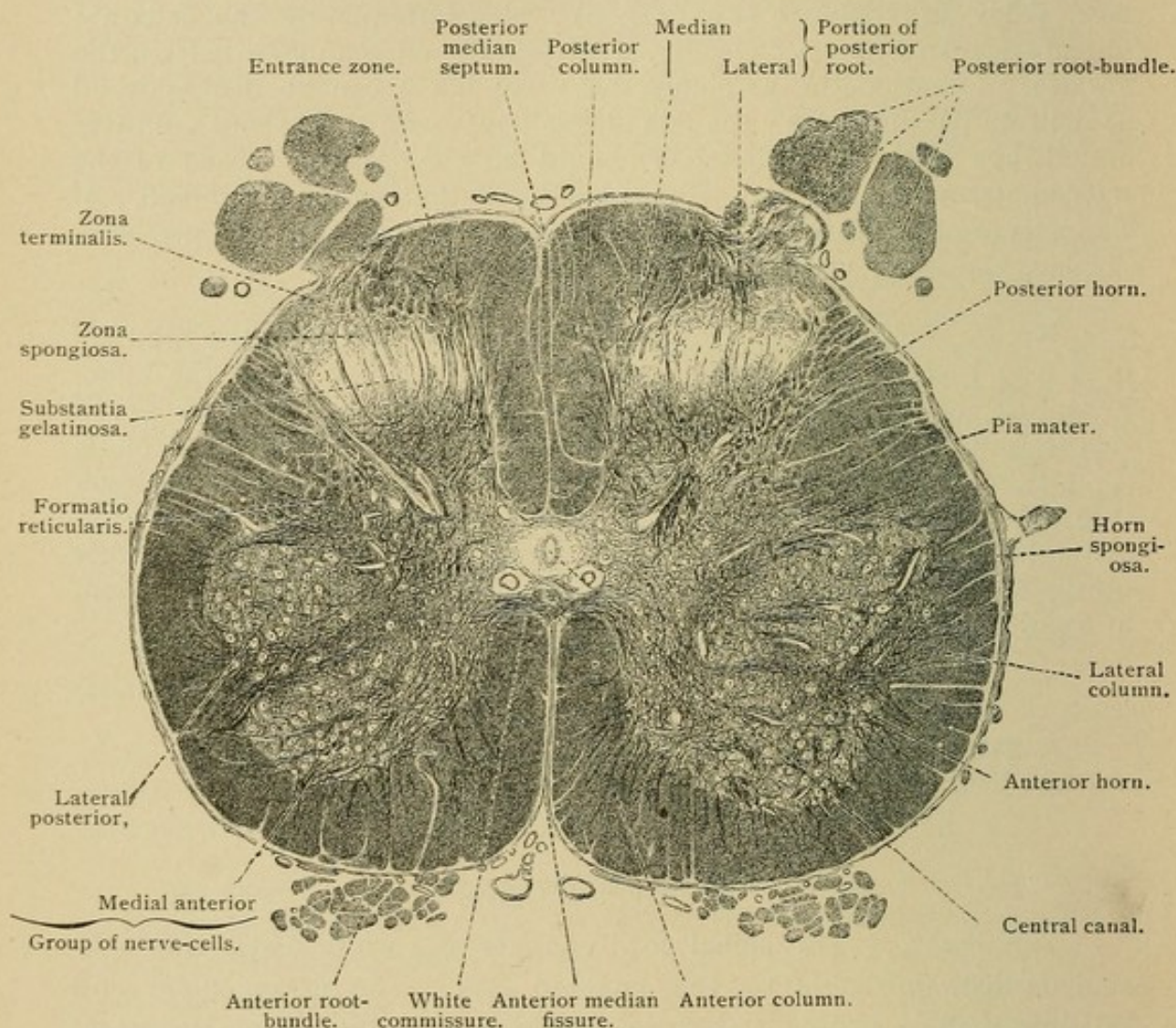


FIG. 115.—CROSS-SECTION OF THE LUMBAR ENLARGEMENT OF THE HUMAN SPINAL CORD. $\times 8$.
Technic No. 74.

the *column of Goll* (funiculus gracilis) and the lateral the *column of Burdach* (funiculus cuneatus). The anterior columns are united by the *white commissure* at the bottom of the anterior median fissure.

The *gray substance* in cross-section appears in the form of an **H** and in its entirety consists of two lateral columns which are connected by a frontally situated lamella, the *gray commissure*. On each column a thick *anterior horn* and a slender *posterior horn* can be distinguished.

At the lateral portion of the anterior horns, in the same frontal plane with the central canal, are the *lateral horns*, which are especially well developed in the upper thoracic region. From the anterior circumference of the anterior horns the *anterior roots* of the spinal nerves emerge in several bundles, while the *posterior roots* enter at the posterior and median side of the posterior horns. Laterally, at the base of each posterior horn a net-like mass of trabeculæ of gray substance, the *reticular process* (*formatio reticularis*) is found; at the median side of each posterior horn, near the gray commissure, lies the *column of Clarke* (dorsal nucleus), visible as a well-defined group in the whole length of the thoracic and in the upper part of the lumbar region and not entirely absent in the remaining portions of the cord. At the summit of the posterior horns a glistening, jelly-like mass, macroscopically easily perceptible, the *substantia gelatinosa* (*Rolando*), can be distinguished. Dorsalward to this is the small *zona spongiosa*, at the dorsal edge of which is found the border-zone, *zona terminalis*, an area of cross-sectioned thin nerve-fibers. In the gray commissure lies the cross-section of the *central canal*, which extends through the whole length of the spinal cord and is surrounded by the *substantia grisea centralis*, diminishing in mass caudalward. The *central canal* is from 0.5 to 1 mm. in diameter; not infrequently it is obliterated. The divisions of the gray commissure in front of and behind the central canal are respectively named the *anterior* and the *posterior gray commissure*. In man the latter is the smaller. From the entire periphery of the gray substance coarser or finer processes, the *septula medullaria* radiate into the white substance. In the cervical and lumbar enlargements of the spinal cord the gray matter is more powerfully developed than in the thoracic region; there is a corresponding variation in the form of the H. The end of the *conus medullaris* consists almost wholly of gray substance.

Minute structure.—The *gray substance* must be first considered, a knowledge of its composition being essential to the comprehension of the structure of the white substance. The gray substance consists of multipolar nerve- (ganglion) cells, that with their dendrites and nerve-processes form a dense nervous tangle, the "nerve-felt" (*neuropilem*). This felt is penetrated by nerve-fibers, coming partly from the white columns, partly from the posterior roots; the whole is supported by a framework of *neuroglia*.

We have therefore to consider first the nerve-cells, then the nerve-fibers; the neuroglia, which also occurs in the white substance, shall be described at the conclusion of the entire recital.

1. The *nerve-cells*, in accordance with the relations and distribution

of their nerve-process, are divided into (1) motor cells, (2) column cells, and (3) internal cells.*

The *motor nerve-cells* (*rhizoneurons*, Fig. 116) lie in two groups † in each anterior horn. They possess a large cell-body (67 to 135 μ) and long dendrites, extending far into the neighborhood; their nerve-process

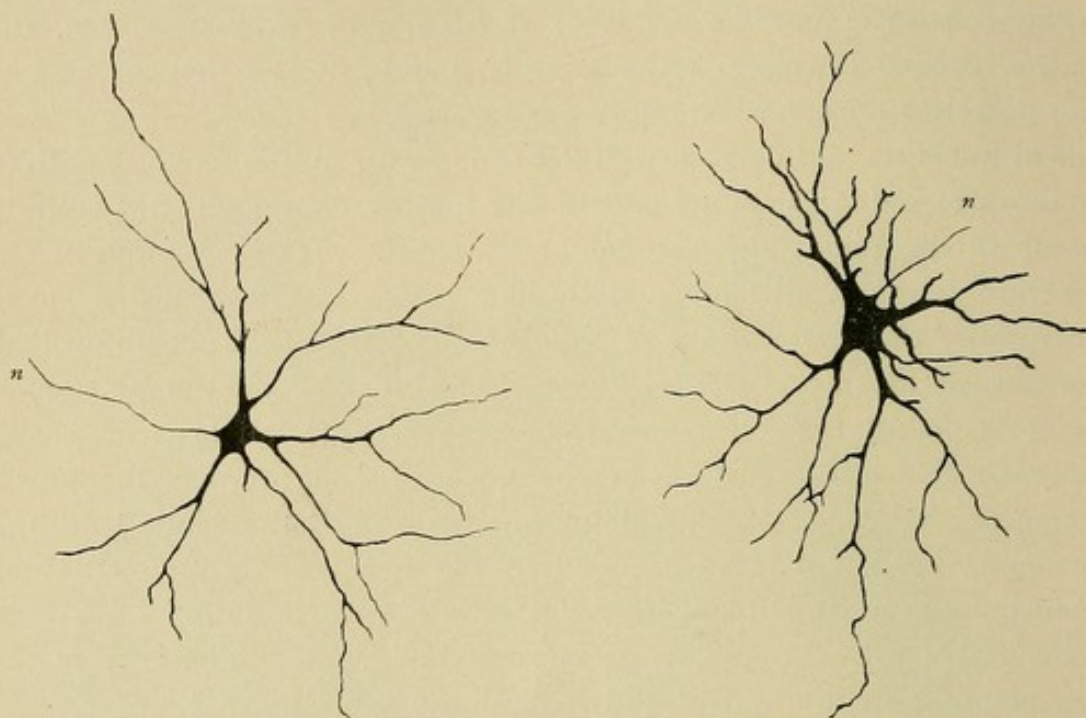


FIG. 116.—TWO FORMS OF MOTOR NERVE-CELLS FROM THE ANTERIOR HORN OF THE SPINAL CORD OF A RABBIT. *n*. Nerve-process. $\times 60$. Technic No. 76. (Schaper.)

* *Editor's remark:* A classification and nomenclature based upon the behavior and distribution of the axis-cylinder have recently been suggested in America that in many respects appear to me to be appropriate and natural, and they have been widely accepted. According to this two chief groups are distinguished, namely: I, *axoneurons*, and, II, *ganglioneurons*.

I. The **axoneurons** embrace all those neurons the cell-body (nerve-cell) of which lies in the interior of the spinal cord or the brain. Corresponding to the different behavior of the nerve-process they are further divided into two subordinate groups, namely:

(a) *Rhizoneurons*, the nerve-process of which leaves the spinal cord through the *anterior root* (they comprise the motor nerve-cells), and—

(b) *Endaxoneurons*, the nerve-process of which does *not* leave the spinal cord. Among these we may distinguish (1) those the nerve-process of which enters the different columns of the white substance (*column cells*), and (2) those the nerve-process of which within the gray substance rapidly breaks up into its terminal ramifications (*internal cells*).

II. The **ganglioneurons** represent those neurons the cell-body of which lies within the *spinal ganglia* or the *cerebral ganglia* and that stand in connection with the central nervous system only by means of their central process.

† A medial-anterior and a lateral-posterior group, separate in the cervical and lumbar enlargements (*cf.* Fig. 115), but in the uppermost cervical and in the thoracic region united in a single colony. In longitudinal sections it may be seen (conspicuously in amphibians) that the cell groups have a segmental arrangement corresponding to the original territory of the individual roots.

emerges from the summit of the anterior horn, makes an oblique descent through the white substance, at the same time receives a medullary sheath and becomes the axis-cylinder of a medullated nerve-fiber. Occasionally the axis-cylinder process gives off a few insignificant lateral twigs (collaterals) before leaving the gray matter. It leaves as a constituent part of an anterior (ventral) root-fiber bundle of the spinal cord. All anterior root-fibers arise from the motor cells of the anterior horns, from those of the same, not the opposite side (Fig. 117).

The *column cells* (*Strangzellen, endaxoneurons*) constitute the chief mass of the nerve-cells of the gray substance and lie everywhere in it (except in the places occupied by the motor nerve-cells), partly scattered, partly in groups in the lateral horn and in the dorsal nucleus (Fig. 117).

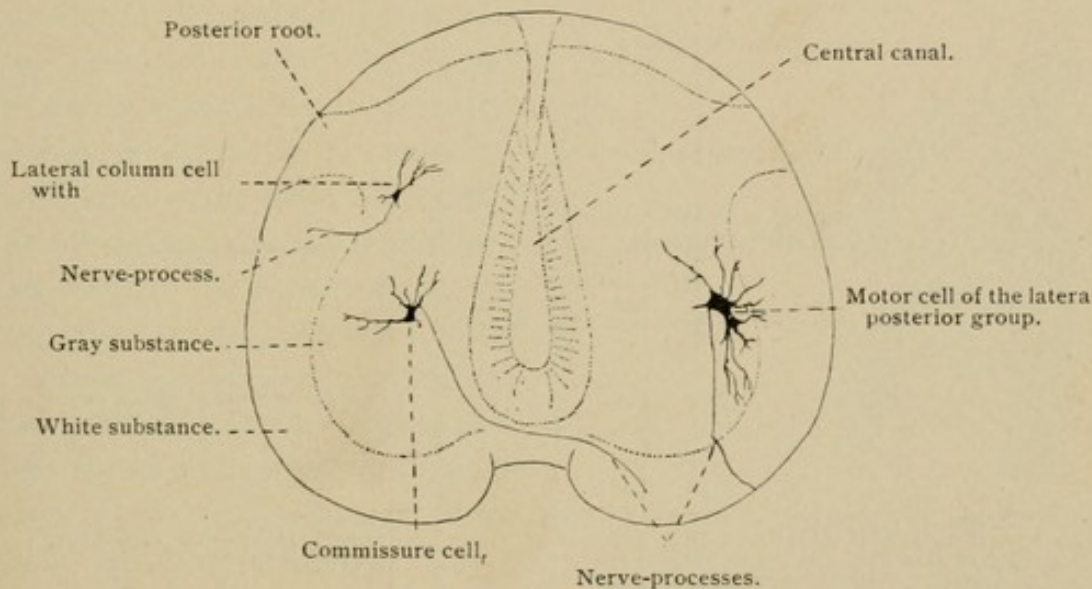


FIG. 117.—CROSS SECTION OF THE SPINAL CORD OF A SEVEN-DAY-OLD EMBRYO CHICK. $\times 80$. The white substance is but slightly developed, the central canal is still very large. Technic No. 76.

They are mostly smaller than the motor nerve-cells and possess few, little-branched, but far-reaching dendrites. Their nerve-process, after sending off numerous collaterals in the gray substance, enters the white substance—in the anterior or lateral column, very rarely the posterior column—either on the same or on the opposite side. Cells of the latter kind are also named *commissure cells*,* because the nerve-process passes through the anterior gray commissure before entering the white substance. Having arrived in the white substance the nerve-process of

* The commissure cells occupy an area which, arch-like, embraces the central canal on the ventral side; here they are of conspicuous size, approaching that of the motor cells of the anterior horns. Also farther back, in the median division of the gray substance, scattered commissure cells occur, but they are wanting in the posterior horns.

the majority of the column cells* divides into a vertical ascending and descending "stem-fiber," that in its course parallel to the longitudinal axis of the spinal cord sends off lateral twigs (collaterals), which return to the gray substance, where they branch and terminate in free fibrils; the stem-fibers themselves finally terminate like the collaterals. The collaterals that enter from the anterior columns penetrate the anterior horns singly or in bundles, where they weave themselves about the large

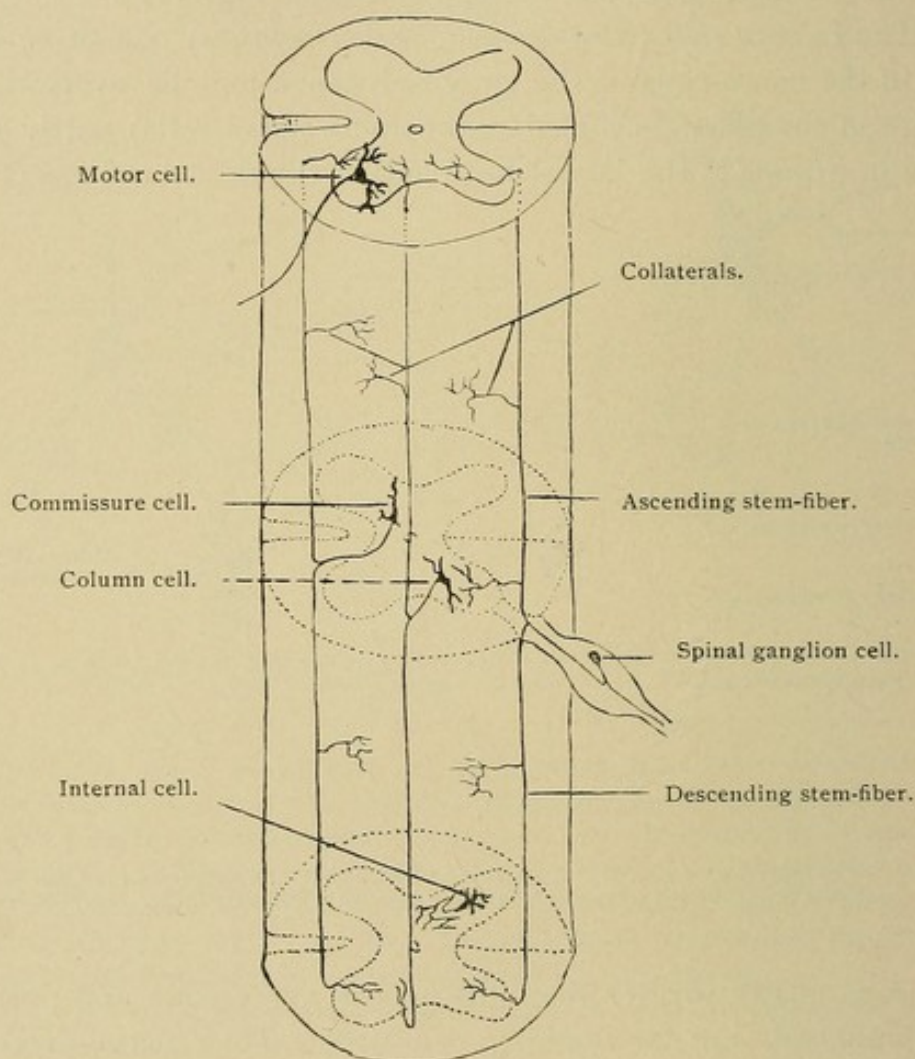


FIG. 118.—SCHEME OF THE LOCATION AND RAMIFICATION OF THE NERVE-CELLS AND OF THE POSTERIOR NERVE-ROOTS OF THE SPINAL CORD.

motor cells; they are especially numerous in the antero-lateral region of the anterior horns; not less numerous are the collaterals coming from the lateral columns. The spindle-shaped "marginal cells" lying in the

* Excepting the nerve-processes coming from the dorsal nucleus, which turn cranialward and proceed to the cerebellum. The nerve-processes of still other column cells enter the white substance and there, *without* dividing, turn upward or downward. Under the name of "pluri-funicular cells" column cells have been described, the nerve-process of which divides in the gray substance into two or three branches and continues in as many fibers in different columns.

zona spongiosa (p. 189) also belong to the column cells. In the adult the nerve-processes of all the column cells are enveloped in a medullary sheath.

The cells so far described belong to the type with the long nerve-process (p. 117). There is another, transitional, variety of cell, the nerve-process of which rapidly divides and remains within the gray substance. Because they do not pass beyond the gray substance these elements are named—

Internal cells; they occur in the posterior horns (Fig. 118), where their terminal ramification spreads out either on the same or on the opposite half of the spinal cord.

2. The *nerve-fibers*, that enter the gray substance from the anterior and lateral columns, partly arise from the medullated collaterals and terminals of the nerve-processes of the column cells, partly from the nerve-processes (likewise invested by a medullary sheath) that come from the brain.* In addition there are the medullated nerve-fibers of the posterior (dorsal) roots, which originate in the centripetal processes of the cells of the spinal ganglia (p. 215). These posterior root-fibers enter the spinal cord in two groups, a lateral, which runs in the zona terminalis, and a larger median, which runs in the posterior column. These fibers do not directly enter the gray substance, but each divides Y-shape into a longer ascending and a shorter descending stem-fiber (Fig. 119), from which numerous collaterals diverge at right angles (Fig. 118). These now enter the gray substance† and with their terminal fibrils distribute themselves over nearly every point of the same. One set terminates principally in the summit of the posterior horn; these fibers take their origin in the lateral root-fiber group and form a very fine-fibered, dense plexus, that also partly lies in the substantia gelatinosa (Fig. 120, c); a second set terminates in the

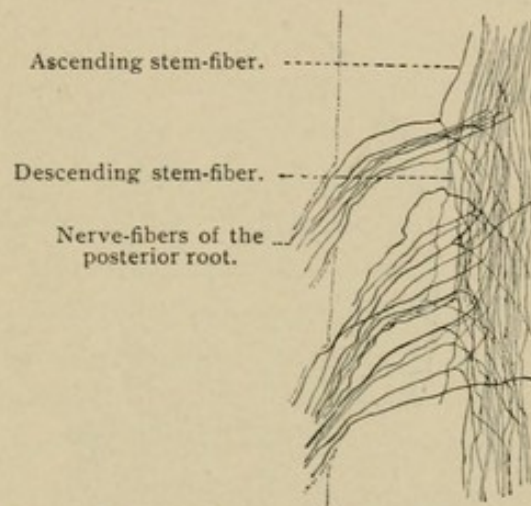


FIG. 119.—FROM A LONGITUDINAL SECTION OF THE SPINAL CORD OF A NEWBORN RAT. $\times 110$. The section shows two posterior nerve-roots. The collaterals are not visible. Technic No. 76.

* For an account of the exact course of these fibers the student is referred to special textbooks.

† An exception occurs in the case of some fiber-bundles which directly enter into the gelatinous substance and partly in this or ventral thereto (in the territory of the posterior horn) divide into ascending and descending stem-fibers.

dorsal nucleus (Fig. 120, *a*); * these originate in the median root-fiber group, as also a third set which, penetrating the median portion of the substantia gelatinosa, passes ventralward into the anterior horn and there, radiating fan-shape, surrounds the motor nerve-cells (Fig. 120, *b*); these latter very robust collaterals ("reflex collaterals") arise from the proximal portions of the stem-fibers, next to the point of bifurcation, and form the reflex bundle.† A fourth, smaller set of collaterals passes through the posterior gray commissure to the posterior horn of the opposite side. A fifth, likewise lesser set crosses transversely through the base of the posterior horns to the lateral column. The stem-

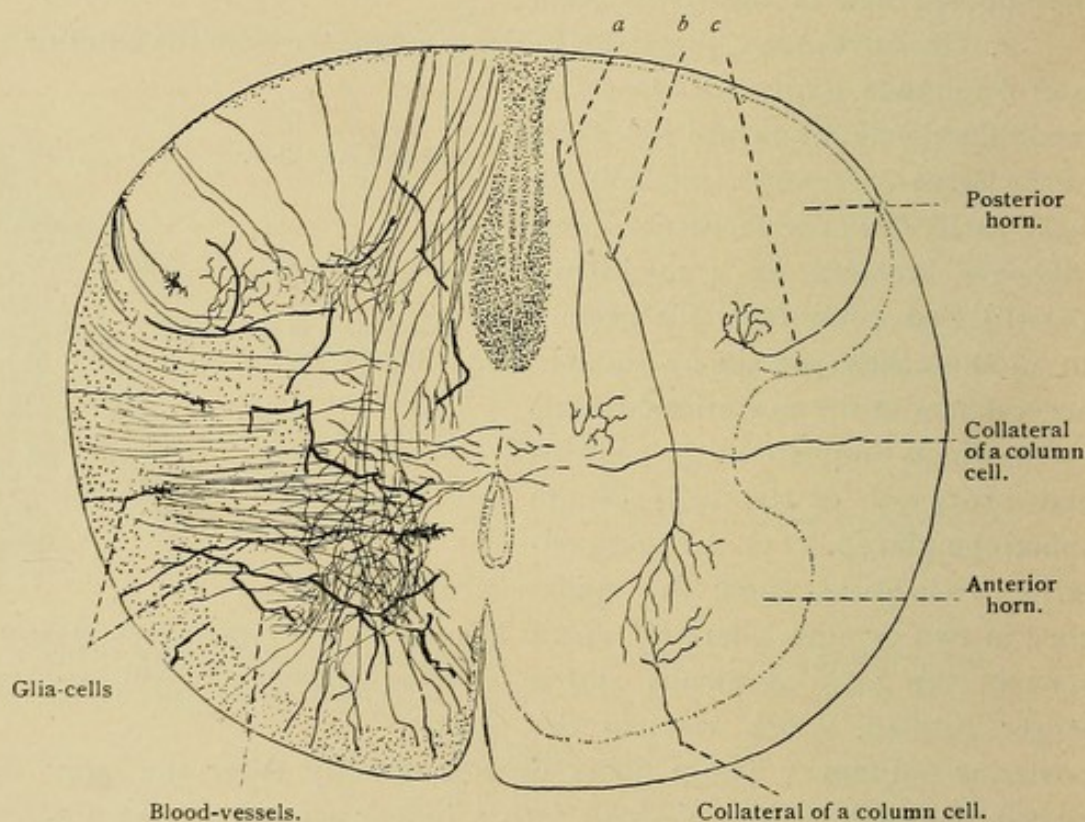


FIG. 120.—CROSS-SECTION OF THE SPINAL CORD OF A NEWBORN RAT, SHOWING COLLATERAL FIBERS. $\times 75$. In the right half only one representative of each variety has been sketched. Technic No. 76.

fibers, probably not until after a long course, sometimes extending into the oblongata, turn into the gray substance, where they terminate like the collaterals.

The peculiarities of the substantia grisea centralis and substantia gelatinosa, which belong to the gray substance, are dependent upon the abundance of the neuroglia and shall be described with this.

* Here the medullary sheaths extend farther than elsewhere—that is, to the last terminal ramifications.

† The reflex bundle and the collaterals of the dorsal nucleus sink into the gray substance in an arch with the concavity lateralward and form a conspicuous mass easily perceived (Fig. 115). The place at which they enter the gray substance has been named "root-entrance zone."

The *white substance* consists exclusively of nerve-fibers, medullated, that do not possess a neurilemma (p. 120), and nonmedullated. The fibers differ greatly in thickness; the thickest are found in the anterior columns and in the lateral parts of the posterior columns, the thinnest in the median parts of the posterior columns and in the lateral columns where the white substance touches the gray. In the remaining portions thick and thin fibers are intermingled. The majority of the nerve-fibers run parallel with the long axis of the spinal cord, hence in cross-sections are cut transversely. In addition there are fibers that take an oblique direction; these are found in large numbers in front of the gray commissure, where they cross and form the white commissure (Fig. 115).

An attempt to classify the nerve-fibers according to their origin will result as follows: 1, fibers which are processes of the posterior roots; the *entire* posterior columns consist of posterior root-fibers, because the latter (or their stem-fibers), entering in the lumbar region of the spinal cord, are pushed toward the median line by the fibers entering at higher levels; 2, fibers which are processes of the column cells (Fig. 118 and 120); 3, fibers which are processes of the nerve-cells of the brain. The latter two occupy the anterior and lateral columns and do not run an interlacing, irregular course, but are united in compact bundles.

The *supporting framework* of the spinal cord is constructed of two genetically distinct formations: 1, *connective-tissue* extensions of the pia, which penetrate the white substance as sheaths for the blood-vessels; this connective-tissue framework grows steadily thinner as it approaches the gray substance, into which it does not extend; 2, the *neuroglia* ("nerve-cement"), which is derived from the same embryonal anlage as the central nervous system. The neuroglia principally consists of nucleated elements, the *glia-cells* (Fig. 121), and (perhaps) of a small amount of homogeneous ground substance. There are two kinds of glia-cells: 1. The *ependymal cells*, which in a single layer line the lumen of the central canal. In youth they are beset with cilia, their cylindrical body is prolonged in an extended process (Fig. 121), that in the embryo reaches to the surface of the spinal cord, where it terminates in a simple or branched end. The cells of the ependyma are phylogenetically the older; they arise also ontogenetically first, but in the further course of development undergo regression in different degrees; the long processes in particular are involved, which retain their original length to the surface of the spinal cord only in the region of the posterior median septum* and opposite,

* The posterior median septum consists for the greater part of processes of ependymal cells.

to the base of the anterior median fissure. In the course of development one division of the ependymal cells wanders peripheryward and becomes transformed into astrocytes. Not infrequently the central canal is completely obliterated. 2. The *astrocytes* (Deiters's cells), in the beginning of their development, all lie in the gray substance; later some retreat into the white substance and then are very differently shaped. Of the numerous processes of the astrocytes one, the "chief process" (Fig. 121), frequently originates earliest, the others, partly finer and partly coarser "secondary" processes, arise later. Many of these cells reach with much-branched processes to the surface of the spinal cord,

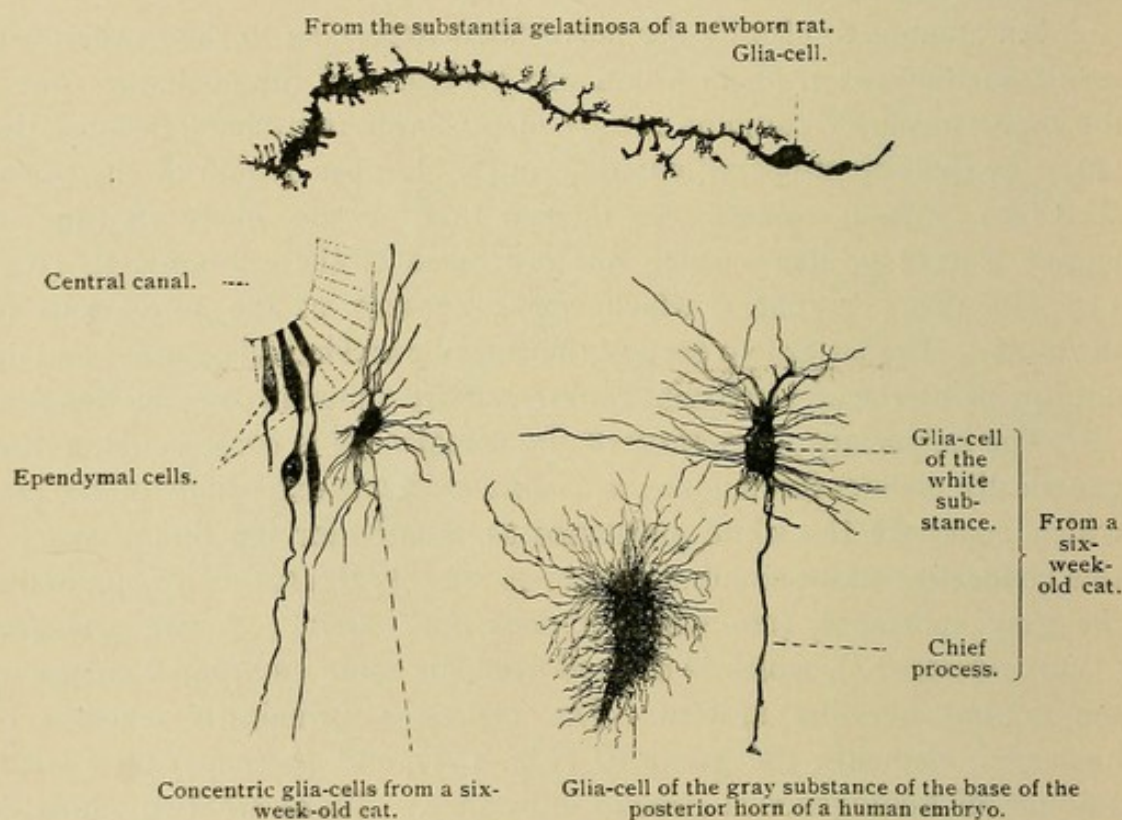


FIG. 121.—GLIA-CELLS FROM THE SPINAL CORD. $\times 280$. Technic No. 76.

where they terminate in expanded ends* and form a conspicuous border, the superficial glia-zone ("gelatinous cortical layer" or "horn-spongiosa"). Of the developed cells two varieties, united by transitional forms, are distinguished: (a) The *short-rayed cells* (mossy-cells) possess shorter, very richly branched processes, that not infrequently are attached to the blood-vessels; they chiefly occur in the gray substance. (b) The *long-rayed cells* (spider-cells), the more usual form, have a small

* These expanded ends stand close beside one another and form a "membrana limitans meningeae," which is not an independent membrane any more than the internal limiting membrane of the retina (see The Visual Organ).

cell-body, from which besides short, also many longer, rigid, less-branched processes radiate (Fig. 121); these chiefly occur in the white substance and are not apt to be confused with the ganglion cells. By the interlacing of the numerous fine processes of neighboring glia-cells (they do not anastomose) a close web is constructed which envelops each individual nerve-fiber.*

In the substantia grisea centralis and substantia gelatinosa the neuroglia assumes a totally different appearance. In the former the astrocytes with their here very long, stiff, unbranched processes are concentrically arranged in a close fiber-wreath (Fig. 121). These and the cells of ependyma are together called "central ependyma filaments." The substantia

gelatinosa consists of a small number of very small ganglion cells, the nerve-processes of which turn into the zona terminalis, of a plexus of delicate nerve-fibrils, and of nerve-fibers (collaterals) passing through; there is besides a granular substance present which has arisen by a transformation of numerous and very delicate processes of the few astrocytes occurring there (Fig. 121).

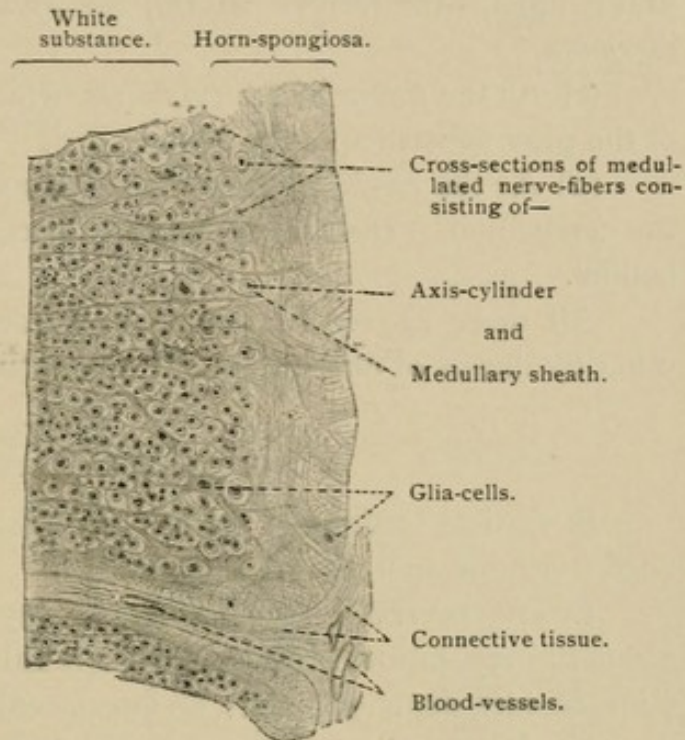


FIG. 122.—FROM A CROSS-SECTION OF THE HUMAN SPINAL CORD IN THE REGION OF THE LATERAL COLUMN. $\times 180$.
Technic No. 75.

THE BRAIN.

The brain, like the spinal cord, is composed of a white and a gray substance, which in their minute structure agree on the whole with the same substances in the cord. But the arrangement of the two substances in the brain is a much more diversified one than in the spinal cord.

*In accordance with this account the neuroglia consists of cells and their processes only; whether also free fibers occur, that have become detached from the cell-body, has not yet with certainty been distinguished. The fact that a portion of the delicate processes (fibers) are differentiated by their chemical nature from the usual cell-processes does not prove that fibers and cells are entirely distinct structures.

The gray substance of the brain occurs in four aggregations :

(a) As the *cerebral cortex*, an expansion covering the entire surface of the cerebral hemispheres.

(b) In the form of discrete masses, which are situated in the cerebral ganglia,—the *corpora striata*, the *optic thalami*, the *corpora quadrigemina*.

(c) As the *lining* of the *ventricles*, which is the direct continuation of the gray substance of the spinal cord.

(d) As the *cerebellar cortex*, an expansion covering the surface of the cerebellum. Discrete masses also occur in the interior of the cerebellum.

All these aggregations have numerous connections with one another by means of fiber-tracts of white substance.

THE CEREBRAL CORTEX.

In vertical sections of the cerebral cortex four zones, *not* sharply defined from one another, are distinguished.

1. The *molecular zone* (neuroglia layer), the most superficial, in ordinary preparations appears finely granulated or reticulated and contains, besides many glia-cells, an interlacement of medullated nerve-fibers running horizontally, the *tangential fibers* (Fig. 123). By means of Golgi's method it may be seen that the reticulum is partly formed by the dendrites of the pyramidal cells of the second and third zones (see sub 2 and 3), and partly by the processes of glia-cells. Besides the latter the *cells of Cajal* occur in the molecular zone; they possess an irregularly shaped cell-body that sends out very long processes running parallel to the surface, from one portion of which vertically to the surface ascending lateral twigs arise* (Fig. 124, 1).

2. The *zone of the small pyramidal cells* (Fig. 123, Fig. 124) is characterized by ganglion cells from 10 to 12 μ in size and of a pyramidal form; the apex of the pyramidal cell is prolonged into a long ramifying protoplasmic process (dendrites),† that after giving off minute lateral twigs enters the molecular zone, where it terminates in numerous, often serrulate branches (Fig. 124, 2); only small dendrites spring from

* In animals four and even more "nerve-processes" of Cajal's cells have been described. It seems as if different unrelated cell forms were grouped under this name. In the forms described as Cajal's cells in man a true nerve-process has not been distinguished; it is probable that these latter elements are glia-cells.

† For this reason it is difficult to determine the size of the pyramidal cells; the considerable differences in the estimated size may be referred to this gradual passage of the cell-body into the apical process.

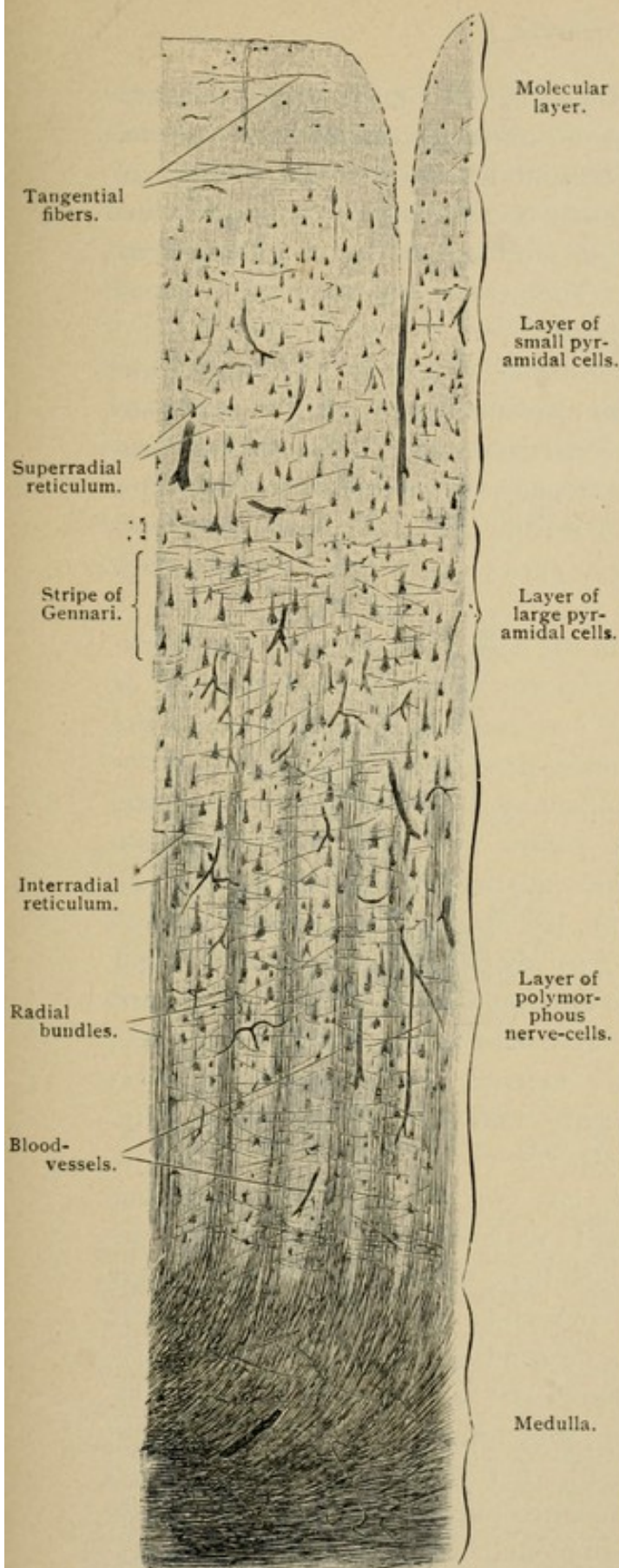


FIG. 123.—VERTICAL SECTION OF HUMAN CEREBRAL CORTEX. $\times 60$. Technic No. 77.

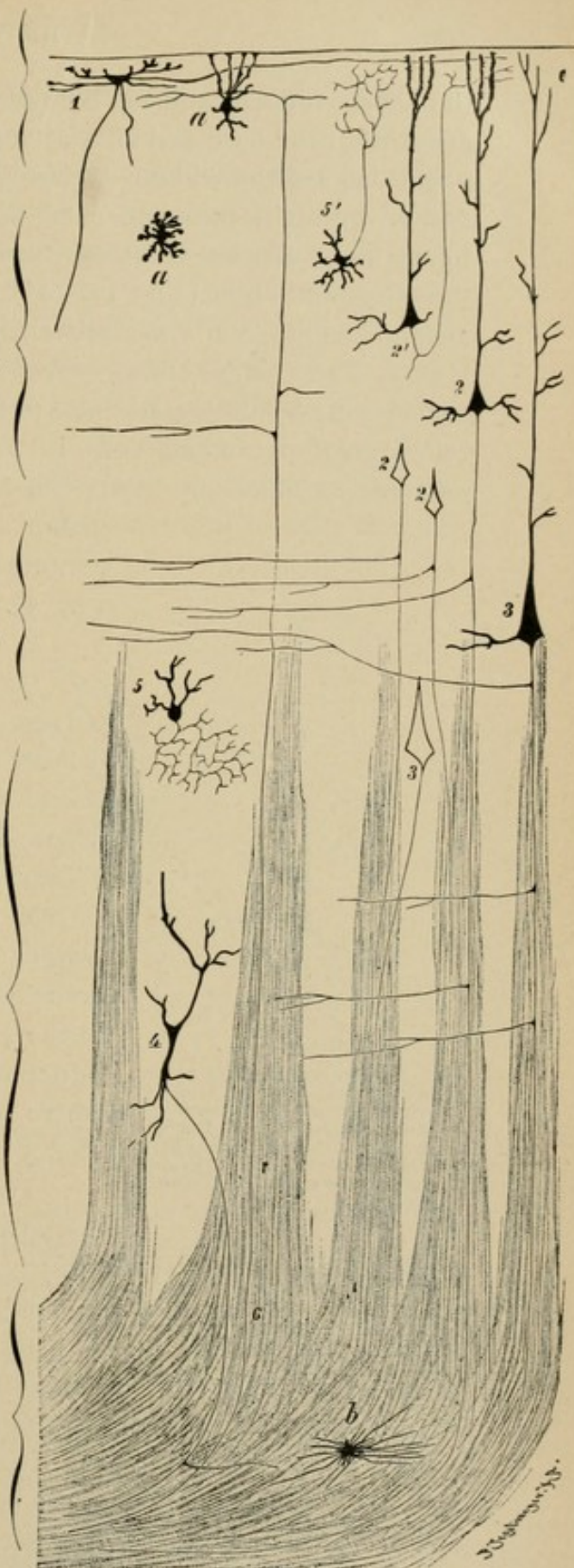


FIG. 124.—SCHEME OF THE CEREBRAL CORTEX, sketched from specimens prepared according to Technic No. 79 b. 1. Cell of Cajal (glia-cell?). 2, 2'. Small pyramidal cells. 3. Large pyramidal cell. 4. Polymorphous cell. 5, 5'. Cells of Golgi's type. 6. Nerve-fiber ending in the superficial zone; a, mossy-cell, b, spider-cell (glia-cells). The ependymal cells are not represented.

the lateral surfaces and the base of the cell. The nerve-process always arises from the base and after giving off branched collaterals, as a rule, passes toward the white substance, there to become the axis-cylinder of one or, by division, of two nerve-fibers; occasionally it turns and runs to the molecular layer, where it divides and enters the web formed by the tangential fibers (Fig. 124, 2'). The nerve-process and the collaterals are enveloped in a medullated sheath.

3. The *zone of the large pyramidal cells* is distinguished from the preceding zone by the greater size of its elements (from 20 to 30 μ , the so-called giant pyramidal cells in the anterior central convolution even measure as much as 80 μ); the extremely robust nerve-process, after giving off several collaterals in the gray cortex, always goes to the white substance (Fig. 124, 3).

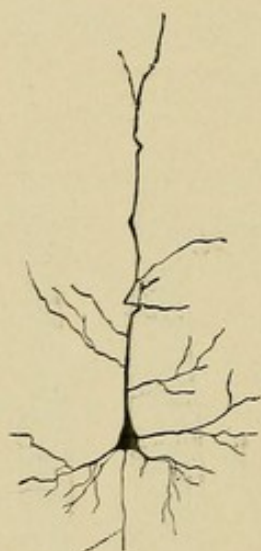


FIG. 125.—PYRAMIDAL CELL FROM A PERPENDICULAR SECTION OF THE CEREBRAL CORTEX OF ADULT MAN. $\times 120$. The terminal branches of the dendrites running toward the molecular layer are not visible. Technic No. 79 b.

4. In the *zone of the polymorphous nerve-cells* the majority of the elements are oval or polygonal; an apical dendrite is wanting; the delicate nerve-process after sending off a number of collaterals enters the white substance, where it passes into one, or dividing into T-branches, into two nerve-fibers (Fig. 124, 4).

In the last three zones ganglion cells of Golgi's type also are found (p. 118). Their branched nerve-process sometimes is confined to the gray cortex in the vicinity of the cell, sometimes extends to the molecular zone, where richly branched it terminates (Fig. 124, 5, 5').

The last two zones contain numerous medullated nerve-fibers. The same are arranged in thick "radiating" bundles, which resolve into single fibers near the zone of the small pyramidal cells (Fig. 123). These bundles are formed by (1) the descending medullated nerve-processes of the large and the small pyramidal cells, by (2) thick medullated nerve-fibers of unknown origin, that ascend from the white substance toward the cortex (Fig. 124, 6), where they repeatedly divide and form the "superradial" and the tangential plexus (Fig. 123), and finally end in free branches. Another set of medullated nerve-fibers runs transversely to the radiating bundles and forms the "interradial" reticulum; this is somewhat condensed toward the superradial reticulum and thus represents the *stripe of Gennari* or *Baillarger* (Fig. 123). This and the

interradial reticulum are composed of the medullated collaterals of the nerve-processes of the pyramidal cells.

The structure of the cerebral cortex is modified in certain localities. In the hippocampal and the uncinate convolution the tangential fibers are present in larger numbers and form an expanded net-like white layer, the *substantia reticularis alba*. In the vicinity of the calcarine fissure the stripe of Gennari is developed into the *bundle* of *Vicq d'Azyr*, which may be seen by the unaided eye. Furthermore, greater* or lesser deviations occur in many localities, which render a classification according to the foregoing description much more difficult.

Finally extensions of the pia, that penetrate in company with the blood-vessels, and neuroglia participate in the construction of the cerebral cortex.

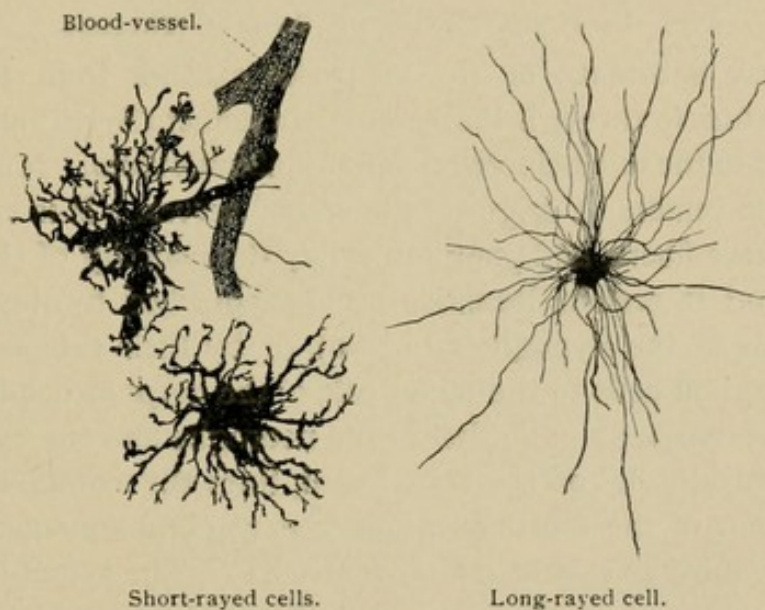


FIG. 126.—FROM SECTIONS OF THE BRAIN OF ADULT MAN. $\times 280$. Technic No. 79 b.

Neuroglia.—This like that of the spinal cord consists of ependymal cells and of astrocytes. In the embryo the peripheral processes of the former extend to the free surface. Of the latter two varieties are distinguished. The elements of the one variety are characterized by their small cell-body and long, rigid, little-branched processes, of which the most delicate rest like a short turf on the cell-body; they are called *long-rayed cells* (Fig. 126), and chiefly occur in the white substance. The elements of the other variety, the *short-rayed cells* (Fig. 126), have short, gnarled, richly branched processes and are mainly found in the gray substance, where they are in intimate relation with the blood-vessels, to

* Regarding the minute structure of the cortex of the cornu ammonis and the bulbus olfactorius, the reader is referred to special text-books.

the walls of which they are often attached by one thicker process (Fig. 126).^{*} On the surface of the cerebral cortex there is a glia-zone formed by the ends of the thitherward extending processes of the glia-cells.

THE CEREBRAL GANGLIA.

The gray substance of the cerebral or basal ganglia consists of ganglion cells differing in size, medullated nerve-fibers, and neuroglia. The macroscopic variations in color depend on the different proportions in which the multipolar ganglion cells and the nerve-fibers are mingled: wealth of ganglion cells is rendered perceptible by a dark red-brown color, profusion of nerve-fibers by a pale yellow-gray color.

THE GRAY SUBSTANCE OF THE VENTRICLES.

The gray substance of the ventricles extends from the floor of the fourth ventricle through the cerebral aqueduct (Sylvii) into the third ventricle, and to the tuber cinereum and the infundibulum. It is of especial interest as the centers of origin of the cranial nerves. It is composed of neuroglia, nerve-fibers, and ganglion cells; the majority of the latter are multipolar and in certain localities are distinguished by their size (*e. g.* in the nucleus of the hypoglossal nerve), or by their peculiar form (the spherical ganglion cells in the upper pair of the corpora quadrigemina).

The ependyma, an extension of the neuroglia and the cylinder cells lining the central canal of the spinal cord, lines the continuation of the canal (the floor of the fourth ventricle, the cerebral aqueduct, the inner surface of the third and of the lateral ventricles). The cylindric or cubical cells of the *ependyma of the ventricles* in the newborn and in part also in the adult possess cilia.

THE CEREBELLAR CORTEX.

The cerebellar cortex consists of three well-defined strata of gray substance, of which the outer and inner are macroscopically, the middle, on the contrary, only microscopically perceptible: they are from within outward, the *granule layer*, the *ganglionic layer*, and the *molecular layer*.

^{*} This relation is regarded as evidence that the neuroglia has not only a mechanical function as supporting apparatus, but also a nutritive function as conveying apparatus for nutrient fluids. The glia-cells are said also to be the means by which the myelin furnished by the blood is transferred from the vessels to the nerve-processes of the central nervous system (*cf.* remark *, p. 112).

1. The *granule stratum*, the innermost, is characterized by its rust color and consists of numerous layers of small cells, that by the ordinary methods exhibit a proportionately large nucleus and a very slightly developed protoplasm. By the aid of Golgi's method it becomes evident that, apart from the glia-cells, two varieties of ganglion cells are present: *small granule-cells* and *large granule-cells*. The former (Fig. 128) are multipolar ganglion cells* with short dendrites (D) with claw-like endings and a delicate nonmedullated nerve-process (N), that

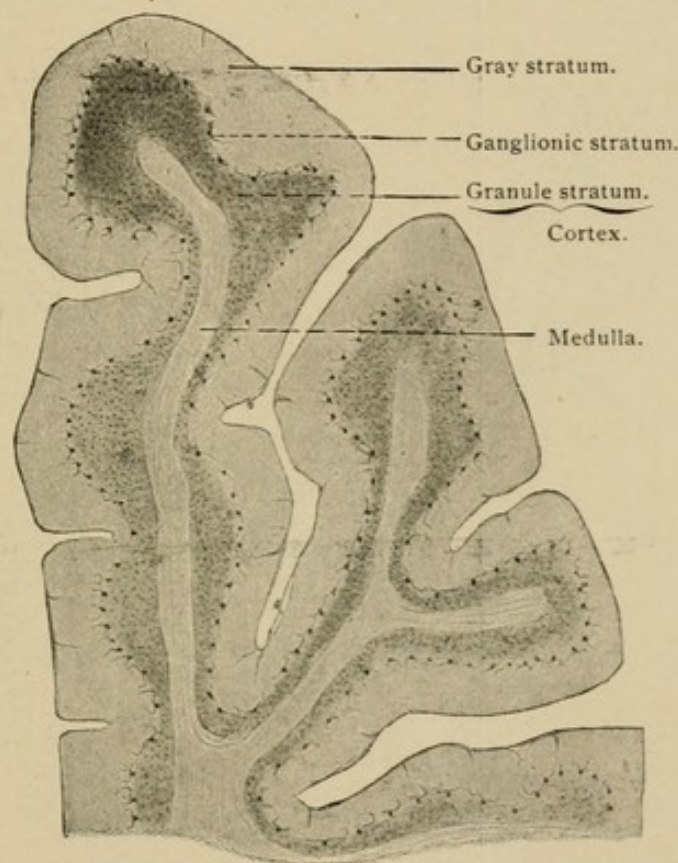


FIG. 127.—FROM A VERTICAL SECTION OF THE CEREBELLUM OF ADULT MAN. $\times 12$. Technic No. 78.

passes vertically into the outermost layer and there divides into two T-branches that run lengthwise to the convolutions, parallel to the surface of the same, and terminate in free unbranched ends. The small granule-cells form the chief mass of the cellular elements of the granule stratum. Less numerous are the *large granule-cells* (Fig. 129), multipolar ganglion cells more than twice the size of the smaller elements, the ramifying dendrites of which extend into the outermost stratum, the nerve-process of which, running in the opposite direction, rapidly

* Their ganglion-cell nature has recently been called into question, because fibrillæ are wanting in the protoplasm and the nucleus does not possess the structure typical for nerve-cells.

divides and terminates in a very rich ramification penetrating the entire granule stratum.

A dense plexus of medullated nerve-fibers occurs in the granule stratum (Fig. 130, 3); the greater part of these fibers come from the white substance of the cerebellum and at the boundary between the granule and the ganglionic stratum form a layer of horizontal bundles (3') running transverse to the longitudinal axis of the convolutions, from which fibers

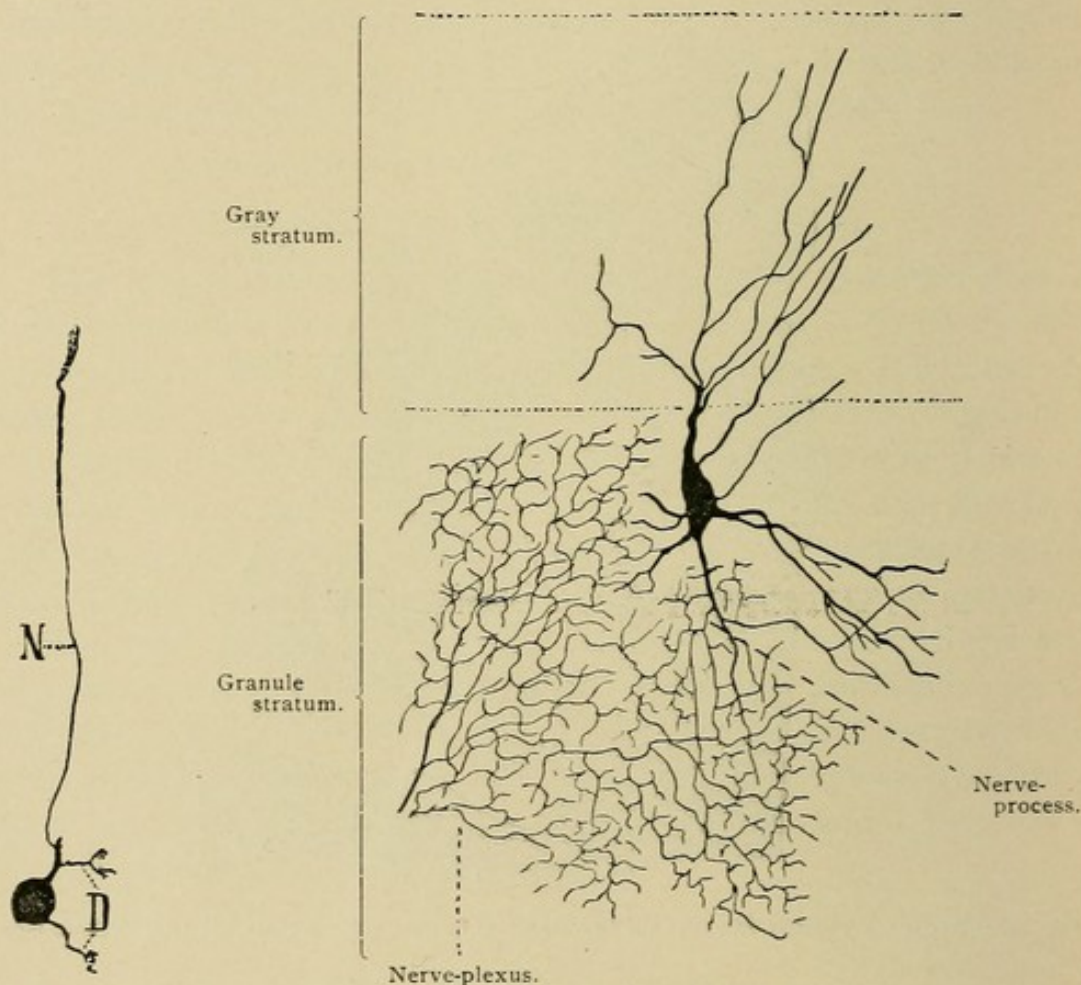


FIG. 128.—SMALL GRANULE-CELL. $\times 400$.

FIG. 129.—LARGE GRANULE-CELL. $\times 200$.

From sections of the cerebellar cortex of a cat six weeks old. Technic No. 80.

ascend into the gray stratum (3''). A small portion of this plexus is formed by the medullated nerve-processes of the cells of Purkinje.

2. The middle, *ganglionic stratum* of the cerebellar cortex consists of a simple layer of very large multipolar ganglion cells, the *cells of Purkinje* (Fig. 64). Their somewhat pear-shaped body sends two robust dendrites into the gray layer, where they terminate in an uncommonly rich arborization extending to the free surface (Fig. 130, 4). The arbori-

zation does not extend in all directions, but only in planes transverse to the long axis of the convolution, therefore the entire ramification can be seen only in transverse sections of the convolution. From the opposite pole of the cell the nerve-process arises, soon acquires a medullary sheath, and passing through the granule stratum enters the white substance of the cerebellum; while still within the granule stratum it sends off collaterals

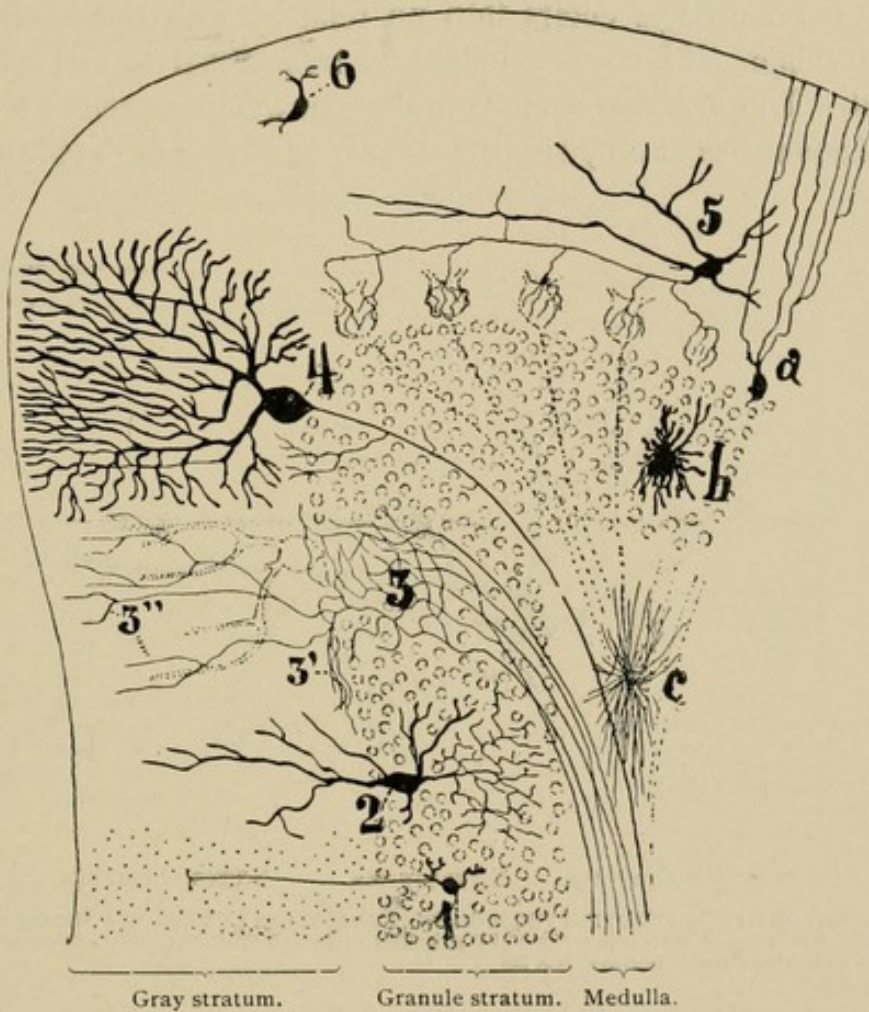


FIG. 130.—SCHEME OF THE CEREBELLAR CORTEX, sketched from specimens prepared according to technic No. 80.

1, Small granule-cell; 2, large granule-cell; 3, plexus of nerve-fibers; 3', horizontal bundle; 3'', fibers of the gray stratum; 4, cell of Purkinje; 5, basket-cell; 6, small cortical cell, the nerve-process is not represented. a, Glia-cell of the gray stratum; b, glia-cell resembling a short-rayed cell; c, long-rayed cell.

that branch there and in part run back between the cells of Purkinje (Fig. 130).

3. The outer, *gray stratum* is characterized by its gray color and contains two varieties of ganglion cells:

(a) The *basket-cells* (large cortical cells), multipolar ganglion cells, the dendrites of which chiefly extend toward the free surface. Their long, at first thin, subsequently thick nerve-process runs horizontally in

the transverse direction of the convolutions and sends a few collaterals toward the free surface, into the depths, at successive intervals, delicate branches that with their terminal ramifications surround, basket-wise, the bodies of the cells of Purkinje (Fig. 131). Frequently the basket also includes in its embrace the initial portion of the nerve-process of the cells of Purkinje.

(b) The *small cortical cells*, which differ from the basket-cells in this that their nerve-process enters into no relation with the bodies of the cells of Purkinje. Two types, united by transitional forms, can be distinguished. The *first type* exhibits multipolar ganglion cells; their cell-body is of equal size or only slightly smaller than that of the basket-cells,

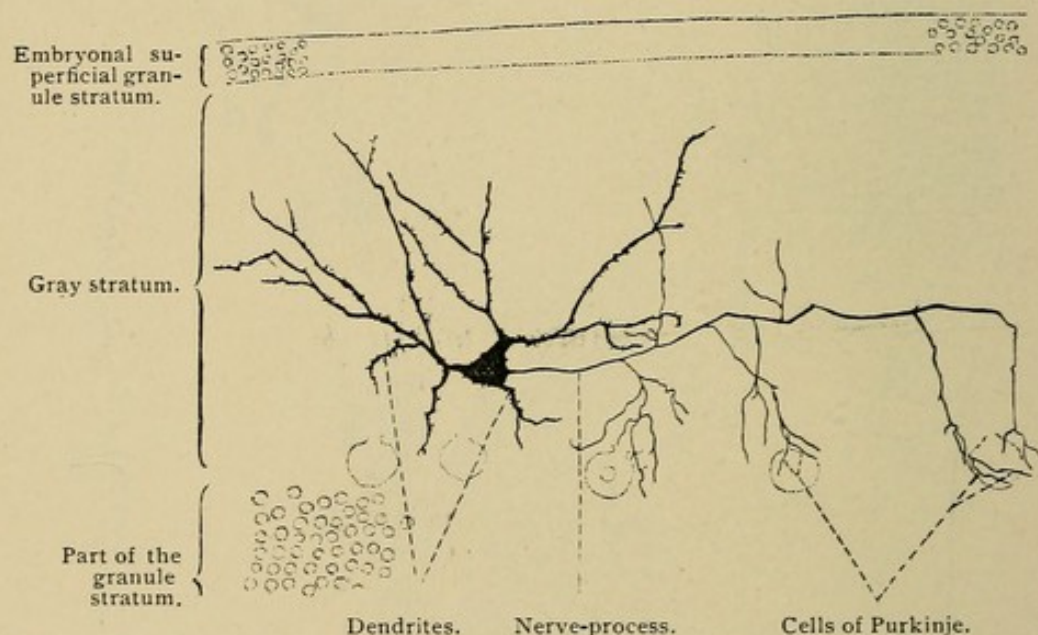


FIG. 131.—BASKET-CELL FROM A SECTION THROUGH THE CEREBELLAR CORTEX OF A SIX-WEEK-OLD CAT. $\times 240$. The five cells of Purkinje were not blackened but were plainly visible; only the outlines of their bodies are sketched. Technic No. 80.

their few (from 2 to 5) dendrites, like those of the Purkinje cells, lie in the transverse planes of the convolutions, their thin nerve-process is very long (1 mm. and over), occasionally exhibits loop formations, and is characterized by a very rich initial ramification (Fig. 132), the terminal ramification being scanty. The cortical cells of the *second type* are in general somewhat smaller and their short nerve-process ramifies in the immediate vicinity.

The elements of the first type form the chief mass of the relatively numerous small cortical cells and occur in the entire thickness of the gray stratum, more profusely in the superficial than in the deep parts. The cells of the second type are found everywhere in the gray stratum.

The medullated nerve-fibers occurring in the gray stratum are processes of the plexus of the granule stratum and partly pass toward the free surface, where after loss of the medullary sheath they terminate in free ramifications between the protoplasmic arborizations of the Purkinje cells, partly they run horizontally between the bodies of the cells of Purkinje, longitudinally to the convolutions (Fig. 130, 3').

The *neuroglia* of the cerebellum consists of (1) cells, the small body of which lies at the outer boundary of the granule layer and sends a few short processes into the depths, but many long processes in a straight course toward the free surface, where they terminate in a triangular expansion (Fig. 130, a) and in this way form a relatively thick peripheral glia-layer; (2) also of stellate elements resembling the short-rayed

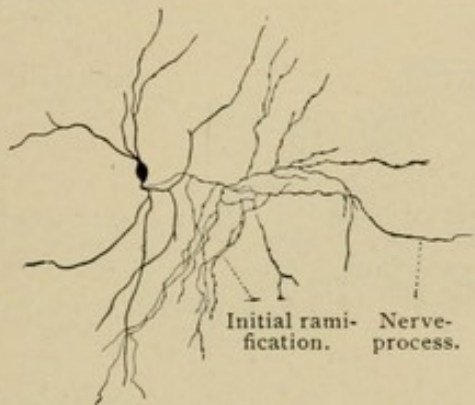


FIG. 132.—A SMALL CORTICAL CELL OF THE FIRST TYPE. FROM A SECTION OF THE CEREBELLAR CORTEX OF ADULT MAN. $\times 173$. Technic No. 80.



FIG. 133.—TWO GLIA-CELLS FROM A SECTION THROUGH THE CEREBELLAR CORTEX OF ADULT MAN. $\times 90$. On the right the body, *P*, and the dendrites, *P*, of a cell of Purkinje are sketched in outline to demonstrate the difference between this element and the glia-cells. Technic No. 80.

cells of the cerebral cortex (Fig. 130, b); they occur in all the strata (Fig. 133). In the white substance typical long-rayed cells are found.

So long as the cerebellar cortex is not fully developed a series of peculiarities exists that is wanting in the adult. In embryos and young animals there is over the as yet slightly developed gray stratum a superficial granule stratum (Fig. 131); the structures in the granule stratum described under the name of "moss-fibers" are developmental forms of medullated nerve-fibers; of like significance are the "climbing plexuses," which are found in the neighborhood of the ramifying protoplasmic processes of the cells of Purkinje.

The union of the elements of the cerebellum is only by contact, not by direct connection.

The *white substance*, the "medulla," of the cerebrum and of the

cerebellum, apart from the elements of the supporting framework (connective tissue and neuroglia), consists throughout of medullated nerve-fibers, without a neurilemma and varying in thickness from 2.5 to 7 μ .

The *hypophysis cerebri* (pituitary body) is composed of two genetically different parts: (1) a *posterior small lobe* that belongs to the brain and is a continuation of the infundibulum; it contains delicate, much-branched nerve-fibers, that form a very fine plexus, and connective tissue, many blood-vessels, and cells that closely resemble bipolar or multipolar ganglion cells, but the nature of which is still uncertain; (2) an *anterior larger lobe* derived from a diverticulum of the embryonal oral cavity; this lobe contains *gland follicles* embedded in loose, vascular

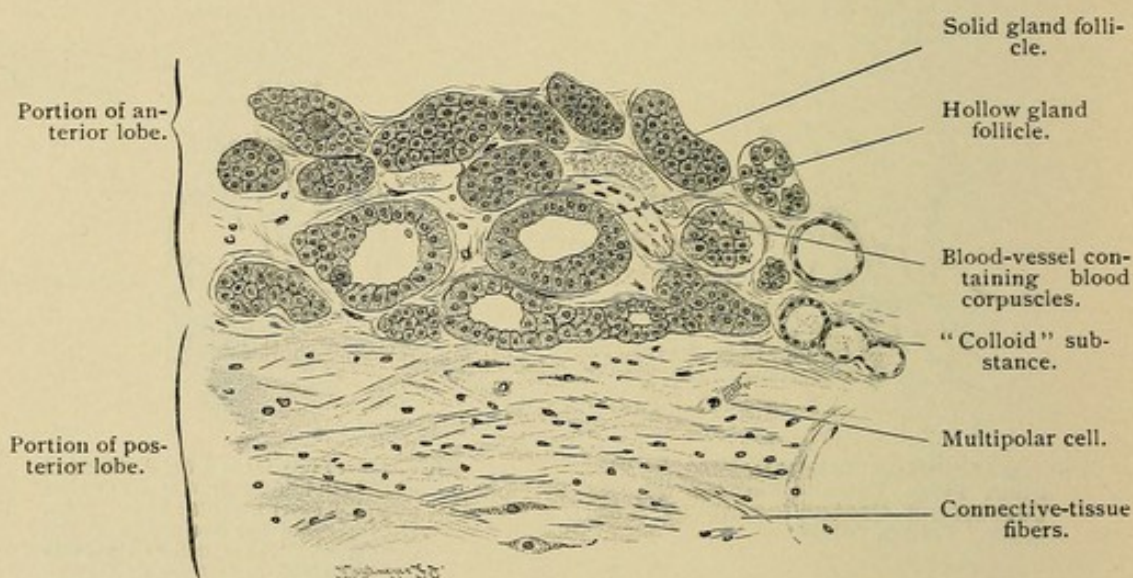


FIG. 134.—PORTION OF A HORIZONTAL SECTION OF A HUMAN PITUITARY BODY, showing the boundary line between the anterior and the posterior lobe. Two gland follicles on the left contain each a dark epithelial cell. $\times 220$. Technic No. 81.

connective tissue, the majority of which are solid and filled with sometimes clear, sometimes granular, cubical epithelial cells, occasionally containing vacuoles (Fig. 134). Only a few of the follicles, toward the border of the smaller lobe, are hollow and occasionally contain a mass resembling the colloid substance of the thyroid, that did not originate in the granules which are to be seen in many of the epithelial cells.

The *pineal body* (corpus pineale, epiphysis) is derived from a fold of the wall of the primitive brain vesicle and consists of (epithelial) cells, some of which have delicate processes, and of a connective-tissue envelope from which processes extend into the interior of the organ.* Almost

* In the pineal body of the ox cross-striated muscle-fibers have been found.

invariably "brain sand" (*acervulus cerebri*) is found in the pineal body, rounded concretions from $5\ \mu$ to 1 mm. in size, with an uneven, mulberry-like surface (Fig. 135). They are composed of an organic basis, calcium carbonate and magnesium phosphate.

Not infrequently (especially in advanced life) there occur in the brain substance round or discoid bodies exhibiting distinct stratification, which stain violet on treatment with iodine and sulfuric acid, therefore are related to amylum (Fig. 136 *a*). These *corpuscula amylacea*, almost constant on the walls of the ventricles of the brain, are also present in many other localities, as well in the gray as in the white substance, and in the optic nerve. Closer investigation reveals the presence of a homogeneous capsule provided with a few processes. They are glia-cells transformed by amyloid infiltration.



FIG. 135.—BRAIN SAND FROM THE PINEAL BODY OF A WOMAN SEVENTY YEARS OLD. $\times 50$.
Technic No. 82.

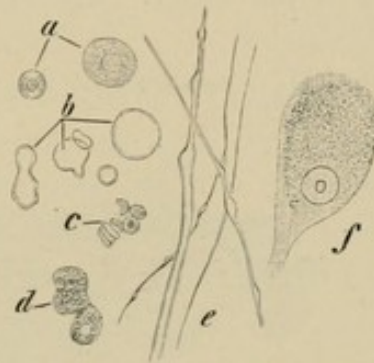


FIG. 136.—FROM A TEASED PREPARATION OF GRAY SUBSTANCE FROM THE WALL OF A VENTRICLE OF THE HUMAN BRAIN. $\times 240$. *a*, Corpuscula amylacea; *b*, myelin drops; *c*, red blood corpuscles; *d*, ependymal cells; *e*, medullated nerve-fibers; *f*, ganglion cell. Technic No. 83.

THE MEMBRANES OF THE CENTRAL NERVOUS SYSTEM.

Two connective-tissue membranes envelop the brain and the spinal cord: the *dura* and the *pia*.

The *dura* of the *spinal cord* (*dura mater spinalis*) consists of compact fibrous connective tissue and numerous elastic fibers, flat connective-tissue cells and plasma cells (p. 93 and Fig. 141). The inner surface is covered by a simple layer of flat epithelial cells. It is poor in nerves and blood-vessels.

The *dura* of the *brain* (*dura mater cerebri*) is at the same time the periosteum of the inner surface of the cranium and consists of two lamellæ: (1) an *inner*, which corresponds to the *dura* of the cord and is of like structure, with the exception that it is richer in elastic fibers, and (2) an *outer* lamella which corresponds to the periosteum of the vertebral canal. The latter is composed of the same elements as the inner

lamella, except that the outer fibers run in a different direction; anteriorly and laterally they run posteriorly and medianward, while the inner fibers run from the anterior median region posteriorly and lateralward. The outer lamella is rich in blood-vessels, which pass from it into the cranial bones. The dura is rich in nerves, of which two varieties may be distinguished, vascular nerves and free-ending *nervi proprii*.

The *pia* of the brain and the spinal cord is a two-layered sack. The outer layer (the *arachnoid* of authors) is covered on its free surface by a simple layer of epithelium and is not closely attached to the dura. The inner layer (the "*pia*") closely envelops the surface of the brain and the spinal cord and sends vascular processes into their substance. The arachnoid and the pia are joined together by numerous bands and trabeculæ extending from the inner surface of the former to the outer surface of the latter. Hernia-like evaginations occur on the outer sur-

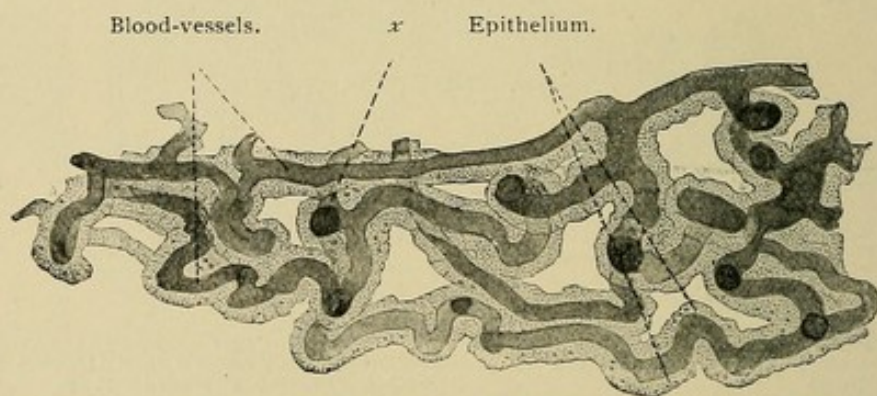


FIG. 137.—PORTION OF THE PLEXUS CHORIOIDEUS OF ADULT MAN. $\times 80$. *x*, Blood-vessel in optical cross-section. The large dots in the epithelium are not nuclei, but pigment and fat-granules. Technic No. 84 *b*.

face of the arachnoid in certain localities, in particular near the superior longitudinal sinus, which push the attenuated dura before them and project into the venous sinus. These are the so-called *arachnoidal granulations* (*Pacchioni*), which were long regarded as pathologic. The pia is composed of delicate connective-tissue bundles and plate-like cells, which cover the inner surface of the arachnoid, the bands and the trabeculæ.

It is the carrier of numerous blood-vessels, that likewise possess nerves. But whether the vessels occurring in the brain and in the spinal cord are encircled by networks of nerves is still questionable.

The *telæ chorioideæ* and the *plexus chorioidei* consist of connective tissue and numerous blood-vessels, the fine ramifications of which are united in lobules that are suspended within the ventricles. They are covered by a simple layer of cubical epithelial cells, ciliated in the newborn, which enclose pigment granules and also oil globules.

THE VESSELS OF THE CENTRAL NERVOUS SYSTEM.

The *blood-vessels* of the central nervous system form a narrow-meshed capillary network in the gray, a wide-meshed network in the white substance, which are everywhere connected with each other. The capillaries of the cerebral cortex open into veins that do not take their origin in the cortex, but beneath in the white substance and from there traverse the cortex and go to the veins lying in the pia. Therefore the blood in the capillaries must traverse the entire cortex before it empties into the veins. All the blood-vessels possess a second so-called adventitial sheath, which often consists of only a simple stratum of epithelial cells (see further below). The walls of the intradural venous sinuses are formed by a membrane of flat epithelial cells.

THE LYMPH PATHS.

1. Between the dura and the arachnoid there is a capillary fissure, the *subdural space*, which communicates with the deep cervical lymph-vessels and lymph-nodes (at least in the rabbit and the dog), with the lymph channels of the peripheral nerves, with the lymph-vessels of the nasal mucous membrane, with the small clefts (juice-canals) in the dura, and finally, round the arachnoidal granulations, with the intradural venous sinuses. The fluid in the subdural space is very scanty.

2. The *subarachnoid space*, that between the two layers of the pia (or arachnoid and pia), communicates with the lymph channels of the peripheral nerves, with the lymph-vessels of the nasal mucous membrane, with the interior of the ventricles of the brain and of the central canal of the spinal cord. The fluid in the subarachnoid space is very abundant; it is called the *cerebro-spinal fluid* (liquor cerebro-spinalis).

3. The spaces occurring within the adventitial sheath of the blood-vessels can be injected from the subarachnoid space. They are called *adventitial lymph spaces*.

The spaces filled only by injecting the brain substance itself cannot be included in the lymph vascular system. These spaces occur as (1) *pericellular spaces*, surrounding the larger ganglion cells of the cerebral cortex, also many glia-cells; as (2) *perivascular spaces* of the blood-vessels, that formed by the adventitial sheath excepted; and between the pia and the cerebrum, as (3) the *epicerebral space*. These may be regarded as an independent juice-canal system.

2. THE PERIPHERAL NERVOUS SYSTEM.

THE NERVES.

The *cerebrospinal nerves* chiefly consist of medullated nerve-fibers differing in thickness and of only a few nonmedullated nerve-fibers; therefore by direct light they appear white. Their mode of union agrees in many respects with that of the striated muscle-fibers. A corresponding sheath formed of loose connective tissue and numerous elastic fibers, often containing clusters of fat-cells, surrounds the entire nerve. It is called the *epineurium* (Fig. 138). Processes of the epineurium in the interior of the nerve envelop the (so-called secondary) nerve-fiber bundles, of which each is encircled by concentrically curved lamellæ of connective tissue, the *perineurium*. From the latter connective-tissue septa extend into the interior of the (secondary) nerve-fiber

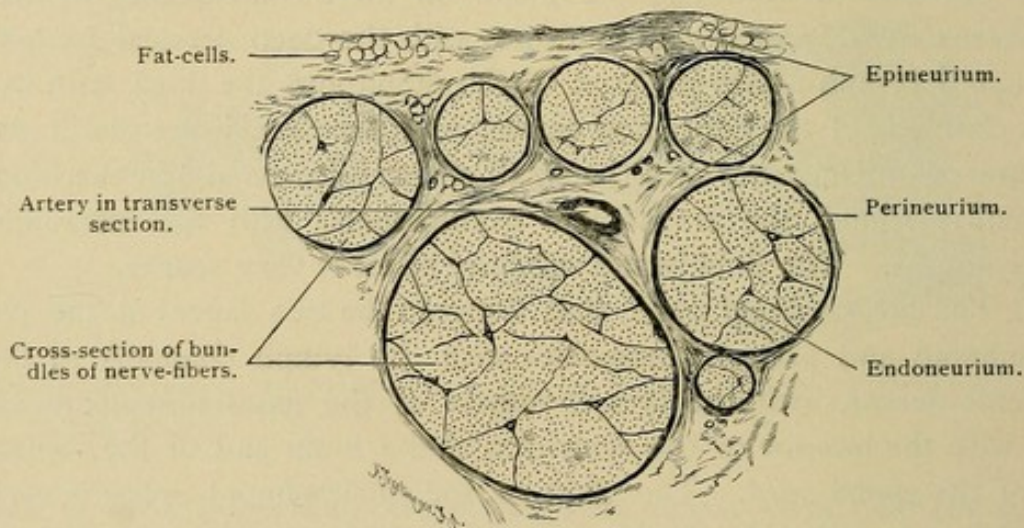


FIG. 138.—PORTION OF A CROSS-SECTION OF THE HUMAN MEDIAN NERVE. $\times 20$. Technic No. 85.

bundles; they have been named the *endoneurium* (endoneural-lamella of the nerve-bundle). Finally, delicate lamellæ from the endoneurium, the *fiber sheaths* (endoneural sheath of the nerve-fiber) corresponding to the perimysium of the single muscle-fiber, surround each individual nerve-fiber. These sheaths are in direct connection with processes of the dura and the pia. The perineurium and the endoneural-lamellæ* consist not only of connective-tissue fibers, but also of elastic fibers and of a variable number of concentric membranes; each membrane is formed by a simple layer of flattened connective-tissue cells, the outlines of which can be

* The two together form the "Henle's sheath" of authors.

demonstrated by silver staining. The fiber-sheaths, in addition to delicate connective-tissue bundles, also consist of plate-like cells. Divisions (namely collaterals) of the peripheral nerve-fibers do not occur during their course, but at the periphery; on the other hand, not infrequently a variable, large number of nerve-fibers branch from one bundle of nerve-fibers to join another bundle. The result of this is an acute-angled plexus of fiber-bundles.

The *sympathetic nerves* are partly more white and partly more gray in color, depending upon the greater or lesser number of fine medullated nerve-fibers present; for example, the splanchnic nerves contain many medullated nerve-fibers, while the gray sympathetic nerves, for example the branches of the abdominal and pelvic plexuses, contain only a very few of the thinnest medullated and, on the other hand, numerous nonmedullated nerve-fibers. One portion of the medullated nerve-

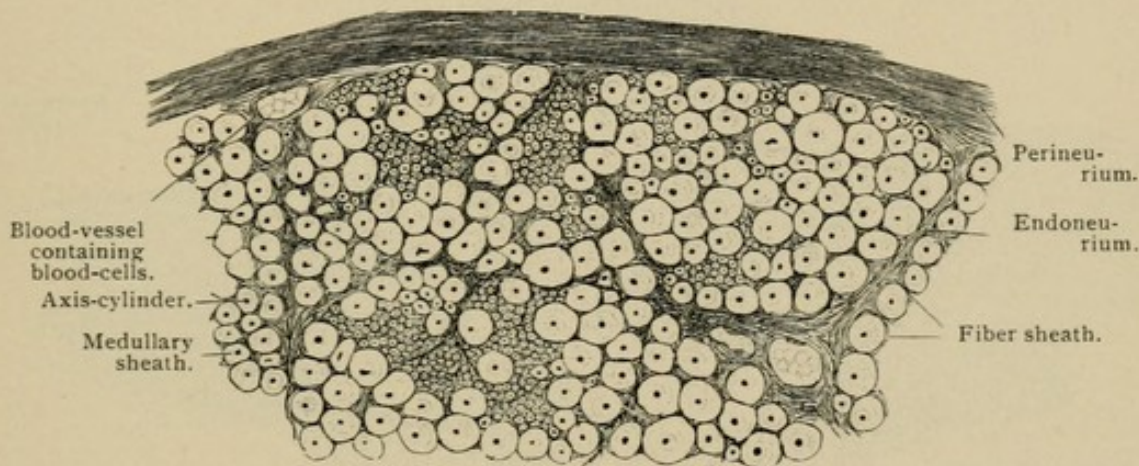


FIG. 139.—PORTION OF A CROSS-SECTION OF THE HUMAN MEDIAN NERVE. $\times 220$. Technic No. 85.

fibers are continuations of the spinal nerves, another portion are nerve-processes of sympathetic nerve-cells; long dendrites of sympathetic nerve-cells occasionally occur in the course of the sympathetic nerves (*cf.* p. 219). The nerve-fibers are grouped together and held in bundles by connective tissue.

The *blood-vessels* run lengthwise within the epineurium and form capillary networks with elongated meshes; they are supported by the perineurium and the endoneurium.

The *lymph channels* occur in the capillary clefts between the lamellæ of the perineurium and between the individual nerve-fibers, so that each nerve-fiber is bathed in lymph. They are in communication with the subdural and the subarachnoid space, but not with the lymph-vessels accompanying the nerve.

THE GANGLIA.

Ganglia are groups of nerve-cells intercalated in the course of the peripheral nerves. Usually they are macroscopically visible. All ganglia consist of small bundles of nerve-fibers, between which lie ganglion cells, partly arranged in rounded groups, partly in longitudinal rows (Fig. 140). A connective-tissue capsule, an extension of the perineurium, envelops the outer surface of the ganglion and sends into the interior processes for the support of the nerve-fibers and the ganglion cells. The ganglia are very rich in blood-vessels, the capillaries of which surround the individual cells. Respecting the minute structure, differences exist between the spinal ganglia and the sympathetic ganglia.

The *spinal ganglia* possess large, spherical, often pigmented nerve-

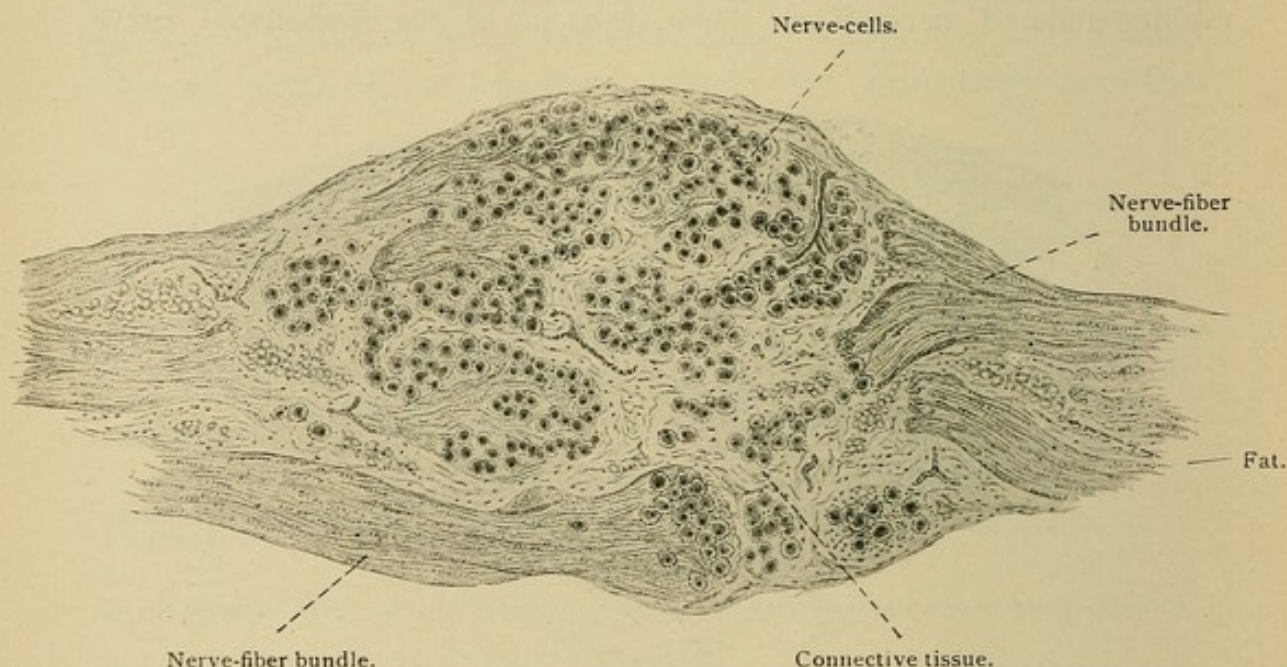


FIG. 140.—LONGITUDINAL SECTION OF A SPINAL GANGLION OF A CALF. $\times 20$. (Schaper.)
Technic No. 86.

cells, the vesicular nucleus of which encloses a large nucleolus. Each cell is enveloped in a "nucleated capsule" (Fig. 141), which consists of flat, concentrically stratified connective-tissue cells and is prolonged on to the process of the ganglion cell as the fiber-sheath. In embryonal life the majority of the nerve-cells of the spinal ganglia are bipolar, the processes springing from opposite poles of the cell. In the course of development the portion of the cell-body from which the processes arise becomes attenuated, stalkwise, to one fiber, one process, from which the primordial two processes proceed; thus the cell becomes unipolar.*

* Isolated bipolar cells also occur in the adult; they may be regarded as elements arrested in their development.

Recent investigations made on domesticated mammals, by the methylene-blue method (p. 42), teach us that we must distinguish different cell-types :

Type I. (*a*) Large, round nerve-cells ; their nerve-process, spirally wound in its initial portion, arises from a conical eminence of the protoplasm and near to its exit from the cell receives a medullary sheath and a neurilemma ; after sending off several delicate collaterals it divides after a shorter or longer interval, uniformly at the niveau of a node of Ranvier, T- or Y-shape (p. 114) in two (Fig. 142, 1) or three (Fig. 142, 3) branches.* One of these, the cellulipetal branch, passes as the axis-

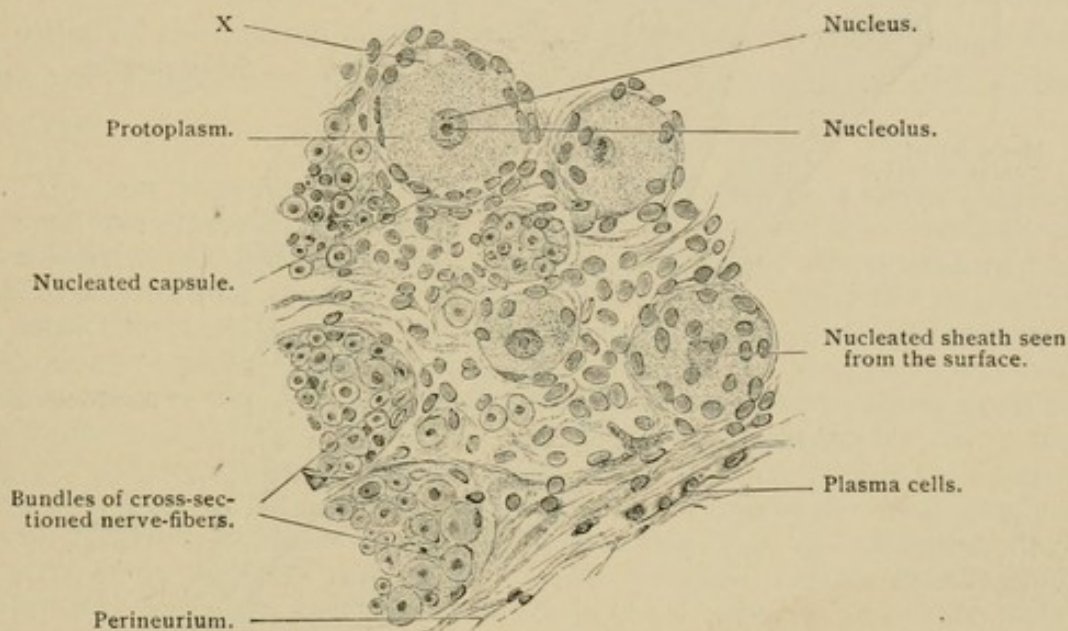


FIG. 141.—FROM A CROSS-SECTION OF THE GASSERIAN GANGLION OF MAN. $\times 240$. The cell-processes cannot be seen. At X the protoplasm of the ganglion cell has retracted and simulates a process. In the axis of the transversely cut nerve-fibers the axis-cylinders are seen in section. Technic No. 86.

cylinder of a sensory fiber to the periphery of the body ; the other, the cellulifugal, usually slighter branch runs as a constituent of a dorsal nerve-root to the spinal cord, in the gray substance of which it terminates in a free ramification (p. 193). Thus in a measure each spinal ganglion cell, by its yet undivided process, is intercalated in the course of its sensory nerve-fiber.

(*b*) Small, round nerve-cells (Fig. 142, 4), that are distinguished from the large cells only by their delicate nerve-process, that receives no medullary sheath or only scattered traces of such a cover.

* Each of the two branches may divide once again ; of the twigs proceeding from the peripheral branch the one runs in the ventral, the other in the dorsal ramus of the spinal nerve (Fig. 142, 2).

Type II. Spherical, unipolar cells, the process of which, after receiving a medullary sheath and a neurilemma, repeatedly divides into a large number of medullated nerves (Fig. 142, 6). These after losing their medullary sheath approach the cells of the first type and form a pericapsular plexus lying upon the nucleated capsule of these cells;

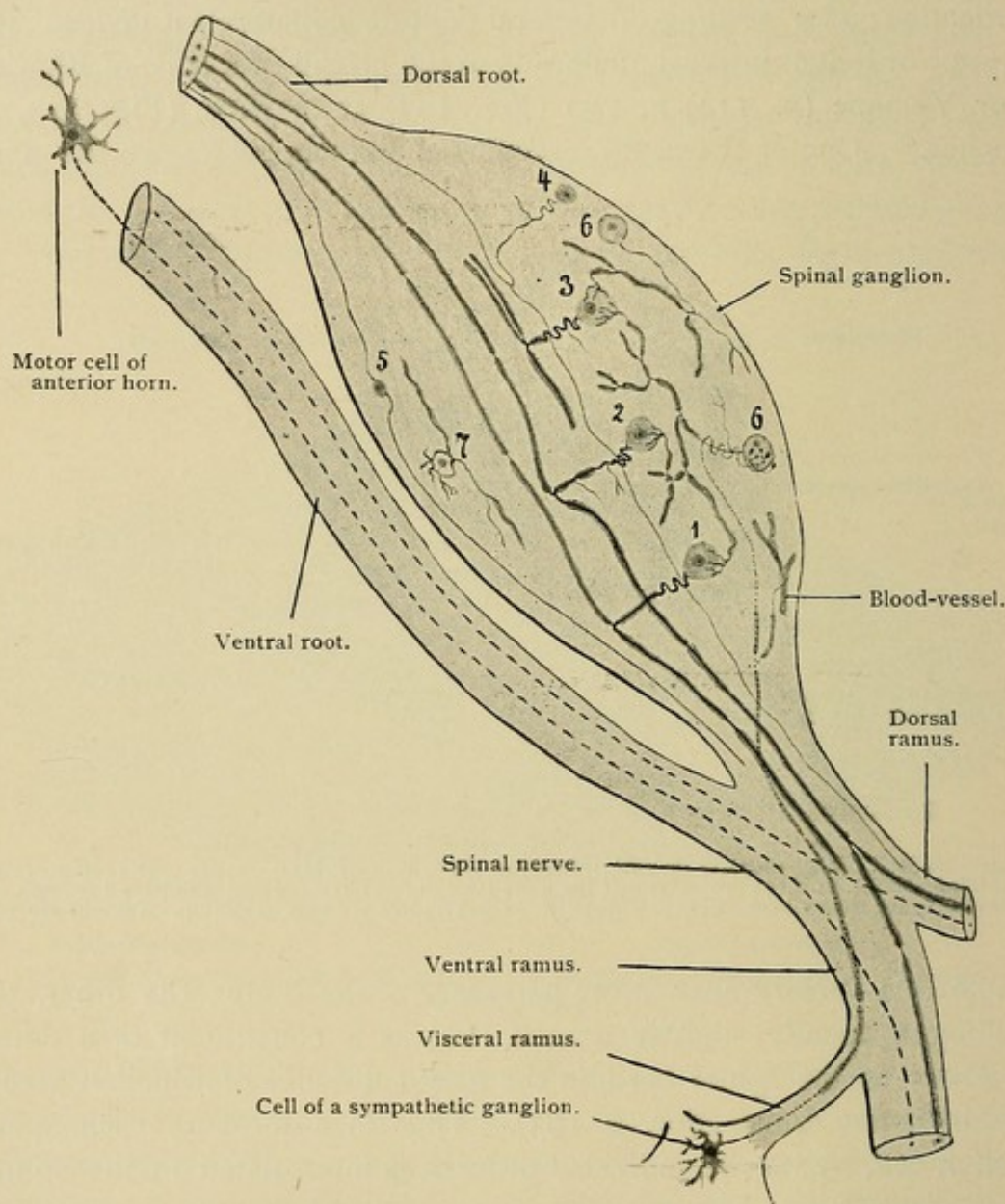


Fig. 142.—SCHEME OF THE NERVOUS ELEMENTS OF A SPINAL GANGLION, PROJECTED FROM PREPARATIONS PRODUCED AFTER TECHNIC No. 186 b. The sensory fibers are represented by continuous lines, the sympathetic fibers by dotted lines, the motor fibers by a linear series of dashes. The medullary sheaths of the motor fibers of the ventral root have not been drawn.

from this arise delicate branches which pierce the capsule and resolve into a pericellular plexus. Each cell of the first type is enveloped in the plexuses of several cells of the second type (Fig. 142, 3). The number of the cells of the second type is relatively insignificant.

Possibly to be regarded as modifications of the second type are *multipolar* nerve-cells, that besides short dendrites possess a centrally and a peripherally running nerve-process (Fig. 142, 7). These processes appear to become medullated nerves, that however do not pass beyond the territory of the ganglion. The multipolar cells are present only in very limited number.

The cells of the second type, possibly also of the first type, are wrapped in varicose pericapsular and pericellular networks of fibers, which are the nonmedullated endings of medullated nerve-fibers coming from a few sympathetic nerve-cells of the sympathetic ganglia. Branches of these fibers also go to the blood-vessels (Fig. 142). Thus through the cells of the second type a small number of entering sympathetic fibers are brought into close relation with a large number of cells of the first type.

The fact ascertained by careful enumeration that in a spinal ganglion there are many more ganglion cells than there are cross-sections of medullated nerve-fibers in the dorsal nerve-root, long ago permitted the conjecture that further complications are hidden in the spinal ganglion. That this conjecture was correct is testified by the brilliant discovery of ganglion cells of the second type, the nerve-process of which does not pass out of the ganglion; but their small number is not sufficient to explain the discrepancy,—of six ganglion cells to one medullated nerve-fiber—even if to them we add the few multipolar ganglion cells regarding the course of the nerves of which little is yet known. This gap is filled by the new fact that the nerve-processes of the small ganglion cells of the first type are chiefly nonmedullated (Fig. 142, 4). Whether there are nerve-fibers that pass through the spinal ganglion without entering into relation with its cells is uncertain. In young chick embryos such fibers, coming from cells of the anterior horns, have been demonstrated; but they have not been found in any mammal.

Other ganglia possessing the same structure as the spinal ganglia are the gasserian, the jugular, the plexus nodosus of the vagus, the petrosal and the geniculate.* The ganglia of the auditory nerve (ganglia nervi cochleae et nervi vestibuli) contain bipolar ganglion cells.

The *sympathetic ganglia* consist of smaller, often pigmented, uni- or dinucleated ganglion cells and of nerve-fibers.

The ganglion cells are multipolar † and fall into three types :

* The latter is said to be partly a sympathetic ganglion.

† Among the cells of type ii are some that have all processes springing from one or from both poles, in the form of one or two bunches correspondingly; such cells were not quite appropriately named uni- or bipolar cells. However, the sympathetic ganglion cells of fishes are actually bipolar; in the amphibia ganglion cells occur of which the single process, subsequently divided T-shape, is embraced by a "spiral fiber" that, ending in a free ramification, weaves itself about the ganglion cell in a fashion resembling the enveloping plexus of the cells of the spinal ganglia.

Type I (Fig. 143, 1). Rounded oval, occasionally flat cells, with numerous short, often flattened dendrites, the ramifications of which, beset with thorn-like excrescences, impart to the cell a quite characteristic appearance. Their nerve-process, provided with very delicate collaterals, emerges from the ganglion as a nonmedullated nerve-fiber

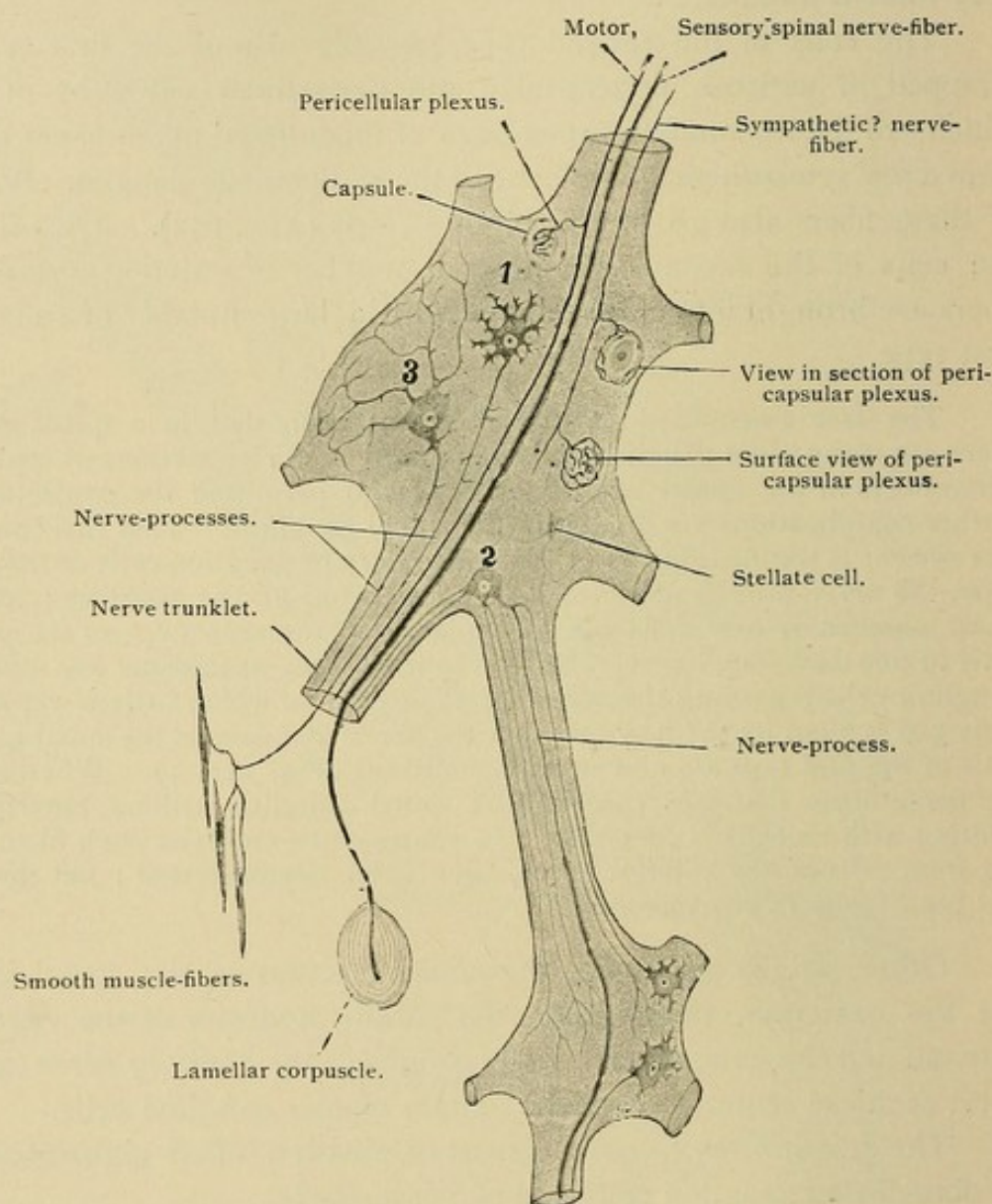


FIG. 143.—SCHEME OF THE ELEMENTS OF TWO SYMPATHETIC GANGLIA, prepared after Technic No. 186 b. 1, 2, 3. Cells of the first, second, and third types.

in a nerve trunklet and finally terminates on smooth muscle-fibers (*cf.* p. 227). The large majority of the sympathetic ganglion cells is composed of these *motor* elements.

Type II (Fig. 143, 2). Polygonal cells, the dendrites of which do not like those of types i and iii remain confined to their ganglion, but always as very slender fibers closely resembling nerve-processes enclosed

in nerve trunklets extend into neighbor ganglia.* Their nerve-process enters with the dendrites into a nerve trunklet, either as a nonmedullated or as a medullated nerve-fiber,† of which the manner of termination is not yet definitely established. The cells of type ii are regarded as sensory elements.

Type III (Fig. 143, 3). The cells resemble those of type ii; their long, ramifying dendrites penetrate between the neighboring nerve-cells through to the periphery of the ganglion, where they form a "general‡ peripheral plexus." Their nerve-process enters into a nerve trunklet as a nonmedullated fiber; the termination is unknown. The number of the cells of type iii is insignificant; they are entirely wanting in the smaller ganglia.

All sympathetic ganglion cells are enveloped in a capsule ("nucleated sheath," Fig. 141), that continues on the nerve-process and also on the coarser dendrites.

The sympathetic ganglia contain, besides these nervous cells, chromaffine cells (p. 220) and many stellate cells provided with long outrunners (Fig. 143), that mostly are applied to the walls of the blood- and lymph-vessels; such cells occur also in many other localities of the body, for example in the intestinal villi, in glands, in the tongue, and very probably are of connective-tissue nature.

The nerve-fibers of the sympathetic ganglia are :

(a) Spinal nerve-fibers, medullated, that simply traverse the ganglion or, after loss of their medullary sheath, with a relatively coarse terminal ramification form a pericellular plexus around the cells (probably of type i). The collaterals of such nerves behave in the same way (Fig. 143). Also sensory nerve-fibers coming from end-organs (lamellar corpuscles, p. 222) pass through the ganglion (Fig. 143).

(b) Nonmedullated nerve-fibers, that with their delicate, varicose terminal ramifications form a pericapsular plexus. It is conjectured that these fibers are sympathetic in nature.

The ciliary, sphenopalatine, otic, and submaxillary ganglia belong to the sympathetic ganglia.

* See also the Nerves of the Stomach and of the Intestines, (plexus of Meissner).

† The medullary sheath does not appear until after the fiber has left the ganglion, often in very great remoteness from the cell-body; this was overlooked by some investigators; the theory they proclaimed, that all nerve-fibers originating from sympathetic cells are nonmedullated, is therefore erroneous.

‡ The expression "general" is opposed to the representation of Cajal, according to which each individual nerve-cell is enclosed in a dendrite basket, which by no means is invariably the case.

In conclusion the *paraganglia* must be considered here ; they are balls or cords of cells that originate in the embryonal anlagen of the sympathetic ganglia, and are distinguished by their staining yellow-brown when fixed in solutions of chromic acid or its salts. For this reason the cells are named *chromaffine* cells. The paraganglia occur in more or less intimate connection with the sympathetic nerve. The recently discovered, macroscopically demonstrable sympathetic accessory organs at the origin of the inferior mesenteric artery belong to the same category. Single chromaffine cells or small groups of them occur as diffuse infiltrations in the interior of sympathetic ganglia and nerves. Finally, the entire medullary substance of the adrenal bodies of the higher vertebrates consists of chromaffine cells.

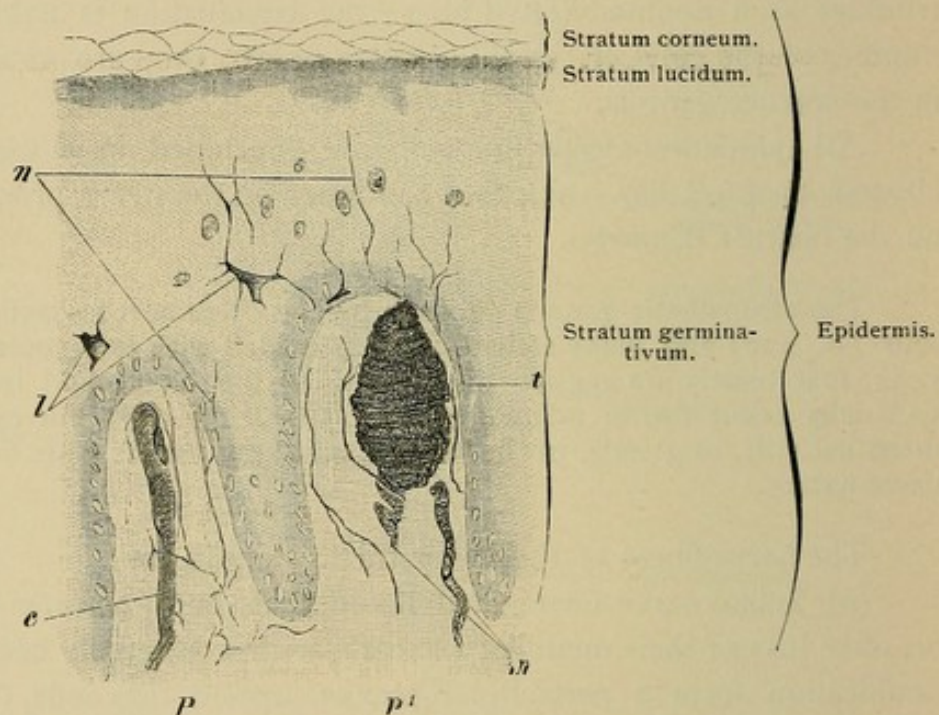


FIG. 144.—VERTICAL SECTION OF THE SKIN OF THE GREAT TOE OF A MAN TWENTY-FIVE YEARS OF AGE. $\times 200$. The cell-nuclei of the stratum germinativum are distinct only in the deepest layer. *l*, Cells of Langerhans; *n*, intraepithelial nerve-fibers. *P*, *P*¹, two papillae of the corium; *P* contains a capillary loop, *c*, of which only one limb is visible; *P*¹ contains a tactile corpuscle, *t*, with two approaching medullated nerve-fibers, *m*. Both papillae contain nonmedullated nerve-fibers. Technic No. 88.

THE PERIPHERAL NERVE-ENDINGS.

TERMINATIONS OF THE SENSORY NERVES.

The peripheral terminal branches of the sensory nerves either are distributed naked, as *free endings*, or they are enclosed by epithelial or connective-tissue cells, with which they form special endings, the *terminal corpuscles*.*

The *free-nerve endings* occur in this manner. The nerve-fibers lose their medullated sheath, divide repeatedly, and form a plexus of primitive fibrils that terminate in pointed or club-shaped ends. These

*The nerve-endings of the *neuro-epithelial cells* are described in the chapters on the special-sense organs.

endings chiefly occur in stratified epithelium. They have been demonstrated with certainty in the cornea (see the Visual Organ), in the oral mucous membrane (see the Gustatory Organ), and in the deeper strata of the epidermis. In the latter cells provided with long, branched processes, the *cells of Langerhans* (Fig. 144), occur; these were formerly regarded as migrated wandering cells (p. 93) from the corium and it is possible that a few of them may really have such an origin; but the majority are transformations of degenerating ordinary epithelial cells, for all the transitional forms, from the typical epithelial cells to the stellate bodies in question, may be found.

Sensory nerves have been found also in the muscles. They divide dichotomously into many nonmedullated fibers, provided with a neurilemma, and terminate in delicate, slender, free fibrils between the muscle-fibers (Fig. 153).

The *terminal corpuscles* may be divided into two main varieties: the *tactile cells* and the *end-bulbs*. In the tactile cells the nerve-fiber terminates in relation with one or two cells; in end-bulbs it terminates in the interior of a finely granular body, the so-called inner bulb.

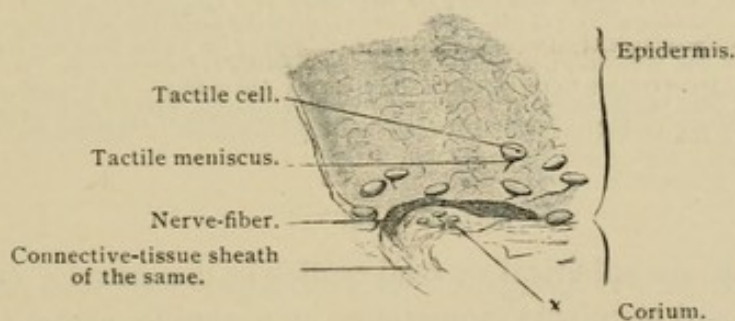


FIG. 145.—FROM A VERTICAL SECTION OF THE SKIN OF THE GREAT TOE OF A MAN TWENTY-FIVE YEARS OLD. $\times 240$. The outlines of the cells and the nuclei of the epidermis can only be indistinctly seen. *x*. Tactile cells in the corium, resting upon the ramifications of a delicate nerve-fiber. Technic No. 88.

I. TACTILE CELLS.

The tactile cells may be either *simple* or *compound*. (*a*) The *simple tactile cells* are oval, nucleated bodies measuring from 6 to 12 μ (Fig. 145), which occur in the deepest strata of the epidermis and in the outer root-sheath of the hairs or in the adjacent portions of the corium. The tactile cells rest on the *tactile meniscus*, a crescentic expansion of a nonmedullated nerve-fiber.

(*b*) The *compound tactile cells* (Grandry's and Merkel's corpuscles) consist of two or more discoid cells, of which each is larger than a simple tactile cell (15 μ high and 50 μ broad), and contains a vesicular nucleus. A medullated nerve-fiber approaches the compound tactile cell (Fig. 146)

and the forks of the divided axis-cylinder clasp a flat disk, the *tactile disk* (*ts*), that lies between two mutually flattened discoid cells (*tz*). The nerve-fiber loses its medullated sheath at the point of entrance and the perineurium becomes fused with the connective tissue of the capsule (*h*) enveloping the tactile cells. The compound forms consisting of two tactile cells are named twin tactile cells (*B 2*), those consisting of three or four tactile cells, "simple tactile corpuscles" (*A, B 1*). The compound tactile cells have only been found in the skin of the beak and of the tongue of birds, especially in web-footed birds; they are almost exclusively situated in the uppermost strata of the corium.

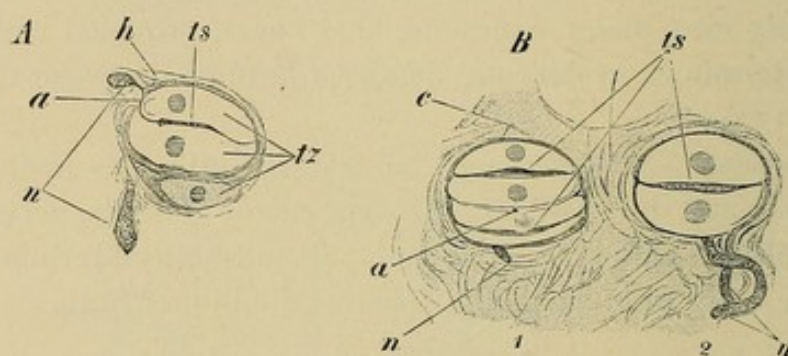


FIG. 146.—FROM VERTICAL SECTIONS OF THE CERE OF THE BEAK OF A GOOSE. $\times 240$. *A*. Compound tactile cell (simple tactile corpuscle), cut parallel to the course of the entering nerve-fiber: *n*, medullated nerve-fiber only partially met by the section; *a*, axis-cylinder: its forks here, in profile, are invisible; *ts*, tactile disk cut vertically; *h*, connective-tissue sheath; *tz*, tactile cells. *B*. Two compound tactile cells cut transversely to the plane of the entering nerve-fiber. 1. "Simple tactile corpuscle," consisting of four tactile cells; 2. twin tactile cells; *ts*, tactile disks; *a*, axis-cylinders in transverse section, before dividing; *n*, medullated nerve-fibers; *c*, corium. Technic No. 89.

2. END-BULBS.

The end-bulbs are spherical or oval bodies in the interior of which nerve-fibers enter and terminate, sometimes in a simple, sometimes in a branched ending. There are various forms of end-bulbs.

(a) The so-called *cylindrical end-bulbs*, the simplest form (Fig. 147), chiefly consist of a modified extension of the entering nerve-fiber and comprise three parts,—the axis-cylinder, the inner bulb, and the capsule. The *capsule* is composed of flattened connective-tissue cells, the continuation of the fiber-sheath (Fig. 139). The *inner bulb* is a finely granular mass which exhibits concentric stratification and has a few nuclei at the periphery. The nerve-fiber loses its medullary sheath on entering the end-bulb, in which the *axis-cylinder* ascends as a flat band and terminates near the upper pole in a free rounded or club-shaped ending. The cylindrical end-bulbs are found in the tunica propria of mucous membranes; for example, in the scleral conjunctiva of mammals and in the oral mucous membrane.

(b) The *lamellar corpuscles* (Vater, Pacini) are transparent, elliptical

structures, from 2 to 4.5 mm. long and 1 to 2 mm. thick, and like the cylindrical end-bulbs consist of a capsule, an inner bulb, and an axis-cylinder. The *capsule* consists of a large number of concentric capsules, one within the other, of which each is separated from its neighbors by a simple layer of flat connective-tissue cells. Each contains fluid and connective-tissue fibers running longitudinally and transversely. Like the capsule of the cylindrical end-bulbs, so these capsules

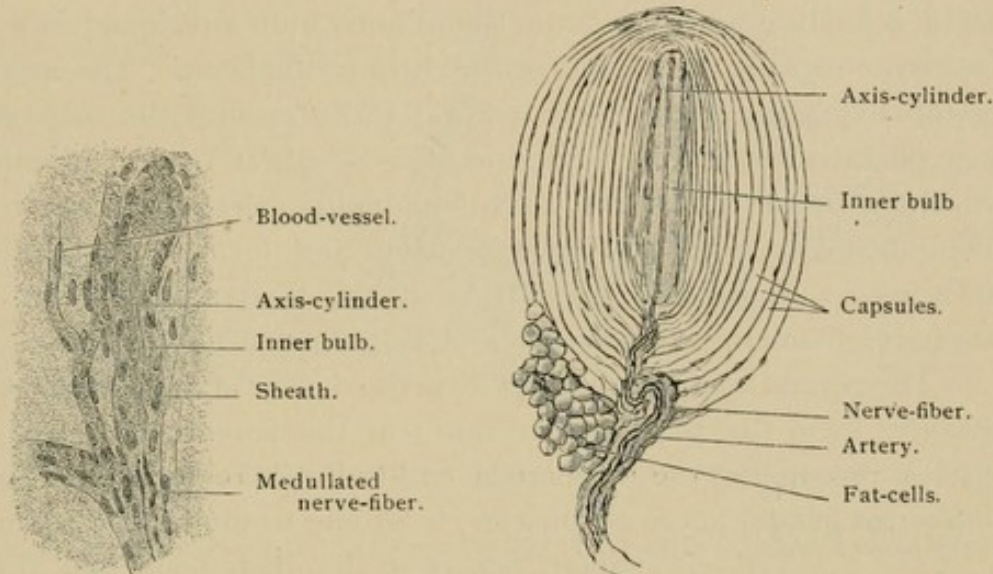


FIG. 147.—CYLINDRICAL END-BULB FROM THE CONJUNCTIVA OF A CALF. $\times 240$. Technic No. 90.

FIG. 148.—SMALL LAMELLAR CORPUSCLE FROM THE MESENTERY OF A CAT. $\times 50$. The cells lining the capsules can be recognized by their shaded nuclei. The medulla of the nerve-fiber may be traced to the inner bulb. Technic No. 91.

originate from the connective-tissue sheath of the entering nerve-fiber. The capsules are the smaller the nearer the inner bulb they lie. At the pole opposite the entrance of the nerve they are not seldom connected by a cord running in the direction of the inner bulb, the *interlamellar ligament*. The inner bulb is like that of the cylindrical end-bulbs; in its axis runs a thick axis-cylinder, which terminates in a simple or forked end, enveloped in the delicate end ramification of a second thin axis-cylinder. The latter is visible only after methylene blue staining. A small artery accompanies the nerve-fiber into the interior of the corpuscle, which breaks up into a capillary network lying between the peripheral lamellæ of the capsule.

The lamellar corpuscles partly occur in superficial situations (abundantly in the subcutaneous connective tissue of the palm of the hand and the sole of the foot, more sparingly in other localities of the skin, on the nipples, in the territory of the pudendal nerve); partly in deeper situations (in the vicinity of the joints, on the nerves of the periosteum and

the bones, in tendons and their sheaths, in fasciæ, in the mesentery, in the neighborhood of the pancreas, and in different parts of the male genital organs of mammals). They transmit simple pressure sensations.

The corpuscles of Herbst and Key-Retzius, occurring in birds, are also lamellar corpuscles; they only differ in being much smaller and in possessing a double row of longitudinally disposed nuclei in the inner bulb.

(c) The *genital nerve corpuscles* of the lower mammals and of man are spherical or oval forms (from 0.06 mm. to 0.4 mm. long), and consist of a finely granular, nonnucleated inner bulb enveloped in a connective-tissue capsule containing cells rich in protoplasm. The approaching medullated nerve-fibers make several turns around the corpuscle, lose their medulla and divide, while the fiber-sheath and the neurilemma pass into the capsule; the naked axis-cylinders penetrate the inner bulb at different points, undergo rapid division and form a dense plexus of fibrils with varicose enlargements.* Each plexus is connected by delicate nerve filaments with plexuses of neighbor corpuscles.

The genital nerve corpuscles lie in the depths of the corium at various distances from the papillary stratum; in the papillæ only smaller corpuscles, resembling the "spherical end-bulbs," are found. The largest number of genital nerve corpuscles, from one to four to the square millimeter, occurs in the glans penis and in the clitoris. The so-called *spherical end-bulbs* (in reality they are sometimes round, sometimes oval) have a similar structure; they are found in the conjunctiva and in the adjoining portions of the cornea of man, and possess a greatest diameter of 0.02 to 0.1 mm. The *articular nerve corpuscles* belong to the same category.



FIG. 149.—TACTILE CORPUSCLE FROM A PERPENDICULAR SECTION OF THE GREAT TOE OF A MAN TWENTY-FIVE YEARS OLD. $\times 560$. *n*, Medullated nerve-fibers; *e*, varicosities; *h*, connective-tissue sheath. The nuclei are invisible. Technic No. 88.

(d) The *tactile corpuscles* (Wagner's and Meissner's corpuscles) are elliptical structures, from 40 to 100 μ long and 30 to 60 μ broad, which are characterized by cross-markings (Fig. 149). They possess a connective-tissue capsule (Fig. 149, *h*) with flattened cells, the boundaries of which, as well as their transversely placed nuclei, produce the cross-striations just mentioned.

One or two medullated nerve-fibers approach each tactile corpuscle (Fig. 149, *n*), make transverse tours encircling the lower pole of the corpuscle, part with their neurilemma and fiber-sheath,

* In imperfect staining the varicosities simulate club-shaped endings.

which blend with the tissue of the capsule, then lose their medullary sheath, and as naked axis-cylinders enter into a granular substance corresponding to an inner bulb; there they form a complicated plexus beset with varicosities (*e*). The tactile corpuscles lie in the papillæ of the

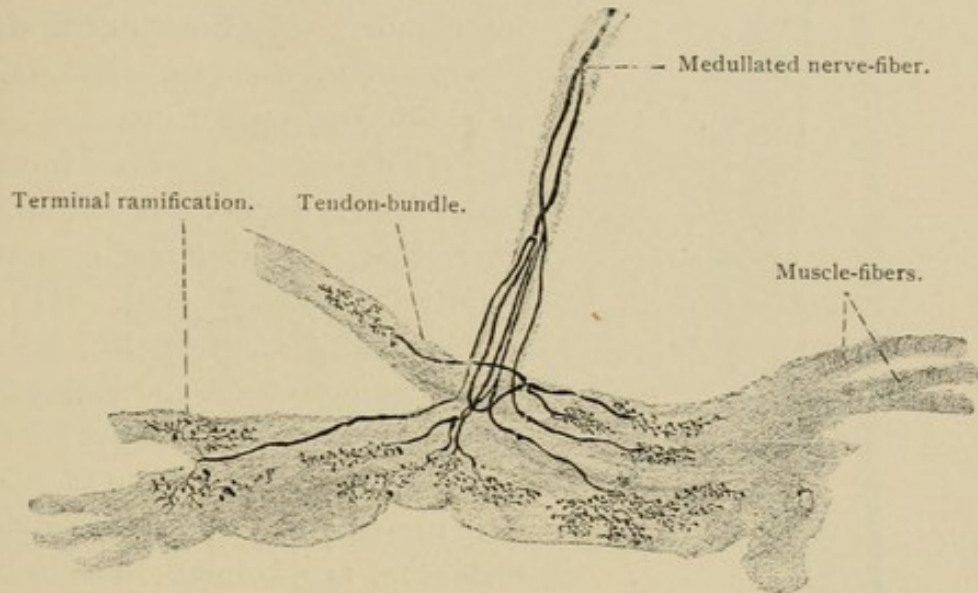


FIG. 150.—TENDON-SPINDLE OF AN ADULT CAT. $\times 80$. Technic like No. 92 *a*.

corium and are most numerous (twenty-three to one square millimeter) on the palm of the hand, on the finger-tips, and on the sole of the foot.

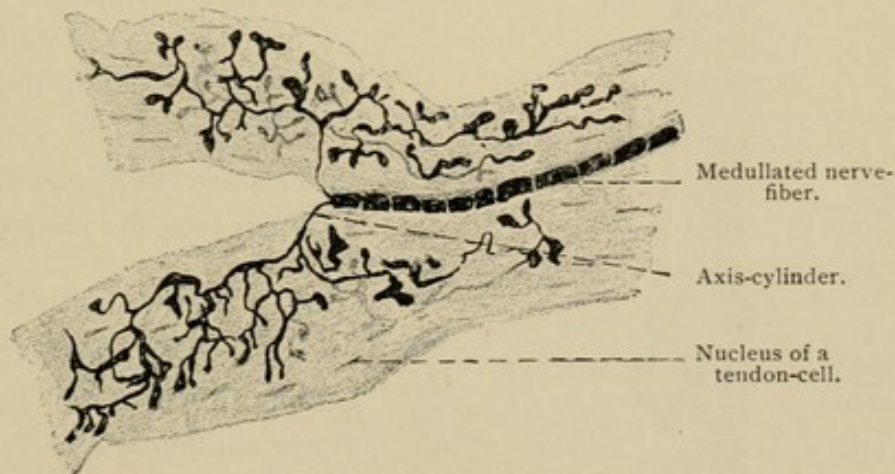


FIG. 151.—A PORTION OF THE PREPARATION OF FIGURE 150. $\times 345$.

In connection with the end-bulbs the tendon- and the muscle-spindle, as well as the terminal cylinder of Ruffini, remain to be considered.

The *tendon-spindles* are usually spindle-shaped expansions of the tendon bundles and are enveloped in a well-developed connective-tissue sheath. The one end of the spindle passes into tendon bundles, the other continues into muscle-fibers (Fig. 150). The nerve-fibers approach

the middle of the spindle, divide repeatedly, lose their medulla, and break



FIG. 152.—MUSCLE-SPINDLE OF AN ADULT CAT.
 X 135. Technic like No. 92 a.

up into a richly developed ramification with often expanded, clavate ends (Fig. 151). The tendon-spindles occur in all tendons in man, but in varying number; they transmit the sensation of extension and enter into activity in coordinated movements.

The *muscle-spindles* (muscle-buds) are clusters of delicate muscle-fibers that are enveloped in a thick perimysium sheath (Fig. 111) and provided with many nuclei; the terminal ramifications of the approaching nerves are arranged either in the form of spirals and rings (Fig. 152, above) or of blossom-like sprays with clubbed ends (Fig. 152, below). The muscle-spindles are wanting in the muscles of the eye, pharynx, esophagus, larynx, in the ischio- and bulbo-cavernosus muscles, in the diaphragm, in the mimetic facial muscles. They react to the pressure exercised by the contraction of neighboring muscles.

The *terminal cylinders* in their end ramifications resemble the tendon-spindles (see the chapter on the skin).

TERMINATIONS OF THE MOTOR NERVES.

The small nerve-trunks supplying striated muscles divide into branches, these subdivide into twigs (nerve-fiber bundles) that anastomose with one another and form a plexus, the *inter-muscular nerve-plexus*. In the territory of this plexus the medullated nerve-fibers undergo numerous divisions, so that the sum of fibers is con-

siderably increased. The twigs of the plexus divide into delicate branches, consisting of *single* nerve-fibers, each one of which finally connects with a

muscle-fiber. At the point where the nerve-fiber comes into contact with the muscle-fiber it tapers, loses its medullary sheath, the axis-cylinder

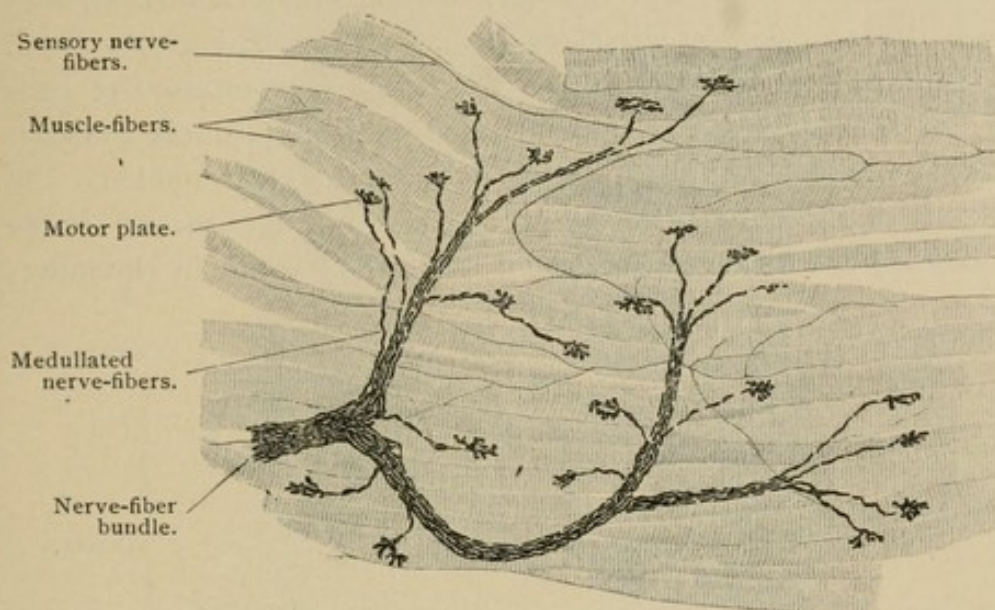


FIG. 153.—MOTOR NERVE-ENDINGS OF INTERCOSTAL MUSCLE-FIBERS OF A RABBIT. $\times 150$.
Technic No. 92 a.

breaks up into slightly tortuous terminal branches with bulbous ends (Fig. 153), which form the so-called *motor (end) plate* and rest upon a rounded, finely granular disk containing numerous vesicular nuclei. Each muscle-fiber possesses at least one motor plate; it lies upon the sarcolemma.

The nerves supplying the *smooth* muscles form a plexus from which bundles of nonmedullated nerve-fibers arise; the latter divide repeatedly and form several networks, from which finally the most delicate nerve-fibers arise. These apply themselves to the smooth muscle-fibers and often are slightly thickened at the point of contact; probably each muscle-fiber possesses a nerve-ending.

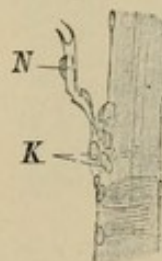


FIG. 154.—MOTOR NERVE-ENDING ON A FIBER OF AN OCULAR MUSCLE OF A RABBIT. $\times 240$.
N, Medullated nerve-fiber; *K*, nuclei of the disk. The transverse striæ are distinct only in the lower half of the muscle-fiber. Technic No. 92 b.

THE SUPRARENAL BODY.

The description of the suprarenal (adrenal) body with the organs of the nervous system is warranted by the profusion of its nervous elements, by its relations to the central nervous system, as established by experiment, as well as by the facts of comparative anatomy.

Each suprarenal body consists of a cellular parenchyma and a connective-tissue capsule, which sends delicate processes into the interior of

the organ, and contains elastic fibers only in the neighborhood of the blood-vessels in the capsule and in the medulla, but not in the cortex. The parenchyma consists of an outer stratum, the *cortex*, which surrounds an inner mass, the *medulla*, on all sides (Fig. 155).* The *cortex* is of fibrous texture, of a yellow color when fresh, and is composed of cells about $15\ \mu$ in size, rounded in shape, that possess a coarsely granular protoplasm, sometimes containing fat particles, and a clear nucleus. In the outer zone of the cortex the cells are grouped in oval masses, in the middle zone they are arranged in cylindric columns, while in the inner-

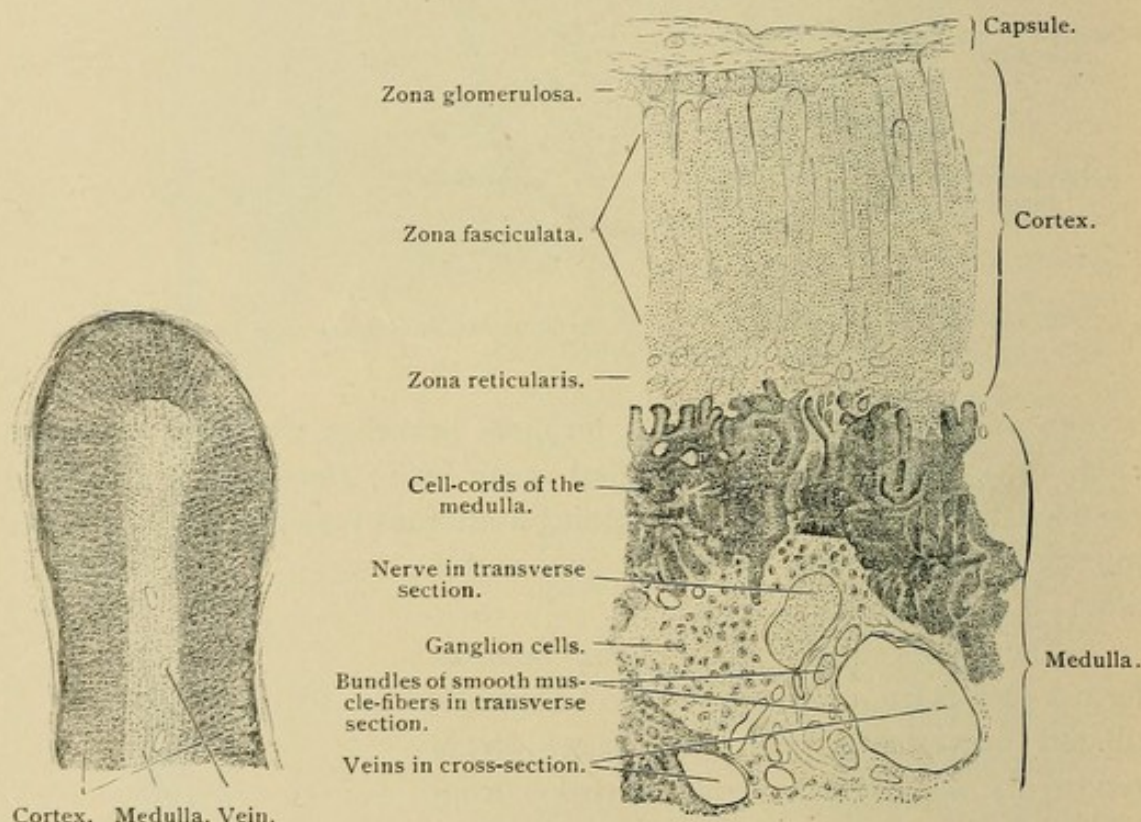


FIG. 155.—SECTION OF THE SUPRARENAL BODY OF A CHILD. $\times 15$. Technic No. 93.

FIG. 156.—SECTION OF A HUMAN SUPRARENAL BODY. $\times 50$. Technic No. 95.

most zone the cells lie irregularly scattered in a reticulum of connective tissue; the cells of the innermost zone are characterized by their pigmentation. According to the described arrangement the cortex is divided into: 1, the *zona glomerulosa*; 2, the *zona fasciculata*; 3, the *zona reticularis* (Fig. 157). The *medulla* in the fresh state is sometimes lighter, sometimes darker than the cortex and consists of chromaffine cells (p. 220), that are arranged in spherical or elliptical cords joined in an irregular network.

* The formations on the ductus deferens and in the broad ligament described as ruptured adrenals consist only of cortical substance.

The *arteries* divide in the connective-tissue capsule into numerous

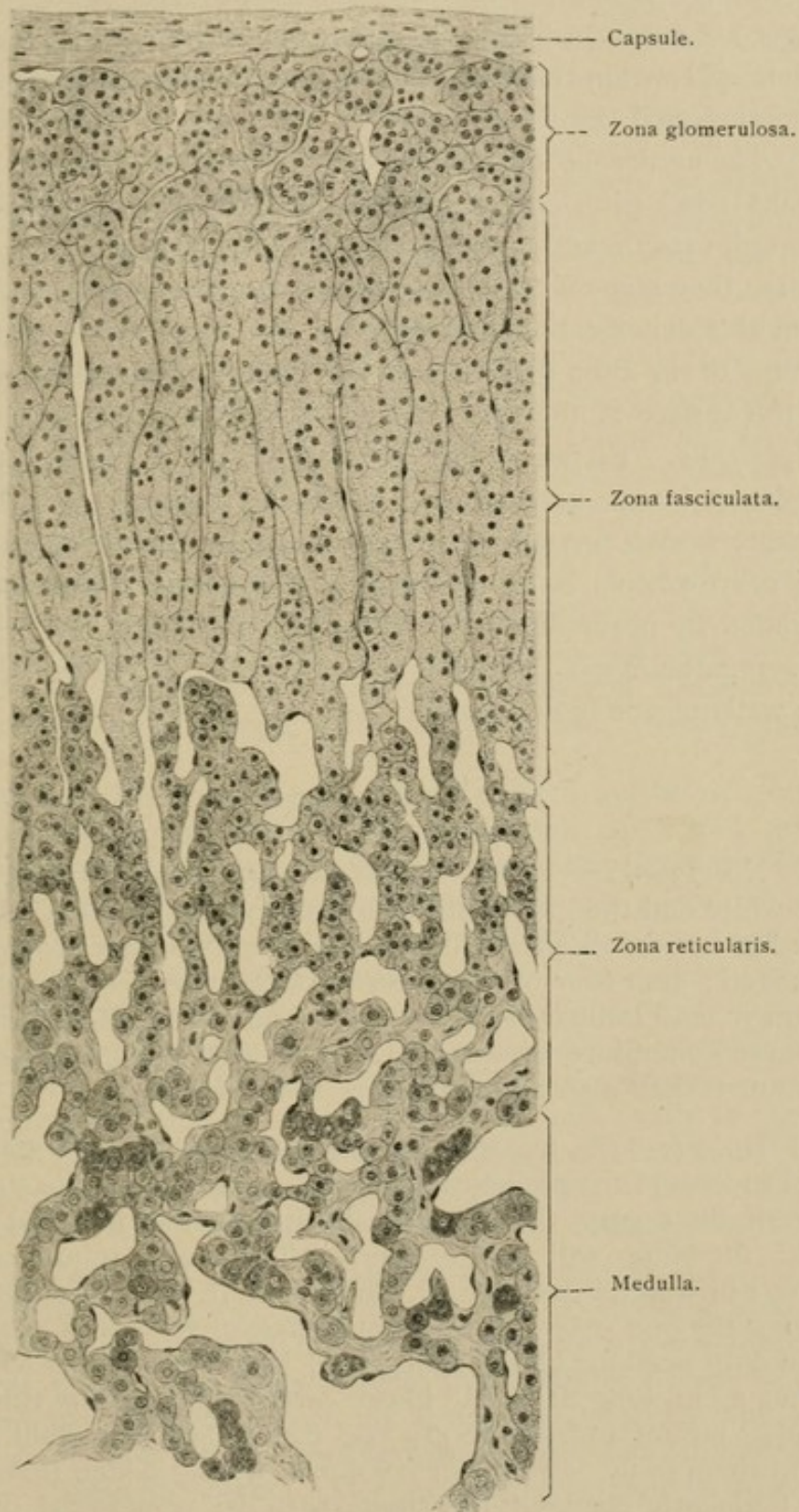


FIG. 157.—SECTION THROUGH CORTX AND MEDULLA OF THE SUPRARENAL BODY OF ADULT MAN.
× 200. (Schaper.)

small branches that penetrate the cortex and there form a long-meshed

capillary network, which passes into the medullary substance where the meshes are round. From the latter the *veins* proceed, of which the larger are accompanied by longitudinal strands of smooth muscle-fibers. While still within the medulla the veins unite and form the chief vein, the suprarenal vein.

The numerous, chiefly nonmedullated *nerves* (in man about 33 small trunks) come principally from the celiac plexus and pass with the arteries through capsule and cortex to the interior of the medulla. During their course they give off a few twigs to the capsule, that form a plexus there; from this delicate branches descend into the cortex between the cell-groups of the zona glomerulosa and the zona fasciculata, which terminate on the surface of the cell-clusters, without penetrating between the individual cells. Richer is the nerve-plexus of the zona reticularis, which originates by the branching of fibers that descend straight through the cortex; it also surrounds only cell-groups. In the medullary substance the nerve-plexus is extraordinarily dense; each individual cell is surrounded by nerve-fibers. In the medulla, seldom in the cortex, groups of sympathetic ganglion cells occur. Some of the nerves terminate in the walls of the blood-vessels.

TECHNIC.

No. 73.—*The spinal cord*.—For the study of the distribution of the white and the gray substance the spinal cord of a child should be fixed in toto in about one liter of Müller's fluid, that should be frequently changed; after four or five months thick cross-sections of the cervical, thoracic, and lumbar regions may be cut, and without further treatment mounted in dilute glycerol (p. 23), or after the customary preliminary treatment they may be mounted in xylol-balsam.

No. 74.—*The spinal cord; staining of medullated fibers after Pal*.—The success of the preparation depends especially on the state of preservation of the organ. The fresher the tissue when it is put into the fixing fluid, the better will be the result. The entire spinal cord should be placed in a large quantity of Müller's fluid, that must be changed daily during the first week and frequently thereafter. If it is desired to investigate only portions of the spinal cord, then place pieces of the fresh cord, about 2 cm. long, from the lower cervical, the middle thoracic, and the lumbar region, in from 200 to 500 c.c. of Müller's fluid or, better, suspend them in it. In four or six weeks, during which time the fluid must be frequently changed, the tissue is to be transferred directly, *without* previous washing, to 150 c.c. of 70 per cent. alcohol and on the following day to the same quantity of 90 per cent. alcohol. The bottle containing the tissue must be placed in the dark (p. 35), and the alcohol frequently changed during the first eight days. Sections may then be made.

The sections are to be placed in a capsule containing 20 c.c. of 70 per cent. alcohol and as soon as possible transferred from this to 30 c.c. of Weigert's hematoxylin to which 1 c.c. of lithium carbonate solution has been added (p. 24). In five or six hours the now very dark, untransparent sections should be transferred to 50 c.c. of distilled water plus 1 c.c. of lithium carbonate solution. In a half-hour, during which time the fluid must be changed several times, the sections will give off no more color and are then to be placed in 30 c.c. of potassium permanganate solution for differentiation (p. 24). In from one-half to three minutes they are to be washed for one minute in distilled water and then transferred to 20 c.c. of the acid mixture (p. 24). The capsule containing the acid mixture should be covered. The decolorization occurs in from ten to fifty seconds; the gray substance becomes light yellow, almost white, the white substance (the medullated nerve-fibers) appears very dark. (Very often the colored blood-cells are also stained a dark color, which is explained by the fact that they contain substances that are also contained in the myelin.) Now transfer the sections to a capsule containing 30 c.c. of distilled water and in five minutes to a second capsule containing the same quantity of fresh distilled water, in which they may remain for two or three days, the water meanwhile to be frequently changed (even running water may be used). Then they are put into 10 c.c. of alum-carmines, in which they may remain from three to fifteen hours. Mount in xylol-balsam. The alum-carmines staining may be omitted.

The foregoing directions are intended for thin, well-fixed preparations. If the sections are thick, if the tissue has lain a long time in alcohol, more time will be required for staining and reduction. Should the staining be unsuccessful, place unstained sections in Müller's fluid for twenty-four hours, wash one minute in distilled water, then stain, and satisfactory results may be obtained. Should the decolorization be insufficient, if the gray substance does not become yellowish-white, the procedure should be repeated; that is, the sections are to be again placed in distilled water one minute, then in potassium permanganate one or two minutes, then in distilled water one minute, and finally in the acid mixture. The given quantities of the permanganate solution and of the acid mixture are sufficient for only about 20 sections. If it is desired to treat more sections larger quantities of these fluids must be used.

No. 75.—*The spinal cord; staining of axis-cylinders and of cells.*—Place pieces at the most 2 cm. long in 200 c.c. of Müller's fluid, that must be changed daily during the first week and once a week thereafter. In four weeks transfer the pieces directly from Müller's fluid to about 50 c.c. of sodium carminate (1 per cent. aqueous solution), in which they should remain for three days. *During this time the bottle containing the pieces must be frequently shaken.* The stained pieces are to be washed for twenty-four hours in running water, then placed in 150 c.c. of 70 per cent. alcohol, and after five hours transferred to the same quantity of 95 per cent. alcohol. Mount the cross-sections in xylol-balsam (Fig. 122).

No. 76.—*Spinal cord, after Golgi.**—Remove the spinal cord along with the (still cartilaginous) vertebral column of newborn rats or mice and treat them according to the method described on page 45. The length of time the objects should remain in the Golgi mixture depends upon the elements it is desired to impregnate.† It requires from

Two to three days for *neuroglia cells*,

Three to five days for *nerve-cells*,

Five to seven days for *nerve-fibers* (collaterals).

Since the pieces must be used as soon as they are taken out of the silver solution only *one* piece at a time should be transferred to the absolute alcohol. Cut the sections through spinal cord *and* vertebral column.

The spinal cord of a three- or seven-day-old embryo chick furnishes still better results, but it is necessary to embed the tissue in celloidin (see Microtome Technic). The spinal cord of kittens, as well as that of human embryos from 20 to 40 cm. long, yields very useful results.

No. 77.—*The brain; staining of medullated nerve-fibers.*—Apply the method given in No. 74. If an entire human brain is to be placed in Müller's fluid, many deep incisions should be made in it and about 3 liters of the fixing fluid should be used.

No. 78.—*The brain; cells.*—Treat pieces 1 or 2 cm. square of the cerebral cortex (central convolution) and of the cerebellar cortex like

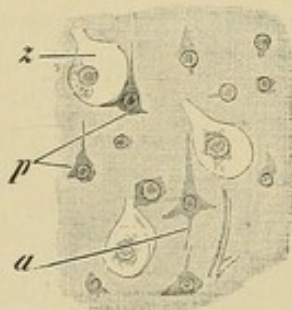


FIG. 158.—PORTION OF A SECTION OF HUMAN CEREBRAL CORTEX. $\times$ 240. *p*, Small pyramidal cells; *a*, the nerve-process of a pyramidal cell.

No. 75. In the cerebral cortex, in addition to the cell-forms described, an extremely variable number of vesicular cavities containing remnants of cells (protoplasm and nucleus) may be seen (Fig. 158, *z*); they are probably pericellular lymph-spaces, which by post-mortem alteration of the brain substance and the influence of the fixation medium have become abnormally enlarged. The sections through the cerebellar cortex must be made transverse to the long axis of the convolutions, since the ramifications of the cells of Purkinje

extend only in planes transverse to the convolution. Only a few cells of Purkinje lie in the depths of the convolutions.

No. 79.—*The brain, after Golgi.**—(*a*) For a *topographic view*, treat the brain of a newborn rat or mouse in the unopened cranium

* *Editor's remark:* The application of the *Cox-Golgi mixture*, in the manner described on p. 45, foot-note, is also highly recommended. Since it can be applied with good results to the central nervous system of *adult* animals it offers in the manipulation of the material and the preparation of the sections valuable advantages, particularly to the beginner. After the treatment with alcohol the larger pieces can be easily cut freehand without being embedded, when *thick* sections are desired.

† If the action of the mixture is too brief the central portions of the sections appear untransparent and are penetrated by abundant precipitates; if the action of the mixture is too prolonged the resulting impregnation of the elements will be unsatisfactory.

according to the method given in No. 76. The cranium may be sectioned with the brain-substance.

(b) For specimens of the cortex the brain of a mouse from eight to thirty days old is most suitable, treated with the Golgi mixture for from two to three days, or of a one- to fifteen-day-old-rabbit or a kitten under six weeks old, treated with the Golgi mixture for five days. Pieces of the brain of adults must remain in the Golgi mixture for from eight to fifteen days. Further treatment like No. 76.

No. 80.—*The cortex of the cerebellum, after Golgi.**—Remove the cerebellum from the cranium of a newborn guinea-pig (or a kitten less than six weeks old) and treat it according to the method given in No. 76. The staining of the elements of the cerebellum is more difficult to accomplish than of those of the cerebrum and the spinal cord. Failures are frequent. The sections should be principally made vertically to the long axis of the convolutions. (For embedding, see Microtome Technic.)

No. 81.—*Hypophysis cerebri*.—Treat like No. 86.

No. 82.—*Brain-sand (acervulus cerebri)*.—Tease the epiphysis in a drop of salt solution. If much brain-sand is present a grating sound will be heard on teasing and the larger concretions can be seen by the unaided eye. Examine with the low power, without a cover-glass (Fig. 135); the granules are not always round, but often oval and dentated; occasionally the irregularity of the surface is indistinct, because the granules are enveloped in concentrically arranged connective-tissue fibers. Push aside the larger granules with a needle, cover a few of the smaller ones with a cover-glass and treat with 2 or 3 drops of hydrochloric acid (p. 53). Bubbles of gas develop and the sharp outlines of the granules disappear.

No. 83.—*Corpuscula amylacea*.—Select the brains of elderly individuals. With a scalpel scrape the mesial surface—that directed toward the third ventricle—of the optic thalamus and distribute the scrapings with a needle in a drop of salt solution; apply a cover-glass. The corpuscles are easily found, and are recognized by their bluish-green color and their stratification (Fig. 136, *a*). They must not be confused with drops of extruded myelin (*b*), which are always clear and have only a double contour. In addition there are found in such preparations numerous red blood corpuscles, ependymal cells (*d*), medullated nerve-fibers, differing in thickness, and ganglion cells; the latter are very pale and often can be detected only by their pigmentation (*f*). Human brains, even though not absolutely fresh, are still useful.

No. 84.—(*a*) Spread out a piece 1 cm. long of the *choroid plexus* in a drop of salt solution and apply a cover-glass. The convoluted red blood-vessels and the epithelium of the plexus can be seen.

* For the application of the Cox-Golgi mixture see p. 45 and p. 232, remark *.

(b) Very pretty permanent preparations can be obtained as follows: Carefully spread out a little piece of the plexus in salt solution; if good fields are visible with the low power let the salt solution flow off and add a few drops of Zenker's fluid (p. 21); then apply a cover-glass, at the edge of which place a little more of the Zenker's fluid. After thirty minutes displace this fluid by distilled water, and after another thirty minutes the water by 50 per cent. alcohol to which a few drops of tincture of iodine have been added. In fifteen minutes take off the cover-glass and transfer the now fixed preparation to a watch-glass with fresh 50 per cent. iodine-alcohol, to which, in case it becomes rapidly decolorized, tincture of iodine is to be added. In from fifteen to thirty minutes transfer the object to pure 70 per cent. alcohol, and after about twelve hours stain it with hematoxylin and eosin (p. 39, 3 b) and mount in xylol-balsam (p. 50).

No. 85.—*Transverse sections of nerve-fiber bundles.*—Treat a piece of nerve, *e. g.* the sciatic, if possible of man, that possesses a well-developed endoneurium, according to the method given in No. 34, p. 123.

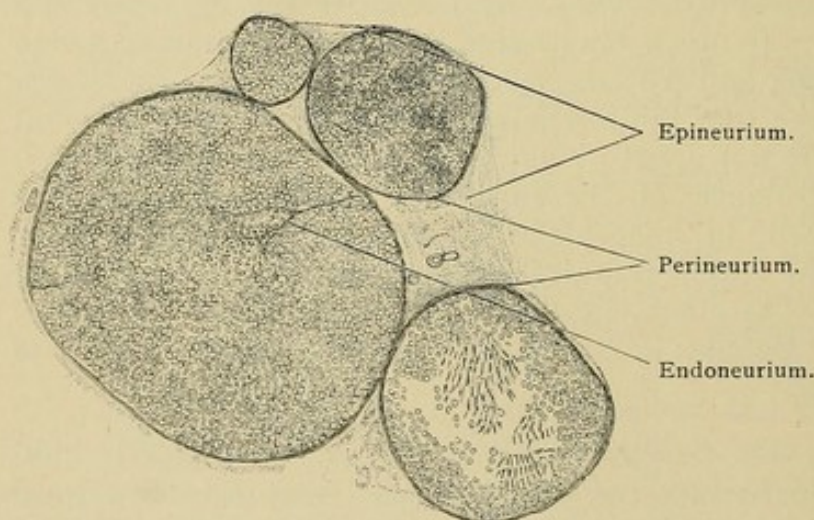


FIG. 159.—FROM A TRANSVERSE SECTION OF A PERIPHERAL (SPINAL) NERVE OF A RABBIT. $\times 50$. In the lower bundle, on the right, some of the transverse sections of nerve-fibers have fallen out, others are lying on their side, as a consequence of pressure. In the rabbit the endoneurium is only slightly developed.

Place it for six days in a 0.1 per cent. solution of chromic acid, then wash it for from three to four hours in running water, and harden it in gradually strengthened alcohols. When the hardening is completed cut thin sections with a sharp razor. It is advisable to embed the tissue in liver, better still, in elder-pith or in

the pith of the sunflower. For this purpose make a hole in the dry elder-pith with a needle and then carefully insert the nerve. Place the whole for about a half hour in water; in this pith swells and firmly clasps the nerve. Stain the sections in picrocarmine and mount in glycerol. The length of time required for staining varies greatly. The sections must be very carefully handled and pressure with the cover-glass must be scrupulously avoided, lest the sections of the fibers, which are not disks but short cylinders, be turned on their sides and not a fiber in section be seen. If successful the section will show the somewhat shrunken axis-cylinder, resembling a red nucleus, surrounded by the yellow medulla, which is enclosed by the reddish neurilemma and the fiber-sheath.

The cross-section of the nerve-fiber has been compared to a picture of the sun (*Sonnenbildchenfigur*) (Fig. 159).

No. 86.—*Spinal ganglia*.—These are very inaccessible. Therefore remove the Gasserian ganglion from the depression in which it is lodged, on the anterior surface of the petrous portion of the temporal bone, and place it in about 100 c.c. of Müller's fluid for fixation. After four weeks wash it for three hours in running water and harden it in 50 c.c. of gradually strengthened alcohols (p. 35). Cut the thinnest possible transverse and longitudinal sections; stain them thirty seconds in hematoxylin, then from two to five minutes in eosin (p. 39, 3 *b*), and mount in xylol-balsam. The ganglion cells are pale red; the axis-cylinders deep red; the medullary sheaths brownish; the nuclei blue (Fig. 141). If the section is not sufficiently thin the large number of deeply-stained nuclei will render it difficult to see the other structures. For this reason it is better to stain the thick sections in picrocarmine for two or three days and mount them in xylol-balsam. The nuclei are then not so intensely stained. Occasionally the protoplasm of the ganglion cell contracts and thus acquires a stellate outline (Fig. 141, x), that may easily lead the beginner to confuse it with a multipolar ganglion cell.

T-shaped branches may be seen in preparations of the spinal cord treated after No. 76. In young embryo chicks the spinal ganglion cells are still bipolar; unipolar cells are found in embryo chicks about seventeen days old. Transition forms occur in chick embryos between the ninth and fourteenth days and in embryo rabbits from 5 to 12 cm. long. Staining with methylene blue (p. 42) is strongly recommended.

No. 87.—*Sympathetic ganglia*.—Fix and harden the large superior cervical ganglion of the sympathetic nerve like No. 86. Here, too, on account of the abundance of nuclei nuclear staining is applicable only to *very* thin sections. The characteristic bundles of nonmedullated nerve-fibers, cut obliquely and transversely, can be recognized with the low power; the ganglion cells are also distinct, but their processes are very unsatisfactory and often cannot be detected; the latter may be better exhibited according to the method given in No. 76 and a suitable object is the cervical portion of a ten or fifteen-day-old embryo chick, while still better results are obtained by staining with methylene blue (p. 42). The intestine of infants (ganglia of the myenteric plexus) is also useful. See further Dogiel (*Arch. f. Anat. und Physiol. Anatom.*, 1899, p. 135).

No. 88.—*Simple tactile cells; intraepithelial nerve-fibers; cells of Langerhans; tactile corpuscles*.—Prepare a mixture of gold chlorid and formic acid (p. 47), boil it and let it cool; then cut from the volar side of a freshly amputated finger or toe (with scissors applied flatwise) several small pieces of the epidermis and uppermost layers of the corium about 5 mm. square and 1 mm. thick. Carefully remove any fat attached to the under surface of the corium and place the pieces in the gold and formic acid mixture for one hour, *in the dark*. Then, with glass rods, transfer the pieces to 10 c.c. of distilled water and in a few minutes to

fresh distilled water to which formic acid has been added (p. 47), and expose the whole to daylight (sunlight is unnecessary). In from twenty-four to forty-eight hours the objects have become dark violet. They are now to be hardened in 30 c.c. of gradually strengthened alcohols. In eight days the pieces may be embedded in liver and sectioned; mount in xylol-balsam. The epidermis is red-violet in different tints; the nuclei are only to be seen in places and often are wholly imperceptible; the corium is white; the capillaries, the excretory ducts of the coil-glands, and the nerves are dark violet to black. For simple *tactile cells* the thinnest possible sections are necessary. They may often be found near the excretory ducts of the coil-glands (Fig. 145). Care must be taken not to confuse them with nuclei of shrunken epithelial cells.

The *intraepithelial nerve-fibers* appear as delicate filaments; their connection with the nerve-fibers in the corium is difficult to trace. Processes of the cells of Langerhans, in thin sections, are apt to be confused with the intraepithelial nerve-fibers (Fig. 144).

The *cells of Langerhans* and the *tactile corpuscles* are easily seen; in thick sections the tactile corpuscles are black (Fig. 144), in thin sections red-violet (Fig. 149).

No. 89.—*Compound tactile cells*.—Cut the yellowish wax-like skin, or cere, from the lateral edges of the upper beak of a duck or goose and treat pieces 1 or 2 mm. thick and 1 cm. long with 3 c.c. of 2 per cent. osmic-acid solution plus 3 c.c. of distilled water; place the whole in the dark from eighteen to twenty-four hours; then wash the pieces for one hour in running water and transfer them to 20 c.c. of 90 per cent. alcohol. In six hours the objects may be sectioned. Embed them in liver and make the sections from the corium toward the epithelium, not the reverse. The sections may be mounted unstained in xylol-balsam. The olive-green tactile cells can be readily seen, but the entrance of the nerve-fiber is difficult to find (Fig. 146). In addition Herbst's corpuscles occur in the sections (p. 224). If it is desired to stain the sections, use a nuclear staining solution (p. 38).

No. 90.—*Cylindrical end-bulbs*.—With scissors and forceps cut from the fresh eye of a calf pieces 1 cm. square of the scleral conjunctiva, up to the corneal margin, taking care not to roll them. It is better to let them lie on the sclera until all are cut. Carefully slip the pieces, epithelial side up, from the sclera on to a cork plate and span them out with needles. Moisten the surface with a few drops of the vitreous humor obtained from the eye; with fine scissors and forceps dissect off a thin membrane consisting of a thin layer of connective tissue and the epithelium resting upon it. This operation must be done with great care; folding and torsion of the membrane must as far as possible be avoided. The membrane, with the epithelial side up, should now be slipped on to a dry slide and spread out. At first it will draw together, but in a moment or two the edges dry somewhat and adhere to the glass and it can then be extended without much difficulty. The slide with the preparation is to be placed in a glass jar containing 65 c.c. of distilled water to

which 2 c.c. of acetic acid have been added. In about an hour (or later), during which time the membrane swells considerably and floats from the slide, by *carefully* touching it with a clean needle endeavor to remove the epithelium; it loosens without much trouble and floats off in fine white shreds. If this is not done *cautiously* the end-bulbs lying close beneath the epithelium may be torn off with it. The more thoroughly the epithelium is removed the better. After it has lain four or five hours in the dilute acetic acid transfer the swollen piece with a few drops of the same fluid to a slide, apply a cover-glass and make slight pressure upon it with the outspread branches of the forceps. On examination with the low power the blood-vessels are plainly seen—they are recognized by their distinct nuclei—and also the medullated nerve-fibers.* Trace such a fiber until the medulla ceases; examine this point with the high power, for there the end-bulbs are most apt to be found. In many cases nothing will be seen but numerous nuclei and even when a favorable situation is found the end-bulbs are so pale that it is very difficult to perceive them (Fig. 147); the axis-cylinder, too, is often very difficult to detect. Only the practised microscopist will succeed in finding them. Beginners are advised not to attempt this preparation.

No. 91.—*Lamellar corpuscles*.—These are best obtained from the mesentery of a cat, where usually they can be seen with the unaided eye. They appear as milky, glass-like, transparent, oval spots between the strands of the adipose tissue of the mesentery. Their number varies greatly. Occasionally they are very scarce and of such small size that to find them requires close searching. Cut out the portion of the mesentery containing a corpuscle, and spread it out in a drop of salt solution on a slide lying on a black background. Endeavor to remove the attached clusters of fat-cells, taking care not to prick the corpuscle. Ascertain with a low power, without a cover-glass, whether the corpuscle has been sufficiently isolated; then cover it with another drop of salt solution and a cover-glass. Pressure must be carefully avoided. The corpuscle represented in figure 148 is very small.

With the high power one can distinctly see the nuclei of the cells between the capsules; the oval nuclei of the inner bulb are often indistinct and pale. If it is desired to preserve the preparation, treat it under the cover-glass with 1 or 2 drops of 1 per cent. osmic acid and after the medulla has become black and the inner bulb has become brown displace the acid with very dilute glycerol. Methylene blue staining (p. 42) is recommended. See further Sokolow (Anat. Anzeiger, Bd. xvi, p. 453).

No. 92.—*Motor nerve-endings*.—(a) *Terminal ramifications*.—Prepare a mixture of 24 c.c. of 1 per cent. gold chlorid solution plus 6 c.c. of formic acid, boil it and let it cool; cut out small pieces 3 or 4 cm. long of the intercostal muscles of a rabbit and treat them like No. 88;

* In the calf some of the nerve-fibers are nonmedullated; these are not recommended for the investigation.

after the dark-violet pieces have lain from three to six days in 70 per cent. alcohol tease a muscle-bundle about 5 mm. broad in a drop of dilute glycerol to which a very small drop of formic acid has been added. It is of advantage to make slight pressure on the cover-glass. To find the terminal ramifications trace with the low power the easily-recognized black nerve-fibers (Fig. 153). The addition of another drop of acetic or formic acid often makes the picture sharper.

(b) *Nuclei of the motor plate.*—Place the anterior halves of the eye-muscles of a recently killed rabbit in 97 c.c. of distilled water plus 3 c.c. of acetic acid. After six hours transfer the muscles to distilled water; with the scissors cut off a thin flat piece and spread it out on a slide; the ramifications of the whitish nerves can be plainly seen with the unaided eye. With low magnification (50 diameters), the anastomoses of the nerve-bundles, as well as the blood-vessels, that are easily recognized by the transversely placed nuclei of their smooth muscle-fibers, can be seen. On account of the large number of sharply contoured nuclei belonging to the intramuscular connective tissue, the end-plates are not easy to find. If a nerve-fiber be traced it will soon be seen that the double-contoured medullary sheath ceases abruptly and loses itself in a group of nuclei; these are the nuclei of the motor plate, the other details of which are not distinctly visible. The cross-striation of the muscle-fibers, which are very pale, is often indistinct (Fig. 154).

No. 93.—*The suprarenal bodies; topographic view.*—Fix the entire suprarenal body of a child in 200 c.c. of 0.1 per cent. chromic acid for eight days and harden it in 150 c.c. of gradually strengthened alcohols; mount unstained sections in dilute glycerol (Fig. 155).

No. 94.—*Elements of the suprarenal body.*—Tease portions of the fresh organ in a drop of salt solution. The elements are very delicate and therefore injured cells are of frequent occurrence.

No. 95.—*For the study of the minute structure of the suprarenal body* place 2 cm. cubes of the fresh organ in 100 c.c. of Zenker's fluid (p. 33) and after from twelve to twenty-four hours harden in an equal quantity of gradually strengthened alcohols; cut thin sections, stain them in Hansen's hematoxylin, and mount in xylol-balsam (Fig. 157). For the exhibition of the nerves treatment with the Golgi mixture for from six to eight days and with the 0.75 per cent. silver solution for from two to three days, with several repetitions of this procedure, is recommended.

V. THE DIGESTIVE ORGANS.

THE MUCOUS MEMBRANE.

The inner surface of the entire alimentary tract, of the respiratory organs, of certain parts of the genito-urinary system, and of some of the organs of special sense is covered by a soft, moist membrane, the *mucous membrane* or *tunica mucosa*. It is composed of a soft epithelium and of connective-tissue. Immediately under the epithelium is a structureless membrane, the *membrana propria* (p. 86); beneath this follows the *tunica propria* (stroma), which passes by a gradual transition into the subjacent, loose-textured *tela submucosa*, that in turn connects the mucous membrane with the underlying structures, for example, muscles or bones. The epithelium of the glands is derived from the epithelium of the mucous membrane (see p. 80).

A. HEADGUT.

THE ORAL CAVITY.

THE MUCOUS MEMBRANE OF THE ORAL CAVITY.

The mucous membrane of the mouth consists of three parts: (1) the epithelium, (2) the tunica propria, and (3) the submucosa (Fig. 160). The *epithelium* is typical stratified squamous epithelium (see page 77). The *tunica propria* is formed of interlacing connective-tissue bundles richly interspersed with elastic fibers. The bundles of the uppermost layers are very slender and form a compact, apparently almost homogeneous felt-work. The surface of the tunica propria is beset with numerous, usually simple papillæ (Fig. 160, 1), the height of which varies greatly in the different regions of the oral cavity. The highest papillæ (0.5 mm.) occur at the edge of the lips and on the gums. The tunica propria passes without sharp limits into the *submucosa*, which consists of somewhat thicker bundles of connective tissue, among which the elastic fibers are not numerous. The submucosa is in general loosely attached to the walls of the oral cavity; only on the gums and on the hard palate is it firmer and here intimately united to the periosteum. It supports the *glands* of the mucous membrane; with the exception of the sebaceous glands occasionally found at the edge of the lips and on the inner surface of the cheeks, these are branched alveolo-tubular glands from 1 to 5 mm. in size. Their main excretory duct (Fig. 160, 2) is somewhat expanded at its lower end and in the greater part of its length is lined with stratified squamous epithelium; the branches and twigs into

which it divides and subdivides are lined with cylinder epithelium, the larger branches with the stratified variety, the smaller branches with the simple. Not infrequently the main excretory duct receives the excretory tubes of small accessory mucous glands (Fig. 160, 3). For the minute structure of the end-pieces, see the next chapter. The numerous *blood-vessels* of the oral mucous membrane are arranged in two networks, situated in two horizontal planes, of which the coarser lies in the submucosa, the other, finer, in the tunica propria. From the latter capillary loops ascend into the papillæ. The *lymph-vessels* similarly form two

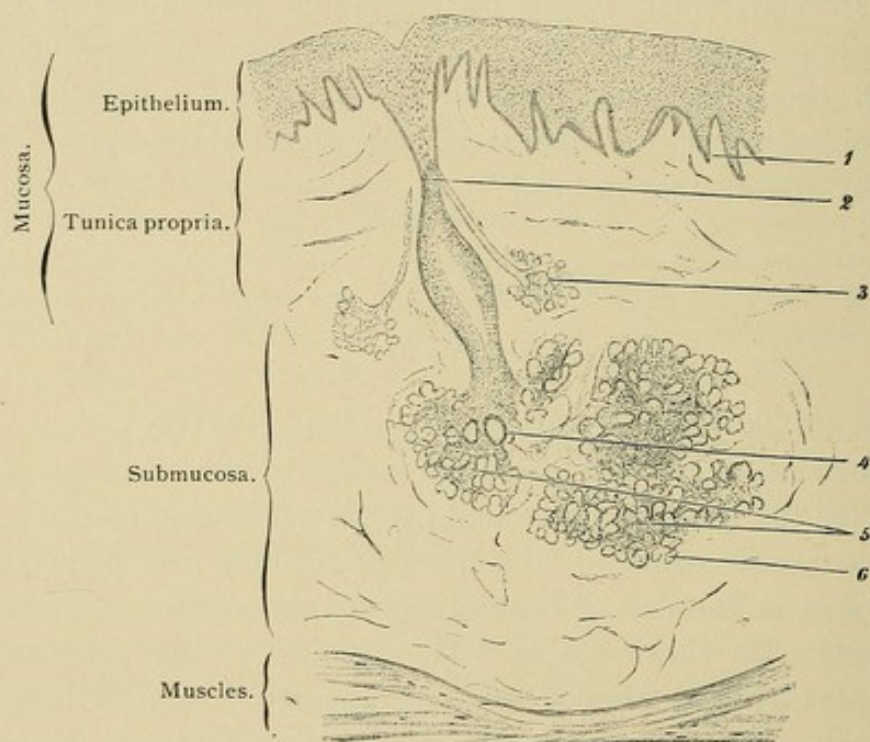


FIG. 160.—VERTICAL SECTION THROUGH THE MUCOUS MEMBRANE OF THE LIP OF AN ADULT MAN. $\times 30$.
1, Papilla; 2, excretory duct; the lumen is cut at only one point; 3, accessory gland; 4, a branch of the excretory duct in transverse section; 5, gland bodies grouped into lobules by connective tissue; 6, a gland-tubule in transverse section. Technic No. 97.

networks, a wide in the submucosa, a narrow in the tunica propria. The medullated *nerve-fibers* form a wide-meshed reticulum in the submucosa, from which many ramifying fibers ascend to the tunica propria. Here they terminate in end-bulbs (p. 222), or they lose their medullary sheath and as nonmedullated nerve-fibers penetrate into the epithelium and after repeated division terminate there in free endings.

THE GLANDS OF THE ORAL CAVITY.

The gland-cells of the oral cavity are of two kinds: (1) cells that yield a secretion rich in albumin, *albuminous* or *serous cells*; (2) cells that produce a secretion consisting of mucin, *mucous* or *mucin cells*.

The *serous cells*, examined when fresh, are characterized by numerous highly refractive granules. In fixed preparations they appear sometimes dark and of slight circumference (empty stage), sometimes a trifle clearer and larger (loaded stage), in correspondence with the functional state (*cf.* Fig. 24, p. 80). The spheric nucleus is situated not quite in the cell center, usually nearer the cell base.

The *mucous cells* in the fresh state are much less refractive. In fixed preparations the typical mucous* cells appear clear; when they are filled with secretion the nucleus is flattened and lies pressed against the cell base and when the contents are discharged it merely becomes oval, without essential change in place and position. The elaborated secretion (not the granular precursors of the same) can be stained by many anilin pigments, also by Delafield's hematoxylin and by mucicarmine (*cf. e. g.* Fig. 25).

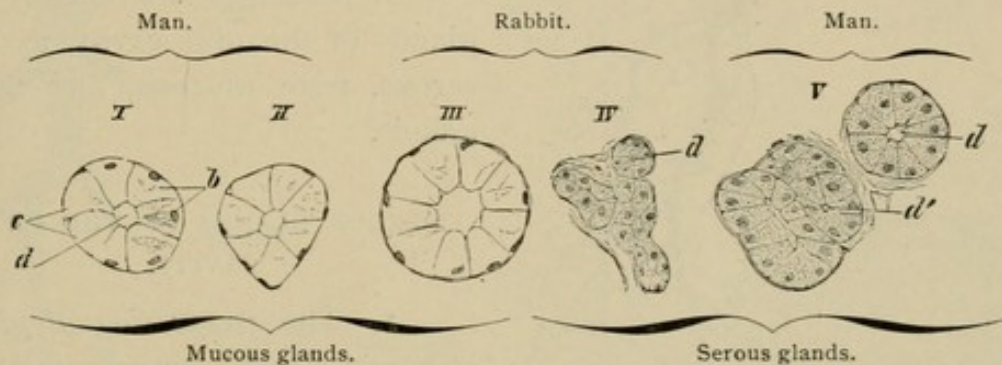


FIG. 161.—FROM SECTIONS OF LINGUAL GLANDS. I. Tubule in cross-section with (*b*) gland-cells empty of secretion and (*c*) gland-cells filled with secretion; *d*, lumen. II. Tubule in cross-section in which all the cells are filled with secretion. III. Cross-section of a mucous gland-tubule. IV. Several tubules of a serous gland; at *d* the very small lumen. V. Tubules with a large (*d*) and a small (*d'*) lumen. All the sections are magnified 240 times. Technic No. 102.

Only a few glands of the oral cavity of man contain exclusively one kind of cells; to these belongs the parotid gland, the end-pieces of which are constructed entirely of serous cells, also the "serous lingual glands" situated in the region of the foliate and the vallate papillæ. The glands of the anterior surface of the soft and of the hard palate, also the "mucous glands" of the root of the human tongue, contain exclusively typical mucous cells. All other glands of the oral cavity are "mixed glands," and mixed in such manner that some end-pieces are clothed by

* By this name I would designate those mucous cells the cell-body of which has for the greater part become a collecting center for secretion (p. 81) and which in different functional phases long retains substantially this center within its circumference. Not all mucous cells share this property; the collecting center of the human olfactory glands is very small and in normal circumstances appears not greatly to enlarge; in the mucous cells of the gastric epithelium, also in those of the lingual glands of the cat, the dimensions of the collecting center vary very considerably, according to the phase of the functional cycle.

serous cells only, while other end-pieces contain chiefly mucous cells, between which single or groups of serous gland-cells are situated. These latter, where their lateral surfaces are in contact with mucous cells, are subjected to compression and may be indeed apparently entirely* pushed back from the axial gland lumen and then form the "demilunes" of Giannuzzi † ("border-cells").

Hereby for the present I accept this interpretation of many demilunes, as serous gland-cells, (1) because they contain granules like those of the serous gland-cells, (2) because of their peculiar relations to the secretory capillaries, and (3) because normally differences between them and the mucous cells, even in varying functional states, are demonstrable. Whether, on the other hand, all demilune cells are of serous nature, is very doubtful. Empty mucous cells, particularly those with collecting centers of variable size, may be pushed from the lumen by neighbors filled with secretion and so become demilunes.

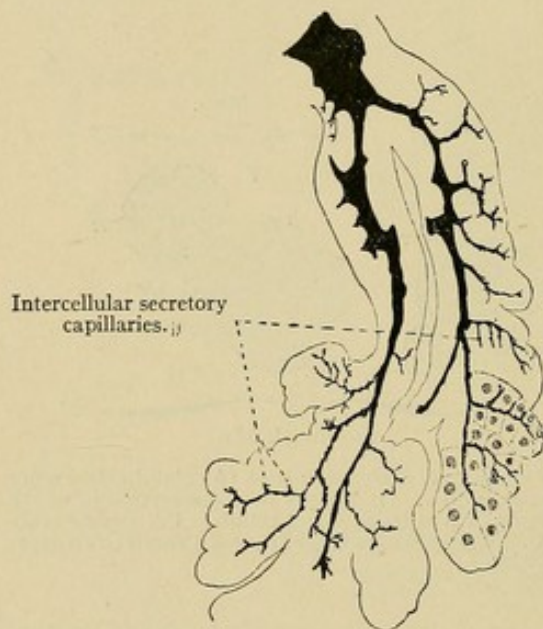


FIG. 162.—FROM A SECTION OF THE ROOT OF THE TONGUE OF A MOUSE. $\times 240$. A serous gland with its tubular system blackened by Golgi's reaction. The tubular character is easily recognized. The right-hand lower portion of the gland is completed by the schematic sketching in of the cells. Technic No. 126.

Accordingly we classify the glands of the oral cavity as pure serous, pure mucous, and mixed glands.

SEROUS GLANDS OF THE ORAL CAVITY.

1. The *serous lingual glands* (Ebner's glands) are compound tubular glands; their aqueous ("serous") secretion is marked by its high content of albumin, hence the name "albuminous glands."

These serous glands are confined to the region of the vallate and foliate papillæ; their excretory ducts open, as a rule, in the furrows

between papilla and wall (Fig. 185) and are clothed in a simple or stratified—not rarely ciliated—cylinder epithelium; the small tubules consist of a delicate membrana propria and short cylindric or conical,

* In reality they stand in communication with the lumen through a secretory capillary (cf. p. 86).

† Not to be confused with these are the so-called demilunes of Pflüger, which are formed by those mucous cells in which the peripheral protoplasmic division is not entirely filled. They are particularly fine in the lingual glands of the cat. Oblique sections of the membrana propria and the stellate cells lying upon it may give rise to deceptive pictures, resembling the demilunes.

membrane-less cells, which in man and sheep exhibit two zones: an inner dark, beset with fine granules, and an outer clear zone, that encloses the round nucleus.* The axial lumen of the tubules (especially in animals) is very narrow (Fig. 161, *d d'*), and takes up still narrower intercellular secretory capillaries (Fig. 162).

2. The *parotid gland* (glandula parotis, auricular salivary gland) is preeminently a compound alveolar gland (p. 85) and of all the oral salivary glands possesses the most highly differentiated duct system; the branches of the excretory duct pass into well-developed salivary tubes, that con-

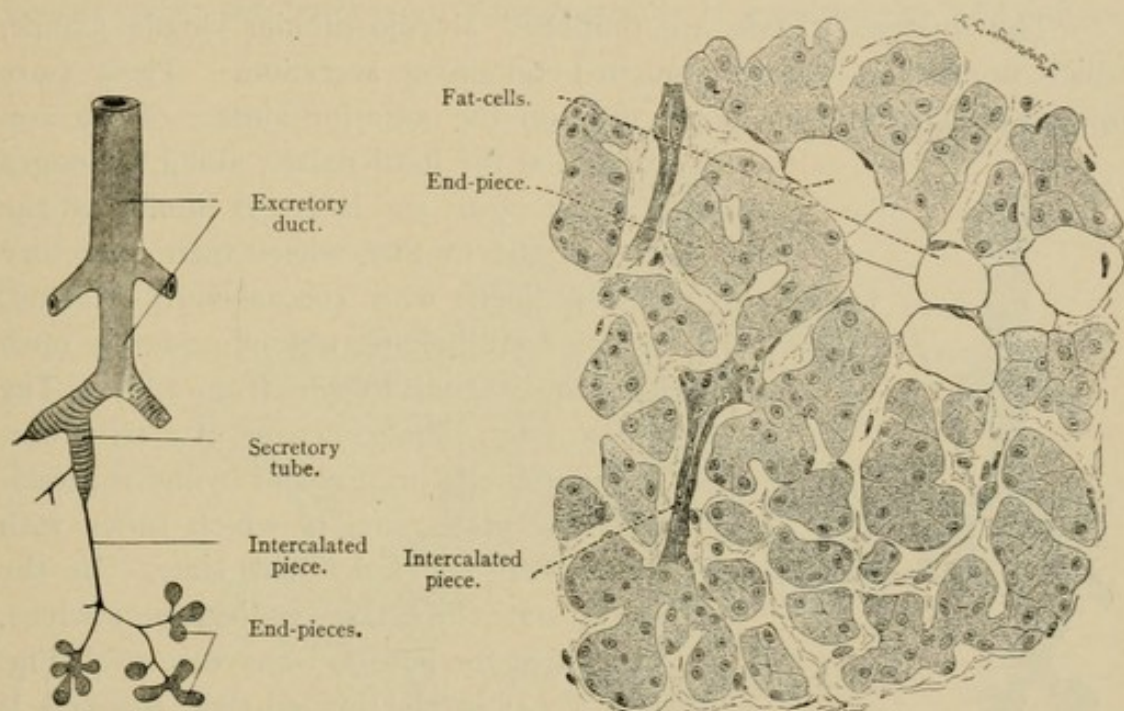


FIG. 163.—SCHEME OF THE HUMAN PAROTID GLAND.

FIG. 164.—SECTION OF THE PAROTID GLAND OF ADULT MAN. $\times 252$. The very narrow lumina are entirely invisible in this preparation. Technic No. 118.

tinue in long, narrow intercalated divisions. The latter lead into short, simple or branched end-pieces (Fig. 163). The excretory duct, parotid duct (Stenoni), characterized by a broad membrana propria lying immediately beneath the epithelium, is clothed in a two-layered epithelium, here and there intermixed with goblet cells, that gradually becomes one-layered in the smaller branches. The tall cylindric epithelial cells of the secretory tubes show distinct longitudinal striation at their base (*cf.* p.

* These differences can be brought out only by special methods and high magnification. The figure 161 shows nothing of this. In the horse, pig, and cat the two zones are in general indistinct, in the rabbit not present. Occasionally between the serous tubules are found a few tubules containing some mucous and some serous cells. In the cat the other lingual glands also are of mixed nature.

88), the intercalated pieces (Fig. 164) are clothed with very slender, often spindle-shaped cells. Finally, the end-pieces consist of a delicate membrana propria with stellate cells and of cubical serous gland-cells; in the empty state these are small and dimly granular, in the loaded state larger and somewhat clearer. (Cf. p. 80.) Free-ending, simple secretory capillaries extend from the axial lumen between the gland-cells, without reaching the membrana propria.

The interalveolar connective tissue often contains groups of fat-cells (Fig. 164).

MUCOUS GLANDS OF THE ORAL CAVITY.

The mucous glands are branched alveolo-tubular simple glands, which produce a mucus- (mucin-) containing secretion. These pure mucous glands, in man, occur only on the anterior surface of the soft

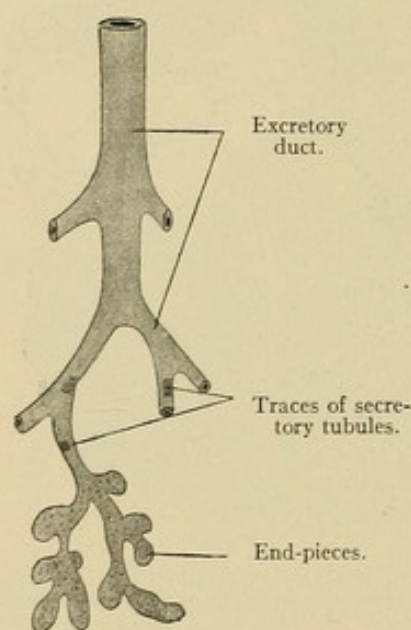


FIG. 165.—SCHEME OF THE HUMAN SUBLINGUAL GLAND.

palate, on the hard palate, along the edges of the tongue, and in larger number at the root of the tongue, where their excretory ducts, lined with (occasionally ciliated) cylinder epithelium, not infrequently open into the lingual tonsils (Fig. 186). The walls of the tubules consist of a structureless membrana propria and cylindric gland-cells, the appearance of which varies with their changing functional state. In the empty state the cell is smaller, the nucleus, situated at the base, is transverse-oval (Fig. 161, I *b*); in the loaded state the cell is broader and the nucleus is pressed flat against the wall (Fig. 161, I *c*, II). Usually one and the same mucous gland, even often one and the same tubule, shows

gland-cells in different secretory phases (Fig. 161, I), which become particularly distinct upon the application of fluids that stain mucin.* The pure mucous glands possess no secretory capillaries.

MIXED GLANDS OF THE ORAL CAVITY.

1. The *sublingual gland* (glandula sublingualis) is a compound alveolo-tubular gland; the canal system consists of an excretory duct, the branches of which continue in very short mucous tubes, which pass

* Rarely does one find in the human lingual mucous glands cell-forms that correspond to those represented in figure 25 *a-c*, page 80.

direct into convoluted end-pieces, which are characterized by their varying caliber—often they are evaginated (Fig. 166). Intercalated tubes are wanting (*cf.* p. 87). The excretory duct, the sublingual duct (Bartholini), and its coarser branches are composed of two-layered cylinder epithelium and connective tissue with elastic fibers. The smaller twigs (0.05 mm. thick and more) possess only a simple cylinder epithelium; they continue in the secretory tubes, whose low cylinder cells show the characteristic striation only in a few places. The end-pieces, enveloped in a *membrana propria* and stellate cells, are clothed with mucous and serous cells; the latter often stand together in groups (Fig. 166), there-

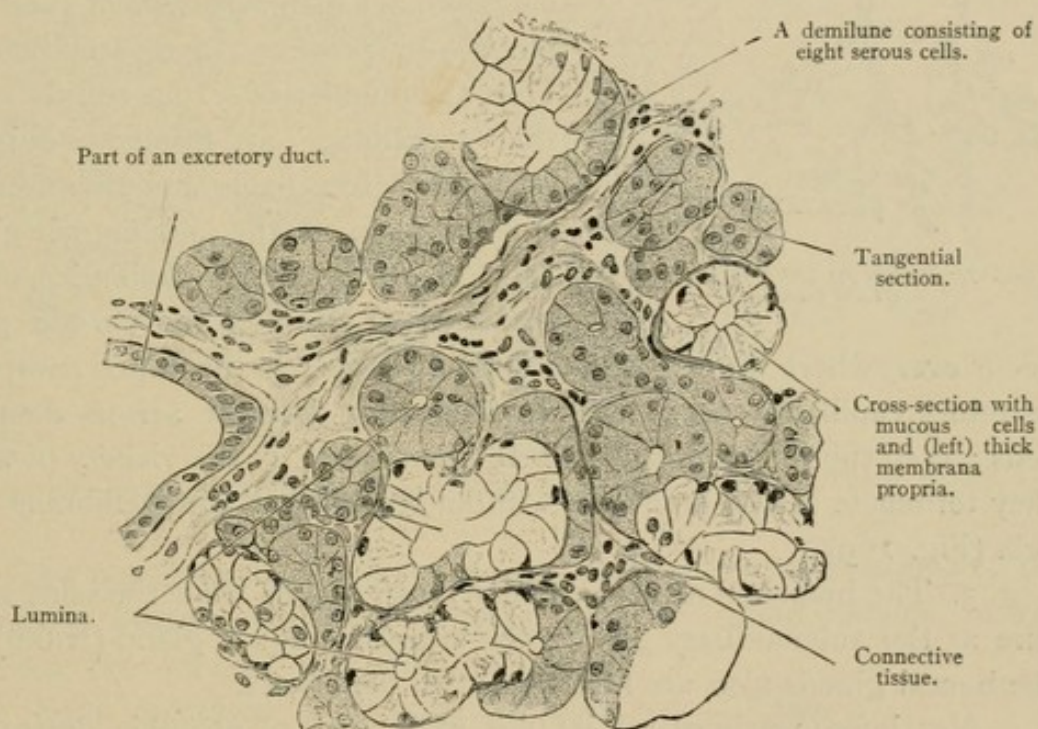


FIG. 166.—THIN SECTION OF THE SUBLINGUAL GLAND OF MAN. $\times 252$. Technic No. 118.

fore the "demilunes" are very large. Only the serous gland-cells are furnished with free, branched intercellular secretory capillaries. The connective tissue lying between the tubules and the lobules is rich in leucocytes.

2. The *submaxillary gland* (*glandula submaxillaris*) is in part predominantly an alveolar (p. 85), in part an alveolo-tubular compound gland. The canal system is more differentiated than that of the sublingual gland, in so far that distinct secretory tubes and short intercalated parts are present. The end-pieces are of two kinds, alveolar and tubulo-alveolar (Fig. 167). The excretory duct, the submaxillary duct (Whar-toni), and its branches, respecting the epithelium are the same as those

of the sublingual gland, but a richly cellular stratum of connective tissue and outwardly to this a thin layer of longitudinally disposed muscle-

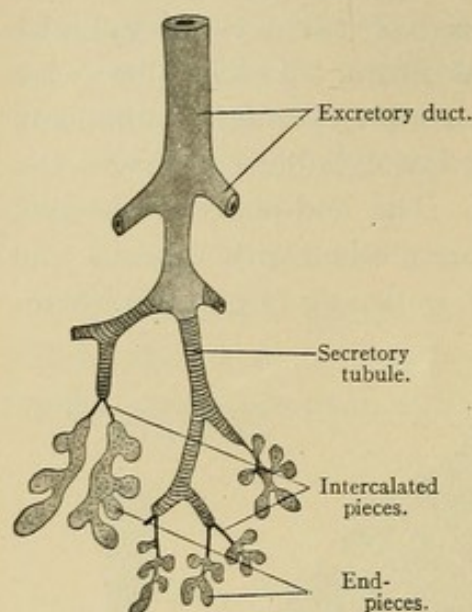


FIG. 167.—SCHEME OF THE HUMAN SUBMAXILLARY GLAND.

fibers constitute a special peculiarity of the submaxillary duct; the epithelial cells of the secretory tubes are marked by the characteristic striation at their base and contain a yellow pigment. The intercalated pieces are clothed with cubical cells and lead into the end-pieces, that either are clothed with serous gland-cells only—the greater portion of the submaxillary gland consists of such end-pieces—or possess a mixed epithelium. Here the number of serous cells is small, the “demilunes” are formed of only one or a few serous cells and therefore are smaller than in the sublingual gland. Inter-cellular secretory capillaries of the character of those of the parotid gland

occur everywhere in the pure serous end-pieces; in the mixed end-pieces secretory capillaries occur only in connection with the serous demilune cells; they run intercellular up to the demilunes, in the vicinity of which they terminate in free branches, without reaching to the membrana propria (Fig. 169).

3. The branched alveolo-tubular labial glands show the same structure as the submaxillary gland; the anterior lingual gland (Nuhn) and the buccal glands also are furnished with demilunes.

Not infrequently gland lobules in process of atrophic destruction are found in the glands of the oral cavity; their end-pieces, characterized by a wide lumen and low gland-cells, are surrounded by abundant connective tissue, occasionally also by many leucocytes.

The foregoing description applies only to the oral glands of men. In the lower mammals very far-reaching differences often exist. The parotid glands of the rabbit, dog, and cat, also the submaxillary gland of the rabbit, agree in structure with the parotid gland of man. The sublingual and submaxillary glands of the dog and cat and the sublingual gland of the rabbit resemble the human sublingual and submaxillary glands.

The *blood-vessels* of the glands of the oral cavity are very conspicuously developed. The arterial stems as a rule run alongside the main excretory duct, where they divide into numerous branches which pass between the gland lobules and finally penetrate within the latter, break

up into capillaries and form close networks around the end-pieces. The capillaries lie in immediate proximity to the gland-cells and are separated from them only by the *membrana propria* (see also p. 86). The larger veins follow the course of the arteries.

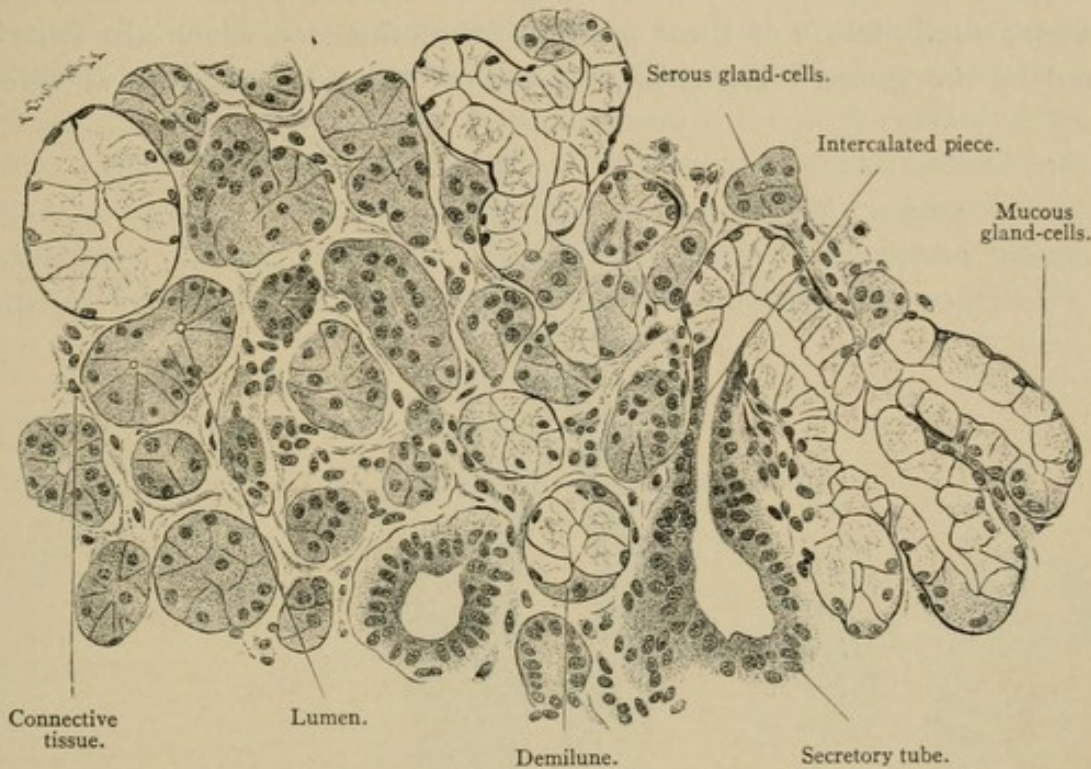


FIG. 168.—SECTION OF THE SUBMAXILLARY GLAND OF ADULT MAN. $\times 252$. Technic No. 118.

The *lymph-vessel trunklets* run with the coarser ramifications of the excretory ducts, without penetrating into the gland lobules. Clefts between the lobules and the end-pieces have been described as lymph channels.

The glands of the oral cavity are profusely supplied with plexuses of medullated and chiefly nonmedullated *nerves*, along the course of which microscopic groups of sympathetic ganglion cells occur (particularly in the walls of the excretory ducts). The fine nonmedullated nerve-fibers partly ramify in the walls of the blood-vessels, partly form an "epilemmal" plexus, lying immediately upon the *membrana propria* of the gland tubules; from this delicate filaments arise, which pierce the *membrana propria* and as "hypolemmal" fibers terminate in short, varicose, simple or branched ends, which lie against the gland-cells.

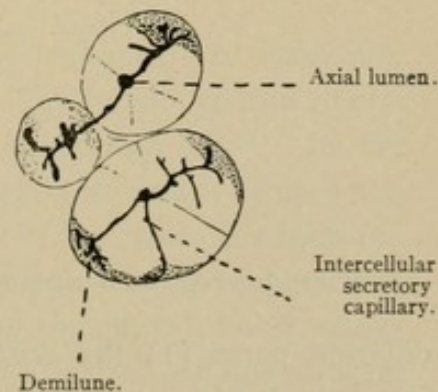


FIG. 169.—FROM A SECTION OF THE SUBMAXILLARY GLAND OF A DOG. $\times 320$. Technic No. 126.

THE TEETH.

The teeth of man and the higher animals are solid structures, which enclose in their interior a cavity, the *pulp cavity*, filled with a soft mass, the *tooth pulp*. The portion of the tooth within the alveolus or socket is called the *root* or *fang*, the free, exposed portion, the *crown*; the juncture of these portions forms the *neck*, which also is covered by the gums. The *solid structures* of the tooth consist of three

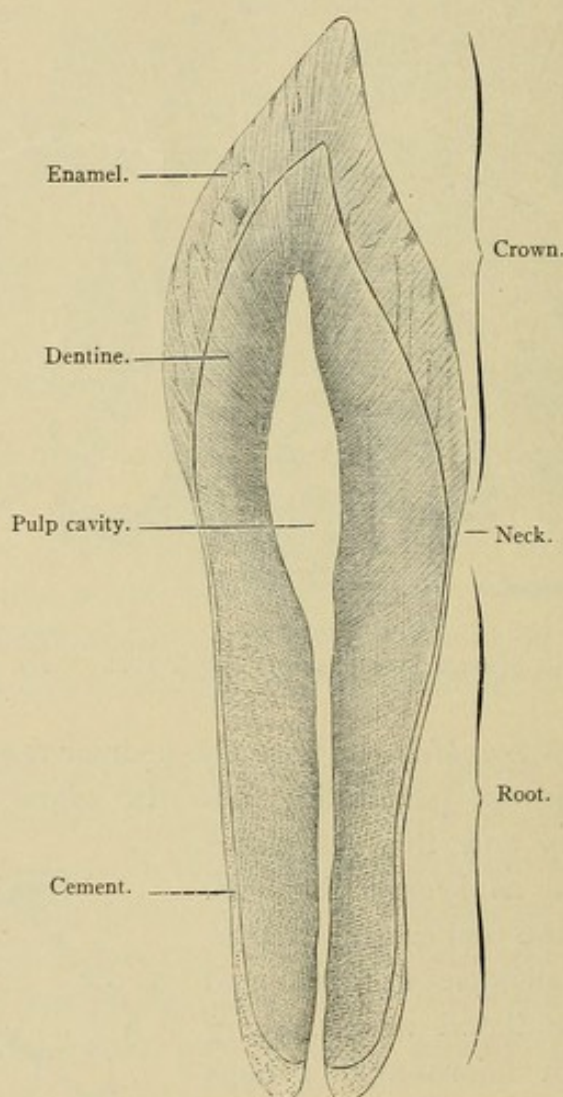


FIG. 170.—LONGITUDINAL GROUND SECTION OF A HUMAN INCISOR TOOTH. $\times 4$. Technic No. 98.

different parts, (1) the *dentine*, (2) the *enamel* with the *enamel cuticle* (*cuticula dentis*), and (3) the *cement*. The arrangement of these parts is as follows: the dentine, which contributes the chief bulk of the tooth and determines its form, encloses the pulp cavity, except on the fang where a narrow nutrient canal admits the nerves and the blood-vessels to the pulp; on the crown the dentine is covered by the enamel, on the fang by the cement, so that its surface is nowhere exposed (Fig. 170).

The *dentine* (*substantia eburnea*) is a white, opaque mass, harder than bone. It consists of an apparently homogeneous calcified ground substance, that in reality contains very delicate, glutin-yielding fibrils, having in general a longitudinal direction, and is pierced by numerous minute canals, the *dental canaliculi* (Fig. 171). The latter begin with a diameter of from 2 to 4 μ at the inner surface of the dentine, describe an S-shaped curve, and then, steadily decreasing in caliber, proceed in a slightly wavy course in a radial direction toward the outer surface of the dentine; there they either terminate at the juncture of the dentine and enamel in tapering ends or they form a loop and turn into a neighboring canaliculus. During their entire course they send off numerous lateral branches, which establish communication with neighboring canaliculi.

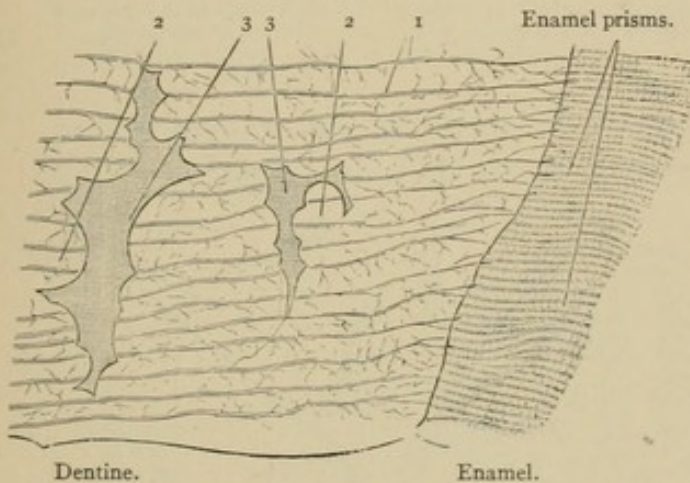


FIG. 171.—FROM A LONGITUDINAL SECTION OF THE LATERAL PART OF THE CROWN OF A HUMAN MOLAR TOOTH. $\times 240$. 1, Dental canaliculi, some extending into the enamel; 2, dental globules projecting toward, 3, the interglobular spaces. Technic No. 98.

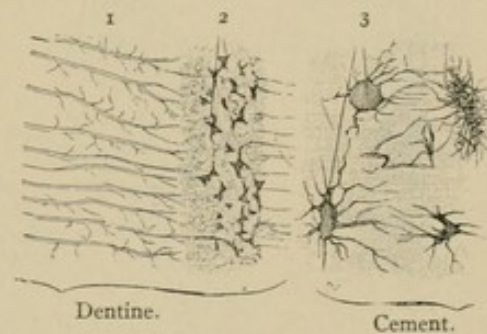


FIG. 172.—FROM A LONGITUDINAL SECTION OF THE ROOT OF A HUMAN MOLAR TOOTH. $\times 240$. 1, Dental canaliculi interrupted by a granule stratum, with many, 2, small interglobular spaces; 3, bone lacunae with many canaliculi. Technic No. 98.

The matrix immediately surrounding the dental canaliculi is especially dense and forms the so-called *dental sheaths*. The lumen of the dental canaliculi is occupied by the soft *dental fibers* (see tooth pulp). In the peripheral parts of the dentine lie the *interglobular spaces* (Fig. 171), uncalcified portions of dentine varying greatly in size, toward which the calcified dentine juts in the form of usually hemispherical protuberances, the *dental globules*. At the neck and in the fang the interglobular spaces are very numerous and very small and form the so-called granule stratum lying immediately beneath the cement (Fig. 172).

The *enamel* (*substantia adamantina*) is still harder than the dentine. It is exclusively composed of long, hexagonal, homogeneous fibers,* from 3 to 6 μ in thickness, the *enamel prisms* (Fig. 173), which are

* The transverse bands do not appear until after treatment with reagents.

firmly united with one another by a scanty amount of irriguous cement-substance. They extend radially, with many undulations, from the surface of the dentine to the free surface of the enamel; this is covered by a very thin but very resistant membrane, the *dental cuticle* (*cuticula dentis*).

The *cement* (*substantia ossea*) coincides in its structure with that of bone. It contains many Sharpey's fibers. Haversian canals are found only in the cement of aged individuals; stratification in lamellæ is seldom well defined. Bone lacunæ are absent near the neck.

The space between the root and the alveolus is occupied by the richly innervated periosteum of the alveolus, the "root membrane," which is firmly united to the cement by Sharpey's fibers, which penetrate from the inferior maxilla through the periosteum into the cement.



Enamel prisms, isolated.

FIG. 173.

FROM THE TOOTH OF AN INFANT. Technic No. 100.



Enamel prisms in transverse section.

FIG. 174.

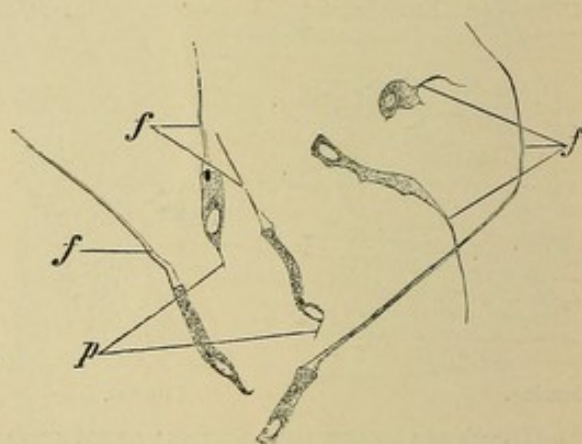


FIG. 175.—SIX ODONTOBLASTS WITH DENTAL FIBERS, *f*; *p*, pulp processes. From the pulp of an infant boy. $\times 240$. Technic No. 99.

The uppermost portion of the alveolar periosteum is called the *circular dental ligament* (*ligamentum circulare dentis*).

The *dental pulp* is formed of a soft connective tissue, containing delicate fibers not united in bundles, the cellular elements of which, partly spherical, partly stellate cells, on the surface are developed into a layer of slender cells, the *odontoblasts*; these send out short processes the pulp processes (Fig. 175), that are connected with other elements in the pulp, and long processes that extend into the dental canaliculi, the previously mentioned *dental fibers* (Fig. 175, *f*). Elastic fibers are wanting in the pulp, as well as in the root-membrane. Vessels and nerves are limited to the pulp of the tooth; the recently revived statement that the nerve-fibers enter the dental canaliculi stands greatly in need of verification.

DEVELOPMENT OF THE TEETH.

The development of the teeth in man begins early, already toward the close of the second month of fetal life * and is first indicated by a

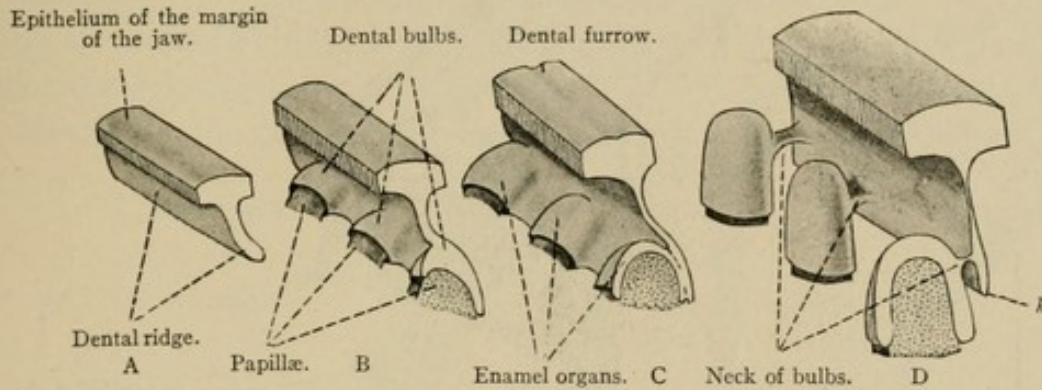


FIG. 176.—SCHEMATIC REPRESENTATION OF THE INITIAL PROCESSES IN THE DEVELOPMENT OF THE TEETH, showing the formation of three teeth. The anlage of each anterior tooth is seen in section; the cut surface of the papilla is stippled. *k*, Free edge of the dental ridge.

proliferation of the epithelium of the margin of the jaw, which in the form of a continuous ridge grows obliquely into the subjacent connective

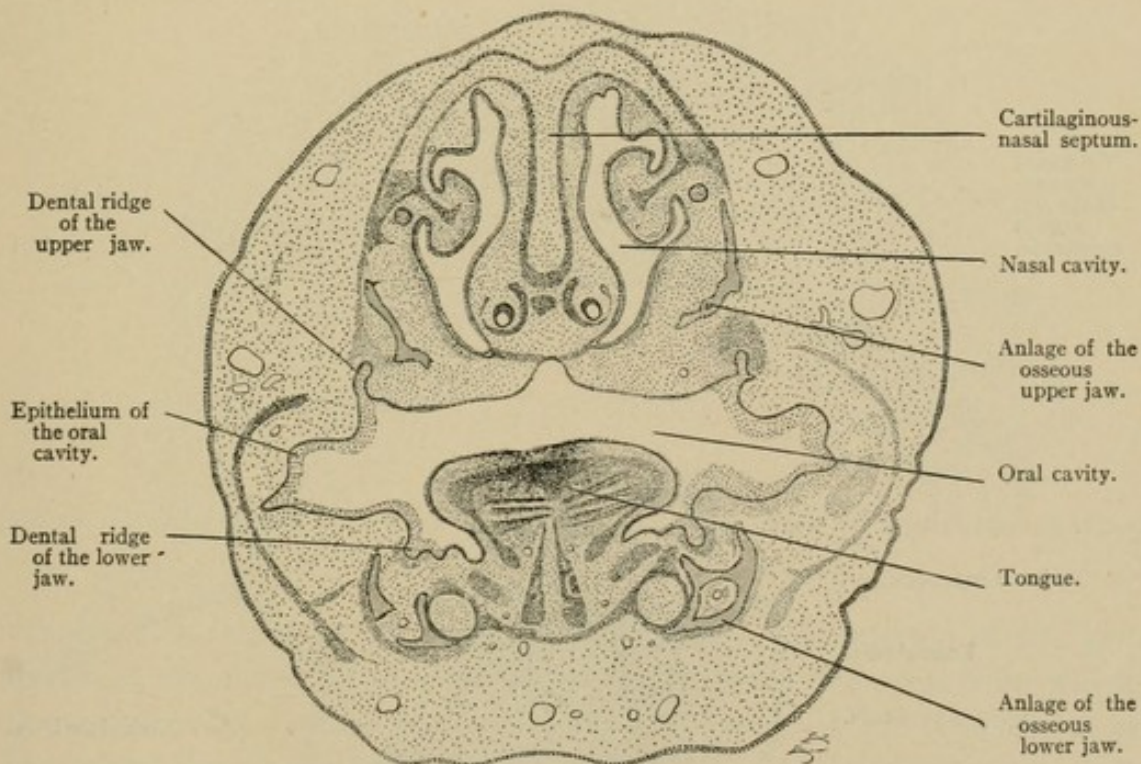


FIG. 177.—FRONTAL SECTION OF THE HEAD OF AN EMBRYO SHEEP, 4 CM. LONG. $\times 15$. Technic No. 101.

tissue. This ridge, the *dental ridge* ("enamel germ") (Fig. 176, A), develops on its lateral (labial) surface knob-like protuberances, the

* That which at an earlier period (the fortieth day) has been described as the anlage of the tooth, is not this alone, but includes the anlage of the labial furrow.

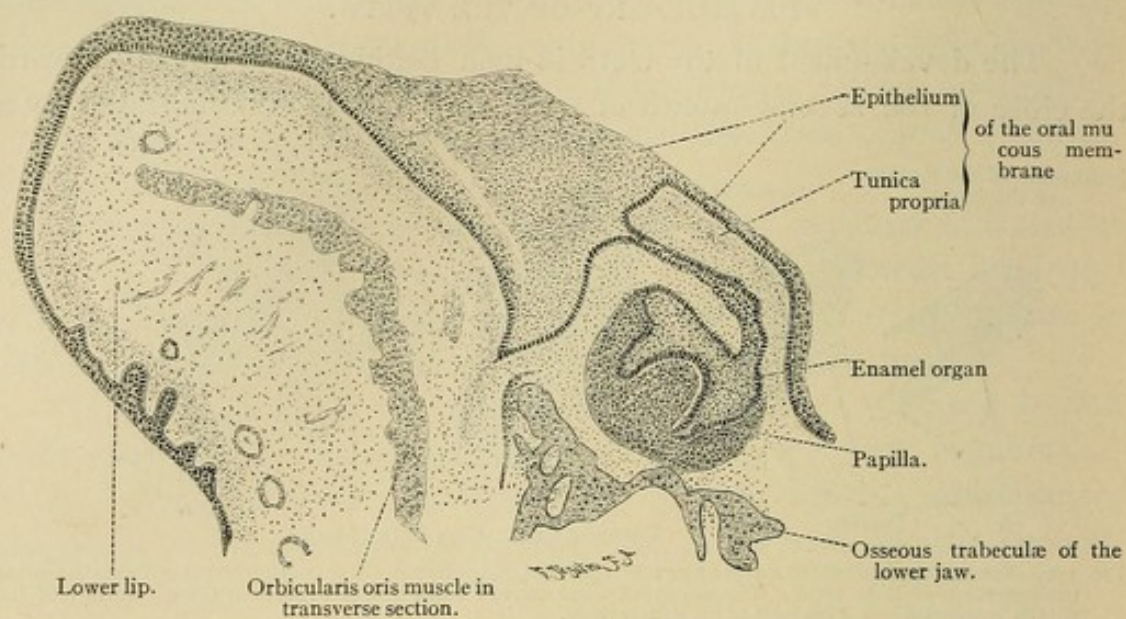


FIG. 178.—CROSS-SECTION OF THE LOWER JAW OF A HUMAN EMBRYO FOUR MONTHS OLD. $\times 42$.
Technic No. 101.

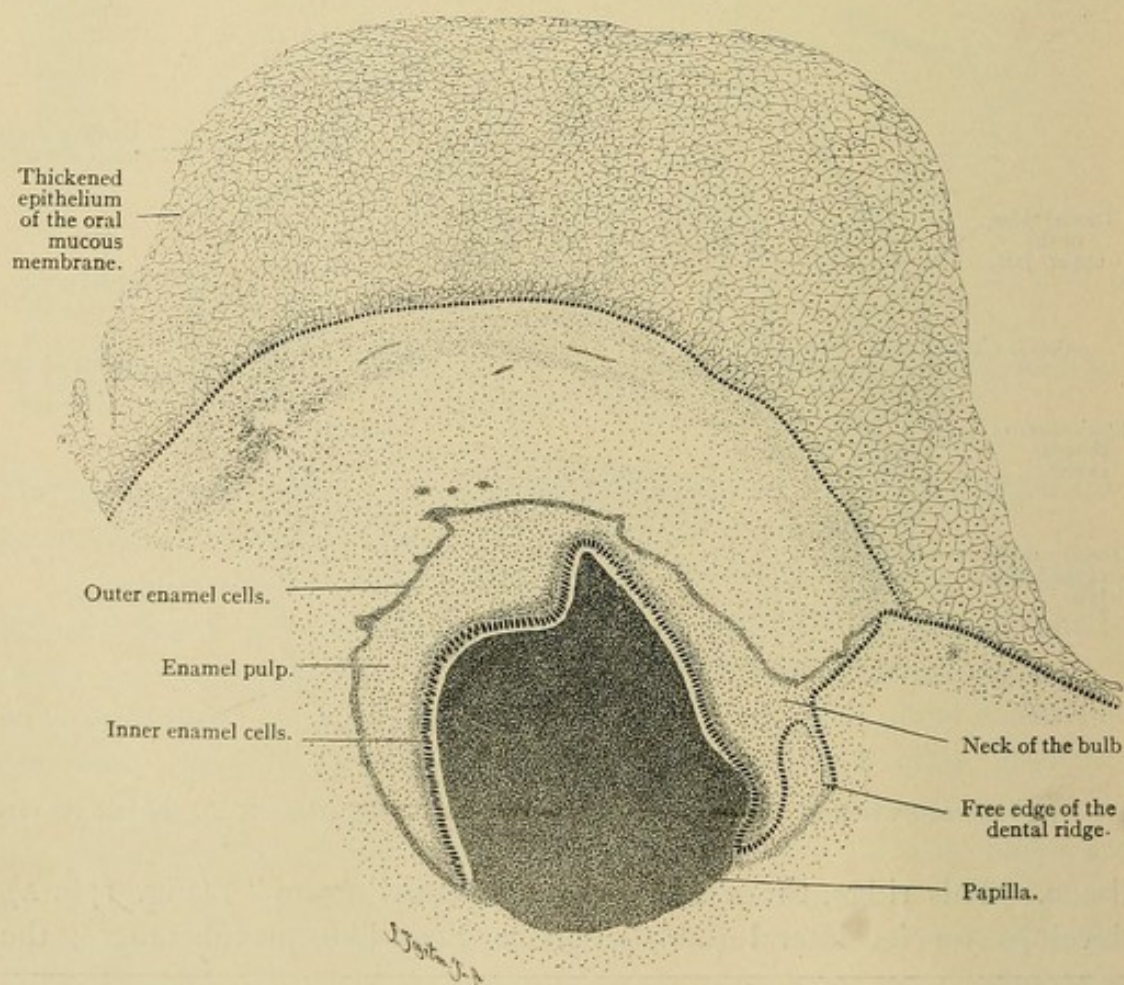


FIG. 179.—FROM A CROSS-SECTION OF THE UPPER JAW OF A HUMAN EMBRYO FIVE MONTHS OLD. $\times 42$.
Technic No. 101

dental bulbs (B), corresponding in number to the temporary teeth, while coincidentally in the tunica propria as many aggregations of closely packed connective-tissue cells arise, the young *dental papillæ* (B) (tenth week). The latter advance obliquely from the external or labial side out of the depths to the inner or lingual side toward the surface and are embraced by the dental bulbs in such a manner that these form an epithelial hood for the dental papillæ. In this way each bulb becomes an *enamel organ*. Meanwhile the dental ridge has taken a more nearly vertical position (c).

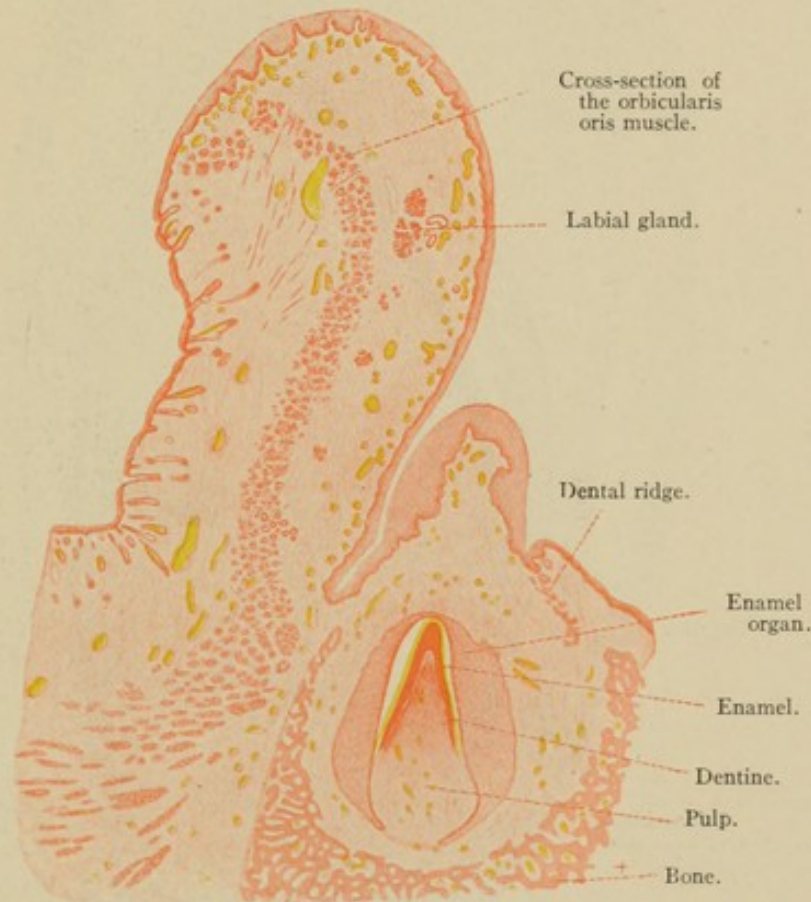


FIG. 180.—VERTICAL SECTION THROUGH THE LIP AND JAW OF A HUMAN FETUS OF SIX AND A HALF MONTHS. $\times 9$. Technic No. 101.

At about this time, too, a longitudinal groove on the margin of the jaw is visible, the *dental furrow*, which exteriorly marks the place from which the dental ridge grew into the depths. The time of the appearance of the dental furrow varies; frequently it is present in the initial stages. It disappears later. The original broad attachment between the dental ridge and the enamel organ becomes diminished by partial constriction (indicated in the scheme c by a stippled line) and finally is reduced to a slender cord, the *neck* of the bulb. Meanwhile the papilla and the enamel organ grow further into the depths, so that the free edge of the

dental ridge does not extend even to half the depth of the enamel organ (Fig. 176 and Fig. 179).

At the same time the elements of the enamel organ undergo further differentiation. The inner cells, resting upon the papilla, develop into

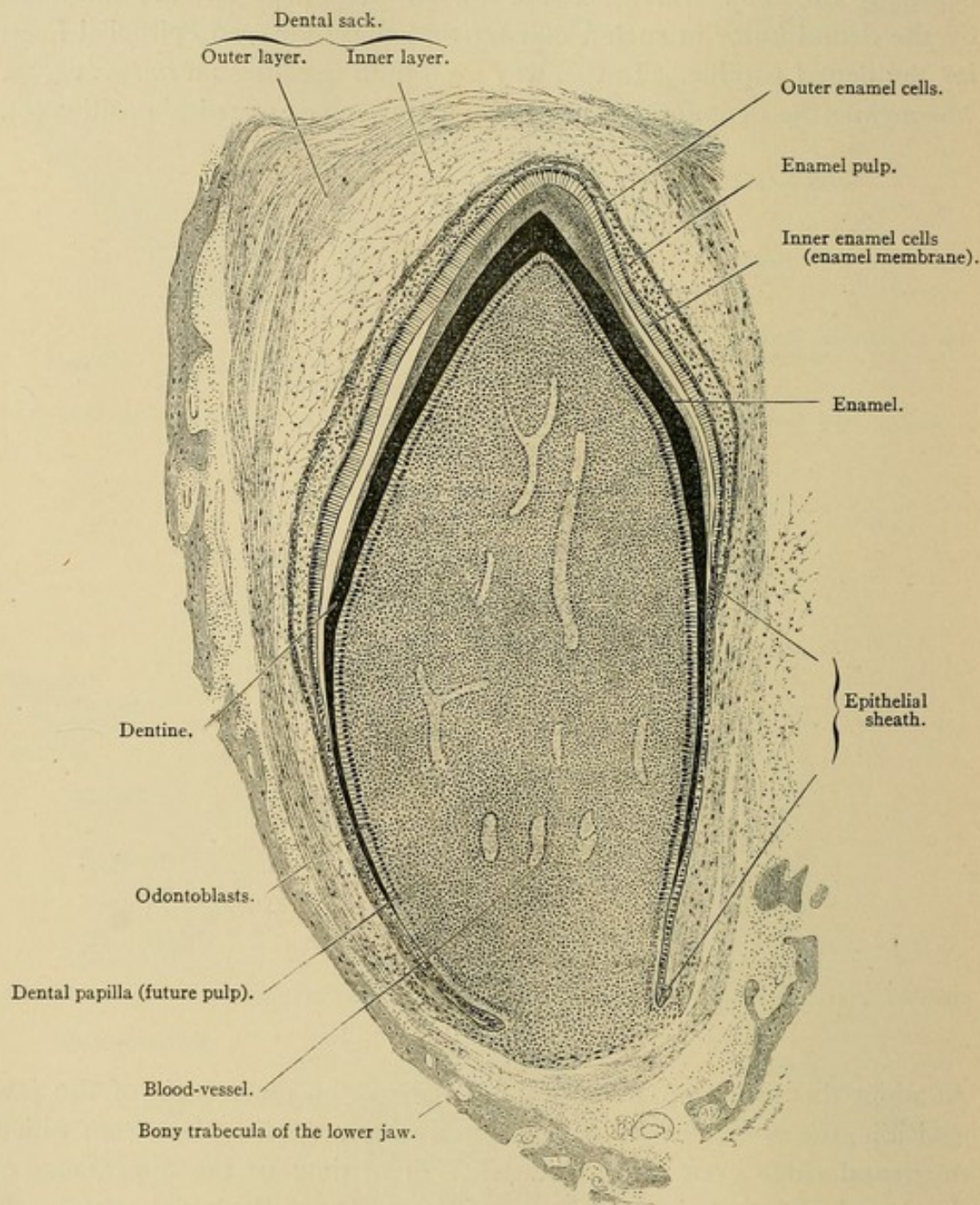


FIG. 181.—LONGITUDINAL SECTION OF A YOUNG MILK-TOOTH OF A NEWBORN DOG. $\times 42$. Technic No. 101.

tall cylinders, called the *inner enamel cells* (Fig. 179); their inner surface is provided with a cuticular border. The peripheral cells, on the contrary, steadily decrease in height (Fig. 182), until finally they are reduced to flattened elements, the *outer enamel cells*; the cells lying between

the inner and the outer enamel cells, by an abundant increase of the intercellular substance, become transformed into stellate, anastomosing elements and constitute the *enamel pulp* (Fig. 182). At the point where the layer of inner enamel cells bends over into the layer of outer enamel cells the enamel organ grows further into the depths, until it has reached the lower end of the anlage of the tooth. In a measure the enamel organ forms the mold, or the matrix, in which the tooth develops. The determination of the shape of the future tooth is the first function of the enamel organ ; the second is the production of the enamel.

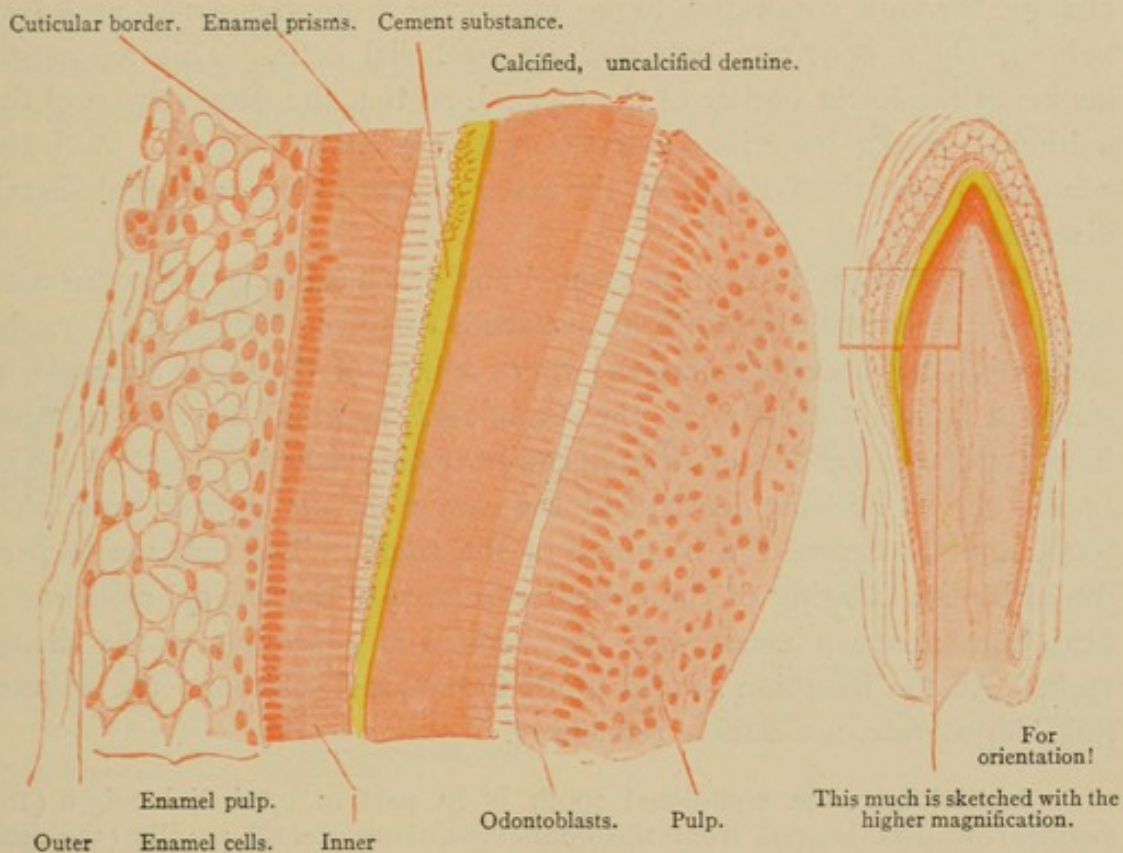


FIG. 182.—PORTION OF A LONGITUDINAL SECTION OF AN INCISOR TOOTH OF A NEWBORN KITTEN. $\times 300$.
Technic No. 101.

In this section the young enamel prisms have been pulled out of their spaces in the cement substance and appear as Tomes's processes of the inner enamel cells.

Enamel is formed only by the *enamel membrane*, that is, by the upper portion of the layer of inner enamel cells ("ameloblasts"), enveloping the crown of the tooth. Each cell of this membrane produces a substance which eventually calcifies and becomes an enamel prism, that is joined to its neighbors by an at first very abundant cement substance. In the further course of development the enamel prisms increase in thickness at the expense of the cement substance.

The lower inner enamel cells, surrounding the root, take no part in

the production of the enamel; they decrease in height and, since here the enamel pulp soon disappears, place themselves directly against the outer enamel cells. The two layers here form the *epithelial sheath* of the root (Fig. 181).

Before the production of enamel has begun the first dentine has been formed (about the twentieth week). The superficial cells of the dental papilla elongate and become the *odontoblasts*, which produce the at first uncalcified dentine (Fig. 182). Development of odontoblasts takes place only so far as the epithelial sheath reaches. As soon as the first dentine is formed, the epithelial sheath at this point undergoes regressive change, through connective-tissue ingrowths from the dental sack (see below), which penetrate between the epithelial cells. This regression begins at the lower border of the enamel, so that the deepest part of the epithelial sheath loses its connection with the enamel organ. With the completed growth of the tooth the last remnant of the epithelial sheath disappears.

Before the production of enamel and dentine begins the connection between the dental ridge and the surface is dissolved * (Fig. 176 D); the connective tissue surrounding the entire anlage of the tooth arranges itself in a compact membrane, the *dental sack*, in which later on an inner looser and an outer denser stratum can be distinguished (Fig. 181). The *enamel cuticle* (cuticula dentis) and the *cement* do not appear until after birth, shortly before the irruption of the tooth. The cuticula is produced by the merging of the cuticular borders of the enamel cells into a firm, homogeneous membrane; the cement is a product of the dental sack. At the irruption of the tooth the enamel cells and the enamel pulp degenerate, not a trace remaining.

Accordingly, the completed tooth is in part of epithelial origin (the enamel), in part derived from the connective-tissue dental papilla (the dentine), which may be compared with a papilla of the mucous membrane, the remains of which persist in the adult as the dental pulp. The cement is in a measure an accessory structure contributed by neighboring tissues.

The permanent teeth develop in the same manner as the temporary teeth; in the twenty-fourth week new dental bulbs arise on the edge of the dental ridge growing further into the depths, which embrace new papillæ penetrating from the side.† The anlage of the permanent tooth at first lies in the same alveolus with the anlage of the milk-tooth and only later is enclosed

* The dental ridge has previously become a much-perforated plate, from which on all sides short, jagged excrescences arise. Remains of the dental ridge may still be found in the gums of newborn children and were erroneously regarded as glands (*glandulæ tartaricæ*).

† The anlages of the permanent molar teeth originate in a lengthening of the posterior end of the dental ridge, which grows in the depths of the mucous membrane backwards toward the angle of the inferior maxilla.

in a separate alveolus. With the exchange of the teeth the septum between these alveoli is resorbed; the dentine and cement of the root of the milk-tooth likewise undergo resorption, which is effected by osteoclasts in the same manner as in the bones.

THE TONGUE.

The bulk of the tongue is formed of striated muscles, the separate bundles and fibers of which freely interlace, that over the greater part of their circumference are covered by a continuation of the oral mucous membrane. The *muscles* are arranged in three planes: (1) *vertically ascending* (in the genioglossus, lingualis, and hyoglossus); (2) *transversely* (in the lingualis), and (3) *longitudinally* (in the lingualis and styloglossus). Since the muscle bundles cross one another for the most part at right angles they form a beautiful network, visible in sections. A

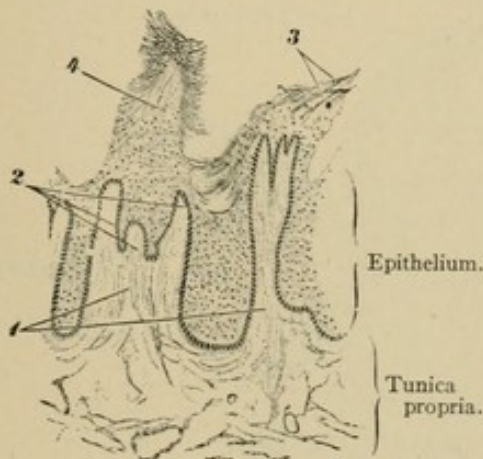


FIG. 183.—LONGITUDINAL SECTION OF THE MUCOUS MEMBRANE OF THE HUMAN TONGUE. $\times 30$. 1, Section of two filiform papillae, each of which bears, 2, three secondary papillae; 3, compound, 4, simple process of epithelium, the surface of which is covered with masses of loosely attached squamous epithelial cells. Technic No. 102.

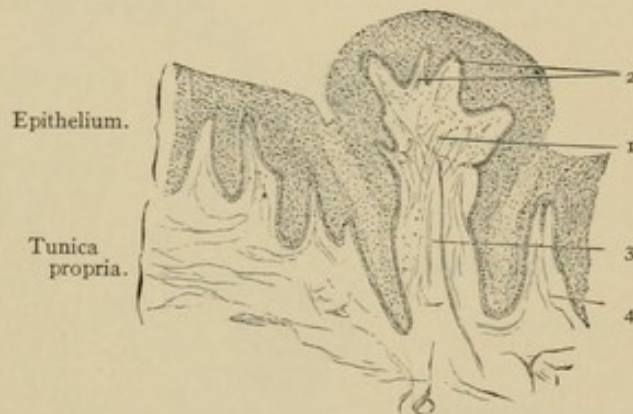


FIG. 184.—LONGITUDINAL SECTION OF THE MUCOUS MEMBRANE OF THE HUMAN TONGUE. $\times 30$. 1, Fungiform papilla with, 2, secondary papillae; 3, stalk of fungiform papilla; 4, small filiform papilla. Technic No. 102.

median septum, the *septum linguae*, divides the muscle masses of the tongue into a right and a left half. The septum begins low at the body of the hyoid bone, gradually increases in height, attains its greatest elevation in the middle of the tongue, then gradually slopes down forward and disappears; it does not extend through the entire thickness of the tongue, but ceases at a distance of about 3 mm. from the surface of the organ. The septum is composed of tough connective tissue fibers.

The *mucous membrane* of the tongue, like that of the oral cavity, consists of an epithelium, a tunica propria, and a submucosa, but is characterized by the conspicuous development and complicated structure of the papillae. Three forms of papillae are distinguished: the *filiform* or *conical*, the *fungiform* or *clavate*, and the *vallate* or *circumvallate papillae*.

The *filiform papillae* (papillae conicae) (Fig. 183) are cylindrical or

conical elevations of the tunica propria, bearing on the summit from five to twenty small secondary papillæ (2). They are composed of distinctly fibrous connective tissue and numerous elastic fibers and are covered with a powerful stratified squamous epithelium, that over the secondary papillæ not infrequently forms a number of filamentous, horny processes. The filiform papillæ are very numerous and are distributed over the entire surface of the tongue; they vary in height from 0.7 to 3 mm.

The *fungiform papillæ* (papillæ clavatæ) (Fig. 184) are spherical structures connected with the tunica propria by a slightly constricted stalk; their entire surface is beset with secondary papillæ (2). They consist of a distinct braidwork of connective-tissue bundles, that contain but few elastic fibers. The epithelial cover is somewhat thinner than on

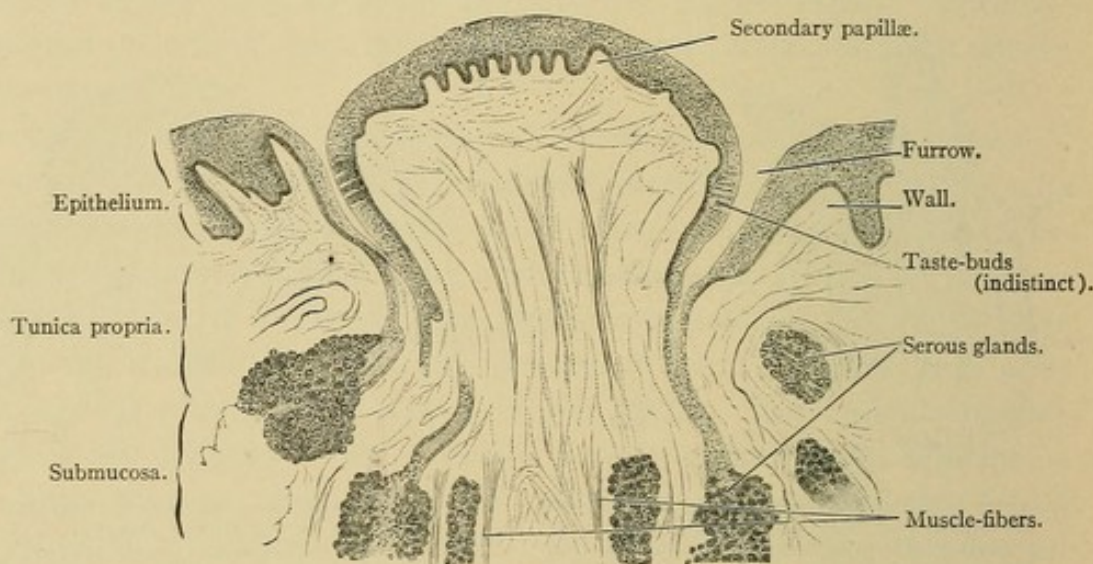


FIG. 185.—VERTICAL SECTION OF A VALLATE PAPILLA OF MAN. $\times 30$. Technic No. 102.

the filiform papillæ and is not cornified. The fungiform papillæ, not so numerous as the filiform, are also distributed over the entire surface of the tongue and in the living they are usually easily distinguished by their red color, due to the capillaries shimmering through the transparent epithelium. Their height varies from 0.5 to 1.5 mm.

The *vallate papillæ* (papillæ circumvallatæ) (Fig. 185) are often very irregularly developed; they resemble broad, flattened, fungiform papillæ and are separated from the remaining mucous membrane by a circular furrow varying in depth; the mucous membrane of the opposite side of the furrow is designated the *wall*. These papillæ are composed of connective tissue like that of the fungiform papillæ, but in man not infrequently contain longitudinally or obliquely disposed smooth muscle-fibers; they are also found in the wall, where their arrangement is

circular. The vallate papillæ* possess secondary papillæ only on the upper, not on the lateral surface. In the epithelium covering their sides, occasionally also in that on the wall, lie the end apparatus of the gustatory nerves, the *taste-buds* (see The Gustatory Organ); in the wall solitary nodules of adenoid tissue are occasionally found (*cf.* p. 146). The vallate papillæ are few in number, from 8 to 15, and only occur at the posterior end of the upper surface of the tongue. They are from 1 to 1.5 mm. high and from 1 to 3 mm. broad. On each posterior lateral margin of the tongue is a group of parallel folds of the mucous membrane, named the *foliate papilla*, that are distinguished by their wealth of taste-buds. The foliate papillæ are especially well developed in the rabbit.

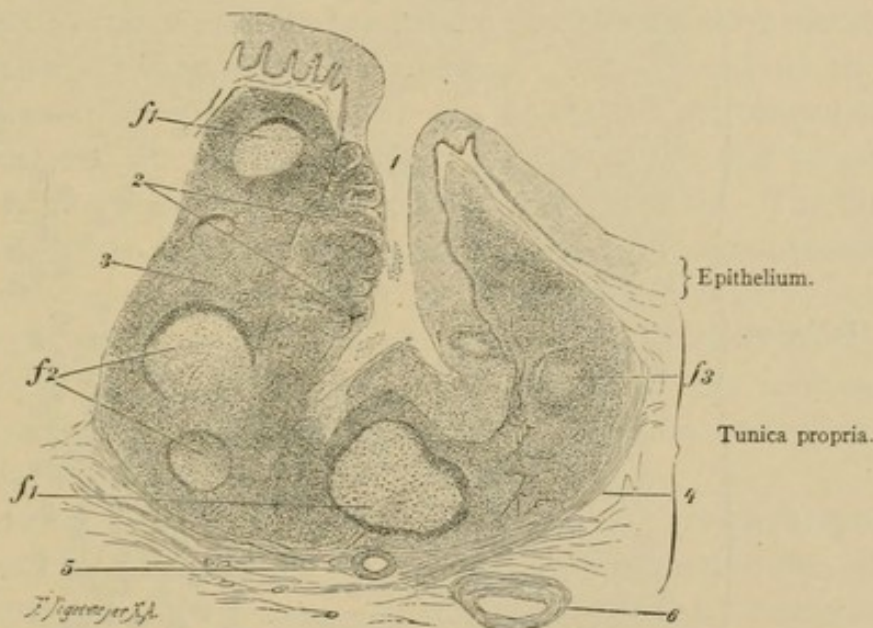


FIG. 186.—VERTICAL SECTION OF A LINGUAL TONSIL OF ADULT MAN. $\times 20$. 1. Crypt of the tonsil, containing migrated leucocytes. 2. Epithelium of the crypt, infiltrated with leucocytes on the left and at the base, almost intact on the right. 3. Nodules of adenoid tissue containing germinal centers: f^1 , nodule cut through the middle, f^2 , through the side, f^3 , at the periphery. 4. Fiber capsule. 5. Section of the excretory duct of a mucous gland. 6. Blood-vessel. Technic No. 102.

The *submucosa* at the tip and on the back of the tongue is firm and resistant (*fascia linguae*), and intimately connected with the underlying parts.

The lingual tonsils (*folliculi linguales*).—The mucous membrane of the root of the tongue extending from the vallate papillæ to the epiglottis is peculiarly modified by the development of the lingual tonsils. They are spherical aggregations of adenoid tissue, from 1 to 4 mm. in size, that, situated in the uppermost stratum of the tunica propria, form easily

*Not infrequently widely branched epithelial proliferations, in the form of deep-reaching pegs, occur in the vallate papillæ, that may separate by constriction from the surface epithelium and then represent concentrically stratified bodies, the "epithelial pearls."

perceptible macroscopic elevations. In the middle of the same a punctate opening* may be seen, the entrance to the narrow, deep *crypt*, which is clothed by a continuation of the stratified epithelium of the oral mucous membrane. Encircling this epithelium lies adenoid tissue, which contains a variable large number of lymph nodules with germinal centers (p. 145)

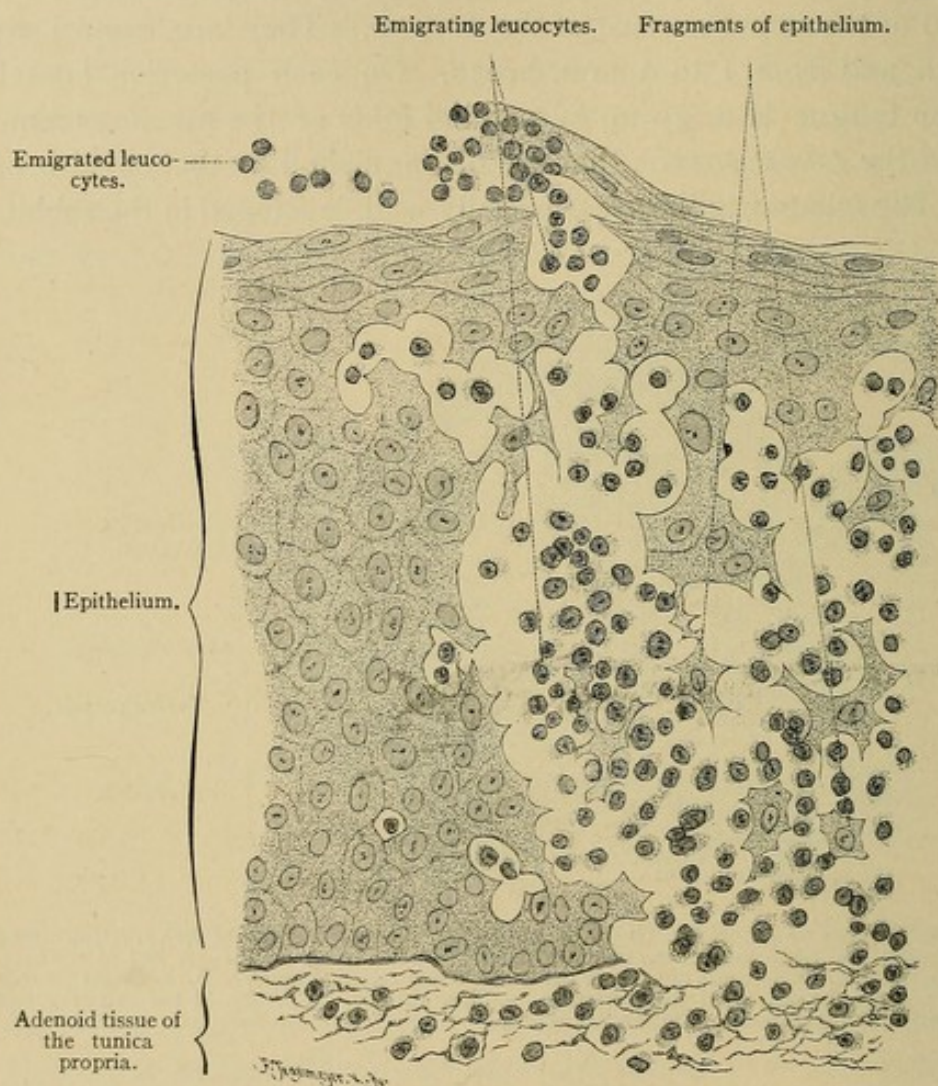


FIG. 187.—FROM A THIN SECTION OF A LINGUAL TONSIL OF MAN. $\times 420$. On the left the epithelium is free from leucocytes, on the right many leucocytes are wandering through. The epithelium is torn and smaller or larger fragments of it are seen lying between the broad passages made by the leucocytes. Technic No. 102.

and is sharply separated from the fibrillar connective tissue of the tunica propria; when the tonsils are well developed the connective tissue is arranged in circular strands about the adenoid tissue and so forms the *fiber capsule* (Fig. 186, 4). Under normal conditions numerous leucocytes of the adenoid tissue continually wander through the epithelium

* This was formerly regarded as the excretory duct of the lingual tonsil, which was regarded as a gland.

into the crypt * and from there into the mouth cavity ; they are readily found in the saliva, as "mucous" and "salivary" corpuscles. The *epithelium* is often much torn † in consequence (Fig. 187), or is infiltrated with leucocytes to such a degree that its boundary toward the tunica propria cannot be definitely determined.

Three kinds of branched *glands* occur in the lingual mucous membrane and in the superficial strata of the lingual musculature. The *serous* glands occur only in the vicinity of the vallate and foliate papillæ, the *mucous* glands in the root and along the edges of the tongue, the *mixed* anterior lingual gland (Nuhn) in the tip of the tongue. (Regarding the minute structure of these glands, see the chapter on The Glands of the Oral Cavity.)

The *blood-vessels* of the lingual mucous membrane form networks spread out parallel to the surface, from which twigs ascend to all the papillæ up into the secondary papillæ. At the root of the tongue small arteries pierce the fiber capsule of the lingual tonsils and break up into capillaries that penetrate to the interior of the nodules. The blood-vessels of the glands form capillary networks around the end-pieces.

The *lymph-vessels* of the tongue are arranged in two nets ; a deep net consisting of larger vessels, and a superficial net, which takes up the lymph-vessels of the papillæ. The lymph-vessels at the root of the tongue are very richly developed ; they form networks encircling the nodules of the lingual tonsils.

The *nerves* of the lingual mucous membrane, the glossopharyngeal and the lingual, contain ganglion cells, that occur scattered in the vallate papillæ and the wall and in groups—the so-called Remak's hemiganglia—beneath nearly every one of the walled papillæ ; the nerve endings behave partly as those of other portions of the oral mucous membrane, partly they enter into intimate relation with the taste-buds (*cf.* The Gustatory Organ).

THE SOFT PALATE AND THE PHARYNX.

The soft palate on its anterior surface is covered with a stratified squamous epithelium ; the tunica propria is furnished with tall papillæ and separated by a continuous layer of thick elastic fibers from the sub-mucosa. In the latter are found adipose tissue, cross-striped muscles, and a powerful, well-guarded stratum of mucous glands, the bodies of which often extend far into the muscles, while their long excretory ducts

* Regarding the rôle of the migrating leucocytes see p. 137, remark *.

† The gaps arising in this way close so soon as the leucocytes have wandered through.

are directed obliquely downward. The minuter structure agrees with that of the lingual mucous membrane. The posterior surface of the soft palate, upward for a distance from the free border, is clothed with a mucous membrane containing no adipose tissue, but otherwise of the same structure; at a level varying individually this changes into typical respiratory nasal mucous membrane with mixed glands (see The Olfactory Organ); the latter occasionally may be traced to the uvula.

The wall of the pharynx consists of three membranes: a mucous, a muscular, and a fibrous membrane. The mucous membrane, consisting of stratified squamous epithelium and a tunica propria with papillæ, is sharply separated from the muscular membrane by a robust layer of longitudinally disposed elastic fibers; this "elastic border-stratum" sends processes into the muscular membrane that embrace the individual muscle-fibers and gradually disappears downward, toward the beginning of the esophagus; upward, too, the border-stratum diminishes in thickness, but where the musculature is wanting it forms a dividing layer between the tunica propria and the submucosa* of the connective-tissue mucous membrane. Numerous alveolo-tubular branched simple glands, mucous glands of the structure of the lingual mucous glands, lie beneath the elastic border-stratum; their excretory ducts are often surrounded by aggregations of leucocytes. In the pharynx also atrophic mucous glands occur. In the upper division of the pharynx (*pars nasalis*) the epithelium changes into the many-row, ciliated cylinder variety, the lower limit of which is subject to considerable variation; the glands occurring here lie above the border-stratum and agree in structure with the mixed glands of the respiratory nasal mucous membrane.

Very richly developed is the adenoid tissue. Between the pillars of the fauces it forms conspicuous aggregations, one on each side, known as the palatine tonsils (*tonsilla palatina*), which in respect to their structure in man and in many animals correspond to a number of large lingual tonsils (p. 259). The leucocytes that wander through the epithelium of the tonsils into the crypts are so numerous that they may be regarded as the most fertile source of the salivary corpuscles. Many mucous glands lie in the neighborhood of the tonsils. The adenoid tissue is also vigorously developed in the nasal portion of the pharynx, where it forms a conspicuous mass, the "pharyngeal tonsil," which agrees in structure with the palatine tonsils, excepting that the adenoid tissue is less sharply circumscribed. Here, too, many leucocytes migrate through the

* Passing upward the submucosa becomes greatly strengthened and as the pharyngo-basilar fascia is attached to the base of the cranium.

epithelium. The development of all the adenoid tissue of the oral cavity and of the pharynx is subject to considerable variation.

The *muscular membrane* (the constrictor muscles of the pharynx) consists of striated muscle-fibers, the description of which belongs to the domain of macroscopic anatomy. The *fibrous membrane* is a dense-fibered connective tissue, richly interlaced with elastic fibers. Blood-vessels, lymph-vessels, and nerves are distributed in the same manner as in the oral cavity.

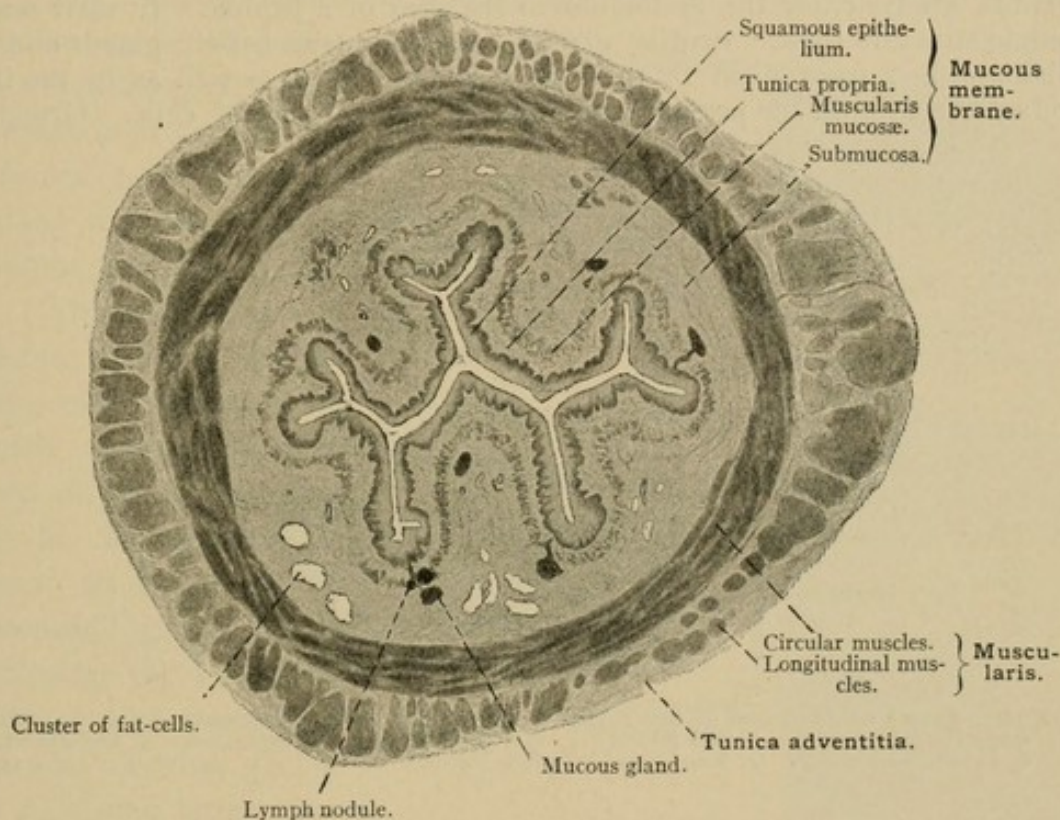


FIG. 188.—TRANSVERSE SECTION OF THE UPPER THIRD OF THE HUMAN ESOPHAGUS. $\times 5$. Technic No. 104.

B. RUMPGUT.

THE FOREGUT.

THE ESOPHAGUS.

The wall of the esophagus comprises a mucous, a muscular, and a fibrous membrane. The *mucous membrane* is composed of a stratified squamous epithelium (Fig. 189), of a tunica propria beset with papillæ, following this of a stratum of longitudinally disposed smooth muscle-fibers, the *muscularis mucosæ*; beneath this is the *submucosa*, which consists of loose bundles of connective tissue and contains small mucous glands of the structure of the lingual mucous glands. Their excretory duct, usually running obliquely cardia-ward, before its passage

through the muscularis mucosæ often is widened ampulla-wise ; attached to it within the territory of the tunica propria is a lymph nodule. The number of these glands fluctuates greatly individually ; as a rule they are more numerous in the upper half of the esophagus. Not seldom these glands, too, exhibit phenomena of degeneration (p. 246).

In addition to these glands of the submucosa the tunica propria of the extreme lower end of the esophagus, in a zone from one to four millimeters broad, contains branched tubular simple glands, with the excretory ducts often widened ampulla-shape, which, in contradistinction to those of the submucous glands, always enter the epithelium at the apex of a papilla. In their microscopic structure these "cardiac glands" resemble true gastric glands and are distinguished from them by their profuse branching, as well as by the individual variation in the presence or absence of the parietal cells. Groups of

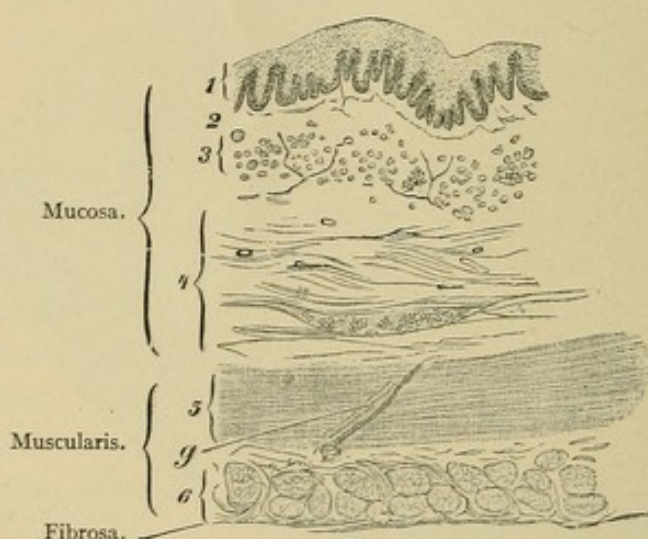


FIG. 189.—FROM A CROSS-SECTION OF THE MIDDLE THIRD OF THE HUMAN ESOPHAGUS. $\times 10$. 1. Stratified squamous epithelium. 2. Tunica propria. 3. Muscularis mucosæ. 4. Submucosa. 5. Circular muscles. 6. Longitudinal muscles. g. Blood-vessel. Technic No. 104.

just such glands lie laterally in the initial portion of the esophagus at the level between the cricoid cartilage and the fifth tracheal ring, occasionally also farther below ; their number, like that of the cardiac glands, is subject to great individual variation.*

The *muscular membrane* in the cervical portion of the esophagus consists of striated muscle-fibers, which in the lower portion are replaced by smooth muscle-fibers. The latter are arranged in two strata, an inner circular, in which the direction of the muscle-fibers is not everywhere exactly transverse, and an outer, not continuous longitudinal layer. The *fibrous membrane* consists of compact connective-tissue interspersed with numerous elastic fibers. The distribution of the blood-

* Examined with the unaided eye such groups have the appearance of erosions, because at these places the surface epithelium is not stratified squamous but gastric epithelium (page 265). The delicate epithelium possibly is the cause of the predisposition of this locality to pulsation diverticula ; the ampullæ favor the tendency to the development of cysts.

vessels, lymph-vessels and nerves is the same as in the pharynx. Between the circular and longitudinal layers of the muscularis the nerves form a net-like plexus, containing small groups of ganglion cells (see plexus myentericus, p. 281).

THE STOMACH.

The wall of the stomach is from 2 to 3 mm. thick and comprises three membranes: a mucous, a muscular, and a serous membrane.

The *mucous membrane*, sharply marked off from the white esophageal mucous membrane by its reddish-gray color, consists of an epithelium, a tunica propria, a muscularis mucosæ, and a submucosa (Fig. 190).

The *epithelium* is a simple cylinder epithelium, the elements of which produce mucus. Two divisions can usually be distinguished in them; an upper chiefly mucous (Fig. 25 c, p. 80), enclosing the centrosome, and a lower protoplasmic division (Fig. 25 p), which latter contains the oval, round, or flattened nucleus. The dimensions of the

mucous division (the secretion collecting center) vary greatly, according to the functional state of the cell (*cf.* Fig. 25). The gastric epithelial cells often closely resemble goblet cells (p. 273).

The *tunica propria* is composed of a mixture of fibrillar and reticular connective tissue, of elastic fibers, and of an extremely variable number of leucocytes, that occasionally lie closely aggregated and form solitary lymph nodules. The tunica propria contains so many *glands* that its tissue is limited to delicate septa between and a thin stratum below the tubules. In the pyloric division the glands are farther apart; there the tunica propria is conspicuously developed and not infrequently is elevated in filamentous or leaf-like villi.

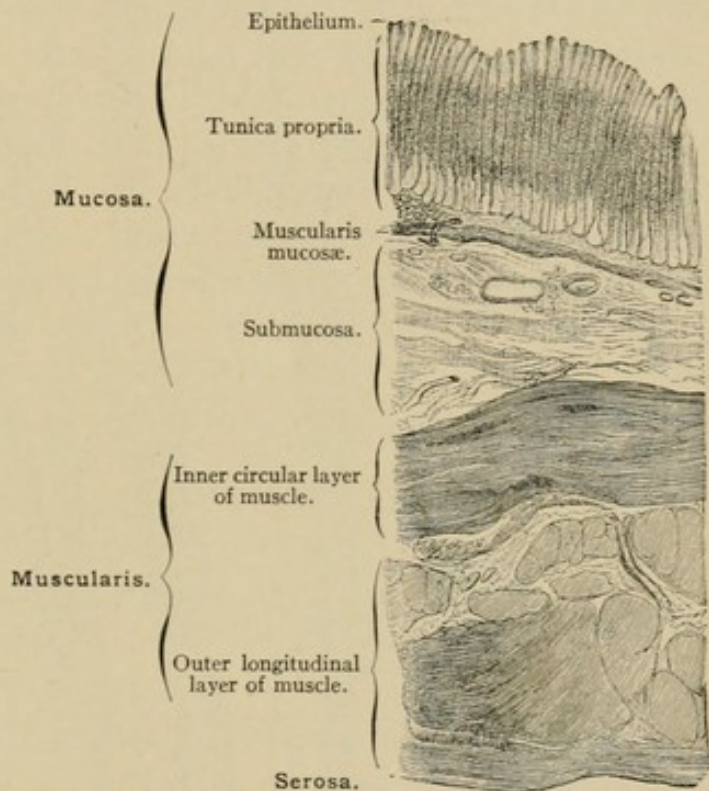


FIG. 190.—TRANSVERSE SECTION OF THE WALL OF A HUMAN STOMACH. $\times 15$. The tunica propria contains glands standing so close together that its tissue is visible only at the base of the glands toward the muscularis mucosæ. Technic No. 105.

The *glands* of the stomach are of two kinds: the one kind is situ-

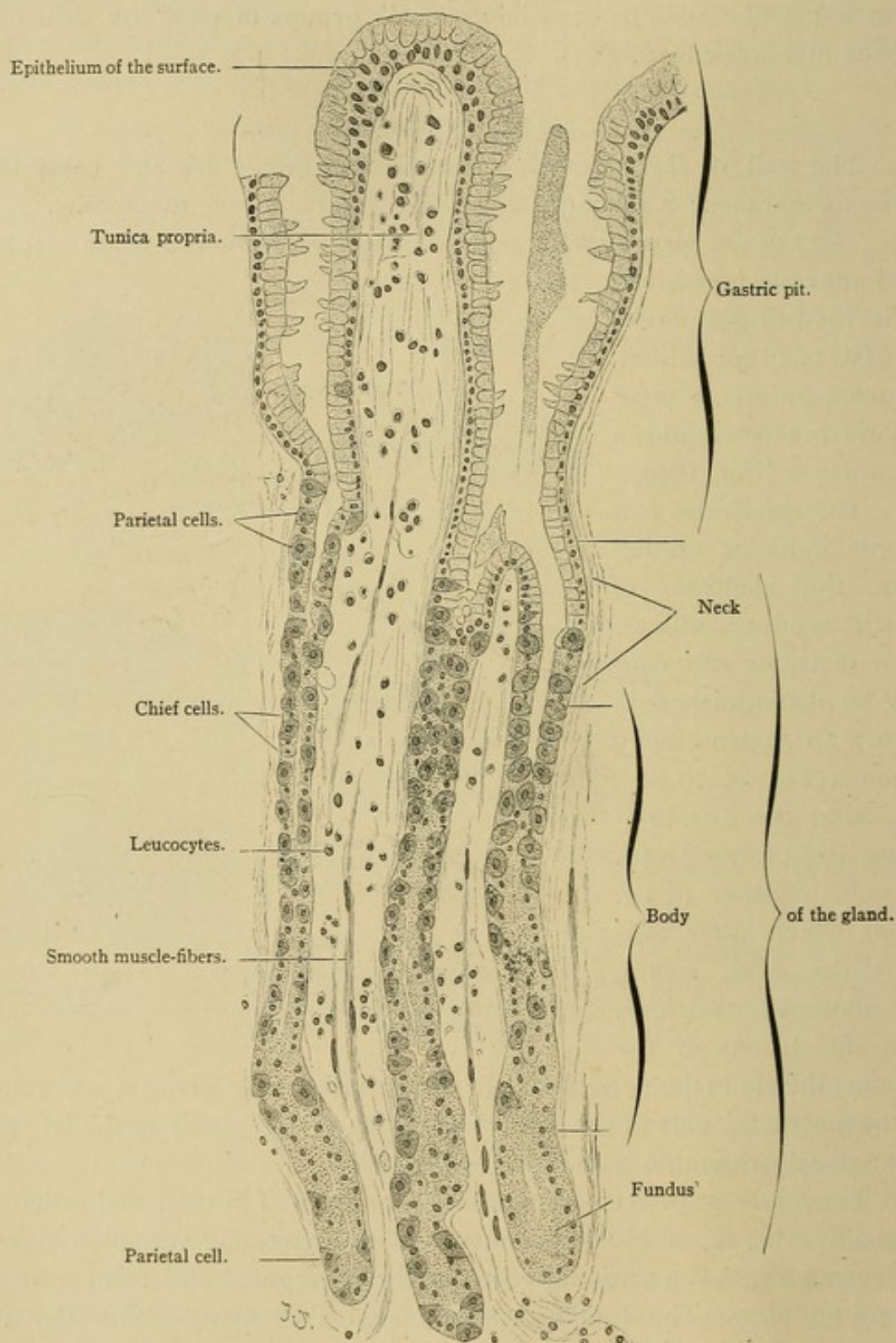


FIG. 191.—VERTICAL SECTION OF THE MUCOUS MEMBRANE OF A HUMAN STOMACH, IN THE VICINITY OF THE FUNDUS. $\times 220$. Technic No. 108.

ated chiefly in the body and the fundus of the stomach and they are

named *gastric glands* or *fundus glands** (*glandulæ gastricae propriae*); the other kind is confined to the small pyloric region and they are called *pylorus glands*. Both are branched (in particular the pylorus glands) or unbranched tubular simple glands (*cf.* p. 83), which, individually or in groups, open in pit-like depressions on the surface of the mucous membrane, the *gastric pits* (*foveolæ gastricae*). The portion of the gland opening into the gastric pit is named *neck*, the succeeding division, *body*, the blind end, *fundus* or base (Fig. 191).† Each gland consists of a *membrana propria* and of gland-cells.

The *fundus glands* have two kinds of cells: chief cells and parietal cells.‡ The chief cells are clear, cubical or short cylindric elements, with a granular protoplasm surrounding a spherical nucleus; they are exceedingly frail and unstable.§ The parietal cells are usually considerably larger, are darker and of polygonal shape; their finely granular protoplasm encloses a somewhat larger, spherical, often double nucleus. The parietal cells are especially marked by their affinity for anilin pigments, with which they stain intensely. The two kinds of cells are not equally distributed. The chief cells form the principal mass of the gland follicles; the parietal cells are irregularly distributed, but are especially profuse in the neck and the body. Here they lie in a row beside the chief cells; toward the gland fundus the parietal cells are pressed out of the line of the chief cells toward the periphery, however, without being wholly removed from the lumen, for a short, simple or multiple transverse canal (an intercellular secretory capillary) passes out from the lumen and extends between the chief cells to the parietal cell (Fig. 192). By the aid of Golgi's reaction, which blackens the secretion, it is most easily recognized that the transverse canaliculi are in connection with a cluster or with a basket-like network of intracellu-

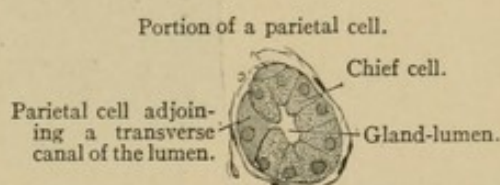


FIG. 192.—TRANSVERSE SECTION OF A HUMAN FUNDUS GLAND. X 240. Technic No. 108.

* In the earlier text-books the fundus glands were called peptic glands, a name based upon a function of the glands now called into question.

† Some authors distinguish the portion of the body of the gland adjoining the neck as the "intercalated division."

‡ The theory supported by different authorities that the chief and the parietal cells are different functional pictures of *one kind* of cells and also the statement that during digestion the parietal cells multiply, but disappear after prolonged fasting, are very much in need of thorough investigation. The stomach of an animal killed after a long winter hibernation still contains parietal cells.

§ The chief cells are said to produce pepsin, the parietal cells hydrochloric acid.

lar secretory capillaries, that is spread out within each parietal cell (Fig. 193 and Fig. 31, p. 86). The chief cells have no intracellular secretory capillaries, but short intercellular secretory capillaries occur between them.

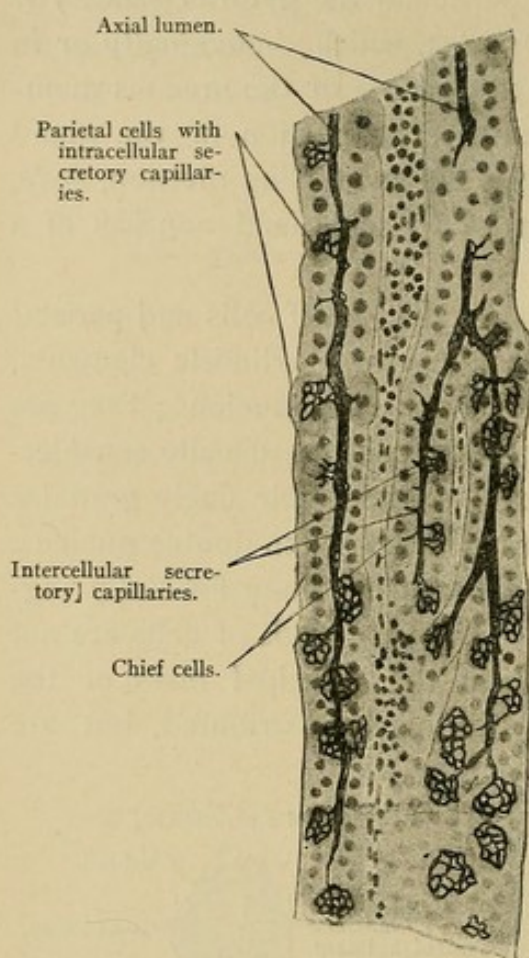


FIG. 193.—FROM A SECTION OF A HUMAN FUNDUS MUCOUS MEMBRANE. $\times 230$. Portions of fundus glands with blackened secretory passages. Technic No. 126.

frequently exhibit vacuoles, which have arisen because of the rapid and profuse formation of secretion, that cannot flow off quickly enough through the usual secretory capillaries. In dogs and cats it has been observed that after a day's fasting some of the parietal cells are without intracellular secretory capillaries, an evidence of their instability. In the territory of the cardia and of the pylorus are islets of mucous membrane, which in their minute structure fully agree with that of the small intestine.

The *muscularis mucosæ* consists of smooth muscle-fibers arranged in two or three superposed layers running in different directions, from which single strands branch off and ascend vertically between the gland follicles (Fig. 191).

The *submucosa* is composed of loose connective-tissue bundles and elastic fibers and occasionally small aggregations of fat-cells.

The *pylorus glands* (Fig. 194) are furnished almost throughout* with cylindric cells provided with a spherical nucleus situated near the cell-base, which in the intermediate zone, that is, the border-zone between the pylorus and the fundus mucous membrane, so very closely resemble the chief cells, they have been compared with them. In the pylorus glands only short intercellular secretory capillaries are found.

The elaboration of secretion in the chief cells as well as in the parietal cells is associated with the formation of granules (p. 80). During digestion the chief cells, also the cells of the pylorus glands, are darker, the nucleus of the latter is pushed more to the middle of the cell; the secretory capillaries of the parietal cells, expanded with secretion, are broader; after abundant meals the latter cells

* In man isolated parietal cells are found; in animals, e. g., the dog, a few dark, conical cells occur, that owe their appearance to the compression exerted by neighbor cells.

It is only in the pyloric region that two separate layers of smooth muscle-fibers can be distinguished in the *muscular membrane*, a thicker inner circular and a thinner outer longitudinal layer. In the other regions of the stomach the arrangement of the muscle tissue is very complicated,

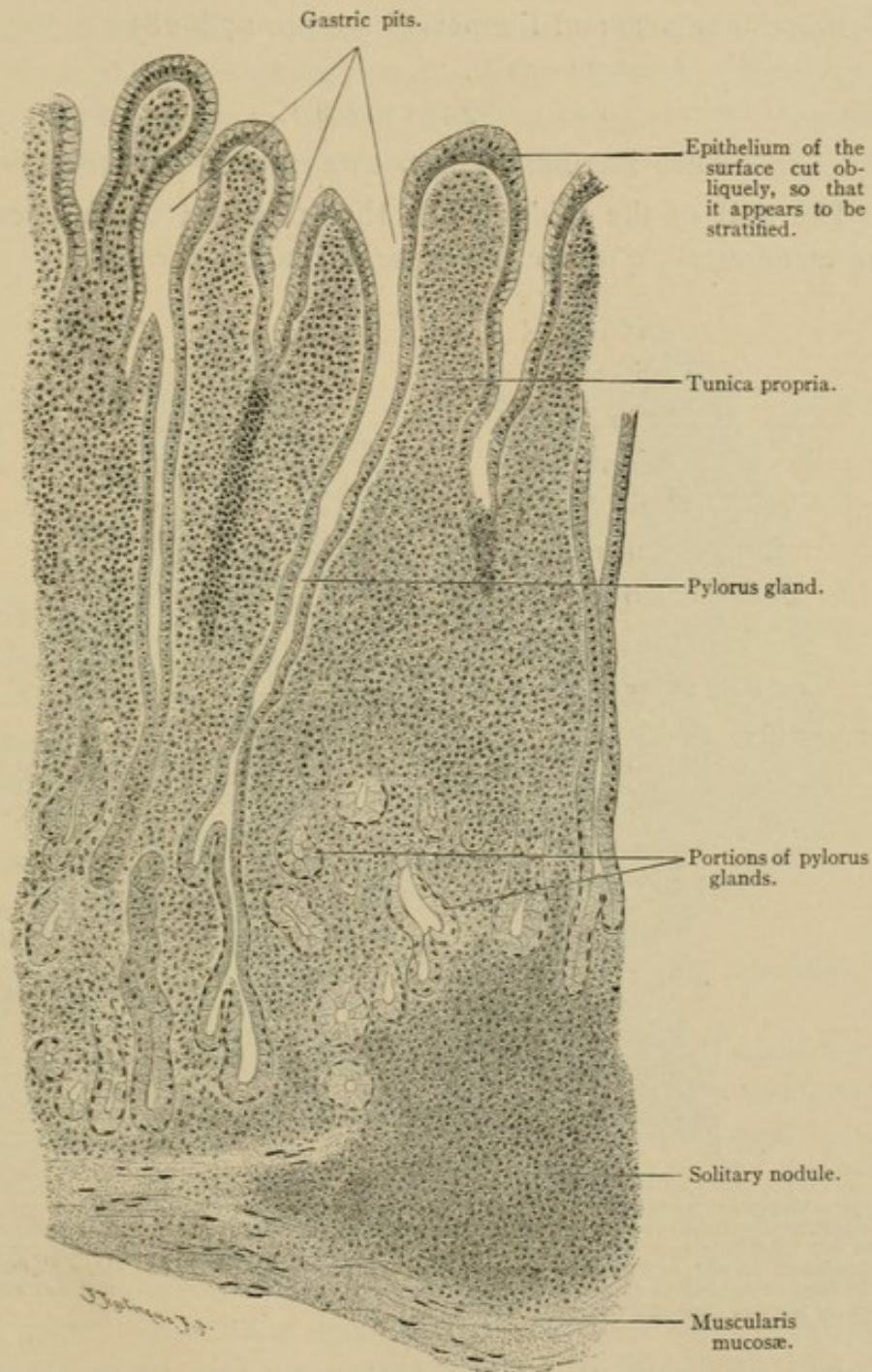


FIG. 194.—VERTICAL SECTION OF THE HUMAN PYLORUS MUCOUS MEMBRANE. $\times 90$. Technic No. 108 b.

owing to the extension of the muscular strata of the esophagus to the stomach, as well as to the curving of the organ that ensues in the course of development; sections exhibit bundles of fibers extending in every

possible direction (Fig. 190). (See further in the text-books on macroscopic anatomy.)

The elastic fibers behave as in the muscular membranes of the midgut (p. 275).

The *serous membrane* will be described with the peritoneum.

For the vessels and the nerves see pp. 278–281.

THE MIDGUT.

THE DUODENUM AND THE SMALL INTESTINE.

The wall of the midgut, like that of the stomach, is composed of three membranes, a mucous, a muscular, and a serous.

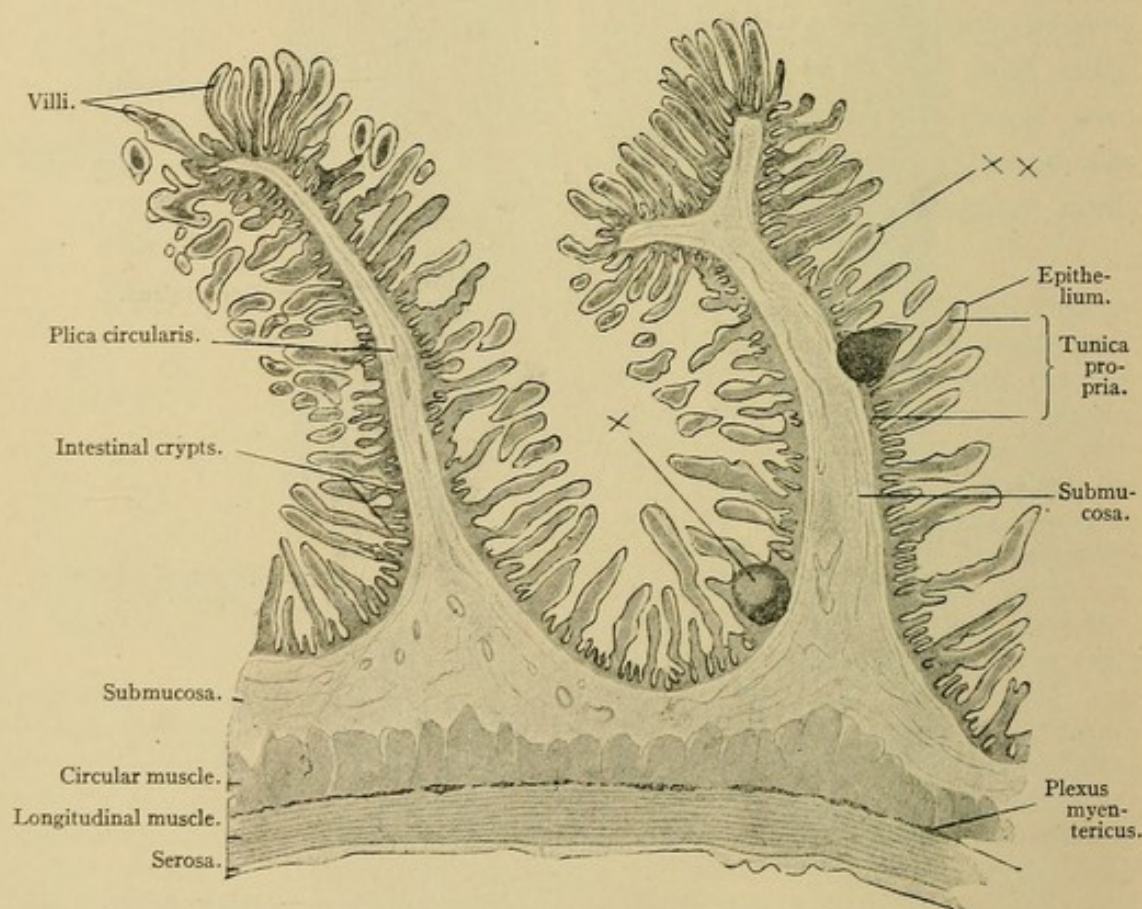


FIG. 195.—VERTICAL LONGITUDINAL SECTION OF THE JEJUNUM OF ADULT MAN. $\times 16$. The plica circularis on the right supports two small solitary nodules, that do not extend into the submucosa and of which the left exhibits a germinal center, X. The epithelium is slightly loosened from the connective-tissue core of many of the villi, so that a clear space, XX, exists between the two. The isolated bodies lying near the villi (more numerous to the left of the plicae circulares) are partial sections of villi that were bent, therefore not cut through their entire length. Technic No. 111.

The *mucosa* is thrown into circular folds, the *plicae circulares*, or *valvulae conniventes* (Kerkring), that are especially well developed in the upper part of the small intestine. In addition to these readily perceptible structures, the object of which is to increase the superficial extent of the *mucosa*, there are still other contrivances serving the same pur-

pose, that stand at the limit of macroscopic perception. These are the elevations and depressions of the mucous membrane. The former, the *villi*, are present only in the duodenum and the small intestine, in the end-gut of man they are wanting; they are about one mm. high, in the duodenum of leaf-like, in the remainder of the small intestine of cylindrical form.* The depressions begin at the pylorus and are found throughout the whole length of the intestine. They exist in their most primitive form in fishes, where they originate in longitudinal parallel folds of the mucous membrane that become connected by small

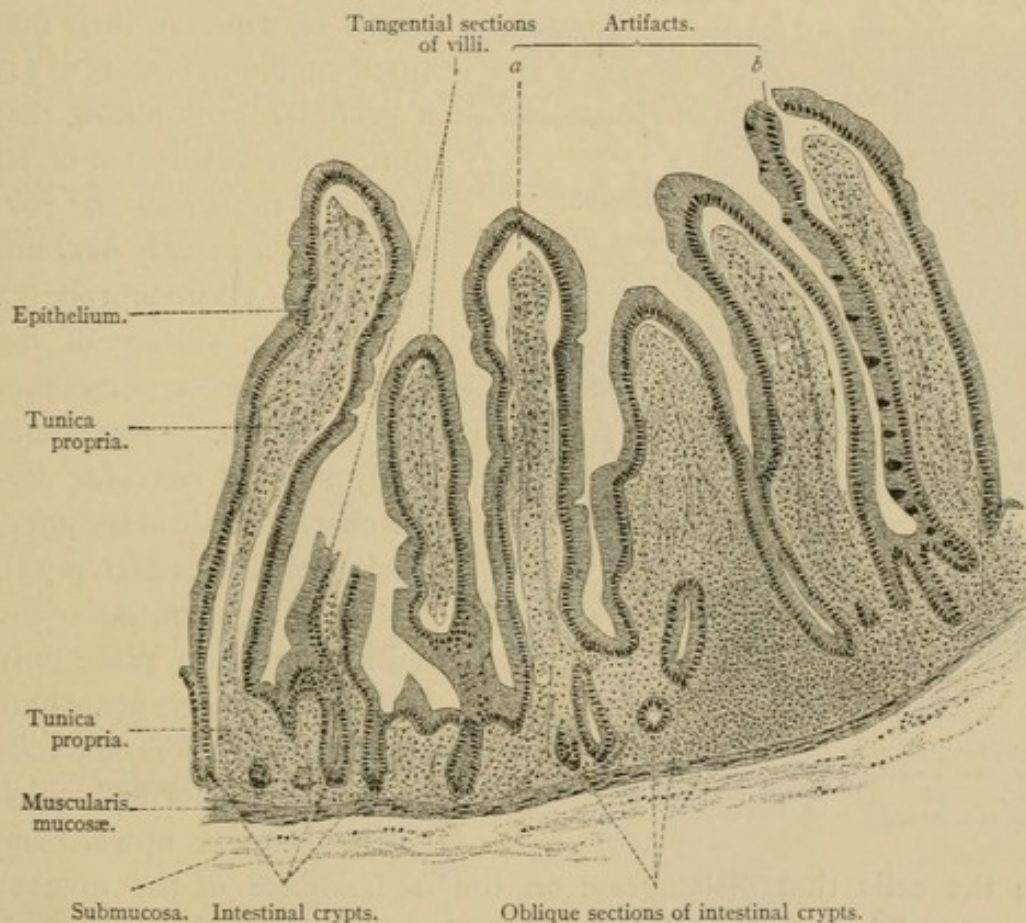


FIG. 196.—VERTICAL SECTION OF THE MUCOUS MEMBRANE OF THE JEJUNUM OF ADULT MAN. $\times 80$. The empty space, *a*, between the tunica propria and the epithelium of the villus is an artifact, the result of the shrinking action of the fixing fluid; not infrequently within the space lie cells that have been squeezed out of the tunica propria. On its retraction the epithelium often tears and then the villus appears to have an opening, *b*, at its apex. The goblet cells have been drawn on one side of the villus on the right. Technic No. 112.

transverse folds. In vertical sections these shallow depressions appear as short, wide sacks, and are called *crypts*. In mammals the crypts are deeper, their lumen is narrower, and in rows close beside one another

* Toward the lower end of the small intestine the villi gradually diminish in height and frequency, at the end of the ileum they are short, stand at greater intervals, and finally on the surface of the ileo-cecal valve directed toward the large intestine they entirely disappear.

they have the appearance of simple tubular glands; but they could only be regarded as such if their epithelial outfit produced a specific secretion, which is by no means the case.* Nevertheless, the name *intestinal glands* (Lieberkühn) has been retained. These glands, better crypts, of the duodenum and the small intestine are from 0.1 to 0.3 mm. long. Their blind end reaches to the muscularis mucosæ.†

The *mucous membrane* consists of an epithelium, a tunica propria, a muscularis mucosæ, and a submucosa. The *epithelium*, which clothes the entire free surface of the mucous membrane, envelops the villi and lines the crypts, is a simple cylinder epithelium (Fig. 17, p. 76), the elements of which in their ma-

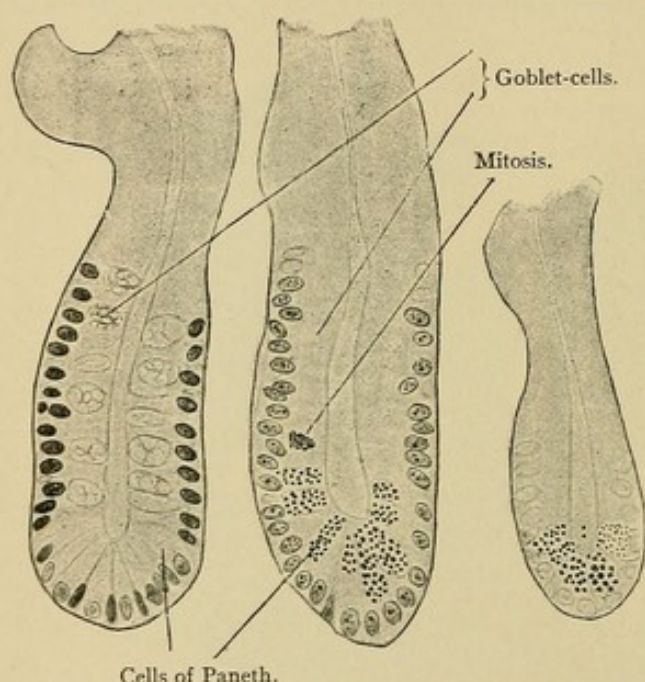


FIG. 197.—THREE INTESTINAL CRYPTS FROM SECTIONS OF THE ILEUM, THE TWO LARGER OF MAN, THE SMALL ONE OF A MOUSE. $\times 300$. The left crypt from a preparation fixed in Zenker's fluid, the other two after technic No. 120.

tured condition consist of (1) a granular protoplasm, that during the resorption of fat contains numerous fat-particles, (2) a usually oval nucleus, and (3) a membrane (?). On the free surface of the cells there is a sometimes homogeneous, sometimes finely striated *cuticular border* characteristic of the intestinal epithelial cell (*cf.* p. 75).

The regeneration of the epithelium takes place only in the intestinal crypts, where by mitotic division new cells are continually formed, which gradually move upward and

replace the cells that disintegrate on the free surface of the mucous membrane. Therefore the youngest generation of epithelial cells is

* In man and in rodents small groups of granule-containing cells (cells of Paneth) occur in the base of the intestinal crypts, that are to be regarded as specific gland-cells (Fig. 197). However, this does not by any means furnish the authority to consider all intestinal crypts as glands, for the cells of Paneth are not only entirely wanting in the carnivora, but even in man it is only in the ileum that they are invariably to be found, while they frequently are absent in the crypts of the duodenum and are altogether wanting in those of the large intestine. But even the existence of Paneth's cells in the ileum crypts gives us no right to regard the *entire* crypt as gland; only the blind end is comparable to a gland, the whole of the large division lying above being the equivalent of the gastric pit of the stomach.

† In a few instances they extend beyond into the submucosa; in this case they always lie in a lymph nodule. Such deep crypts are often found in the cat.

found in the crypts, the oldest on the free surface, in the small intestine on the tips of the villi. *Goblet-cells* in extremely variable numbers occur in the intestinal epithelium; they possess an elliptical, not infrequently a chalice-like form, their upper portion, that directed toward the surface of the intestine, is occupied in varying extent by the mucus into which the protoplasm is transformed, the nucleus with the remainder of the unaltered protoplasm lies at the base of the cell; a



FIG. 108.—INTESTINAL EPITHELIUM. $\times 560$. A. Isolated goblet-cells of a rabbit. x. Escaping mucus. Technic No. 110 a. B. From a section of the mucous membrane of the human intestine. b. A goblet-cell between cylinder cells. Technic No. 111.

cuticular border is wanting, in place of which a sharply defined circular orifice is found (Fig. 198 A) through which the mucus is poured out on

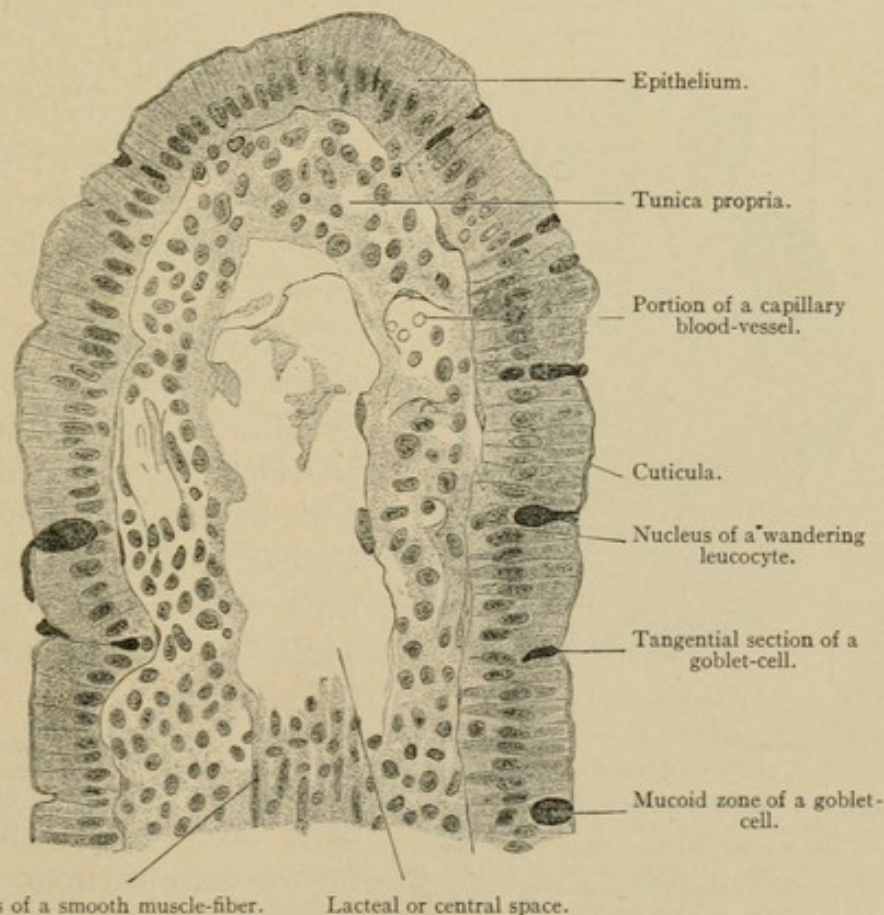


FIG. 199.—LONGITUDINAL SECTION THROUGH THE APEX OF THE VILLUS OF A DOG. $\times 360$. The goblet-cells contain the less mucus the nearer they lie to the summit of the villus. Technic No. 112.

the surface of the intestine. The goblet-cells are derived from the ordinary epithelial cells of the intestine. In suitable conditions each young

young intestinal epithelial cell can become a goblet-cell* and produce mucin.

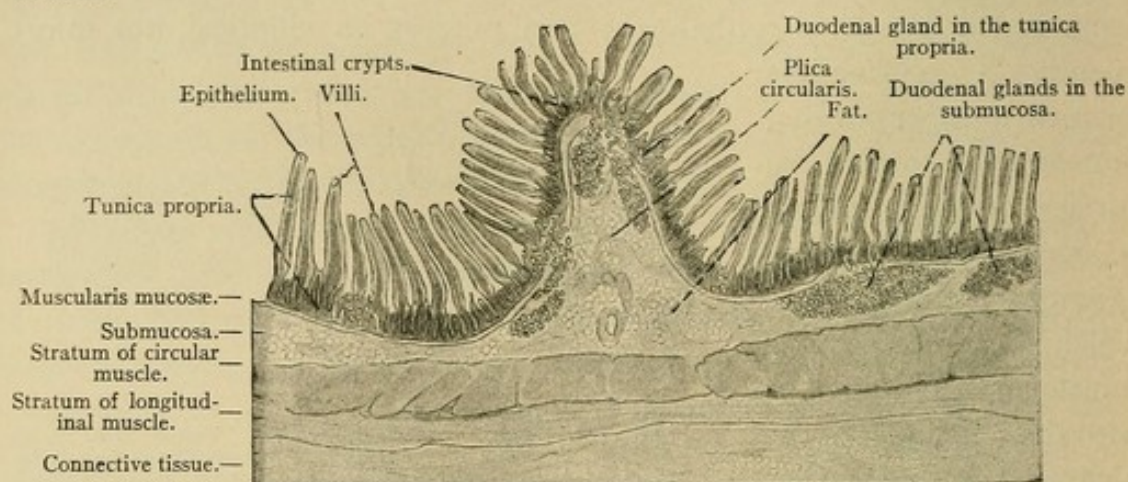


FIG. 200.—VERTICAL LONGITUDINAL SECTION OF THE HUMAN DUODENUM. $\times 16$. Technic No. 109.

The separate phases of secretion appear in regular sequence and so that the later stages are always higher, near the tips of the villi (Fig. 199), than the initial stages, which are found in the intestinal crypts.

Between the epithelial cells migratory leucocytes from the underlying tunica propria are found in varying number.

The *tunica propria* forms the bodies of the villi and fills the spaces between the intestinal glands, at the blind end of which it is arranged in a thin stratum. It consists chiefly of reticular and fibrillar connective tissue intermingled with elastic fibers, that contains a widely varying quantity of leucocytes (cf. p. 145).

The *muscularis mucosae* consists of an inner circular and an outer longitudinal layer of smooth muscle-fibers. Fibers derived from the muscularis mucosae ascend within each villus

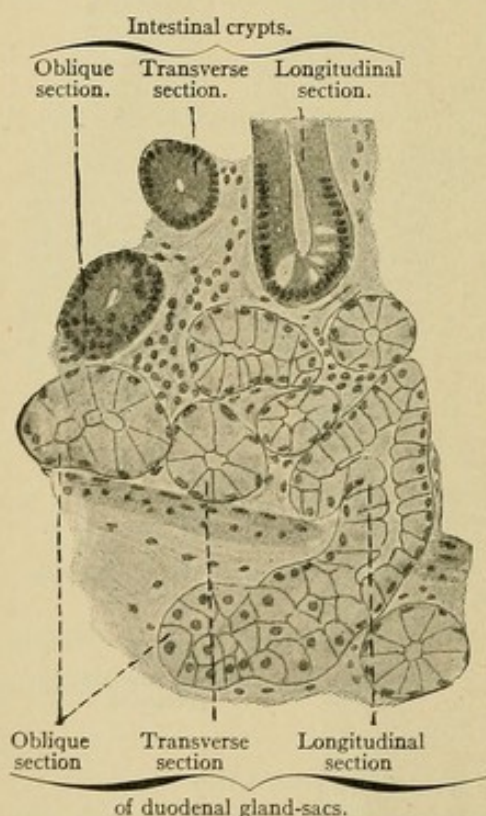


FIG. 201.—FROM A SECTION OF A HUMAN DUODENUM. $\times 240$. Only the lower half of the mucosa and upper half of the submucosa are sketched. A large portion of the duodenal gland lies above the muscularis mucosae. Technic No. 109.

nearly to its apex. Their contraction effects a shortening of the villus.† The elastic fibers behave as in the muscle membrane.

* In regard to the mode in which the goblet-cells produce secretion, see p. 81.

† Cf. Technic No. 110, p. 301.

The *submucosa* consists of loose fibrillar connective tissue with a sparing admixture of elastic fibers; in the territory of the duodenum it contains compound alveolo-tubular glands, from 0.2 to 3.4 mm. in size, the *duodenal glands* (Brunner).^{*} In man they lie densely crowded at the sphincter of the pylorus, but diminish in number down therefrom. In the vicinity of the cystic duct they are again more profuse; toward the end of the duodenum they vanish entirely. Their excretory duct, clothed with simple cylinder epithelium, pierces the muscularis mucosæ and opens either into the base of an intestinal crypt or, running parallel with the latter in the tunica propria, on the inner surface of the intestine. The walls of the alveolo-tubules are formed of a structureless membrana propria and of cylindric gland-cells, resembling the cells of the pylorus glands.

The muscle tunic of the intestine consists of an inner, robust circular and an outer, slighter longitudinal stratum of smooth muscle-fibers. Numerous elastic fibers lie, not only on the outer and inner surfaces of both muscle strata, but also within the strata. Their number stands in direct relation to the thickness of the musculature.

For the structure of the *serosa*, see the Peritoneum, p. 295.

THE ENDGUT.

I. THE LARGE INTESTINE.

The wall of the large intestine likewise consists of a mucous membrane, a muscular membrane and a serosa.

The mucous membrane is smooth, villi are wanting, and the crypts are twice as long (0.4 to 0.6 mm.) as those of the small intestine. The epithelium, tunica propria, and muscularis mucosæ are the same as those of the small intestine, with which they also agree in their microscopic structure and in the regeneration of the epithelium. The glands contain a relatively large number of goblet-cells.[†]

^{*} The bodies of the duodenal glands do not all lie exclusively in the submucosa; often portions, in fact near the end of the duodenum entire bodies of duodenal glands are found in the territory of the tunica propria. In the cat duodenal glands in process of atrophy are frequently found.

[†] The reason for this lies in the fact that the young epithelial cells originating in the glands of the small intestine move more rapidly to the surface; for the superficies of the small intestine, so greatly augmented by the villi, requires a larger reparative supply to replace the cells perishing there; therefore the elaboration of mucus often does not take place within the crypts, but first begins in the cells on the villi. In the large intestine, where the villi are absent, the transit to the surface takes place slowly and the cells have time to produce secretion during their sojourn in the crypts. It is this that gave rise to the erroneous impression that the glands of the small intestine yield a serous fluid, the glands of the large intestine mucus.

The muscular coat of the large intestine consists besides elastic fibers of an inner annular and an outer longitudinal layer of muscle; the latter is well-developed only within the territory of the *tæniæ*, being extremely thin in the intervals. The serosa agrees in its minute structure with that of the small intestine.

The vermiform process of man is characterized by a large number of round, in old persons flat, lymph nodules (see below) and is in many cases—25 per cent.—partially obliterated; this process occurs with increasing frequency

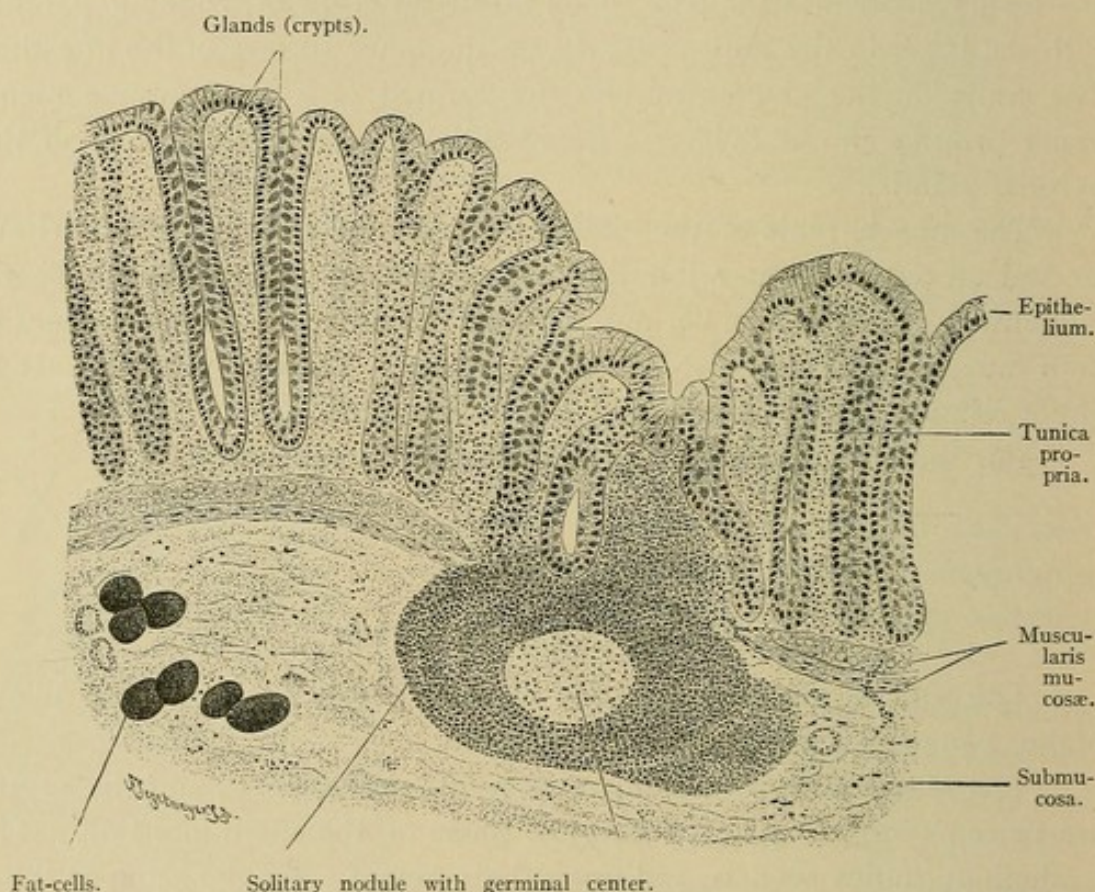


FIG. 202.—VERTICAL SECTION OF THE MUCOUS MEMBRANE OF THE DESCENDING COLON OF ADULT MAN. $\times 80$. Compare the length of the glands with those of the small intestine (Fig. 196), that are from the same individual and drawn under the same magnification. Technic No. 114.

in advanced age—in 50 per cent. of persons over 60 years. The obliteration is not the result of a pathologic process, but the effect of the elsewhere familiar involution. Epithelium and glands perish, lymph nodules disappear, and the tunica propria is transformed into an axial connective-tissue strand, that is enclosed in the unaltered submucosa and muscularis. These post-embryonal processes must not be confused with the kataplastic changes in the intestinal glands, occurring from the 5th to the 6th embryonal month, which so far have been observed only in man.

2. THE RECTUM.

In composition and structure the rectum in general agrees with the large intestine, but is distinguished by its longer glands (0.7 mm.) and

by a thick longitudinal layer of muscle. At the upper end of the columnæ rectales begins the transition of the mucous membrane into the skin; instead of the simple cylindrical epithelium, a powerful stratified squamous epithelium appears, which covers a tunica propria with vascular papillæ. The intestinal glands may be traced for a short distance into the territory of the stratified squamous epithelium, but farther on they are wholly wanting. The columnæ rectales contain smooth muscle-fibers.

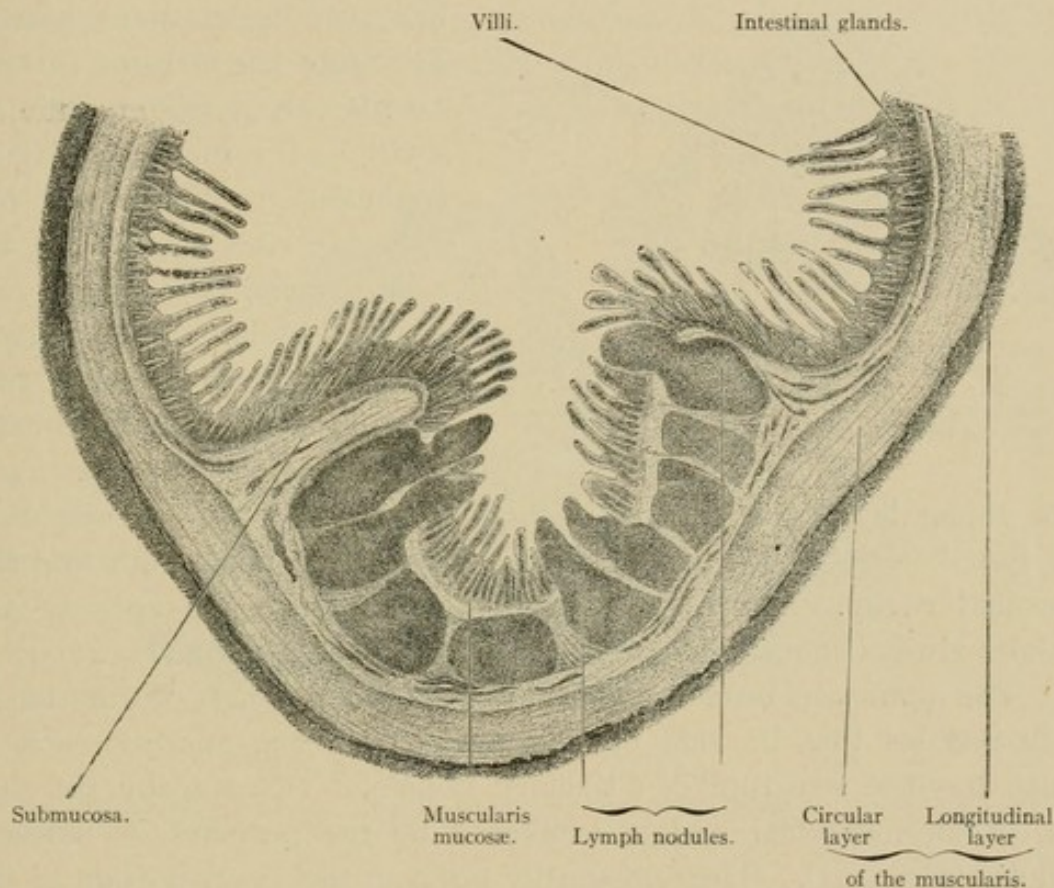


FIG. 203.—TRANSVERSE SECTION OF A PATCH OF PEYER OF THE SMALL INTESTINE OF A CAT. The crests of four nodules were not within the plane of the section. $\times 10$. Technic No. 113.

THE LYMPH NODULES OF THE STOMACH AND THE INTESTINES.

It has been previously mentioned (p. 145) that the tunica propria of the mucous membranes contains leucocytes in variable numbers, either distributed as diffuse adenoid tissue or balled together in circumscribed masses. In the latter case they form nodules from 0.1 to 2.5 mm. large, which stand isolated, as the *solitary nodules*, or united in groups as the *agminated nodules*.

The *solitary nodules* ("solitary follicles") occur in greatly varying number in the gastric mucous membrane, in larger number in the intes-

tines. They usually possess an oval form and in the beginning of their development always lie in the tunica propria; * their summit extends up close under the epithelium, their base is directed toward the muscularis mucosæ. With progressive growth (in cats at the time of birth) they break through the muscularis mucosæ and expand in the submucosa, where the loose tissue offers but little resistance. The part of the nodule lying in the submucosa has a spherical form and soon becomes considerably

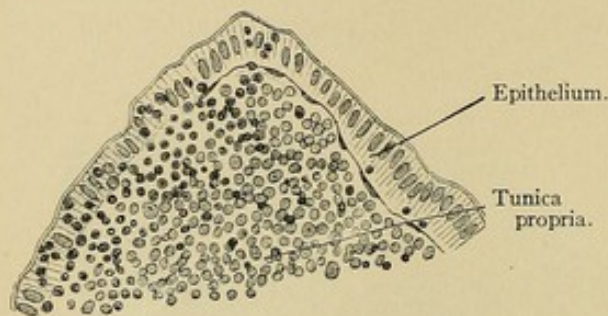


FIG. 204.—FROM A SECTION OF THE SMALL INTESTINE OF A SEVEN-DAY-OLD KITTEN. $\times 250$. Crest of a solitary nodule. The epithelium on the left contains many wandering leucocytes. The epithelium on the right contains but three leucocytes. Technic No. 113.

larger than the division lying within the tunica propria. Therefore the matured solitary nodules are pyriform, with the small end turned toward the epithelium. Where the nodules are situated the villi are wanting and the crypts are pushed aside. The solitary nodules are composed of adenoid tissue and usually contain a germinal center (p. 146).

The young leucocytes formed in them in part pass into the neighboring lymph-vessels and in part wander through the epithelium into the intestinal cavity. The cylinder epithelium covering the apex of the nodules always contains leucocytes in course of migration (Fig. 204).

The *agminated nodules* (patches of Peyer) are groups of from ten to sixty nodules that lie side by side, never over one another, each of which has the structure of a solitary nodule. Occasionally the outline of an individual nodule is altered by the pressure of adjacent nodules (Fig. 203). They principally occur in the lower portion of the small intestine, always on the side opposite to the attachment of the mesentery, either distinctly isolated from one another or transformed into a diffuse mass of leucocytes, in which only the germinal centers can be distinguished. This is not infrequently the case in the vermiform process of man.

THE BLOOD-VESSELS OF THE STOMACH AND OF THE INTESTINES.

The blood-vessels of the stomach and of the large intestine have a precisely similar distribution, which is modified in the small intestine by the presence of the villi. In the stomach and in the large intestine the entering arteries first give off small branches to the serosa, then pierce the muscularis, which they supply, and then in the submucosa

* This is also their usual seat in the human small intestine, while in the large intestine they also extend into the submucosa (cf. Fig. 195 with Fig. 202).

form a network extending parallel to the surface. From this small twigs ascend through the muscularis mucosæ into the tunica propria, where at the base of the glands they form another network spread parallel to the surface. Fine capillaries (from 4.5 to $9\ \mu$ wide) develop from the latter, form plexuses that envelop the glands and pass into capillaries twice as wide (from 9 to $18\ \mu$), which latter form a plexus that lies wreath-like about the mouths of the glands. Venules take their origin from the wide capillaries, which without taking up other branches pass vertically down between the gland tubulæ and open into a venous plexus spread parallel to the surface in the tunica propria; in their further course the veins run alongside the arteries. The veins

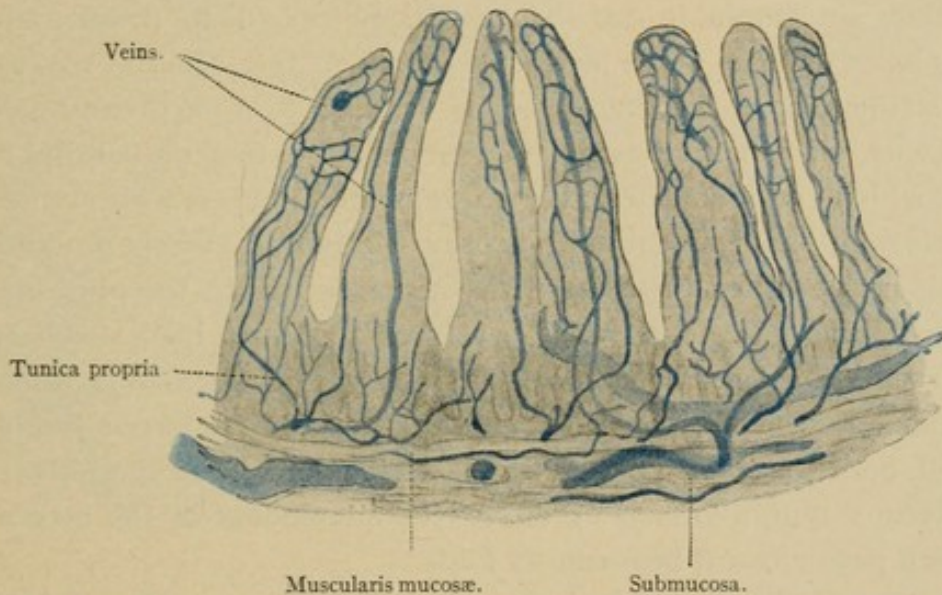


FIG. 205.—VERTICAL SECTION OF THE MUCOUS MEMBRANE OF THE HUMAN JEJUNUM. $\times 50$. The blood-vessels are injected with Berlin blue. The vein of the first villus on the left is cut transversely. Technic No. 116.

arising from the venous plexus in the submucosa are furnished with valves to the point where they empty into the approaching, parallel collecting veins of the intestine. The remaining branches and the trunk of the portal vein are without valves.

In the small intestine only the arteries supplying the crypts are distributed in the same manner as in the large intestine. The villi are provided with one artery (several when the villus is broad), which lies opposite the vein; from the former capillaries arise that lie close under the epithelium and obliquely or vertically to the long axis of the villus pass into the veins.* The further course of the veins is the same as in the large intestine.

* The distribution is the same in the dog, but in the rabbit and the guinea-pig the arteries going to the villi break up into fine branches that run to the base of the villus and then form a

The duodenal glands are enveloped in a capillary plexus which is supplied by the blood-vessels of the submucosa.

The lymph nodules ("follicles") are surrounded by a superficial capillary network, from which fine capillaries extend into the interior; often these do not penetrate to the center, in which case a non-vascular spot exists in the middle of the nodule.

THE LYMPH-VESSELS OF THE STOMACH AND OF THE INTESTINES.

The lymph- (chyle) vessels of the stomach and of the large intestine begin in the mucous membrane as blind capillaries, about 30μ wide, and descend between the gland follicles. In the mucous membrane of the small intestine the lymph-vessels begin in the axis of the villi; in cylindrical villi they are simple, in leaf-shaped villi multiple ducts (from 27 to 36μ wide) closed at their upper end, and represent the "central space" of the villus (Fig. 199). All these vessels descend to a narrow-meshed capillary plexus lying at the base of the glands and extending parallel to the surface, which communicates by numerous anastomoses with a wide-meshed horizontal plexus in the submucosa; the lymph-vessels proceeding from this network are provided with valves, penetrate the muscular coat and here take up the efferent vessels of a plexus lying between the circular and longitudinal muscular strata. This plexus is called the interlamina lymph-vessel plexus, and takes up the numerous lymph capillaries of both muscular layers. Beneath the serosa the lymph-vessels ("subserous lymph-vessels") run to the attachment of the mesentery and then pass onward between its folds.

The course of the lymph-vessels just described is modified in the mucosa of certain localities. These places are the patches of Peyer; the nodules, which never contain lymph-vessels, press aside the capillaries, which run in the interstices between them, as canals diminished in number, but increased in caliber. It is probable that the lymph-sinuses of the rabbit (p. 146, remark*) are nothing else than such immensely widened, flattened capillaries.

THE NERVES OF THE STOMACH AND OF THE INTESTINES.

The numerous nerves, mainly consisting of nonmedullated, sympathetic fibers, form a plexus beneath the serosa, then pierce the longitudinal layer of the muscular tunic and spread out between this and the

capillary network that lies close under the epithelium. At the summit of the villus the capillaries open into a small venous trunk, that in the course of its vertical descent takes up the capillaries surrounding the mouths of the glands. I have found the same arrangement in the broader villi of man.

circular muscle layer in a conspicuous network, the *plexus myentericus* (Auerbach), which is furnished with numerous groups* of multipolar ganglion cells, usually found at the nodal points of the network. The meshes of the plexus are polygonal. From this network bundles of nonmedullated nerve-fibers are given off, usually at right angles, part of which supply the longitudinal and circular strata of the muscular tunic, part of which pierce the latter and enter the submucosa. In the musculature the nerves form a rich, rectangular-meshed network, from which nerve-fibers turn aside and after repeated division approach the muscle-fibers, on

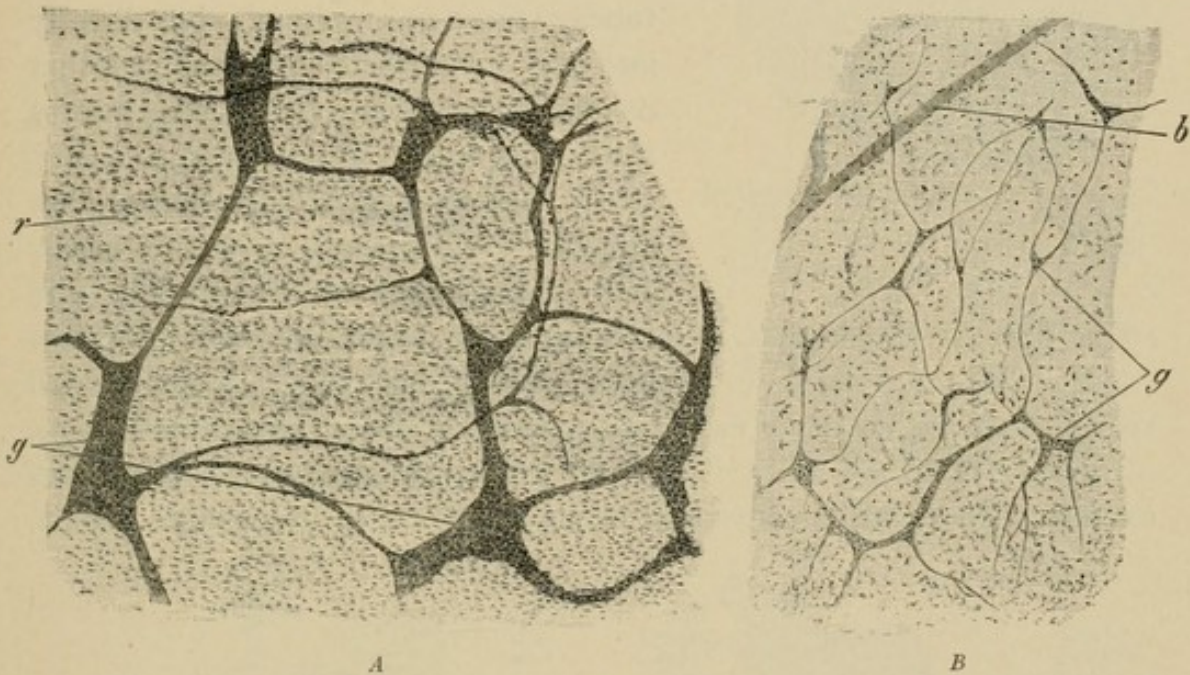


FIG. 206.—A. SURFACE VIEW OF THE PLEXUS MYENTERICUS OF AN INFANT. $\times 50$. g, Groups of ganglion cells; r, layer of circular muscle-fibers, recognized by their rod-shaped nuclei. Technic No. 117 a.
B. SURFACE VIEW OF THE PLEXUS SUBMUCOSUS OF THE SAME INFANT. $\times 50$. g, Groups of ganglion cells; b, blood-vessel shimmering through the overlying tissue. Technic No. 117 b.

which (not within) they terminate in free, slightly swollen endings. The nerves that go to the submucosa there form a second, delicate plexus, the *plexus submucosus* (Meissner), the meshes of which are narrower and the groups of ganglion cells† smaller. From this spring numerous fibers, which enter the tunica propria and in part weave a nervous net about the glands, in part enter the villi, where they terminate free in the parenchyma of the villus or close beneath the epithelium, without connection with the epithelial cells.

* These groups—small ganglia—behave similarly to the sympathetic ganglia and contain chiefly cells of type i (cf. p. 218).

† Their elements belong mainly to type ii (p. 218) and with their dendrites may reach to beneath the epithelium.

A plexus corresponding to the plexus myentericus also occurs between the layers of the muscular membrane of the esophagus.

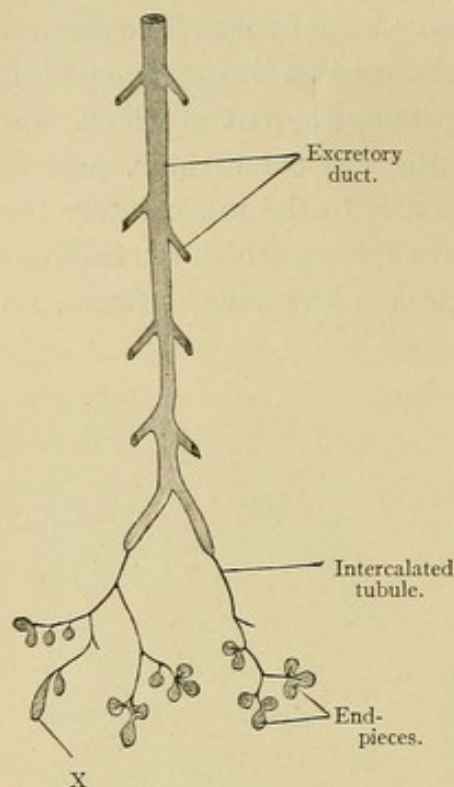


FIG. 207.—SCHEME OF THE HUMAN PANCREAS. X, tubular end-piece.

THE PANCREAS.

The pancreas is for the smaller part a tubular, for the greater part an alveolar gland (*cf.* p. 84). Its canal system consists of an excretory duct, the branches of which do not lead into secretory tubes,—these are wanting,—but directly into very long intercalated tubules, that dividing repeatedly pass into mostly short end-pieces.

The excretory ducts, the pancreatic duct (Wirsungi) and the accessory pancreatic duct (Santorini), are composed of a simple cylindric epithelium and of connective tissue, which latter is denser beneath the epithelium, looser toward the periphery. The chief excretory duct and its coarser branches carry in their walls small glands,

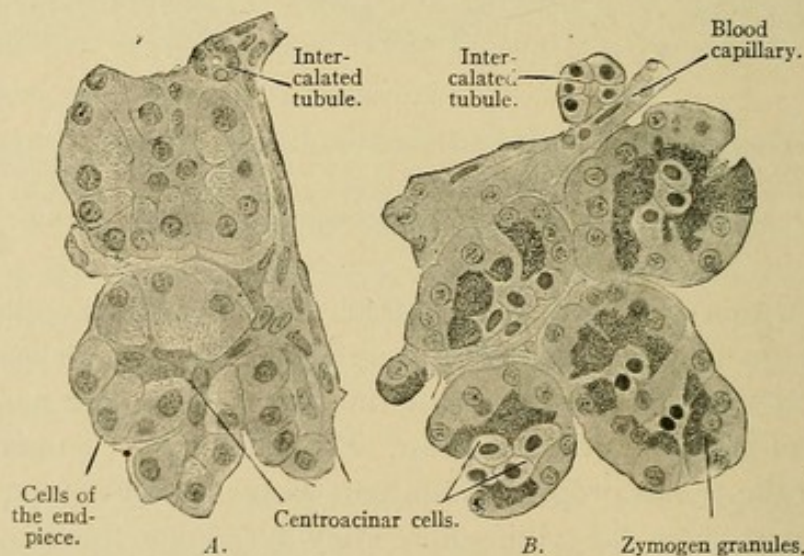


FIG. 208.—FROM SECTIONS OF A HUMAN PANCREAS. $\times 500$. In section A the granules are wanting, the elements of the intercalated tubule are flat and dark; in section B the granules are distinct, the cells of the intercalated tubule are cubical and clear. Technic No. 119.

the elements of which resemble mucous cells.* The cylinder epithelial cells of the smaller branches steadily decrease in height and finally pass

* Regarding the musculature, see remark *, p. 287.

into the longitudinally disposed cubical or plate-like cells of the intercalated tubules. These cells of the intercalated tubules do not directly annex themselves to the epithelium of the end-pieces, as, for example, is the case in the submaxillary gland (Fig. 168, right), but as the so-called *centroacinar cells* shove themselves into the interior of the end-pieces, whereby they come to lie upon the inner surface of the secreting cells* (Fig. 208). The latter are small conical cells, that in the zone directed toward the lumen contain numerous highly refracting granules, the *zymogen granules*, precursors of the secretion. In fresh preparations they are visible even with relatively low magnification (Fig. 232); the clear peripheral division of the cell contains the round nucleus. The relative proportions of the granular and clear zones vary according to the functional state of the cell. In the beginning of digestion the granules disappear, while the clear zone of the cell becomes larger. Then the granular zone again enlarges, to such extent that it occupies nearly the entire cell. In

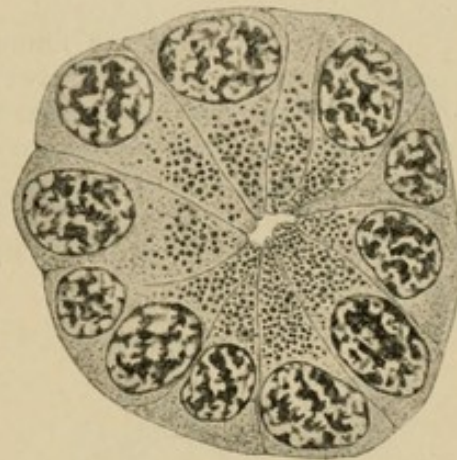


FIG. 209.—TRANSVERSE SECTION OF A GLAND-TUBULE OF THE PANCREAS OF NECTURUS; showing zymogen granules. $\times 400$.—(Schäper.)

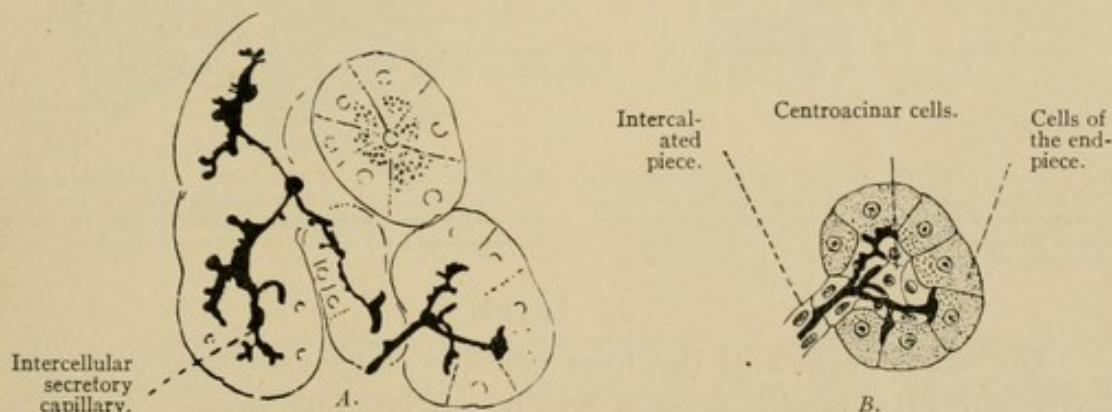


FIG. 210.—A. FROM A SECTION OF THE PANCREAS OF ADULT MAN. $\times 320$. Technic No. 126.
B. SCHEMATIC ELABORATION OF THE RIGHT LOWER PORTION OF A.

the fasting state the two zones are of equal size (*cf.* p. 80).† Intercellular secretory capillaries extend from the axial lumen between the gland-cells, without reaching to the membrana propria; where the centroacinar cells shut off the gland-cells from the central lumen the latter pour their secre-

* Owing to this the microscopic picture is a very complicated one and, with the lumen not always visible and the many unavoidably oblique sections, very difficult to understand (*cf.* in particular Fig. 208 B). The centroacinar cells cannot be demonstrated in all end-pieces.

† The cells of the intercalated tubules also show changing states (Fig. 208).

tion into secretory capillaries which penetrate between the centroacinar elements and open into the axial lumen (Fig. 210 B).

The blood- and lymph-vessels, as well as the nerves, behave as in the glands of the oral cavity.

In the pancreas are found groups of epithelial cells arranged in solid cords, varying in number, always small,—measuring up to 0.3 mm.,—called the *intertubular cell-groups*, that usually are separated from the remaining tissue of the pancreas by a scanty amount of connective tissue poor in elastic fibers. They are penetrated by wide capillaries and have a certain resemblance to liver tissue. Gland lumina have not yet been demonstrated in mammals. The meaning of these structures is still uncertain.

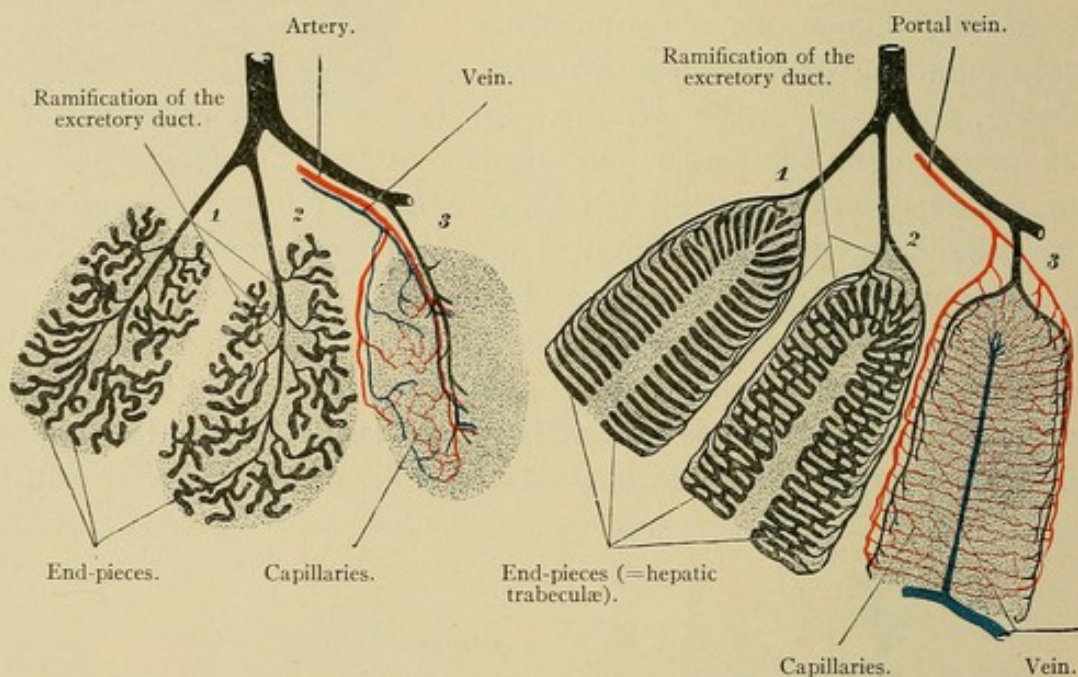


FIG. 211.—SCHEME OF AN ORDINARY COMPOUND TUBULAR GLAND. In lobule 3 only the ramifications of the excretory duct, without the end-pieces, are sketched.

FIG. 212.—SCHEME OF THE LIVER. In lobule 1 only the direction, in 2 only the branching, of the end-pieces is sketched; in 3 only the excretory ducts are drawn.

THE LIVER.

The canal system of the liver consists of an excretory duct (the hepatic duct) the ramifications of which pass into end-pieces. Separate divisions of the excretory system, corresponding to secretory tubes or to intercalated pieces, cannot here be distinguished. The liver is a compound tubular gland*; this structure is recognized with difficulty on account of the following peculiarities:

1. In the other glands† the end-pieces are convoluted (Fig. 211), in the liver they are nearly straight (Fig. 212).

* In this form the organ persists apparently only in one vertebrate (myxine); in other vertebrates it changes during embryonal life to a net-like gland, by the union of its branched tubules.

† In the entire ensuing comparison the compound tubular glands are meant.

2. In the other glands the end-pieces course in every possible direction and surround on all sides the ramifications of the excretory duct; hence the latter lie in the *interior* of the gland lobules (p. 85). In the liver the end-pieces run in a definite direction, toward the axis of the lobule; all ramifications of the excretory duct lie *external* to the gland lobules (*cf.* Fig. 211 with Fig. 212).

3. In the majority of other glands the end-pieces terminate blindly, without anastomosing with one another, in the liver the end-pieces are freely connected with one another and form a net (Fig. 212, 2). Hence the term "end-piece" is inadequate, for blind ends have not yet been with certainty established in the liver; instead of the name "end-piece," the phrase "trabecula of hepatic cells" or "hepatic trabecula" will be adopted in the description of the liver.

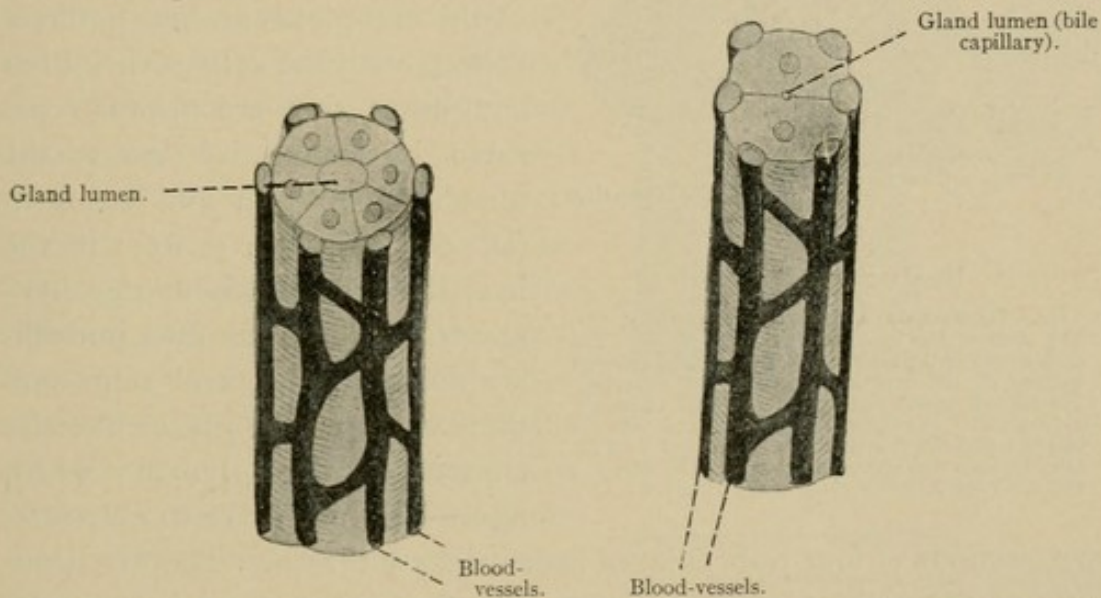


FIG. 213.—SCHEME OF A SEGMENT OF THE END-PIECE OF AN ORDINARY TUBULAR GLAND.

FIG. 214.—SCHEME OF A SEGMENT OF AN END-PIECE (HEPATIC TRABECULA) OF THE LIVER. THE UNION WITH NEIGHBOR TRABECULÆ IS HERE NOT TAKEN INTO ACCOUNT.

4. In other glands the arteries and veins proceed together with the ramifications of the excretory duct and like these lie in part within the lobule (Fig. 211, 3). In the liver the portal vein, which corresponds to the artery of other glands, follows the branches of the excretory duct and like these lies external to the lobule. But the veins course independently of the branches of the portal vein; even their origin lies in the interior of the lobule (Fig. 212).

In addition to these relatively gross distinctions there are minute differences.

5. In other glands the axial lumen of the end-piece in cross-section is surrounded by many gland-cells—six or more (Fig. 213); in the liver by only two gland-cells (Fig. 214). This difference is conditioned

by the relatively large size of the gland-cells (hepatic cells), on the one hand, and by the extreme narrowness of the gland lumina of the liver, on the other hand; two hepatic gland-cells are exactly enough to circumscribe the lumen.

6. In other glands each gland-cell attains to contact with a blood-vessel only on *one* side (Fig. 213); in the liver each hepatic cell has *several* sides touching blood-vessels (Fig. 214), a circumstance likewise brought about by the size of the hepatic cells.

All these peculiarities would not so greatly obscure the tubular gland character of the liver were it not for the existence of a still greater difference:

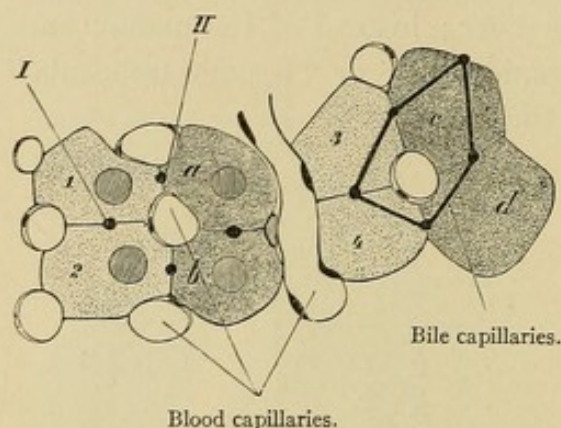


FIG. 215.—SECTION OF A RABBIT'S LIVER. $\times 570$. The outlines were made with a camera lucida. The dark nuclei of the blood capillaries and the different shading of the hepatic trabeculae are schematic. The section passes through the hepatic trabeculae 1/2 and a/b in such wise that the gland-cells are halved; through the trabeculae 3/4 and c/d exactly between contiguous gland-cells. The cells 3, 4 and c, d exhibit their surface to the observer.

7. In other glands the cells of the end-pieces are not in direct contact with the cells of neighbor end-pieces, they are always separated by connective tissue, the membrana propria and so forth (*cf. e. g.* Fig. 30, p. 85); in the liver the cells of neighboring hepatic trabeculae come into immediate contact on several sides and these contiguous surfaces likewise embrace a gland lumen; which figure 215 may serve to elucidate.

Cross-sections of four trabeculae of liver cells are drawn. The first, consisting of the cells 1 and 2, touches directly on the second trabecula, consisting of the cells a and b. 1 and 2 enclose a gland lumen (I), likewise a and b. Between the contiguous surfaces of 1 and a there is also a lumen (II). Thus the gland-cells of the liver, not only on one surface but on several surfaces touch on lumina; these lumina may be united with one another by lateral branches that run between the gland-cells and thereby form actual meshes. The right half of the figure shows such a mesh; it embraces the cross-section of a blood-vessel and therefore may be named *vasozonal mesh*, in contradistinction to meshes that girdle a single liver cell and are called *cytozonal meshes*. The arrangement by which the gland-cells of the liver are embraced on different sides by gland lumina also occurs in other gland-cells, for example, in the serous cells of the salivary glands, that are surrounded by an entire network of secretory capillaries (Fig. 169); the gland lumina of the liver may be directly compared with the secretory capillaries of

other glands and named *bile capillaries*. But while in other glands the secretory capillaries open into a larger axial chief lumen, such axial lumina are wanting in the domain of the hepatic lobule; the bile capillaries open at the periphery of the lobule directly into the interlobular bile-ducts.

The microscopic structure of the liver. The main excretory duct, the *hepatic duct*, and its larger branches consist of a simple stratum of cylinder epithelium, occasionally containing goblet-cells, and of connective tissue separated into a tunica propria and a submucosa. The tunica propria is the carrier of the glands of the bile-duct, chiefly short, pear-shaped follicles clothed with mucous gland-cells, and of isolated longitudinally and transversely disposed plain muscle-fibers. The *cystic duct*, the *common bile-duct*,* and the *gall-bladder* exhibit the same structure; their tunica propria is elevated in anastomosing folds, the *rugæ*; here there is also a thin continuous layer of interlacing smooth muscle-fibers. The cylinder epithelial cells of the gall-bladder are distinguished by their height (0.05 mm.) from those of the common bile-duct (0.024 mm.).† The branches arising from the further division of the hepatic duct, the *interlobular bile-ducts*, with decrease in caliber exhibit diminishing thickness of the wall; the larger consist of simple cylinder epithelium and fibro-elastic tissue, the smallest possess only a structureless membrana propria and a simple layer of low epithelial cells provided with a cuticular border, which as they approach the lobule annex themselves directly to the trabeculæ of liver cells.‡

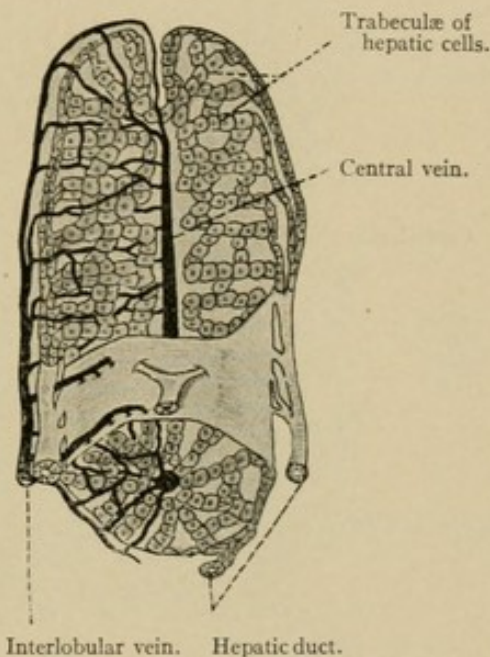


FIG. 216.—SCHEME OF AN HEPATIC LOBULE, represented in transverse section below and in longitudinal section above. In the left half the blood-vessels are drawn, in the right half only the cords of hepatic cells. $\times 20$.

* The opening of the common bile-duct is encircled by smooth muscle-fibers, that are partially connected with the musculature of the intestine; they may be designated a sphincter. Similar sphincters occur at the opening of the two excretory ducts of the pancreas.

† The vasa aberrantia are blind-ending bile-ducts running outside of the parenchyma of the liver. They are chiefly found at the left border of the liver (lig. triangul. sinistr.), at the portal fissure, and in the vicinity of the vena cava. They represent the last remnants of liver substance occurring at these places in embryonal life.

‡ This transition is *very* difficult to see and can be distinctly perceived only in sections in which the bile-ducts have been injected or have been blackened by Golgi's silver method.

The *lobules* of the liver (hepatic lobules, liver islands, also erroneously named acini) can be seen with the unaided eye, on examining the outer surface or a cut surface of the organ, as irregular polygonal fields, that sometimes are distinct, as in the hog, sometimes ill-defined, as in man and the majority of mammals. Their true form is somewhat like that of a prism rounded above, transversely blunted below; they have a height of 2 mm. and a breadth of 1 mm. (Fig. 216). Close under the exterior of the liver the lobules often are arranged with their apex looking toward the surface and a section made parallel to the surface will pass through the lobules transversely (*cf.* Fig. 217); but in the interior

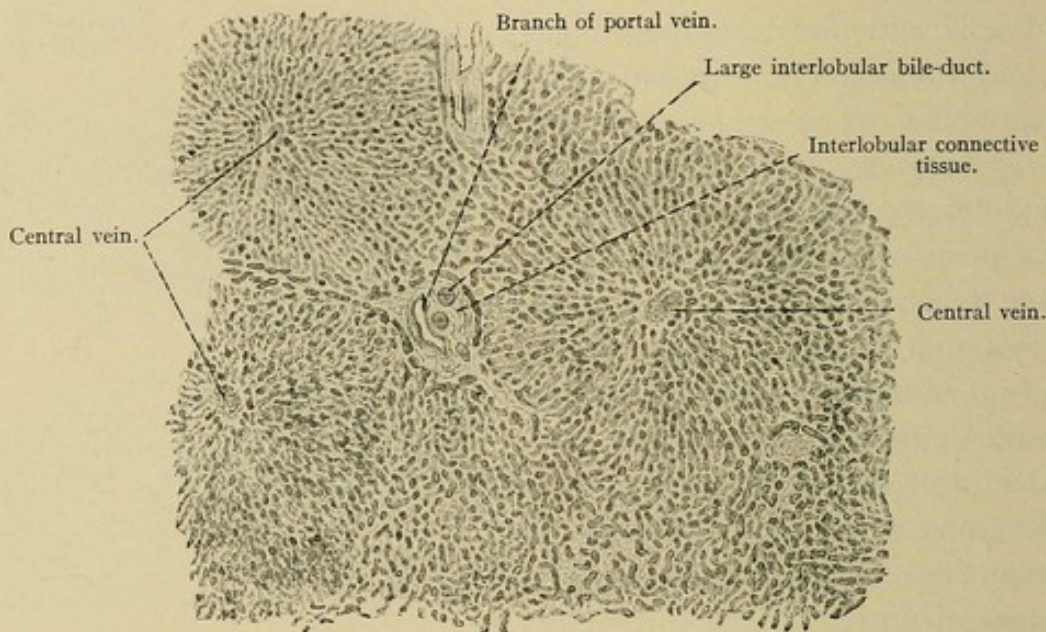


FIG. 217.—FROM A HORIZONTAL SECTION OF THE HUMAN LIVER. $\times 40$. Three central veins, cut transversely, represent each a center of as many hepatic lobules, that at the periphery are but slightly defined from their neighbors. Below and to the right of the section the lobules are cut obliquely and their boundaries cannot be distinguished. Technic No. 123.

of the liver the lobules stand in various directions. Each lobule consists of hepatic trabeculæ and blood-vessels and is separated from its neighbors by the *interlobular* connective tissue,* which supports the branches of the excretory duct (the hepatic duct), the branches of the portal vein and the hepatic artery, of the lymph-vessels and the nerves.

On examining a cross-section of a lobule of the liver with low magnification, the trabeculæ of hepatic cells may be recognized as cords and small lamellæ that extend from a little vein, the central vein, situated in the axis of the lobule, radially toward the periphery, and by means of lateral branches connect with neighbor trabeculæ (Fig. 216 and Fig. 217). By the usual methods the gland lumina in these trabeculæ cannot be seen; only by injection of the canal system through the hepatic

* The sharp demarcation of the lobules depends on the quantity of the same.

duct or by the method of Golgi, which blackens the bile, can they be successfully demonstrated. It is then evident that the lumen of the smallest interlobular bile-duct continues directly into the hepatic lobule and there lies in the axis of the hepatic trabecula. Longitudinal section

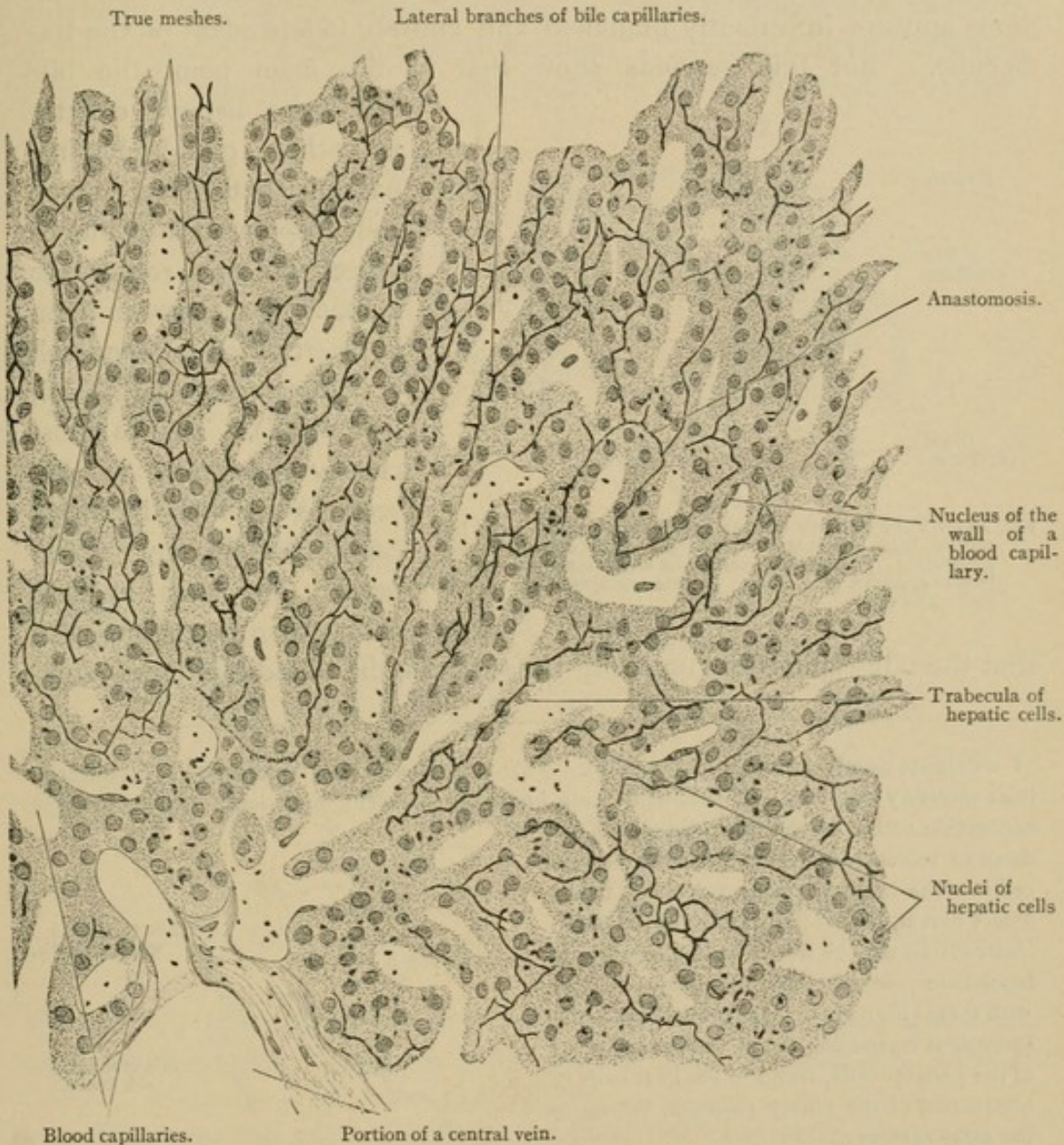


FIG. 218.—FROM A CROSS-SECTION OF A HUMAN HEPATIC LOBULE. $\times 300$. The boundaries of the hepatic cells could not be seen in the preparation. The black dots are foreign matter due to precipitation of the silver. Technic No. 126.

of the lumen shows that it runs zigzag and is beset with small lateral branches* (Fig. 218), which, where several trabeculae of liver cells are in

* These intercellular lateral branches must not be confused with short lateral twigs of the bile capillaries, that terminate in a minute knob-shape enlargement. The knob corresponds to a small vacuole occurring in the liver cell, which communicates with the bile capillary by means

direct contact, by union with other lateral branches may form true meshes* (Fig. 218). All the lumina lying in the interior of the lobule are named *bile capillaries*. The entire system of bile capillaries is freely united, not only through the meshes, but also through anastomoses brought about by the union of neighbor hepatic trabeculae (Fig. 218) and in thick sections appears luxuriantly branched and entirely independent of the trabeculae. But thin sections show that in the main point the bile

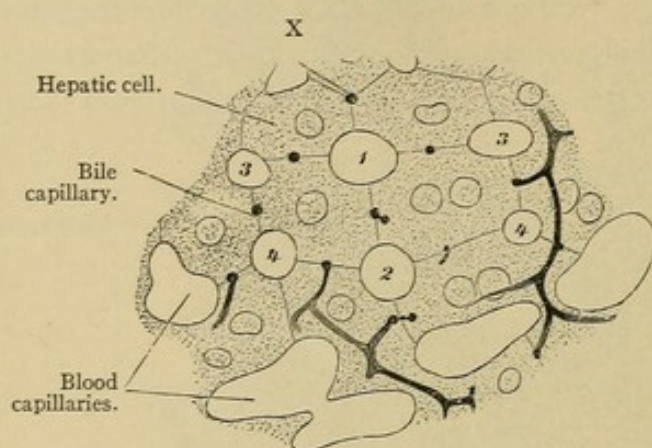


FIG. 220.—THIN SECTION OF THE LIVER OF A RABBIT, WITH INJECTED BILE CAPILLARIES. $\times 560$. (The drawing is *not* schematic.) Two of the hepatic cells are in contact with four blood capillaries (1, 2, 3, 4). X. Bile capillary at the edge of an hepatic cell.

capillaries behave exactly as other gland lumina, namely that gland lumen (bile capillary) and blood-vessel do not come into contact,† but between them is intercalated a gland-cell or a portion of such a cell (see p. 86). This is most clearly recognized in thin sections in which the blood capillaries are cut transversely (Fig. 220); in these it may also be plainly seen

that the bile capillaries run along the surfaces, the blood capillaries along

of a delicate canal (the small lateral twig). This lateral twig may be regarded as an intracellular secretory capillary. Undoubtedly these knobs are transient formations, only occurring in connection with a certain functional cycle, are drops of secretion that pass from the hepatic cell into the capillary; the evidence of this I detect herein, that entire sections of the canalicular system may be free from knobs, while in immediate proximity each canaliculus is beset with them (Fig. 219). It is probable that the formations resembling the secretory capillaries of the parietal cells, found in the liver cells in obstruction of the biliary passages, belong to the same category.

* The number of meshes is by no means so large as one might infer from not very thin sections examined with low powers. Very frequently meshes are simulated by the very zigzag canaliculi with their lateral branches crossing at different planes (Fig. 221). One may search entire sections, in particular such as pass transversely through an hepatic lobule, without finding a single true mesh.

† Whether this is invariably the case appears to me latterly doubtful; in very thin injected sections of rabbit's liver I have, in isolated places, seen bile capillaries close beside blood capillaries; the same is said to occur in the dog and in man.

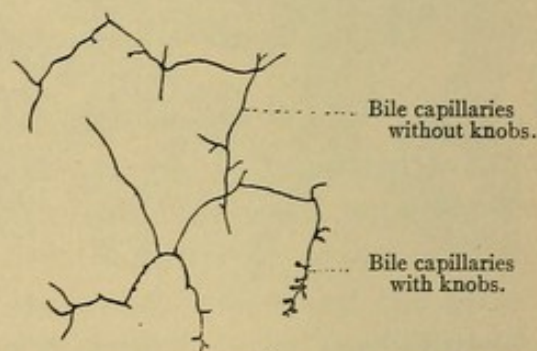


FIG. 219.—FROM A SECTION OF THE LIVER OF A DOG. $\times 490$. Technic No. 126.

the edges of the hepatic cells; still this is not an invariable rule, for bile capillaries are found that also follow the edges of the cells (Fig. 220, x), a relation existing notably in man.

The gland-cells of the liver, the *liver cells*, *hepatic cells*, are irregular, polyhedral structures, which consist of a granular protoplasm and one or more nuclei; a membrane is wanting.* The protoplasm contains granules of pigment and glycogen,† as well as fat droplets of various

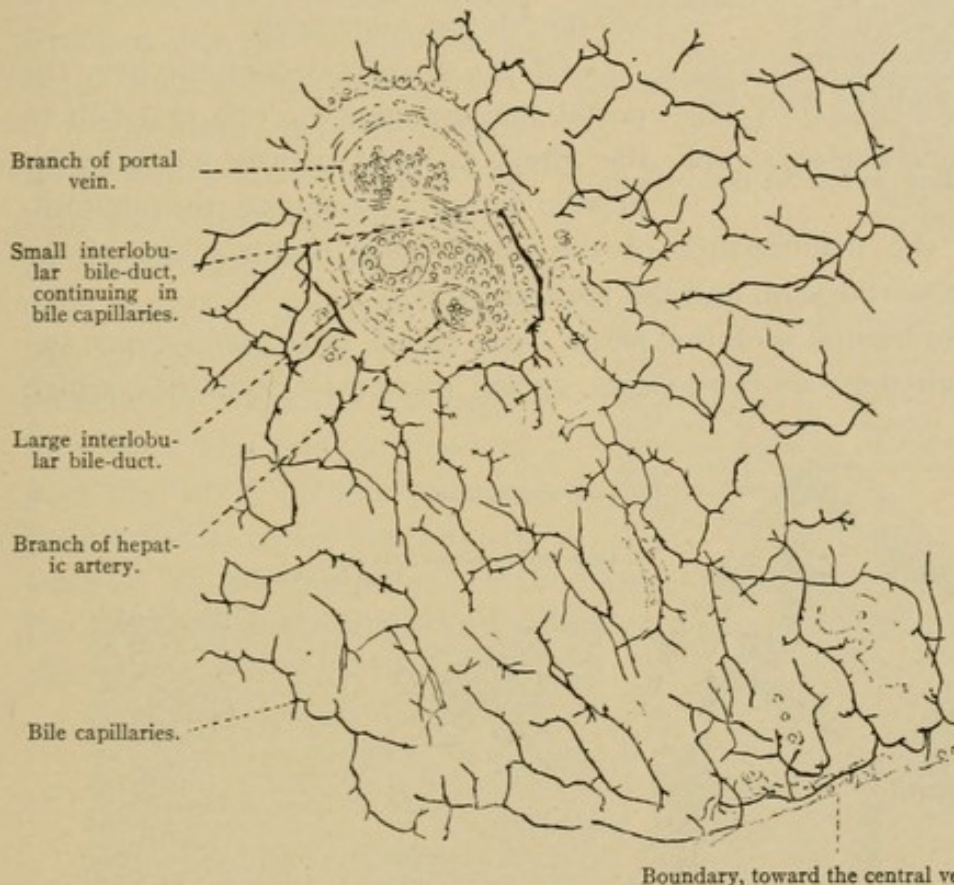


FIG. 221.—TRANSVERSE SECTION OF THE LIVER OF A DOG. $\times 240$. Bile capillaries blackened according to the method of Golgi. Technic No. 126.

sizes, which latter are invariably found in mammalian animals and well-nourished persons. The cells have a size of from 18 to 26 μ .‡ Visible

* Where the liver cells bound the bile capillaries their exoplasm (p. 63) is somewhat modified and is connected with the cuticular border of the epithelium of the interlobular bile-ducts (p. 287). This modified stratum has been inaccurately described as a special wall of the bile capillary. With equal reason a special wall other than that formed by the gland-cells should be ascribed to all gland lumina.

† The latter can be demonstrated in alcohol preparations that have not been treated with aqueous solutions.

‡ Individual hepatic cells are distinguished by their greater diameter, of the body as well as of the nucleus; such large nuclei divide in the mode of amitosis; frequently one of the two nuclei goes to destruction, in other cases the nuclei arising in this way persist—as many as seven have been observed.

functional differences also exist in the liver cells (Fig. 222 *B*). They either are small, dull, and indistinctly contoured,—such conditions occur principally in the fasting state,—or larger, clear in the center, at the periphery provided with a coarsely granular belt—appearances that occur chiefly during digestion. In man the two states often are met in the *same* liver.

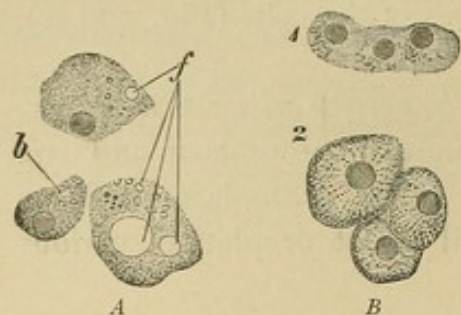


FIG. 222.—LIVER CELLS OF MAN. $\times 560$.
A. Isolated liver cells containing smaller and larger fat-drops, *i*; *b*, imprint from contact with a blood-vessel. Technic No. 121.
B. From a section; 1, empty cells; 2, cells filled with secretion. Technic No. 123.

Of the *blood-vessels* of the liver the portal vein assumes the rôle that falls to the artery in other glands, while to the hepatic artery is assigned the subordinate task of the maintenance of the interlobular ramifications of the bile-duct, the portal vein, and the hepatic veins.

From the branches of the *portal vein*, called *interlobular veins*, because they run between the lobules, spring numerous capillaries, which

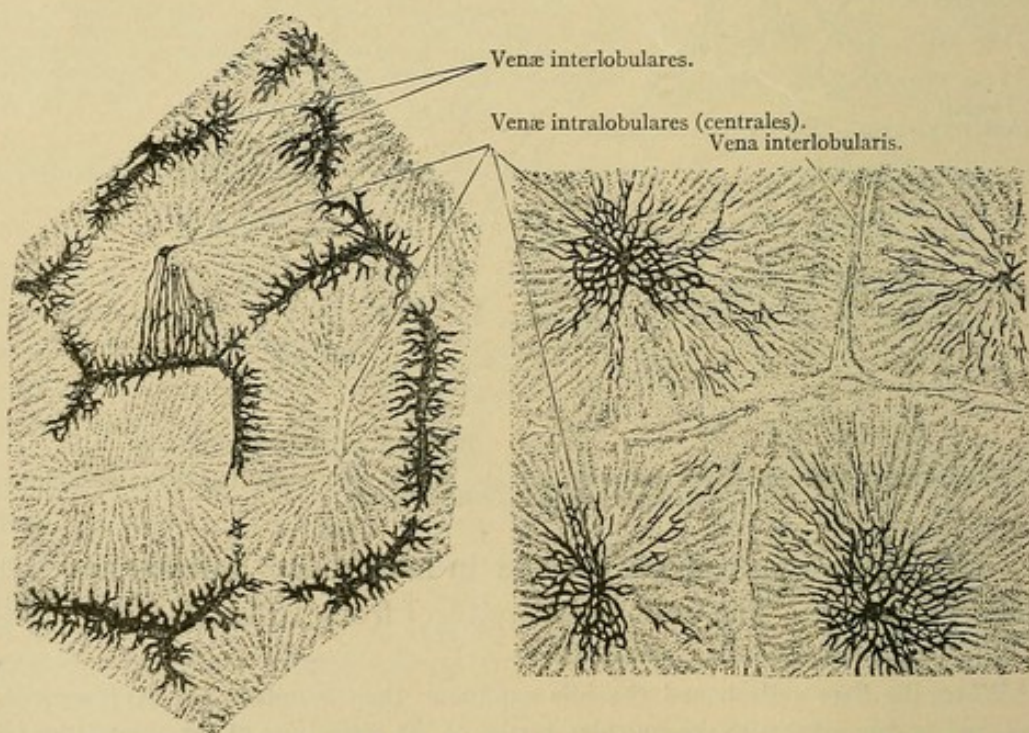


FIG. 223.—HORIZONTAL SECTION OF THE LIVER OF A RABBIT. Injected through the portal vein. $\times 40$. Three hepatic lobules are represented. The injection mass filled only the branches of the portal vein (interlobular veins); in the upper lobule it penetrated to the central vein. Technic No. 125.

Fig. 224.—HORIZONTAL SECTION OF THE LIVER OF A CAT. Injected through the vena cava inferior. $\times 40$. Four hepatic lobules are shown. The injection mass filled the central veins and the capillaries emptying into it, but did not penetrate to the interlobular veins. Technic No. 125.

possess the conspicuous width of from 10 to 12 μ . They penetrate within the lobules, where they lie between the hepatic trabeculae (Fig. 225); during their course they repeatedly anastomose with one another

and finally empty in a small vein lying in the axis of the lobule, the *central vein*, or *intralobular vein*, the transverse and longitudinal section of which is visible even in sections of the uninjected liver (Fig. 217). The central veins represent the radicles of the hepatic veins and empty into the *sublobular veins*, which run along the one slightly flattened side, the so-called base, of the hepatic lobule (Fig. 226).

The branches of the *hepatic artery* follow those of the portal vein and ramify only in the interlobular tissue, where they form capillary networks about the larger bile-ducts and the branches of the portal and hepatic veins. The veins proceeding from the artery or its capillaries open into the portal interlobular veins or into the beginnings of the portal capillaries. In the capsule of the liver (see below) the hepatic artery forms a wide-meshed capillary plexus.

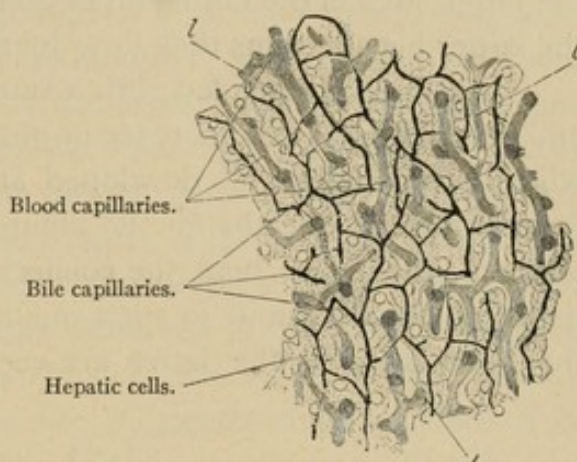


FIG. 225.—FROM A SECTION OF THE LIVER OF A RABBIT. $\times 240$. The portal capillaries were injected with a red mass, the bile capillaries with a blue mass. The hepatic cells are in contact with blood capillaries on both sides. At a few points the red mass has retracted and given rise to a space (I), between the hepatic cells and the portal capillaries. The dark spots on the portal capillaries are optical cross-sections of blood capillaries which run vertically through the thickness of the section.

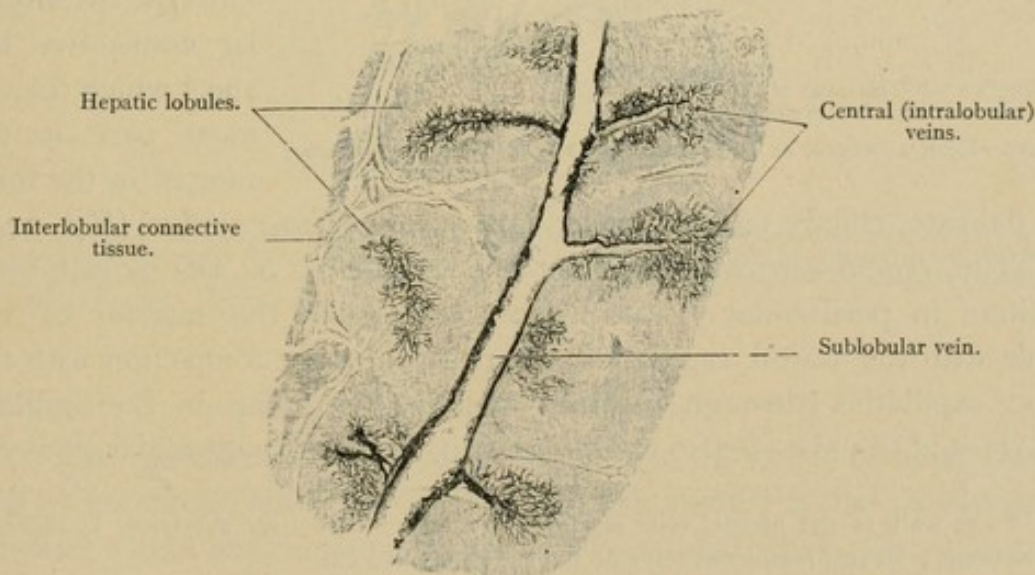


FIG. 226.—FROM A VERTICAL SECTION OF THE LIVER OF A CAT. Injected through the vena cava inferior. A sublobular vein cut longitudinally; it takes up the central veins. The greater part of the injection mass has fallen out of the wide blood-vessels. $\times 15$. Technic No. 125.

The course of the blood-vessels therefore is as follows: the portal vein enters at the transverse fissure, repeatedly divides into branches that

steadily decrease in size and run in the connective tissue between the lobules as the interlobular veins. From these capillaries arise, which pass toward the axis of the lobule and terminate in the central vein. Several of the latter unite in the formation of each of the sublobular veins, which like the larger hepatic veins they form by their union run between the lobules.

The liver is provided with a capsule consisting of connective tissue and elastic fibers (that increase in old age), the *capsula fibrosa* (Glissoni), which is especially well developed at the transverse fissure and in the form of special sheaths for the different vessels* penetrates into the interior of the liver; here the connective tissue (interlobular connective tissue) is usually found in such small amount between the lobules that the boundaries of the latter are very imperfectly defined (cf. Technics

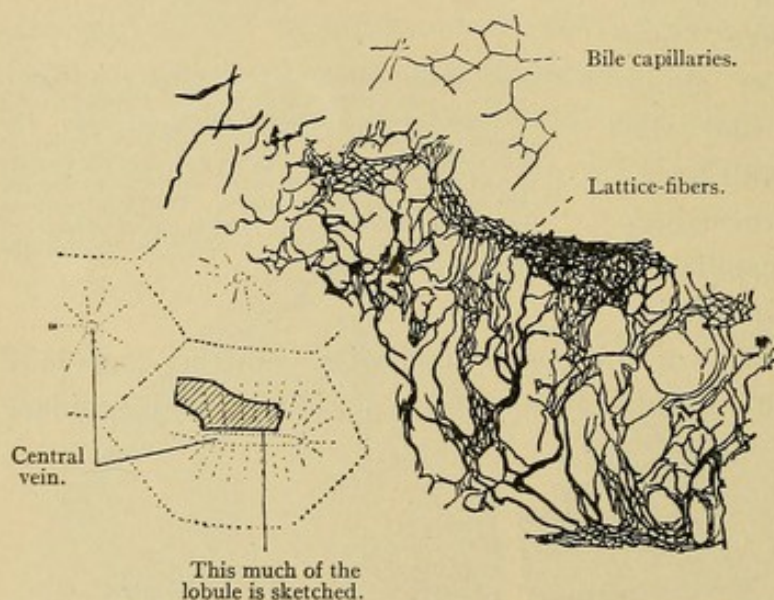


FIG. 227.—FROM A SECTION OF HUMAN LIVER. $\times 214$. Technic No. 126.

No. 122 and No. 123). Delicate fibers ("lattice-fibers," *Gitterfasern*) derived from the interlobular connective tissue—but no elastic elements—penetrate into the interior of the lobules; they form the intralobular connective tissue,† which for the most part is arranged in the form

of a delicate, chiefly radially placed latticework (*Gitterwerkes*) (Fig. 227).

The *lymph-vessels* accompany the branches of the portal vein, continue in perilobular lymph spaces and enter the interior of the lobule with the portal capillaries, where an intimate connection with the blood capillaries (through normal openings occurring in the capillary wall) is said to exist;‡ also the larger veins, from the sublobular veins on,

* The walls of the hepatic veins are firmly attached to the liver substance by this connective tissue; for this reason the veins do not collapse when cut.

† The so-called stellate cells do not belong to the connective tissue, but are epithelial elements of the portal capillaries. The stellate form is caused by the peculiar arrangement of the protoplasm around the nucleus. The stellate cells can be seen only in gold preparations.

‡ Such connection is supported by the fact that in injections of the portal vein (in the rabbit) the lymph-vessels become visible; indeed the trophospongium (p. 64) of the hepatic cells has been successfully injected through the portal vein, which doubtless could happen only in an indirect way, through the simultaneous injection of the lymph-vessels.

are accompanied by lymph-vessels. These *deep* lymph-vessels are freely connected with a narrow-meshed network of lymph-vessels occurring in the capsule of the liver.

The *nerves* chiefly consist of nonmedullated nerve-fibers, with which only a few medullated nerve-fibers are mingled; they supply the capsule of the liver and enter the interior of the organ with the hepatic vessels, the ramifications of which they follow; according to investigations on mammalian animals they terminate for the greater part on the hepatic vessels, accompanying them into the interior of the lobules; a lesser portion end as sensory fibers in the interlobular tissue and as secretory fibers between the liver cells (this has been observed in the dove). Ganglion cells occur only in the course of the nerves in the wall of the gall-bladder.

The secretion of the liver, the *bile*, frequently contains drops of fat, also granular masses of bile pigment. Cylinder cells from the bile-ducts are to be regarded as incidental admixtures.

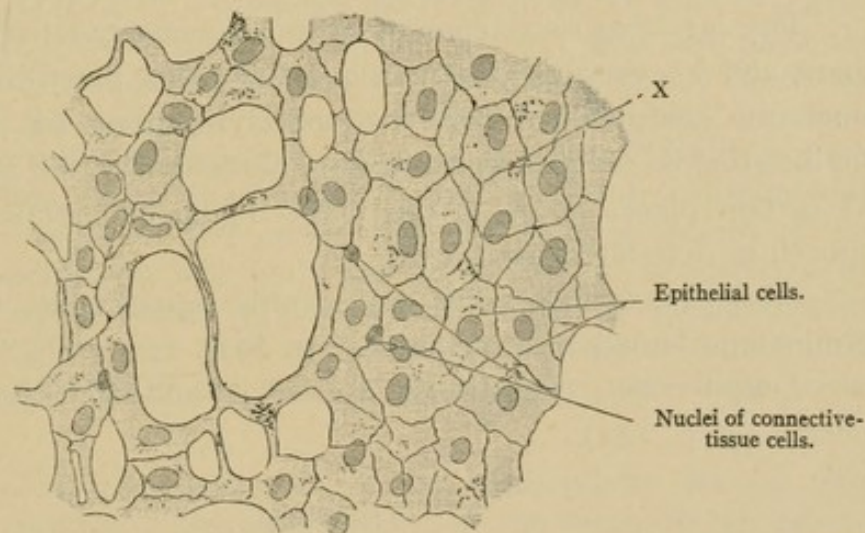


FIG. 228.—FROM THE GREATER OMENTUM OF A RABBIT. $\times 240$. The meshes are formed by thick and thin bundles of connective tissue. The wavy striation of the bundles can only be indistinctly seen, because the preparation is mounted in xylol balsam. At X the epithelial cells of the opposite surface can be seen shimmering through. Technic No. 127.

THE PERITONEUM.

The peritoneum principally consists of connective tissue bundles and of numerous elastic fiber-nets; the free surface of the peritoneum is covered with a simple layer of flat, polygonal epithelial cells; these cells consist of superficial divisions, very thin plates (in the dog and the rabbit they are covered with a delicate hair border), the contiguous edges of which are very accurately approximated (Fig. 228), and of deep divisions enclosing the nucleus, that are connected with one another by delicate processes. The size of the plates varies according to the stretching to

which they are subjected. The connection with the subjacent parts (the parietes, the viscera, etc.) is effected by loose (subserous) connective tissue.

The *connective-tissue bundles* are arranged in thinner (in the visceral peritoneum) or thicker (in the parietal peritoneum, in the mesentery) layers, chiefly parallel to the surface, and interlace in various directions; in certain localities (in the greater omentum, in the middle of the lesser omentum) the bundles form a beautiful network with polygonal or rectangular meshes. The strands of the network also are covered with flat epithelial cells (Fig. 228).

The number of connective-tissue cells among the bundles is on the whole not large; only in young animals are larger groups of cells found; they resemble plasma-cells and probably all stand in close relation to the formation of vessels (*cf.* pp. 94, 135).

The *elastic fibers* in the deeper layers of the peritoneum, particularly in the parietal lamella, are profuse and vigorously developed.

The *subserous tissue* consists of loose connective tissue, many elastic fibers, and fat, varying greatly in quantity; it is plentiful where the peritoneum is easily shifted over the underlying parts, but on the liver and the intestine so reduced that it cannot be demonstrated as a special layer. At certain places, *e. g.*, in the broad ligaments, numerous strands of smooth muscle-fibers are found.

Blood-vessels and *nerves* are scantily represented; the latter partly terminate in lamellar corpuscles (*cf.* p. 222).

Lymph-vessels occur in the superficial and deep layers of the peritoneum (*cf.* p. 141).

TECHNIC.

No. 96.—*Isolated squamous cells from the oral cavity.*—With a scalpel gently scrape the upper surface of the tongue and mix the scrapings on a slide with a drop of salt solution; apply a cover-glass; in addition to isolated, pale, squamous epithelial cells (Fig. 15, p. 75), leucocytes ("salivary corpuscles") may be found, also, with more vigorous scraping, the tips of filiform papillæ, which not infrequently are surrounded by finely granular, dark masses of micrococci, to which tufts of *leptothrix buccalis* are attached. The preparation may be stained under the cover-glass (p. 53) with picrocarmine and then treated with dilute acidulated glycerol, provided too many air bubbles do not make the preservation of the preparation impossible.

No. 97.—*Mucous glands of the lips.*—These are millet-sized nodules perceptible to touch and accessible for macroscopic preparations. For microscopic preparations cut from the mucous membrane of a human lower lip (not the margin of the lip) 1 cm. cubes; fix them in 50 c.c. of

potassium-bichromate-acetic acid (p. 32) and in twenty-four hours harden in 50 c.c. of gradually strengthened alcohols (p. 35). Cut many sections, not too thin, and stain them with Hansen's hematoxylin (p. 38); place the sections in water and with the naked eye select those which include the excretory duct and preserve them in xylol-balsam (p. 50); examine with a low power (Fig. 160).

No. 98.—*Dried ground tooth*.—For the preparation of dried ground sections of teeth the latter should be obtained immediately after they are extracted, sawed into transverse disks 2 mm. thick, glued with sealing-wax upon cork and treated like No. 61 (p. 177). If longitudinal sections are desired the entire tooth should be glued to the cork. Longitudinal sections are to be preferred, since they show all parts of the tooth in a *single* preparation (Figures 170, 171, 172).

If it is desired to decalcify the teeth of an adult, proceed as with No. 63 (p. 178). The enamel consisting of earthy salts and only from 3 to 5 per cent. of organic substances dissolves completely, hence only the dentine and cement remain.

No. 99.—*Odontoblasts*.—Remove the teeth from the jaws of a newborn child; place them in 60 c.c. of Müller's fluid; after six days the pulp can be easily withdrawn in toto by means of forceps. With the scissors cut from the surface of the pulp a piece the size of a lentil and tease a little the tolerably tenacious tissue in a drop of Müller's fluid; apply a cover-glass, press lightly upon it, and examine with the high power. At the edges of the preparation the long processes of the odontoblasts standing out like hairs will be seen; also scattered, completely isolated odontoblasts (Fig. 175). In order to preserve, treat under the cover-glass with distilled water for two minutes, then with picrocarmine (p. 53); when the staining is completed, add dilute acidulated glycerol.

No. 100.—*Enamel prisms*.—These are obtained by teasing fragments from the surface of the lateral parts of the teeth of No. 99 in a drop of Müller's fluid. Examine with a high power. The enamel prisms will be found in groups of three or more; they are distinguished by their dark outlines and (usually indistinct) cross-striation (Fig. 173). Mount in glycerol.

The prismatic form of the enamel prisms can be seen in thin sections cut parallel to the upper surface of the tooth. Only portions of a section exhibit regular hexagonal prisms, that is, only true cross-sections of the prisms are hexagonal (Fig. 174). The enamel of young teeth may be sectioned without previous decalcification.

No. 101.—*Development of teeth*.—For the study of the early stages select pig and sheep embryos; these are the most easily obtained at the slaughter houses (*cf.* No. 67, p. 180); for the first stage (Fig. 177) the pig embryos should have a size of about 6 cm.,* for the second stage a size of

* Measured from the tip of the snout to the root of the tail.

about 10 or 11 cm. For later stages (Fig. 181) the inferior maxilla of newborn dogs or cats is very suitable. Place the heads (or the lower jaws) in 100 c.c. of potassium-bichromate-acetic acid * (p. 32) and harden in from 80 to 120 c.c. of gradually strengthened alcohols (p. 35). After the heads have lain six or eight days in 90 per cent. alcohol, they are to be decalcified in 100 c.c. of distilled water plus 1 or 2 c.c. of nitric acid (p. 36). When the decalcification is completed, in from three to eight days, harden again in alcohol. In five or six days cut off the lower jaw and divide it in front in the middle (larger jaws should be cut vertically into pieces 1 or 2 cm. long); stain the pieces in bulk in borax-carmines.† When the staining and decolorization are completed the tissue is to be transferred to absolute alcohol, in which it must remain for several days; it is then embedded in liver and sectioned. It is necessary to cut many (20 to 40) thick sections, since only those which pass through the middle of the tooth, or its anlage, can be used. Mount in xylol-balsam. Not infrequently in sectioning the enamel organ is lifted from the papilla, so that a free space exists between the two. The dentine is often stained in different tones of red; this is due to the different ages of the calcified and uncalcified strata of the dentine.

No. 102.—*Papillæ filiformes, fungiformes, vallatæ; folliculi linguales*.—Cut pieces 2 cm. square from the mucous membrane of the upper surface of a human tongue. Each piece should have some of the muscle tissue attached to its lower surface; for fungiform papillæ cut the piece from the tip of the tongue; for filiform, from the middle of the tongue; for vallate, from the root of the tongue and for lingual tonsils, the punctiform openings of which can be seen with the naked eye, from the root of the tongue, and place them in 100 or 200 c.c. of Müller's fluid (p. 33). The fluid must be changed several times; after two weeks wash the tissue and harden it in 50 c.c. of gradually strengthened alcohols (p. 35). For filiform papillæ cut thick sagittal sections of the tongue and do not stain them; stain the other sections in Hansen's hematoxylin (p. 38) and mount in xylol-balsam (Figs. 183, 184, 185). For the preparations represented in Fig. 161 and Fig. 186 the tissue was fixed and hardened in 55 c.c. of absolute alcohol. Rabbits' tongues may be placed in toto in 200 c.c. of Müller's fluid; the subsequent treatment is the same. Thick cross-sections through the anterior half of the entire tongue are suitable for the study of the arrangement of the lingual muscles. Thin sections of the root of the tongue show beautiful mucous and also serous glands.

No. 103.—*The tonsils*.—The tonsils of adult man do not furnish instructive preparations. They should be treated according to technic

* Objects fixed in Müller's or in Zenker's fluid are also useful.

† Bulk staining, despite the length of the procedure, is preferable to individual staining of sections with hematoxylin, because too many sections must be stained which on investigation are found to be useless.

No. 102. The tonsils of the rabbit and the cat are recommended; to find them proceed as follows:

Dissect the skin from the anterior surface of the throat and with a pair of stout scissors cut through the trachea and esophagus above the sternum, grasp the cut end of the trachea with forceps and with the scissors dissect out both tubes up to the head of the esophagus, keeping close to the anterior surface of the vertebral column; at the same time the cornua of the hyoid bone will be divided. Here the wall of the pharynx is to be divided; then cut through the musculature close to the median edges of the inferior maxilla up to the angle, also through the ligaments of the tongue. (In the rabbit it is advisable to divide both angles of the mouth and with scissors introduced within the mouth cleft to sever the ligaments and the genioglossus muscle). Draw the trachea and attached structures downward, press the tongue down between the rami of the inferior maxilla and divide its remaining attachments (to the palate) close to the bone. Put the tongue down with its free surface looking upward; with delicate scissors divide the posterior wall of the pharynx in the median line down to the larynx and pull the walls apart; the tonsils will then be seen as a pair of oval prominences, about 5 mm. long, on the lateral walls of the pharynx. They may be fixed in 60 c.c. of potassium-bichromate-acetic acid (p. 32), hardened in 50 c.c. of gradually strengthened alcohols (p. 35), stained with Hansen's hematoxylin (p. 38) or with hematoxylin and eosin (p. 39), and mounted in xylol-balsam.

No. 104.—*The esophagus*.—Pieces from 2 to 6 cm. long of the entire tube are to be fixed in 60 c.c. of Müller's fluid (p. 33) and in two weeks hardened in 50 c.c. of gradually strengthened alcohols (p. 35); stain with Hansen's hematoxylin (p. 38); mount in xylol-balsam (Fig. 188).

No. 105.—*The membranes of the stomach*.—For topographic preparations place pieces from 2 to 5 cm. square for six hours in 100 c.c. of 3 per cent. nitric acid (p. 32). Detach the gastric contents adhering to the mucous membrane by moving it slowly to and fro in the acid. In a half hour renew the acid; harden in 60 c.c. of gradually strengthened alcohols (p. 35). Mount thick unstained sections and thin sections stained with Hansen's hematoxylin (p. 39) and with eosin in xylol-balsam (Fig. 190).

No. 106.—*Fresh gastric glands*.—From the fundus of the stomach of a rabbit just killed cut pieces about 2 cm. square and separate the loosely attached muscular coat from the mucous membrane. Grasp the latter with forceps at the left edge and with fine scissors cut a very thin strip, from 0.5 to 1 mm. broad; tease it well in a drop of 0.5 per cent. salt solution. The body and fundus of the glands can be satisfactorily isolated without much trouble. The protoplasm of the parietal

cells can be distinctly seen (Fig. 229 *B*), the chief cells are invisible. The nuclei may be stained with picrocarmine (p. 53) and the preparation mounted in dilute glycerol. The isolation of the pylorus glands can be accomplished only by very careful teasing.



FIG. 229.—LOWER HALF OF AN ISOLATED FUNDUS-GLAND OF A RABBIT. $\times 240$. *B*, Parietal cell; *M*, membrana propria.

No. 107.—*Isolated gastric epithelium*.—Place a piece 1 cm. square of gastric mucous membrane for about five hours in 30 c.c. of Ranvier's alcohol (see further p. 29). In the majority of the cells the mucous portion occupies a large division and they have the appearance of those pictured in Fig. 25 *c*, p. 80. The preparation may be stained under the cover-glass with picrocarmine and mounted in diluted acidulated glycerol (p. 53).

No. 108.—*Gastric glands*.—The stomach of a cat or dog that has been fasting for one or two days is especially recommended. The stomach of the rabbit, on account of the very small size of the chief cells, is less suitable. Dissect off the mucous membrane from the muscular coat and place pieces of the former about 1 cm. square in 10 c.c. of absolute alcohol. In about a half hour transfer them to 20 c.c. of fresh alcohol. The *form* of the glands can be recognized in moderately thin sections; the only difficulty is the circumstance that the gland-sacks stand very close together. The beginner may not recognize the glands and may mistake for them the gastric pits lined with clear epithelium. The stomach of man, which however is suitable for use only for a few hours after death, exhibits this disadvantage in a lesser degree. For the study of the *minute structure* of the glands and of the surface epithelium embed the tissue in liver and cut the thinnest possible sections.

(a) *For fundus glands, chief and parietal cells*, cut vertical or, better, horizontal sections of the mucous membrane and stain them with Hansen's hematoxylin for two or four minutes. Wash the sections thoroughly in 30 c.c. of distilled water, which must be changed as often as it becomes bluish—about once or twice. Transfer them to 5 c.c. of a 0.03 per cent. solution of Congo red (p. 25), for from three to six minutes, wash two minutes in distilled water and mount in xylol-balsam. If the sections are too thick everything appears red; the large red parietal cells cover the smaller chief cells; examine the thinnest parts of the section, especially the fundus of the glands, where the parietal cells are not so exceedingly profuse. The parietal cells can be recognized with the low power as isolated red spots on a rose-red ground. With the high power the pale blue smaller chief cells can be seen. The very narrow lumen of the fundus glands can be best seen in cross-sections (sections parallel to the surface of the mucosa). The lateral twigs of the chief lumen can only be perceived in very choice sections (Fig. 192). Figure 191 is a combination of several thin longitudinal sections.

(b) For pylorus glands stain vertical and horizontal sections of the mucosa with Hansen's hematoxylin and mount in xylol-balsam. The lumen of the pylorus glands is wider (Fig. 194). Owing to the extreme sinuosity of the glands, thin sections contain but few glands cut in their entire length, mostly only parts of glands.

No. 109.—*Duodenal glands*.—Cut open lengthwise the stomach and duodenum of a cat, remove the contents by swaying gently to and fro in salt solution (p. 20), and fasten the pyloric end of the stomach and the upper half of the duodenum, in all a piece 5 or 6 cm. long, to a cork plate by means of quills. Place the whole for six hours in 100 c.c. of 3 per cent. nitric acid with the tissue side downward. Further treatment like No. 111. Cut longitudinal sections, which simultaneously pass through pylorus and duodenum. Stain with Hansen's hematoxylin. Mount in xylol-balsam. The preparations represented in figures 200 and 201 were fixed after technic No. 120.

No. 110.—*Epithelium and villi of the small intestine*.—From the middle portion of the small intestine of a rabbit just killed cut a piece one cm. long, open it along its length and remove the contents by carefully pouring over it 0.7 per cent. salt solution. Then grasp the piece at the left edge with the forceps, with fine scissors cut off a small strip and spread it out in a drop of salt solution on a slide placed on a black background. With the unaided eye one can see the villi projecting from the edge of the preparation. Examine the preparation *without* a cover-glass, with the low power. The villi will be seen partly extended, partly contracted; the latter condition may be recognized by transverse folds running across the villus (Fig. 230). Details cannot be detected. Apply a cover-glass; the villi become flattened and appear clearer; the cylindrical epithelium and close beneath this the loops of the capillary blood-vessels can be distinctly seen. If the epithelium contains goblet-cells, these appear as bright, shining, rounded spots. For the investigation of the epithelium proceed as follows:



FIG. 230.—INTESTINAL VILLUS OF A RABBIT. $\times 70$.

(a) Tease the piece a little; in this way cylinder cells, singly and in groups, are loosened, which are to be examined with the high power. Not infrequently some cylinder cells are found inflated to a spherical form. The cuticular border is sometimes separated in very distinct rods. Goblet-cells when present may be recognized by their homogeneous appearance and if carefully focused the sharply outlined orifice may be perceived. Occasionally the epithelial cells are difficult to loosen from the basement membrane; in such cases make a second investigation an hour later, when the epithelium will be sufficiently macerated to be brushed off.

(b) For permanent preparations place pieces 1 cm. square of the intestine opened lengthwise in 30 c.c. of Müller's fluid. In three or five days take the tissue out, scrape it with the tip of a scalpel, and distribute a

little of the scraping in a drop of diluted glycerol; cover-glass; high power (Fig. 198 *A*).

No. 111.—*Sections of the small intestine*.—Place pieces from 2 to 4 cm. long of the intestine of a rabbit, better of a puppy or a kitten, in 100 or 200 c.c. of 3 per cent. nitric acid.* After six hours the pieces are to be hardened in 100 c.c. of gradually strengthened alcohols. Cross-sections can be made through the entire intestinal tube; in most cases only fragments of the villi are thus obtained; to obtain entire villi cut open with a razor the hardened intestine along its length, pin it with needles on a cork plate, with the mucosa uppermost. The villi can be seen spreading apart, with the unaided eye. Cut thick sections of the mounted intestine and stain them for one minute with Hansen's hematoxylin and mount in xylol-balsam. Goblet-cells are very frequently found in the epithelium (Fig. 198 *B*). Staining in bulk with borax-carminc is strongly advised.

The human intestine before being placed in the nitric acid must be cut open and washed in the same fluid. It is advisable to pin pieces about 5 cm. square to a cork plate and thus to place them in the fixing and hardening fluids. If the intestine is not absolutely fresh the entire superficial epithelium loosens, so that the naked connective-tissue villi are exposed.

Horizontal sections of the intestine furnish very beautiful pictures. Not infrequently the cross-sections of the glands drop out and then only the connective-tissue tunica propria remains. In these preparations the goblet-cells all appear as clear bodies of equal size and therefore afford no clue in regard to the topography of the secretory phases of the cell.

For the latter purpose the following is recommended:—

No. 112.—*Triple staining of the intestine*.—Small pieces of tissue are to be fixed in Flemming's mixture (p. 34), hardened in gradually strengthened alcohols, and subsequently treated according to No. 11, p. 43.

No. 113.—*Agminated nodules (patches of Peyer)*.—These can be seen shimmering through the uninjured fresh intestinal wall of the rabbit, but in the dog and the cat, on account of the thickness of the musculature, they are often imperceptible. In the latter animals patches are constant at the point where the small intestine opens into the large. Cut out the portion of the intestine of a rabbit containing the Peyer's patches and proceed according to the method given in No. 111. In the cat take the lowermost portion of the ileum (about 2 cm.) with a piece of the cecum of the same length; slit them open lengthwise and span them out on a cork plate, with the mucosa uppermost. Usually the mucosa is covered with a tenacious excrement, difficult to remove by washing,

* If the intestine is placed in the reagent *immediately* after death the muscles of the villi contract and a separation of the connective tissue from the epithelium is then the invariable result (cf. Figs. 195 and 196). Therefore it is advised to let the intestine grow cold before putting it in the fixing fluid.

which glues the villi together, so that only oblique sections of the latter can be obtained. Further treatment like No. 111 (Fig. 203).

Closely crowded nodules are found in the blind half of the vermiform process of the rabbit, which encroach upon the mucosa and compress it within such narrow areas that cross-sections exhibit very complicated pictures, scarcely intelligible to the beginner.

Fixation in 0.1 per cent. chromic acid (p. 32), with hardening in gradually strengthened alcohols, makes the germinal layers very distinct, but it is not as useful for the remaining elements as the nitric acid.

No. 114.—*The large intestine.*—Treat empty pieces like No. 111 or No. 112 (*cf.* Fig. 26, p. 81). Pieces filled with feces must be cut open, washed, and spanned on cork (Fig. 202).

No. 115.—*Fresh crypts of the large intestine of the rabbit.*—Cut a piece 1 cm. long from the lowermost portion of the large intestine (between two spherical masses of feces), place it on a dry slide, open it with the scissors, and spread it out with the mucous surface uppermost; add a drop of 0.7 per cent. salt solution, grasp the piece with forceps at the left edge and with fine scissors cut off an *extremely thin* strip. Transfer this with a drop of salt solution to another slide; by means of needles separate the muscularis from the mucosa and tease the latter a very little; apply a cover-glass with slight pressure. With a low power the crypts can be readily seen, but it is difficult to detect their orifices (Fig. 231). The epithelial cells are often granular in the portion bordering the lumen.

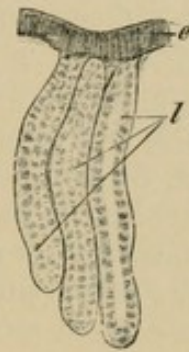


FIG. 231.—e, Epithelium; l, crypts. $\times 80$.

With the high power the cylinder epithelium of the surface can be well seen, from the side and from the surface. The content of the goblet-cells often is not clear, as in sections, but dark and granular.

No. 116.—*Blood-vessels of the stomach and the intestines.*—A stomach and intestine injected (p. 48) from the descending aorta are to be fixed in from 50 to 200 c.c. of Müller's fluid and hardened in gradually strengthened alcohols. One portion of each should be cut into thick (up to 1 mm.) sections and mounted unstained in xylol-balsam (Fig. 205), and another part used for horizontal preparations, which with the low power and change of focus are very instructive. For this purpose pieces of the large intestine 1 cm. square can be transferred from absolute alcohol to 5 c.c. of turpentine, instead of carbol-xylol, for vigorous clearing and mounted in xylol-balsam. It is also easy to strip the muscularis from the mucosa and mount the separate membranes in xylol-balsam.

No. 117.—*Nerve plexuses.*—For this purpose intestines with a thin muscularis are preferable, therefore the intestine of the rabbit and guinea-pig (not of the cat) are especially suitable. It is not necessary that the object be absolutely fresh; the small intestines of children several days after death can still be used. Prepare 200 c.c. of a dilute solution of

acetic acid (10 drops of glacial acetic acid or 25 drops of the ordinary acetic acid to 200 c.c. of distilled water). Then separate a piece from 10 to 30 cm. long of the small intestine from the mesentery. Cut it off and with the finger lightly press out the contents; tie one end of the piece of intestine and fill it at the other end with the dilute acetic acid; tie the open end and place the whole piece in the remainder of the diluted acetic acid. In one hour change the fluid. In twenty-four hours transfer the intestine to distilled water, with scissors open it along one side of the line of attachment of the mesentery and cut off a piece 1 cm. long. The muscularis can be readily separated from the mucosa with the aid of forceps; the two membranes are only firmly united at the attachment of the mesentery.

(a) *Plexus myentericus*.—If a piece of black paper be placed under the glass dish containing the tissue the white nodal points of the plexus can be seen by the unaided eye. Transfer a piece of the muscularis, about 1 cm. square, in a drop of the dilute acetic acid to a slide; examined with the lower power it furnishes a very pretty picture (Fig. 206 A). If it is desired to preserve the preparation place the tissue for one hour in 30 c.c. of distilled water, which must be changed several times, and then for from eight to sixteen hours in 5 or 10 c.c. of a 1 per cent. osmic acid solution, *in the dark*; wash the piece quickly in distilled water and mount in diluted glycerol. The osmium preparations are not as beautiful as the fresh ones in the acetic acid. In the guinea-pig both strata of the muscularis can be readily separated* (if the intestine is absolutely fresh on being filled with the dilute acid); the plexus remains attached to one stratum. Pieces of this should be placed for one hour in distilled water, then treated with gold chlorid (p. 47), and mounted in xylol-balsam. The gold-chlorid treatment is less adapted to human intestines, since both the muscular layers are also stained red and partially conceal the plexus.

(b) *Plexus submucosus*.—With a scalpel scrape the epithelium from the isolated mucosa; place a piece about 1 cm. square on a slide; apply a cover-glass, press upon it slightly, and examine with low powers (Fig. 206 B). To preserve the preparation proceed as in No. 117 (a); except that it is advisable to span the pieces on cork and before transferring them from the ninety-five per cent. alcohol to the carbol-xylol to press them somewhat, in order that the alcohol may be completely removed from the spongy mucosa.

In addition to nerves many blood-vessels are present, which are easily recognized by the structure of their walls, in part by the transversely placed nuclei of the muscle-fibers.

No. 118.—*The parotid, submaxillary, and sublingual glands*.—Cut from the named glands (human glands in winter are useful three or four days after death) a number of pieces from 0.5 to 1 cm. square and place

*Possibly the cause of the firm attachment of the muscle strata in man lies in the age of the object.

them in 30 c.c. of Zenker's fluid* (for further treatment see p. 33). Stain one piece in bulk in borax-carmines (p. 40). Embed another unstained piece in liver and cut the *thinnest* possible sections; small fragments about 2 mm. long can be used; stain them in Hansen's hematoxylin (p. 38), two or three minutes; the transfer of the sections to the staining solution must be done slowly, or the delicate structures will be torn to pieces; then stain with eosin (No. 3 b, p. 39), and mount in xylol-balsam. (Very thin sections should be examined in water after the staining in hematoxylin is completed, since the cell boundaries are then very much more distinct.) If the staining is successful the salivary tubules and the crescents are red. In the sublingual gland and in the mucous cells of the submaxillary gland the membrana propria also stains red; it must not be confused with the sections of the crescents, which latter are granular, while the membrana propria has a homogeneous appearance (Fig. 166). The mucous cells in the borax-carmines preparations are clear throughout. In the sections stained with hematoxylin they are sometimes clear, sometimes a pale blue of different shades; the portion which stains is a reticulum which occurs in certain functional stages of each mucous cell. The very short intercalated pieces of the submaxillary gland are difficult to find (Fig. 168); on the other hand, they may be easily seen in the parotid gland (also in that of the rabbit). Of the end-pieces only those which have been accurately halved are suitable for study. The numerous oblique and tangential sections are often very difficult to understand.

No. 119.—*The Pancreas*.—The human pancreas as a rule cannot be used. The treatment is the same as for the parotid gland, Technic No. 118. By this method the characteristic granular zone of the gland-cells, bordering the lumen, can only be seen with high powers and then not always, for the granules are exceedingly sensitive to water and very difficult to preserve. If a pin-head sized piece of the fresh pancreas of a cat or other mammal is teased in a drop of 0.75 per cent. salt solution and examined with a low power the end-pieces will appear spotted; the spots are the partly clear, partly granular divisions of the cells. With high powers the picture is like Fig. 232.



FIG. 232.—GLAND-CELLS FROM THE PANCREAS OF A CAT. Above groups of cells as they usually appear; below two isolated cells. $\times 560$.

No. 120.—*Granules of the salivary glands, the pancreas, and the cells of Paneth*.—Fix perfectly fresh pieces in potassium-bichromate-formol (p. 33) and harden in ascending alcohols (p. 35). Stain very thin sections with Heidenhain's iron-hematoxylin (p. 44) or with acid fuchsin. In staining with the latter add from 1 to 3 drops of the solution to from 5 to 10 c.c. of absolute alcohol, and leave the sections in this diluted stain for 24 hours. In many cases the granules can be plainly distinguished only with an immersion lens.

*The preparations represented in figures 164, 166, 168 were fixed and hardened after technic No. 108.

No. 121.—*Liver cells*.—Make an incision in a fresh liver and with the blade of a scalpel obliquely placed scrape the cut surface. Transfer the brown liver tissue adhering to the blade to a slide and add a drop of salt solution. Apply a cover-glass. Examine first with the low power, then with the high (Fig. 222 A). In addition to the liver cells the preparation contains numerous colored and colorless blood-cells.

No. 122.—*Hepatic lobules*.—Place small pieces (about 2 cm. cubes) of a pig's liver in from 30 to 50 c.c. of absolute alcohol. The majority of the lobules are hexagonal; they can be seen on the exterior of the liver with the unaided eye and after a moment become distinctly visible on the cut surface. The section of the central vein also becomes visible. In about three days sections can be cut; stain them with Hansen's hematoxylin (p. 38). The division into lobules can be well seen with the low power, but the hepatic cells, as well as the bile ducts, are less satisfactory for study. Better for this purpose is the following.

No. 123.—*Human liver*.—Place pieces about 2 cm. square, as fresh as possible, for four weeks in 200 c.c. of Müller's fluid for fixation and then in 100 c.c. of gradually strengthened alcohols for hardening. Examine unstained sections, cut parallel and also vertical to the surface, and stain others with Hansen's hematoxylin or with this and eosin (p. 39); mount in xylol-balsam. The demarcation of the lobules is indistinct, because of the slight development of the interlobular connective tissue. The division into lobules can be more readily perceived on macroscopic inspection, than on investigation with the microscope. For orientation the beginner should recall that *isolated* sections of blood-vessels always

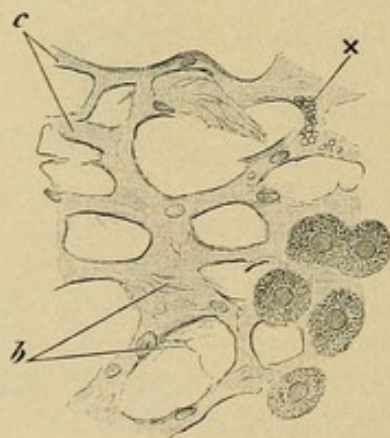


FIG. 233.—FROM A SHAKEN SECTION OF A HUMAN LIVER. $\times 240$. *c*, Blood capillaries, at *x* still containing blood corpuscles. *b*, Interlobular connective tissue. On the right side are five hepatic cells; the others have fallen out of the meshes of the capillary network.

represent intralobular veins; while groups of such sections represent branches of the portal vein, the hepatic artery, and the bile-duct, and always correspond to interlobular structures. Exact transverse sections of central veins can also be recognized by the trabeculae of hepatic cells radiating from them (Fig. 217).

No. 124.—For the demonstration of the *capillaries* and the *intralobular connective tissue*, which in ordinary preparations is scarcely visible, shake a number of thin, double-stained sections of human liver (No. 123) for from two to three minutes in a test-tube half filled with distilled water. The liver cells in part fall out; examine the edges of the preparation in a drop of water (Fig. 233). This preparation can be mounted in xylol-balsam, but the more delicate connective-tissue fibers disappear therein.

* For the study of the structure of the gall-bladder, as well as of the larger bile-ducts, only absolutely fresh tissue can be used, since the alkaline bile soon after death saturates the wall of the gall-bladder, stains it yellow, and makes it unfit for microscopic investigation.

No. 125.—*Blood-vessels of the liver.*

(a) Chloroform a rabbit and quickly place a 2 cm. cube of liver (without allowing much blood to flow from it) in 50 c.c. of absolute alcohol. In two days the natural injection can be seen on the surface; it is indicated by brown spots at the center of the lobules. Cut thick sections parallel to the surface and mount them unstained in xylol-balsam. Examine with a low power. Very frequently only the superficial strata of the liver contain filled blood-vessels.

(b) Of all injections that of the liver is most easily accomplished. Inject Berlin blue (p. 48), either through the portal vein or the inferior vena cava; in the latter case it is advisable to make an incision above the diaphragm; allow the heart to rest upon the latter and insert the canula through the right auricle into the inferior cava. The injected liver is to be placed in toto in about 500 c.c. of Müller's fluid; after six days pieces about 2 cm. square of the portions best injected are to be cut out, again placed for two or three weeks in about 150 c.c. of Müller's fluid, and finally hardened in 100 c.c. of gradually strengthened alcohols. Cut thick sections and mount them unstained in xylol-balsam (Figs. 223, 224, 226).

No. 126.—*Exhibition of gland lumina by Golgi's "black reaction."*—Place small pieces of the root of the tongue, of the stomach, of the salivary glands, and of the liver for three days in the potassium-bichromate-formol mixture and then in the silver solution. For further treatment see page 45. Very often the staining does not succeed until after the procedure has been repeated once or twice. After-staining (p. 47) is strongly advised. In the liver the "lattice-fibers" occasionally stain.

No. 127.—*Epithelium of the peritoneum.*—Proceed as in No. 40, p. 153, but instead of taking the mesentery, which also yields instructive pictures, use the greater omentum. The pieces may be stained in Hansen's hematoxylin (p. 38) and mounted in xylol-balsam (Fig. 228).

VI. THE RESPIRATORY ORGANS.

THE LARYNX.

The *mucous membrane* of the larynx is a continuation of the pharyngeal mucous membrane and like this is composed of an epithelium, a tunica propria, and a submucosa, which latter connects the mucous membrane with the underlying parts. The epithelium over nearly the whole of the organ is a many-row (p. 77) ciliated epithelium; the ciliary wave is directed toward the cavity of the pharynx. On the true vocal cords, on the anterior surface of the arytenoid cartilages, and on the laryngeal surface of the epiglottis the epithelium is of the stratified squamous variety. The *tunica propria* consists of numerous elastic fibers and of fibrillar connective tissue, which in the lower animals is condensed to a membrana propria immediately beneath the epithelium. The tunica propria is the site of a varying number of leucocytes; even solitary nodules (p. 146) are found in the mucous membrane of the ventricle of the larynx (Morgagni). Papillæ mainly occur in the mucous membrane clothed with stratified squamous epithelium; on the free border and on the lower surface of the vocal cords the papillæ are merged in longitudinal ridges. On the laryngeal surface of the epiglottis only isolated papillæ are present, on which are short taste-buds. The *submucosa* contains branched alveolo-tubular glands, from 0.2 to 1 mm. in size; the middle of the vocal cords for a certain distance from the free edge is without glands.

The *cartilages* of the larynx principally consist of the hyaline variety, which in a measure exhibits the peculiarities of the costal cartilages (p. 97). The hyaline cartilages are the thyroid, the cricoid, the greater portion of the arytenoids, and often the triticeous cartilages. The epiglottis, the cuneiform cartilages (Wrisbergi), the cornicular cartilages (Santorini), the median portion of the thyroid, and the apex and vocal process of the arytenoid cartilages are of the elastic (fiber-net) variety. Occasionally the triticeous cartilages are composed of fibro-cartilage. Between the twentieth and thirtieth years of life ossification (chiefly endochondral) begins in the thyroid and cricoid cartilages.

The larynx is richly supplied with *blood-vessels* and *nerves*. The blood-vessels form two or three networks extending in planes parallel to the surface and a close subepithelial capillary plexus.

The *lymph-vessels* form two anastomosing networks also extending in horizontal planes, of which the superficial consists of narrower vessels and lies beneath the blood capillary network.

The *nerves* include microscopic ganglia in their course and form a deep and a superficial plexus. The nonmedullated nerves end partly as little subepithelial terminal trees, the twigs of which are provided with enlargements, or in end-bulbs, and partly intraepithelial in free branches and in tastebuds (see the Gustatory Organ). Below the vocal cords subepithelial nerve-endings and buds are wanting; but many intraepithelial nerve-fibers are present, that spin networks about the individual gustatory cells.

THE TRACHEA.

The ciliated mucous membrane* of the trachea possesses a structure like that of the larynx, excepting only that the elastic fibers form a close network in which the fibers pursuing a longitudinal direction predominate. This network lies immediately beneath the epithelium and above the mixed glands. The cartilages are of the hyaline variety. The posterior wall of the trachea is composed of a layer of transversely arranged smooth muscle-fibers, that usually is covered by a stratum of muscle-fibers extending longitudinally.† The glands of the posterior wall are distinguished by their size (2 mm.); they not infrequently penetrate the muscular layer, so that they lie in part in the fibrous tissue behind it.

The behavior of the blood-vessels, lymph-vessels, and nerves is the same as in the larynx; the nerve-fibers ending on the smooth muscle-fibers of the trachea are nonmedullated and come from the nerve-cells of the small (sympathetic) ganglia; the sensory nerve-fibers are medullated and of cerebrospinal origin (? cf. remark †, p. 219).

THE BRONCHI AND THE LUNGS.

The lungs may be regarded as compound alveolo-tubular glands, in which, as in all glands, excretory and secretory (in this case respiratory) divisions are distinguished. The *excretory division* comprises the larynx and the trachea with its branches, the bronchi. Each bronchus on entering the lung divides repeatedly and within the same undergoes continual subdivision, by the direct giving off of small lateral twigs and by the branching at acute angles with gradual decrease in the caliber of the large branches; in this way each bronchus breaks up into minutest twigs, that nowhere anastomose with one another and that retain the character of the excretory duct to a diameter of 0.5 mm.

* The mucous membrane which covers the posterior wall of the trachea appears to vary; at least I have found there, in the mucous membrane of a healthy man, stratified squamous epithelium and a tunica propria with papillæ.

† The smooth musculature of the trachea and its branches is as richly provided with elastic fibers as that of the intestine (p. 275).

At this point the *respiratory division* begins. Isolated hemispherical evaginations, the *alveoli*, appear at irregular intervals on the walls of the minute bronchial branches. Such bronchial branches are called *respiratory bronchioles*. These divide and lead into the *alveolar ducts*, which differ from the bronchioles only in being completely encircled by alveoli. The alveolar ducts divide at right or acute angles and pass without sharp demarcation into the slightly expanded, blind *alveolar sacks* or *terminal vesicles* (less correctly, infundibula), the walls of which

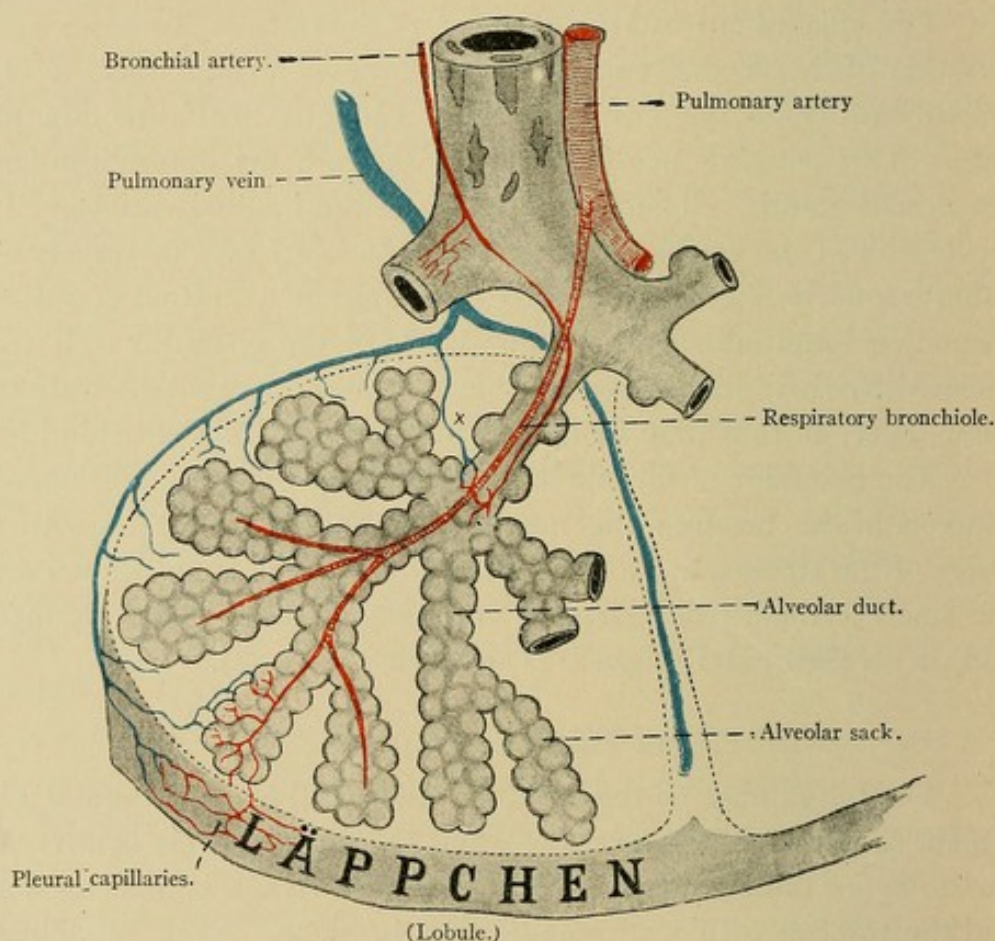


FIG. 234.—SCHEME OF THE TERMINAL RAMIFICATIONS OF THE HUMAN BRONCHIAL TREE AND ITS BLOOD-VESSELS.

are thickly beset with alveoli.* Each alveolus is open, not only toward the alveolar sack,—this broad opening is termed *base*—but also is in direct communication with neighboring alveoli by means of a widely varying number of minute canals, the so-called *pores* (Fig. 237 B).

The entire respiratory division is separated by connective tissue into *lobules* from 0.3 to 3 sq. cm. in size. All the branches of the ex-

* To describe as atrium a special division between the alveolar duct and the alveolar sack appears to me superfluous; it cannot be distinguished in good molds of the human lung.

cretory division down to a diameter of from 1.5 to 1 mm. lie *between* the lobules, are interlobular.

The *minute structure of the bronchi* and their largest branches does not differ from that of the trachea. Gradually modifications appear, which first involve the cartilages and the musculature. The *C-shaped ring cartilages* are replaced by irregular plates, lying on all sides of the bronchial wall. They diminish in size and thickness with the decrease in the diam-

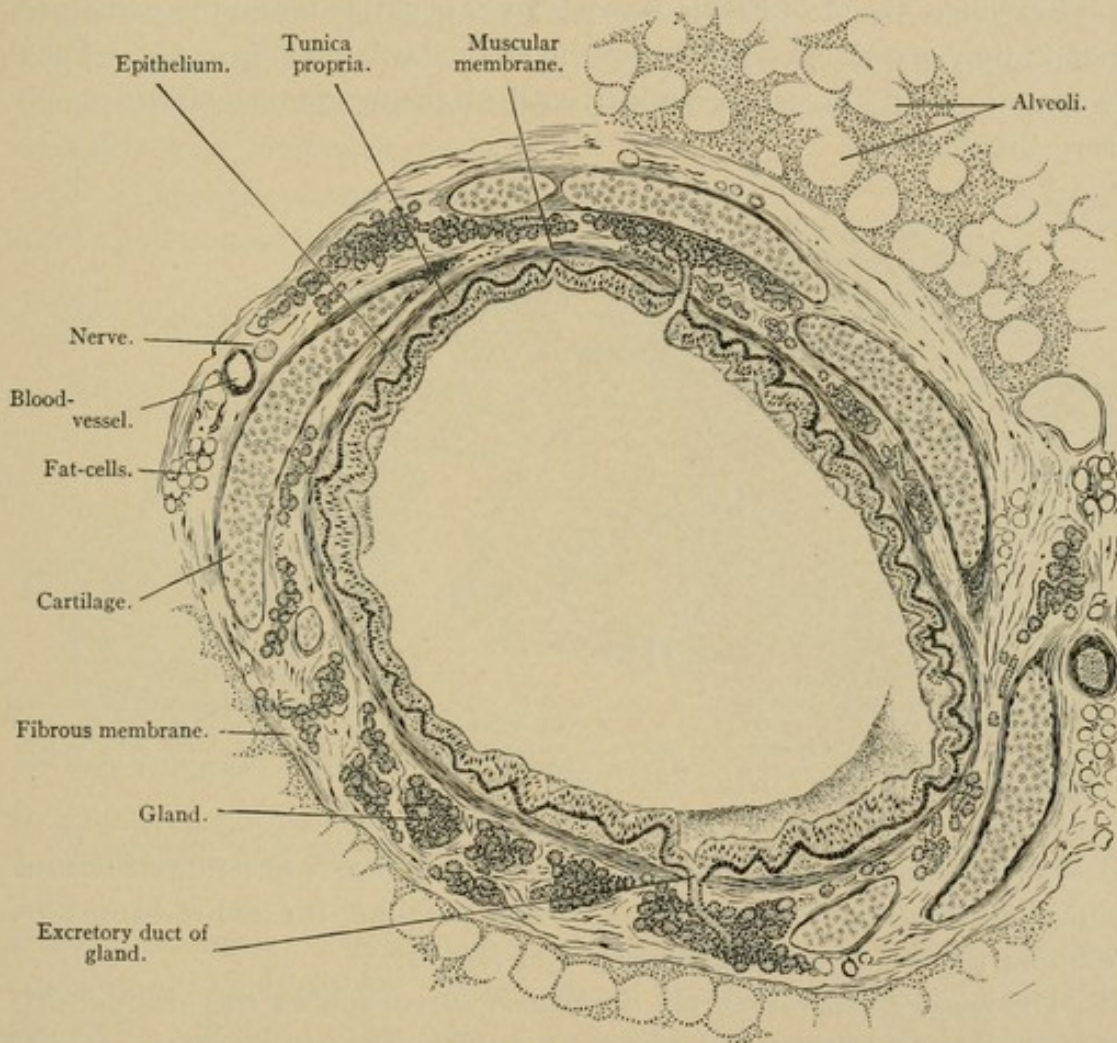


FIG. 235.—CROSS-SECTION OF A BRONCHIAL BRANCH, TWO MILLIMETERS THICK, OF A CHILD. $\times 30$. Technic No. 128 d.

eter of the bronchial branches and disappear in the branches 1 mm. in diameter.

The *smooth muscle-fibers* are circularly disposed in a continuous layer lying within the cartilages and embracing the *entire* circumference of the tube. The thickness of the muscular layer decreases with the diameter of the bronchial branches; but muscle-fibers are still present in the alveolar ducts. They are wanting in the alveolar sacks.

The *mucous membrane* is thrown into longitudinal folds and consists of a many-rowed ciliated epithelium containing goblet-cells, that in the minute bronchial branches becomes gradually reduced to a single-row epithelium, and of a connective-tissue tunica propria. The latter contains a network of numerous longitudinally disposed elastic fibers and leucocytes in greatly varying number. Occasionally the latter form solitary nodules, from the crest of which leucocytes wander through the epithelium into the bronchial tube.

Branched alveolo-tubular *mixed glands* occur as far as the cartilages extend; they are situated outside of the muscular layer (Fig. 235). They are numerous and do not disappear until at the beginning of the respiratory bronchioles.

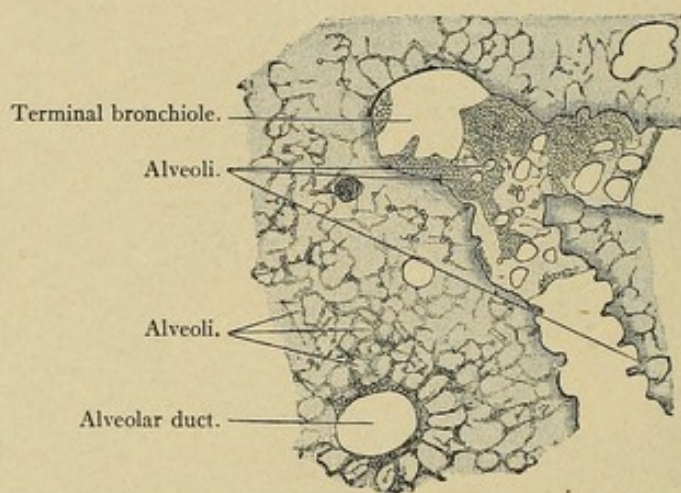


FIG. 236.—FROM A SECTION OF THE LUNG OF ADULT MAN. $\times 50$. The terminal bronchiole divides into two branches (on the right). A portion of the wall of the bronchiole fell within the plane of the section; here the entrance to the alveoli is seen from above; in the lower branch the alveoli are viewed from the side. The epithelium of the bronchiole is mixed. The epithelial outfit of the alveoli is only partially visible with this magnification. Technic No. 129.

External to the cartilages is a *fibrous membrane* consisting of fibrous connective tissue and elastic fibers, which envelops the entire bronchus, including the accompanying vessels and nerves.

The *minute structure of the respiratory division*, after the gradual disappearance of cartilages and glands, is distinguished in particular by the nature of the epithelium.

The *respiratory bronchioles*, succeeding the smallest bronchial branches, at their beginning still contain a layer of single-row ciliated epithelium; in their further course the cilia are lost, the cells become cubical, and between these another kind of epithelial cell appears, in the form of thin nonnucleated plates of different sizes. These plates and isolated or small groups of cubical cells form an epithelium called *respiratory epithelium*. The transition of the cubical epithelium into the respiratory epithelium is not abrupt, but occurs in such wise that on the one side of the

bronchiole cubical, on the other side respiratory epithelium is found, or that groups of cubical cells are surrounded by respiratory epithelium and the reverse. Hence the respiratory bronchioles contain a mixed epithelium (Fig. 236 and Fig. 237 *A*). Since the respiratory epithelium steadily gains in extent and the groups of cubical cells become steadily less frequent, the epithelium of the bronchioles changes into that of the alveolar ducts.

The epithelium of the *alveolar ducts* and of the *alveoli* is the same as the respiratory epithelium of the bronchioles. The developmental history teaches that the smaller nonnucleated plates originate from cubical epithelial cells, that become flattened by inspiration, that is, by the complete stretching of the alveolar wall. The larger plates are formed by the subsequent blending of sev-

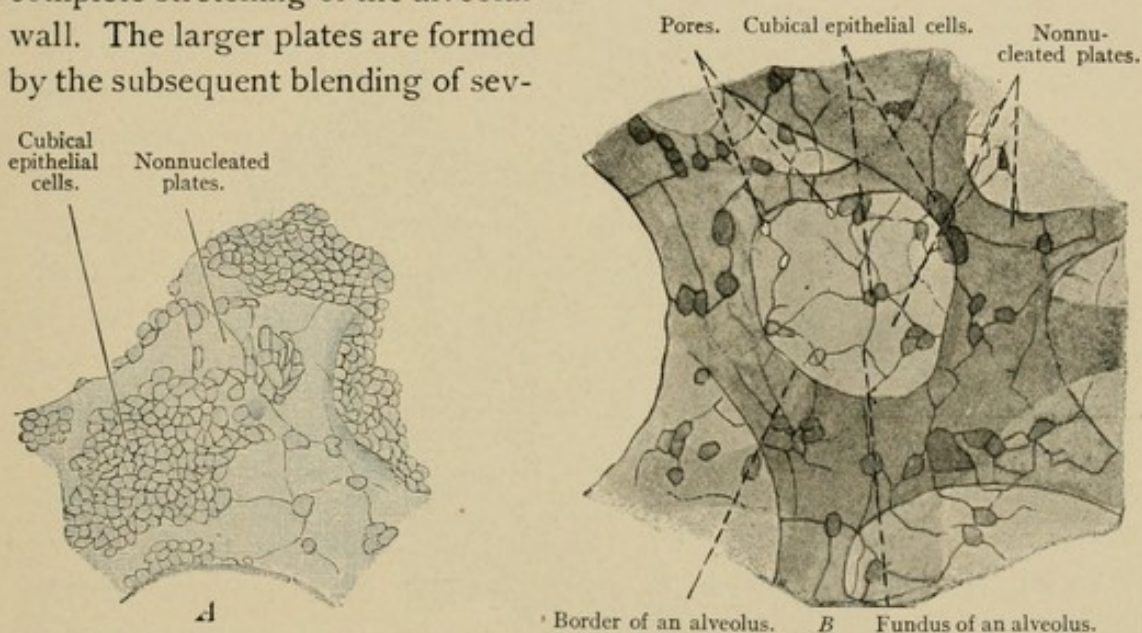


FIG. 237.—FROM SECTIONS OF A HUMAN LUNG. $\times 240$. *A*. Mixed epithelium of a respiratory bronchiole. *B*. An alveolus sketched with change of focus. The border of the alveolus is shaded; it is covered by the same epithelium as that of the (clear) fundus of the alveolus; the nuclei of the cells are invisible. Technic No. 129.

eral smaller ones. The alveoli of old embryos and of stillborn children contain only cubical cells. The walls of the alveolar ducts and of the alveoli, in addition to the previously mentioned muscle-fibers in the former, are composed of a delicately striated ground layer and many elastic fibers. The latter are circularly arranged in the alveolar ducts; at the entrance ("base") to the alveolus the elastic fibers form a thick ring, while delicate, convoluted little fibers occur in the entire wall of the alveolus (Fig. 238). The elastic rings of neighboring alveoli grow together at the points of contact and form the *alveolar septa*.*

* This wealth of elastic fibers enables the alveolus to expand during inspiration to three times its usual diameter and during expiration to return again to its original diameter of from 0.1 to 0.3 mm.

The *interlobular connective tissue* occurring between the lobules of the lungs supports the larger blood- and lymph-vessels and, besides fine elastic fibers and a few connective-tissue cells, in the adult contains black pigment granules and minutest particles of carbon, that have come there by inhalation. In children the interlobular connective tissue is more richly developed and therefore the demarcation of the lobules is more distinct.

The surface of the lung is covered by the *visceral pleura*; this is composed of connective-tissue, numerous fine elastic fibers, and on its

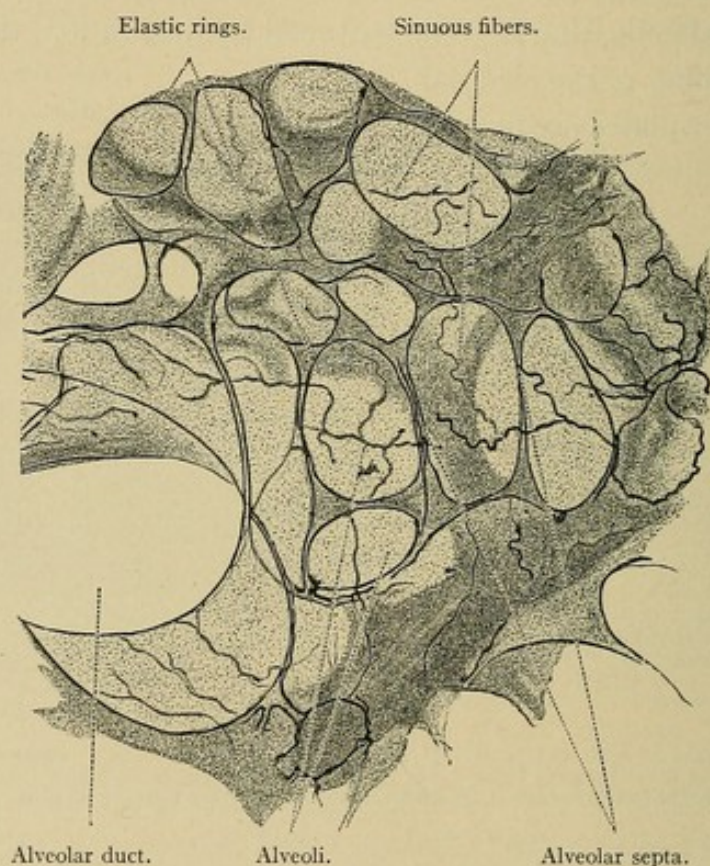


FIG. 238.—SECTION OF THE LUNG OF A RABBIT. $\times 220$. Stained elastic fibers. Technic No. 130 b.

free surface is clothed with a simple stratum of flat, polygonal epithelial cells.* The *parietal pleura* has the same structure, but contains fewer elastic fibers.

The lungs have two *systems of blood-vessels*: 1, the system of pulmonary arteries and veins, serving the purposes of respiration; 2, the system of bronchial arteries and veins. The branches of the *pulmonary artery* penetrate at the hilus of the lung and run beside the bronchial branches in the lobules to the bronchioles, alveolar ducts, and alveolar

* In the dog and the rabbit they are provided with a delicate hair border.

sacks; where they break into a very narrow-meshed capillary plexus, lying immediately beneath the respiratory epithelium of the alveoli, alveolar ducts, and respiratory bronchioles and communicate with a wide-meshed capillary net lying under the pulmonary pleura. The *pulmonary veins* arise, one each at the base of an alveolus (Fig. 239), take up on the surface of the lung veins from the capillaries of the pleura, and collect in small trunks that run at the periphery of the lobules and only later approach the larger bronchial branches. The *bronchial arteries* provide for the bronchial ramifications up to the respiratory bronchioles and supply a deep capillary plexus for

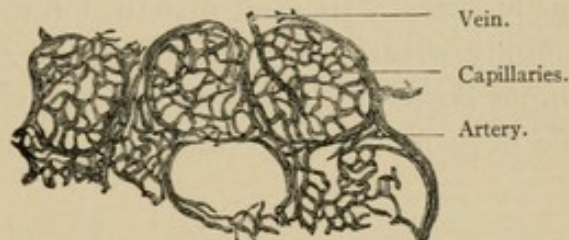


FIG. 239.—FROM A SECTION OF THE LUNG OF A CHILD, INJECTED THROUGH THE PULMONARY ARTERY. $\times 80$. Of the five alveoli drawn the three upper ones are fully injected. Technic No. 131.

the glands and muscles and a superficial plexus for the tunica propria; also the walls of the pulmonary arteries and veins, the bronchial lymph-glands, and the pulmonary pleura receive branches from the bronchial arteries. The *bronchial veins* empty their blood partly into the pulmonary veins (Fig. 234 $\times$), partly in the territory of the azygos vein. Large and small anastomoses exist between the pulmonary and bronchial arteries.

The *lymph-vessels* form a superficial and a deep plexus; the well-developed *superficial plexus*, lying beneath the pleura, is connected with pea-sized lymph-nodes irregularly distributed under the pulmonary pleura, and opens by several small, valved trunks into the bronchial lymph-glands. The wide-meshed *deep plexus*, situated in the interlobular connective tissue, collects the lymph-vessels of the bronchial mucous membrane* and of the walls of the blood-vessels; from this small, valved trunks proceed which, running with the bronchial ramifications, pass out at the hilus and there open in the bronchial lymph-glands (cf. p. 141).

The numerous *nerves* of the lungs, originating from the sympathetic and the vagus, contain medullated and nonmedullated nerve-fibers and small groups of ganglion cells. The nerve endings stand chiefly in relation to the blood-vessel walls.

THE THYROID GLAND.

The thyroid gland arises essentially from a median proliferation of the ventral wall of the esophagus and at first has the appearance of a

* Lymph-vessels are not present in the alveolar ducts and alveolar sacks (of the dog).

tubular, compound, retiform gland; its excretory duct, the *thyro-glossal duct*, opening at the foramen cæcum of the tongue, becomes obliterated in an early embryonal period and, excepting a few fragments, atrophies and disappears; the network of gland tubules, that at first are not hollow, becomes constricted into short pieces, "follicles," which are bound together in lobules by loose connective tissue interlaced with elastic fibers. In adult man the follicles are oval sacks blind at both ends, differing greatly in diameter (from 40 to 120 μ), and clothed with a simple layer of sometimes cubical, sometimes cylindric epithelial cells, that in man contain granules of a partially fatty nature. The lumen of

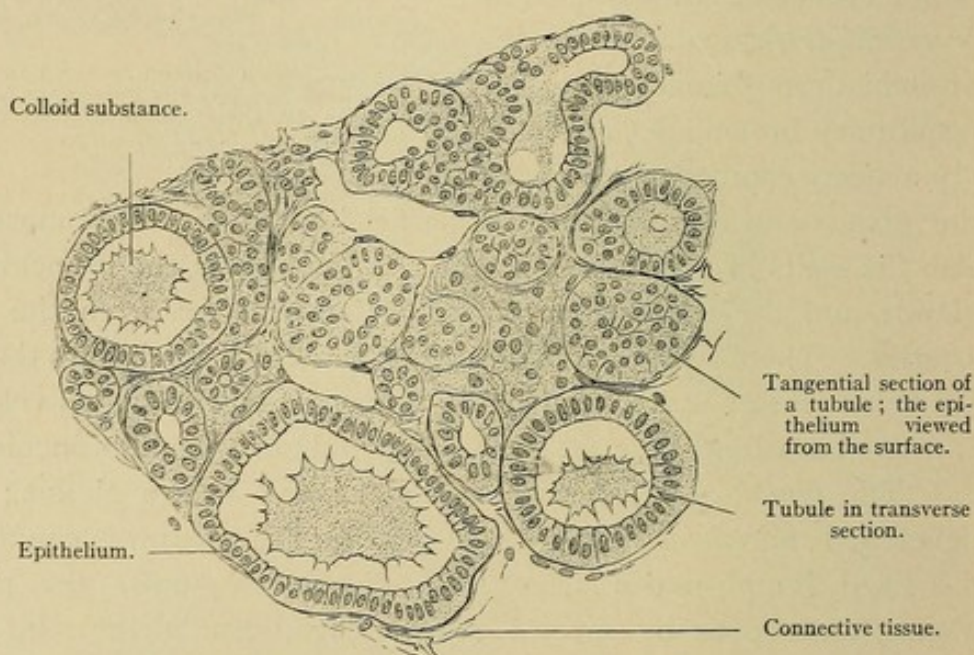


FIG. 240.—A LOBULE FROM A THIN SECTION OF THE THYROID GLAND OF ADULT MAN. $\times 220$. Note the difference in the diameter of the tubules. Technic No. 132.

the sack is filled with a homogeneous, viscid mass, the *colloid substance*, a product of the epithelial cells.* The very numerous *blood-vessels* form a capillary plexus enveloping the follicles and lying close beneath the epithelium. The equally numerous *lymph-vessels* form a network lying between the follicles. The *nerves* run with the ramifications of the blood-

* Formerly the colloid substance was credited as a characteristic of the thyroid gland; but since masses resembling the colloid have been found in the hypophysis (p. 208), and since in the blood- and lymph-vessels of the throat the coagulated blood may very closely resemble the colloid, this sign loses its diagnostic value. The manner in which the secretion produced by the thyroid gland is discharged is not yet clear. It has been observed that at all the nodal points of the net of terminal bars the cement substance is wanting; perhaps this is an instance of the sundering of the epithelial cells to permit the passage of the secretion to the lymph channels. Possibly the secretion is taken up by the blood-vessels. That the secretion plays an important part in the economy of the body, by rendering poisonous products of metabolism innocuous, has been established by experiment.

vessels and form an enveloping plexus, chiefly for the vessels, partly also for the gland sacks. The penetration of terminal twigs into the epithelium has not been observed.

On the posterior surface of each lateral lobe of the thyroid gland are found from one to four "epithelial corpuscles," two millimeters in size; they consist of cords or nests of epithelial cells, that surrounded by blood capillaries and connective tissue have arisen from the visceral clefts. In different mammalian animals, embryos and adults, there has been found in each lateral lobe of the thyroid gland a duct lined with squamous to cylinder ciliated epithelium, the *central canal of the thyroid gland*, that is connected with the surrounding gland lobules and the epithelial corpuscles.

THE THYMUS.

The thymus arises paired out of the epithelium of the third visceral cleft; the originally hollow evagination sends out solid branching

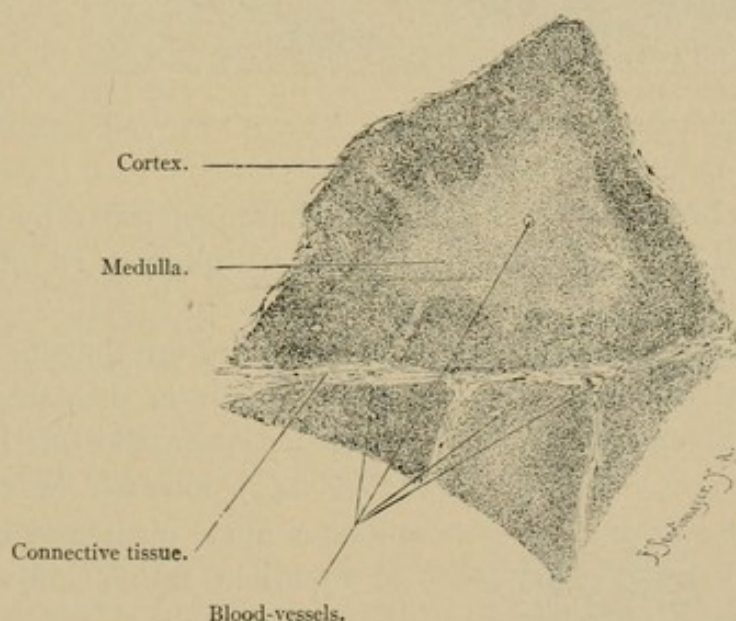


FIG. 241.—SECTION OF SEVERAL SECONDARY LOBULES OF THE THYMUS OF A SEVEN-DAY-OLD RABBIT. $\times 50$. The lower lobules are sectioned tangentially, so that chiefly cortex is visible. Technic No. 133.

sprouts, in the vicinity of which conspicuous masses of adenoid tissue develop; numerous leucocytes wander from here into the epithelium and make rents through which also the blood-vessels and connective tissue penetrate. During childhood the thymus consists of lobes from 4 to 11 mm. in size; fibrous connective tissue intermixed with delicate elastic fibers sends septa in each individual lobe, whereby a subdivision in smaller, one millimeter square ("secondary") lobules is effected. The lobules are connected by means of a "medullary cord," from 1 to 3 mm. in thickness. The appearance of these lobules varies greatly; occasionally each lobule consists wholly of adenoid tissue, which is more densely developed at the periphery than in the center, so that a darker cortical

portion can be distinguished from a lighter medullary substance (Fig. 241). In these cases, owing to the extensive wandering in of the leucocytes, the epithelium has almost entirely disappeared. In other cases, particularly in man, the medullary substance contains larger or smaller

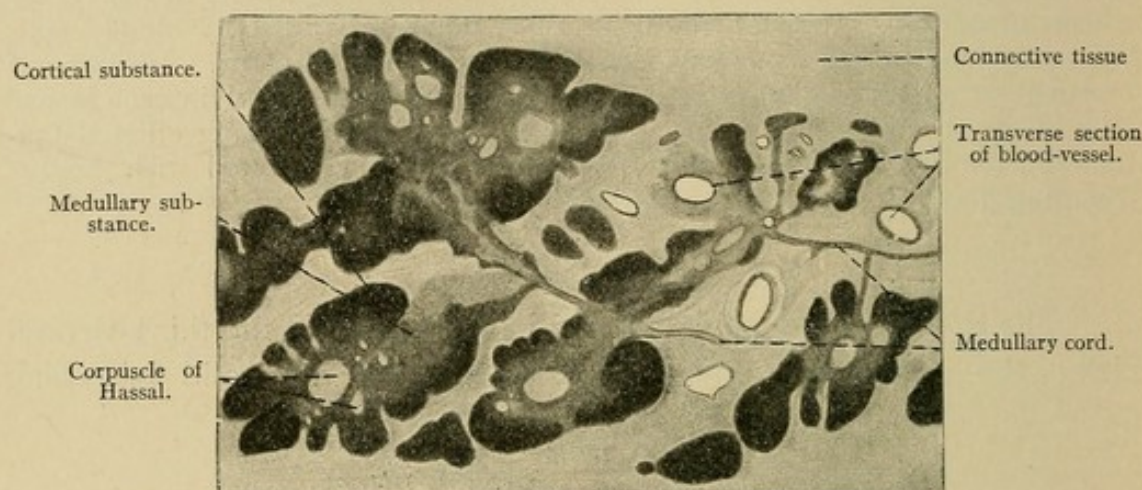


FIG. 242.—TRANSVERSE SECTION OF A PORTION OF THE THYMUS OF A CHILD ONE AND THREE-QUARTER YEARS OLD. $\times 21$. Technic No. 133.

groups of epithelial cells, that not infrequently are penetrated by wandering leucocytes, and also the branches of the medullary cord often consist of distinct epithelial cells (Fig. 242).

The medullary substance and medullary cord are the site of concentrically striated corpuscles, of from 15 to 180 μ in diameter, which are altered balls of epithelial cells.* These "corpuscles of Hassal" (Fig. 243) increase in size and number after birth and can still be found in the remains of the thymus in adults.

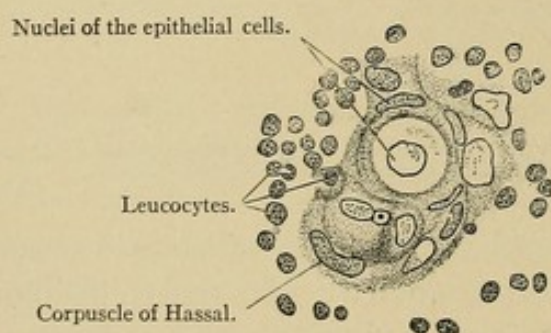


FIG. 243.—CORPUSCLE OF HASSAL FROM A SECTION OF THE THYMUS OF A YOUNG DOG. $\times 560$. Technic No. 133.

The *arteries* run between the cortex and the medulla and feed capillaries which chiefly develop in the former, in the latter only where it is not composed of epithelium. The *veins* collecting from the latter partly run in the medulla,

partly they empty in large venous stems lying between the lobules. The numerous *lymph-vessels* arise from wide lymph spaces lying immediately on the surface of the lobules and form large stems lying in the inter-

* Also structures resembling the epithelial corpuscles of the thyroid gland are said to occur.

lobular connective tissue, that subsequently proceed as valved vessels alongside the larger blood-vessels. The *nerves* end chiefly on the blood-vessels, only an extremely few small fibers penetrate the medulla, where they terminate in free ends.

The atrophy of the thymus, of which the period of onset is extremely variable, takes place in such wise that the larger portion of the adenoid tissue vanishes and fat appears in its place. Not only leucocytes, but also colored blood-cells arise in the adenoid tissue. Hematoblasts (p. 140) also have been found in it.

TECHNIC.

No. 128.—*The larynx, the bronchi, and the thyroid gland.*—Of animals the adult cat is particularly recommended. Expose the trachea above the manubrium; cut it and the esophagus through transversely and dissect both loose upwards (see No. 103, p. 298). The tongue may be removed with these parts. The thyroid gland should be allowed to remain attached to the larynx. The whole is placed for from two to six weeks in 200 or 400 c.c. of Müller's fluid, then washed for one hour in running water and hardened in 200 c.c. of gradually strengthened alcohols (p. 35). In about eight days cut sections, transverse and longitudinal, through the vocal cords and through pieces of the trachea; stain them for five minutes in Hansen's hematoxylin (p. 38) and mount in xylol-balsam. Particularly instructive are sections taken transversely through the vocal cords, in which the mucous membrane, glands, muscles, blood-vessels, nerves, and cartilage afford the most varied study. Very beautiful pictures are obtained by staining the sections with borax-carmin (p. 40) and with resorcin-fuchsin (No. 13, p. 43).

No. 128 a.—*The bronchi.*—From an animal just killed (rabbit)* remove the lungs, fix them in Müller's fluid and harden them in gradually strengthened alcohols, like No. 128. In eight days cut out of the lung 1 cm. cubes that contain a portion of a longitudinally disposed bronchus. With the scissors remove the greater part of the attached lung tissue; embed the bronchus in liver and make thin transverse sections, which may be stained in Hansen's hematoxylin (p. 38) or after No. 13, p. 43, and mounted in xylol-balsam (Fig. 235). This method is also applicable for the exhibition of the alveoli and the alveolar passages.

No. 129.—*The respiratory epithelium.*—For the demonstration of this tissue only animals just killed can be used. Young kittens (*not* newborn) are suitable; they should be killed by decapitation. The trachea and lungs should be carefully taken out and filled by means of a glass pipet with a previously prepared solution of silver nitrate (50 c.c.

* The lungs of cats are less suitable, because of the conspicuous masses of fat that often accompany the bronchial branches.

of a 1 per cent. solution to 200 c.c. of distilled water). The trachea should then be tied fast and the whole placed for from one to twelve hours in the remainder of the silver solution and stood in the dark. On removing them from the silver solution the lungs should be quickly washed with distilled water and transferred to 150 c.c. of gradually strengthened alcohols, in which they may remain (in the dark) indefinitely. The reduction can be undertaken in an hour after the silver injection or later. For this purpose the lungs in the alcohol should be exposed to *sunlight*, in which in a few minutes they become a deep brown. Then with a *very sharp* razor cut sections, taking care not to compress the tissue. Despite the hardening in alcohol the lung tissue is still soft and allows only thick sections to be cut. Sections are most easily cut in a direction parallel to the surface. Place the sections for from ten to sixty minutes in 5 or 10 c.c. of distilled water to which a crystal of common salt about the size of a lentil has been added, and mount them unstained in xylol-balsam. (It is not advisable to employ nuclear staining, since not only the nuclei of the epithelial cells, but also those of the capillaries and other tissues are colored, and consequently the picture becomes very complicated.) Orientation in such sections is not altogether easy. The investigation should be begun with the low power. The small alveoli are easily recognized; the somewhat larger spaces correspond to alveolar ducts. The outlining of the epithelium is on the whole finer with medium magnification (80 diameters), and by no means equally good in all places. The cubical epithelial cells are usually colored a somewhat deeper brown. Find a good place, study it with the high power (240 diameters), and by changing the focus (elevating and depressing the tube) practice orientation in the relief of the preparation; with high magnification, either only the interior or the margin of an alveolus can be distinctly seen. Fig. 237 *B* was drawn with change of focus. The pores of the alveoli can not be demonstrated on each alveolus.

No. 130.—*Elastic fibers of the lungs, (a) fresh.*—With the scissors placed on a freshly cut surface of the lung (the lung need not be fresh), cut a flat piece about 1 cm. square, spread it out with needles on a dry slide, apply a cover-glass and treat it with two drops of potash lye (p. 23) diluted one-half with water; the diluted lye destroys all parts excepting only the elastic fibers, the thickness and arrangement of which can be easily investigated with the high power (240 diameters).

(*b*) *For permanent preparation.*—Fix 1 or 2 cm. cubes of lung in absolute alcohol (§ 4, p. 31) for forty-eight hours, stain thick sections with resorcin-fuchsin (p. 43) and mount in xylol-balsam (Fig. 238).

No. 131.—*Blood-vessels of the lungs.*—Inject the lung from the pulmonary artery with Berlin blue (p. 48); fix it in Müller's fluid, and harden it in alcohol. Cut thick sections, principally parallel to the surface of the lung (Fig. 239).

No. 132.—*The thyroid gland.*—Thin sections of the gland, hardened in toto (see No. 128), are to be stained with picrocarmine (p. 41) and mounted in xylol-balsam (Fig. 240). The retracted colloid masses

stain an intense yellow. Examine thick sections in glycerol, in which the lymph-vessels filled with colloid substance are often distinctly visible. Vacuoles in the colloid are artifacts produced by fixation.

No. 133.—*The thymus*.—Place the thymus of a newborn infant in potassium-bichromate-acetic acid (p. 32) and harden it in gradually strengthened alcohols. Stain sections with Hansen's hematoxylin; mount them in xylol-balsam (Fig. 241). Care should be taken not to confuse the cross-sections of the blood-vessels, the lumina of which when they are not true transverse sections change in elevating and depressing the tube, with the concentric corpuscles of Hassal. The preparation represented in Fig. 242 is from a thymus fixed in Flemming's mixture and stained with safranin.

VII. THE URINARY ORGANS.

THE KIDNEYS.

The kidneys are compound tubular glands, which consist wholly of minute tubes, the *uriniferous tubules*. The macroscopically perceptible differences between the peripheral and the central portions of the organ, the so-called *cortical* and *medullary* regions, are principally determined by the course of the uriniferous tubules, the divisions within the cortex pursuing a tortuous, those within the medulla a straight course.

Each *uriniferous tubule* begins in the cortex as a spherical dilatation enveloping blood-vessels, the *renal corpuscle* (Malpighi, Fig. 244), which occasionally is marked off by a constriction, the *neck*, from the greatly convoluted succeeding division, the *convoluted tubule* (*tubulus contortus*), at first invariably directed corticalward. This passes into a straight portion, that is at first centrally directed, but soon turns back and forms a loop, *Henle's loop*, in which a *thin descending* and a *thick ascending limb* may be distinguished.* The latter passes into a shorter convoluted portion, the *intercalated piece*,† that by means of a narrower connecting piece empties into a *collecting tubule*. These collecting tubules during their centrally directed course take up other connecting pieces, farther on unite under acute angles with neighboring collecting tubules, and converge toward the apex of a renal papilla, where, diminished in number but greatly increased in diameter, they open as the *papillary duct*. Henle's loop-tubules and the collecting tubules are named *straight tubules* (*tubuli recti*). Each uriniferous tubule pursues a completely isolated course

* Since the transition of the thin into the thick division does not always lie at the curve of the loop, the designation "ascending" or "descending limb" is less advisable.

† The intercalated division is always in contact with the renal corpuscle to which it belongs, owing to the mode of development of the uriniferous tubule (Fig. 244).

until it is taken up by a collecting tubule. The loops of Henle and the peripheral portions of the collecting tubules are grouped together in

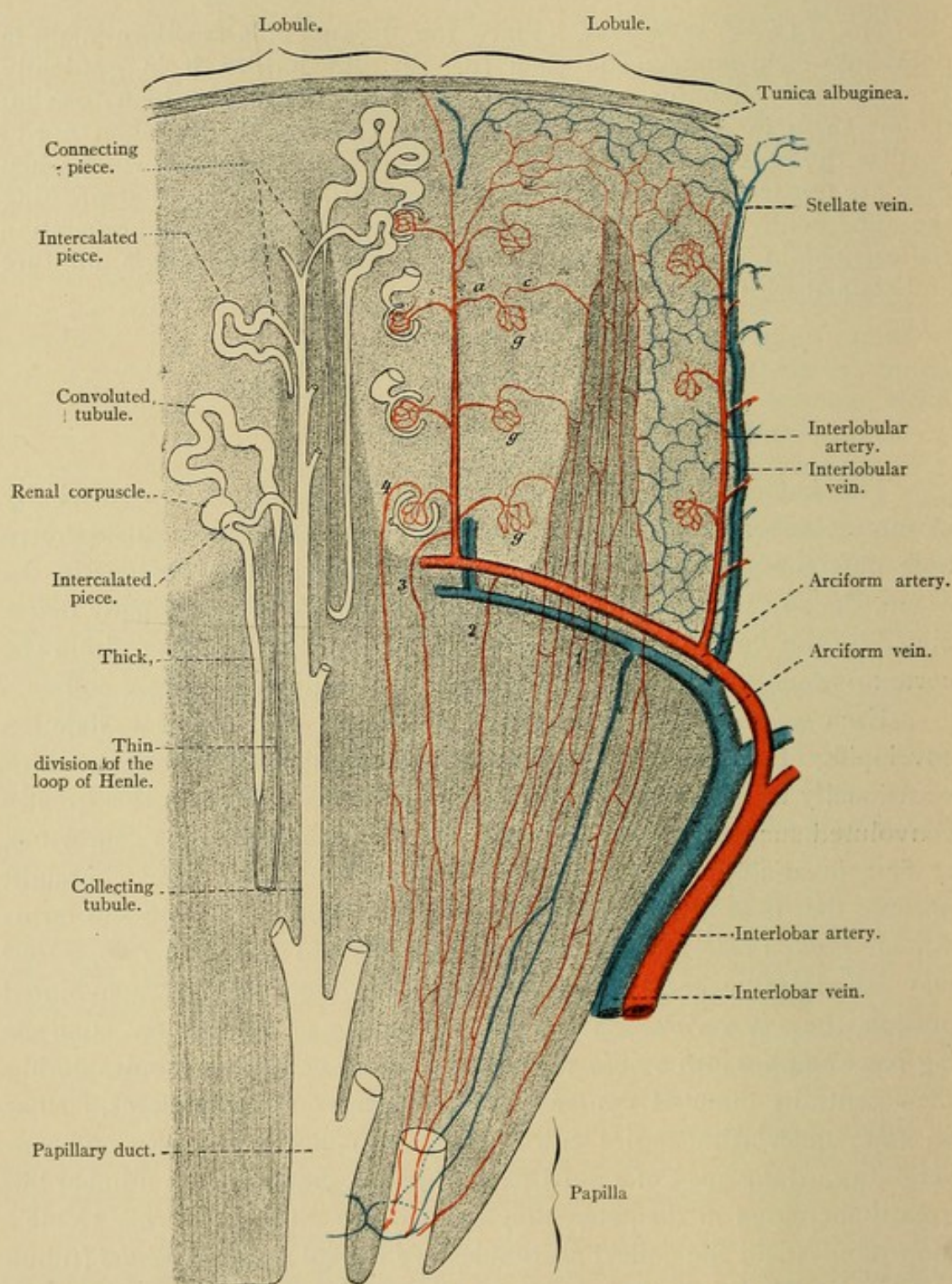


FIG. 244.—SCHEME OF THE COURSE OF THE URINIFEROUS TUBULES AND THE RENAL VESSELS.

bundles as they pass toward the medulla and form the structures in the cortex known as the *medullary rays* or the *pyramids of Ferrein*.

The uriniferous tubules possess in their entire length a single-layered, single-rowed epithelium, but their minute structure differs so greatly in the

several divisions that a separate consideration of each division is necessary. The *renal corpuscle*, from 0.13 to 0.22 mm. in size, consists of a spherical plexus of blood-vessels, the *glomerulus*, and the expanded, invaginated, blind initial piece of the uriniferous tubule, the *capsule of the glomerulus* (Bowman). The glomerulus lies within the invaginated portion of the capsule, and is almost completely enveloped by it. The invagination is similar to that on a large scale of the heart in the pericardium. Accord-

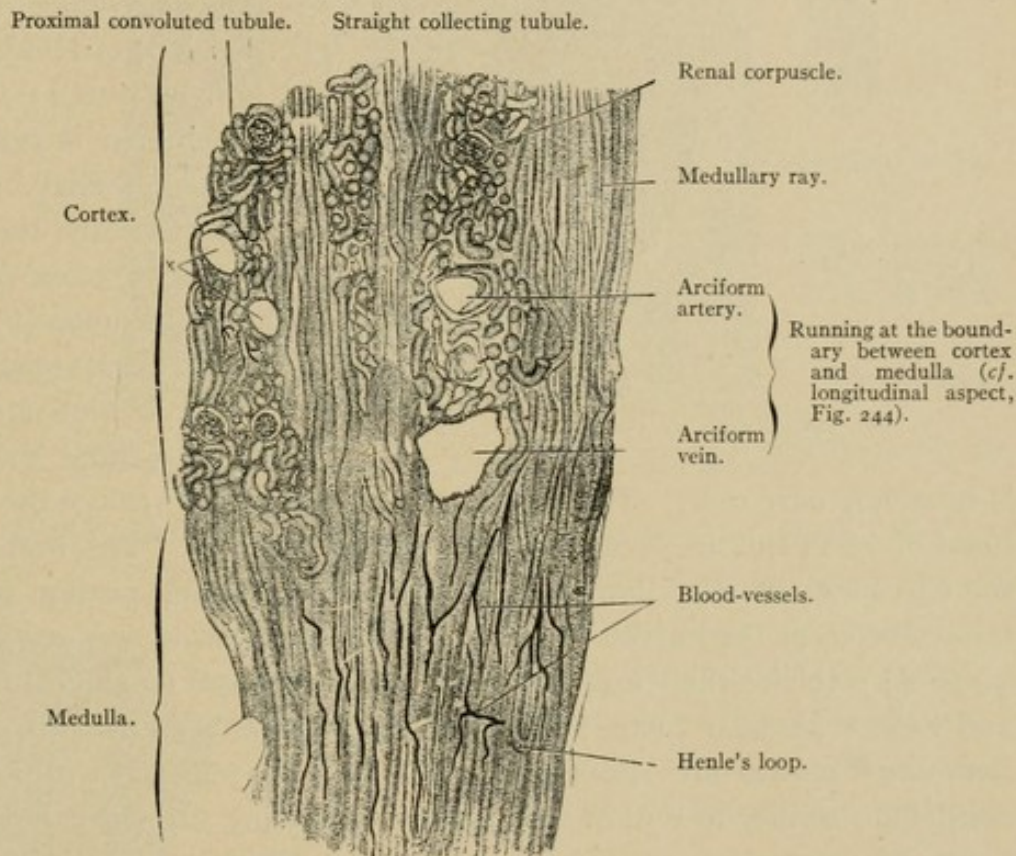


FIG. 245.—FROM A SECTION OF HUMAN KIDNEY, INCLUDING A PORTION OF THE CORTIX AND THE MEDULLA. At $\times$ two renal corpuscles have fallen out. $\times 20$. Technic No. 135.

ingly two layers are distinguished in the capsule, an inner (quasi visceral) lying close upon the glomerulus, which in young animals consists of cubical cells, that later become more and more flattened and fuse into a syncytium (p. 73), and an outer (quasi parietal) layer, that is composed of flat, polygonal cells (Fig. 246).*

At the neck the outer layer of the capsule passes over into the wall of the *convoluted tubule*, which is from 40 to 60 μ thick. The protoplasm of the cells of this division, the boundaries of which are not sharply defined, consists of granules that by means of protoplasmic filaments are bound together in rows radially placed to the lumen; these rows are most distinctly seen at the outer end or base of the cell and with medium

* The network of terminal bars (p. 79), occurring in all divisions of the uriniferous tubules, has not yet been successfully demonstrated on the epithelium of the capsule.

magnification have the appearance of minute rods (Fig. 247). The nucleus of the cells always lies near the base; the surface of the cells di-

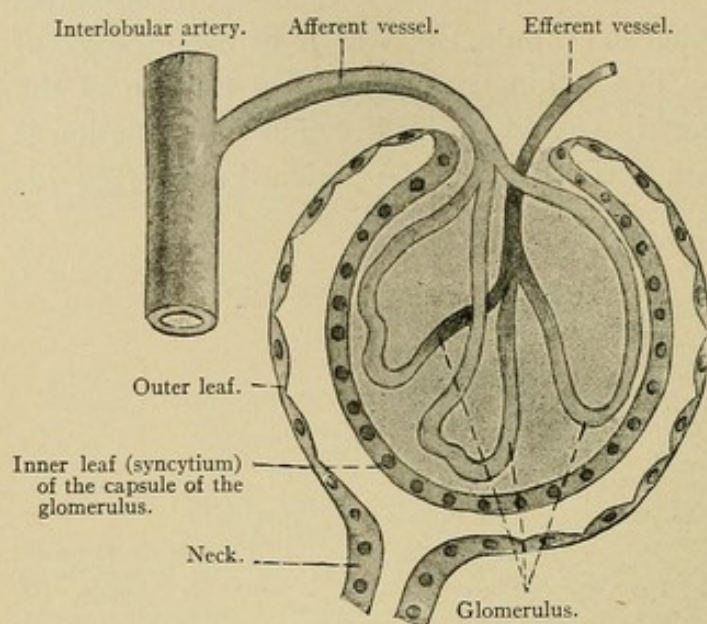


FIG. 246.—SCHEME OF A RENAL CORPUSCLE.

rected toward the lumen is provided with an extremely unstable striated border (p. 76, remark *), the "brush-border."* The *descending limb* (thin division) of Henle's loop is from 9 to 15 μ thick; the lumen is very wide. The epithelial cells are flat elements, the nuclei of which often protrude into the lumen (Fig. 249, 1). The *ascending limb* (thick division) is from 23 to 28 μ thick, the lumen

is relatively narrower. The epithelial cells resemble those of the convoluted division, but are somewhat lower (Fig. 249, 2). The transition of the narrow descending limb into the thicker ascending portion does not always occur at the curve of the loop. The *intercalated portions* are from 39 to 44 μ thick; their epithelial cells are cylindrical or conical in shape and have a peculiar luster; here, too, rods have been described. The *connecting tubules* are about 25 μ thick and are clothed with a cubical epithelium similar to that of the smallest collecting tubules. The *collecting tubules* increase in thickness as they approach the apex of the papilla; the thinnest have a diameter of 45 μ , the thickest, the papillary ducts (ductus papillares), a diameter of from 200 to 300 μ . Their epithelial cells are in part clear, in part dark cylinder elements (Fig. 249, 3), that increase in height with the increase of the caliber of the tubule.

The renal corpuscles let the urine water pass through, the rodded epithelial cells furnish the pigment and the uric acid; the thin division of Henle's loop, the connecting piece, and the collecting tubules are simply excretory canals.

* According to recent investigations undertaken on winter hibernating animals the epithelial cells of the convoluted tubules at the beginning of secretion enlarge, in place of the brush-border a clear, homogeneous crest appears (Fig. 247 X), while the granular protoplasm with the nucleus lies at the cell base. Then the crest empties its contents, whereby the cell becomes smaller and the lumen of the tubule wider. After a period of rest the cells again become tall (consequently the lumen of the tubule appears narrow), develop a brush-border, and up to the beginning of the next secretion are wholly granular.

The capsule of the glomerulus and the uriniferous tubule are covered in their entire length with a structureless membrana propria situated

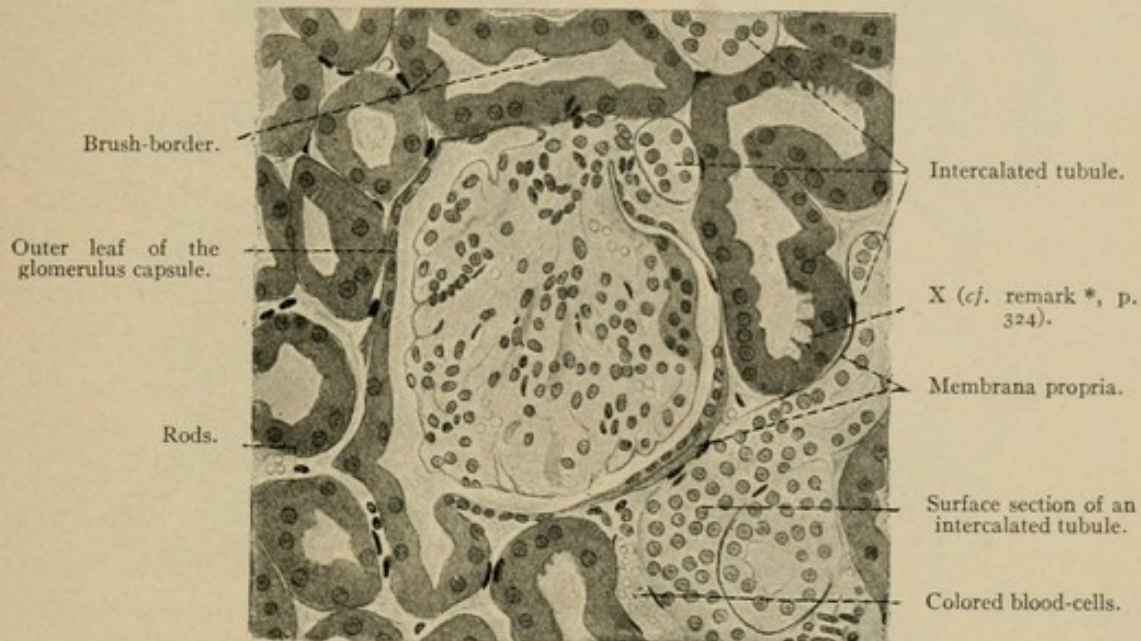


FIG. 247.—FROM A SECTION OF A HUMAN KIDNEY. $\times 240$. The epithelium covering the glomerulus (i. e., the inner leaf of the capsule) cannot be distinctly seen. Technic No. 135.

outside of the epithelium, which is thickest in the descending limb of Henle's loop and gradually disappears toward the papillary duct. The tubules are enveloped in a small amount of loose connective tissue, *interstitial connective tissue*, which on the surface of the kidney is condensed

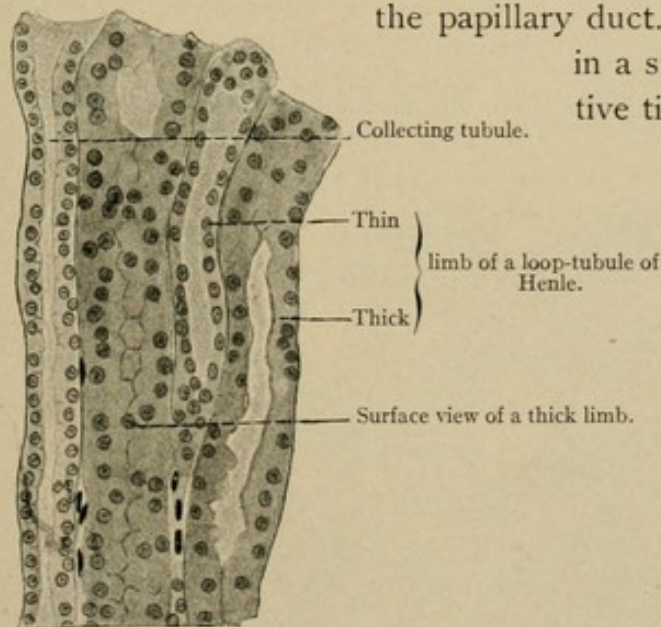


FIG. 248.—TUBULES OF A MEDULLARY RAY. FROM A LONGITUDINAL SECTION OF A HUMAN KIDNEY. $\times 240$. Technic No. 135.

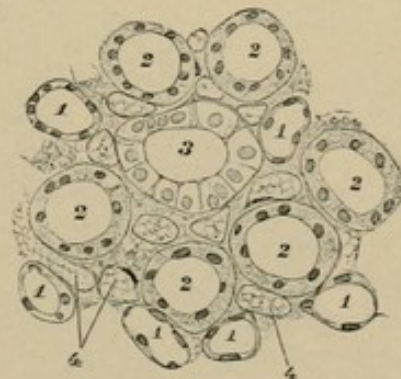


FIG. 249.—FROM A CROSS-SECTION OF THE MEDULLA OF A HUMAN KIDNEY. $\times 240$. The section is through the base of the papilla. 1, Thin, 2, thick division of Henle's loop. 3, Collecting tubule. 4, Blood-vessels filled with blood-cells. Technic No. 135.

to a fibrous membrane, the *tunica albuginea*, containing smooth muscle-fibers and elastic fibers, which increase in old age. The vessels run in the interstitial connective tissue, which is relatively poor in elastic fibers.

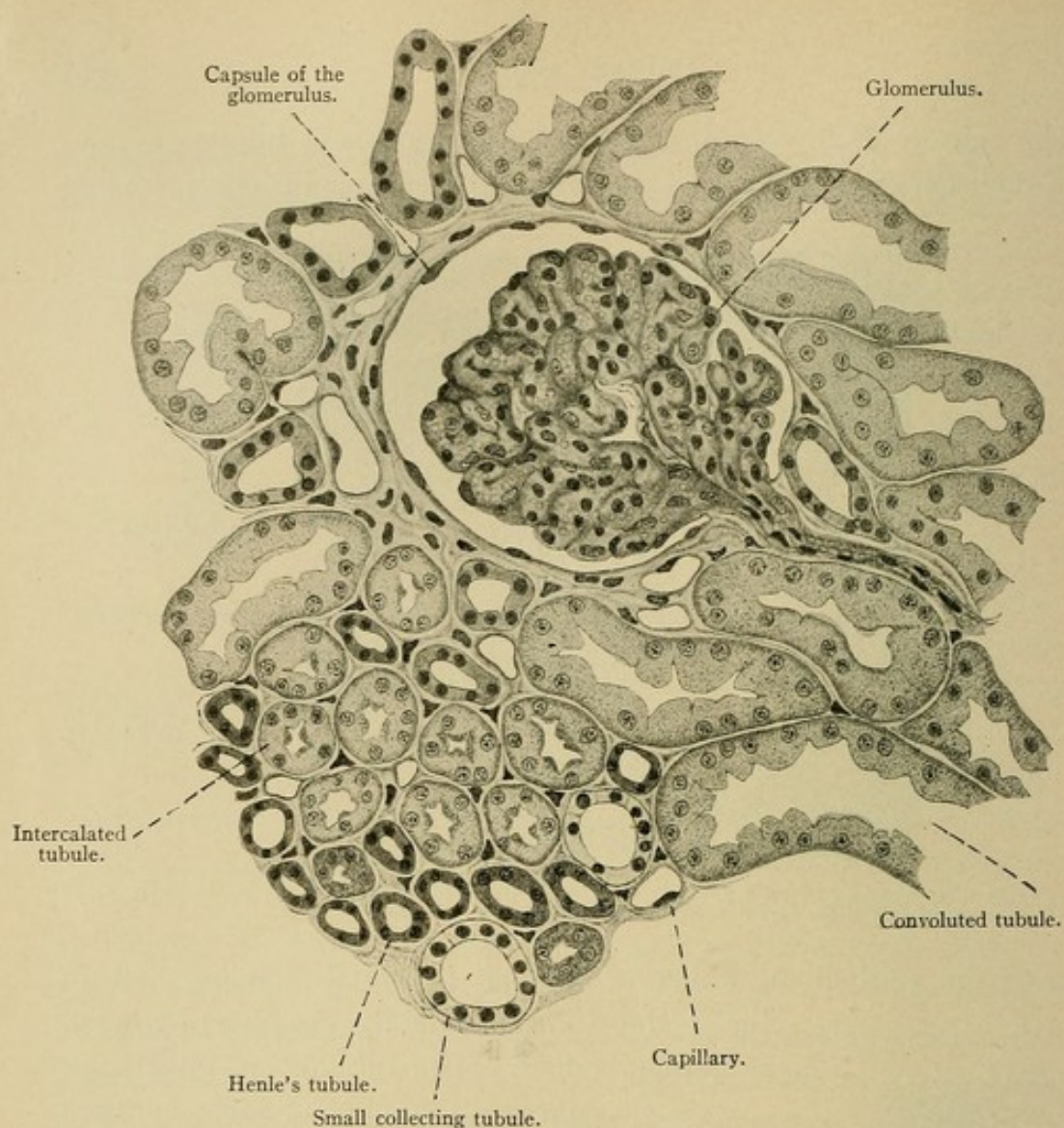


FIG. 250.—FROM A SECTION THROUGH THE CORTEX OF A HUMAN KIDNEY (parallel to the surface). At the left lower corner there is a cross-sectioned medullary ray. $\times 200$. (Schaper.) Technic No. 135.

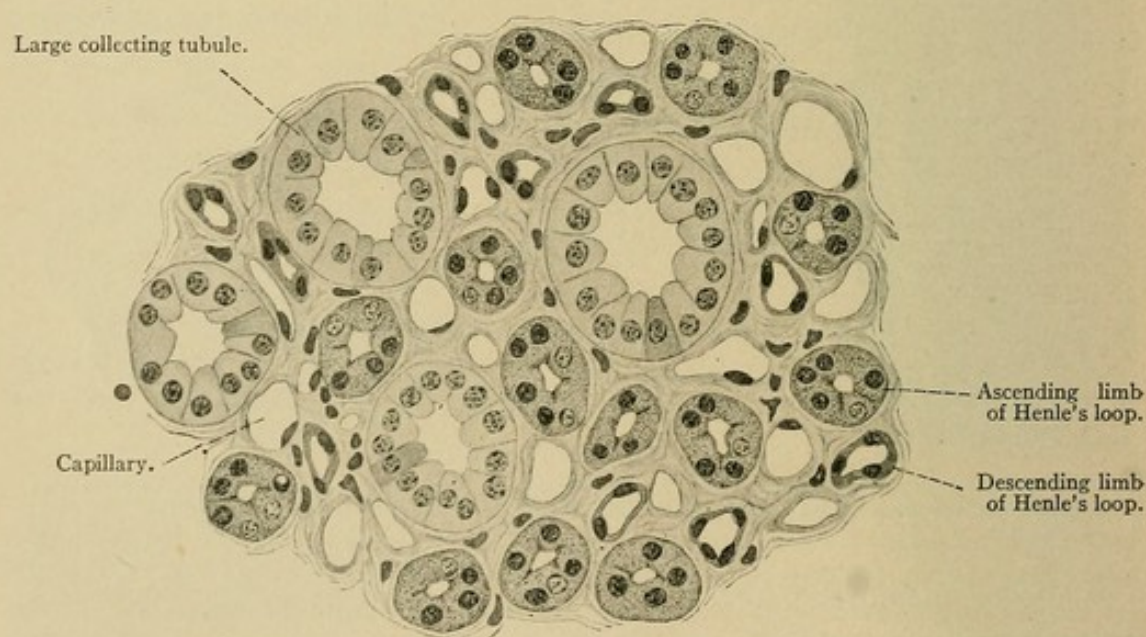


FIG. 251.—FROM A TRANSVERSE SECTION THROUGH THE MEDULLA OF A HUMAN KIDNEY. $\times 320$. (Schaper.) Technic No. 135.

The blood-vessels of the kidneys. The renal artery divides in the hilus of the kidney into branches, which after giving off small twigs to the fibrous capsule and the tunica albuginea, and to the renal calices enter the parenchyma of the organ at the circumference of the papillæ and as the *interlobar arteries* (*arteriæ interlobares*) pass without branching to the boundary between the cortex and the medulla (Fig. 244). Here the arteries bend at right angles and form very irregularly curved arches (*arteriæ arciformes*) along the boundary line, with the convexity toward the periphery. From the convex side of the arches and from their terminal ramifications branches spring at regular intervals, that run toward the periphery, the *interlobular arteries* * (*arteriæ interlobulares*) (Fig. 244, 252), which give off short lateral twigs, each of which supplies a glomerulus (Fig. 244, g).

Each interlobular artery breaks up into terminal branches, of which some supply the tunica albuginea, some continue as the capillaries of the cortex or form the afferent vessel of a glomerulus. Each glomerulus arises by the rapid division of an artery into a number of small capillary twigs,† that immediately reunite in a single (arterial) vessel;‡ this latter is called the *efferent artery* (Fig. 244 c, 252); it is somewhat smaller than the entering vessel of the glomerulus, which is called the *afferent artery* (Fig. 244 a, 252). The efferent artery breaks up into a capillary network, with elliptical meshes in the region of the medullary rays,

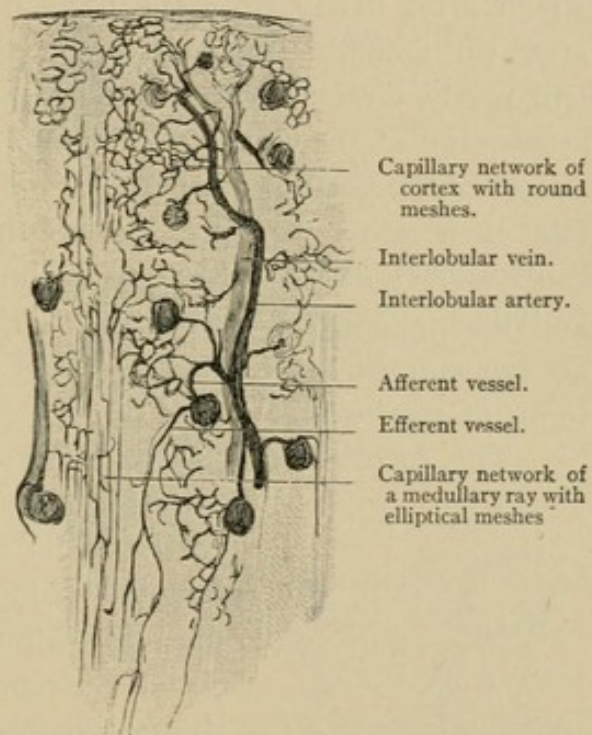


FIG. 252.—FROM A LONGITUDINAL SECTION OF THE INJECTED KIDNEY OF A GUINEA-PIG. $\times 30$. Technic No. 137.

* Microscopic regions of the kidney with ill-defined boundaries, in the axis of which lies a medullary ray and at the periphery of which interlobular arteries ascend, are designated lobules. In Fig. 244 two lobules are indicated. These lobules have no relation whatever to the lobules of the kidney during fetal life.

† The wall of these capillaries is said to consist of a common protoplasmic mass without nuclei; possibly the syncytium of the inner lamella of the capsule (p. 323) contains elements of the vascular wall.

‡ Consequently each glomerulus is an arterial rete mirabile (p. 143, remark *). In dogs and cats retia mirabilia occur in the kidneys that do not stand in any relation to uriniferous tubules, that is, they are not enveloped in a "capsule."

with round meshes in the region of the convoluted tubules. From the latter veins arise, the *interlobular veins* (*venæ interlobulares*) (Fig. 244, 252), which lie close beside the interlobular arteries, in their further course continue alongside the arteries, and open into the *venæ arciformes*; the latter also take up small veins that arise from the confluence of capillaries situated in the deeper portions of the cortex. The veins of the peripheral zone of the cortex converge to points where they unite in radicles arranged in a stellate form, the *venæ stellatæ* (Verheyinii), which are connected with the interlobular veins. The foregoing account of the distribution of the blood-vessels applies only to the cortex and to the medullary rays.

The medulla receives its blood supply from (1) the *arteriolæ rectæ*, which arise partly from the efferent vessels of the most deeply situated (in animals relatively large) glomeruli (Fig. 244, 4 and 252), and partly direct from centrally running branches of the interlobular arteries or of the arciform arteries (Fig. 244, 3, 2); and (2) from offshoots of the cortical capillaries (Fig. 244, 1). The veins of the medulla take their origin from the wide-meshed capillary network surrounding the papillary ducts and empty in the *venæ arciformes*. The renal vein and its branches have no valves. Direct communication between the arteries and the veins occurs both in the tunica albuginea and in the interior of the kidney.

The *lymph-vessels* originate from a network of closed capillaries occurring in the cortex (a similar network appears to be present in the medulla); the small trunks arising from them run with the blood-vessels, without enveloping them (p. 136), and make their exit at the hilus. Besides these deep lymph-vessels there are two superficial capillary nets, one in the capsula adiposa and one in the capsula fibrosa (the latter is in communication with the cortical capillary plexus). The small trunks originating from them empty into neighboring lymph-glands.

The partly medullated *nerves* run either with the blood- and lymph-vessels in the connective-tissue capsule of the kidney or they form in the hilus a plexus intermingled with sympathetic nerve-cells, in the construction of which the branches supplying the renal pelvis, as well as the nerves that accompany the blood-vessels participate. In the interior of the kidney the

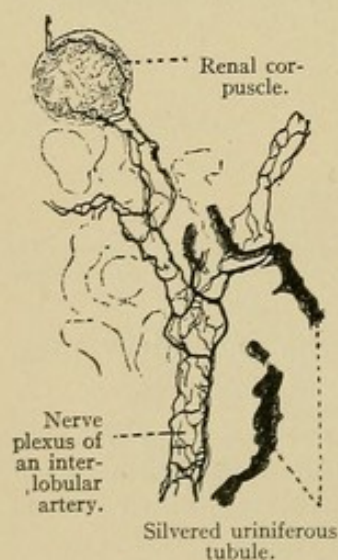


FIG. 253.—SECTION OF THE KIDNEY OF A MOUSE. $\times 180$.
Technic No. 138.

nerves form networks which envelop the arteries up to the renal corpuscle (Fig. 253). The walls of the veins and the capillaries are also encircled by nerve networks, delicate branches of which form epilemmal and hypolemmal networks (p. 247) on the straight and particularly on the convoluted uriniferous tubules, from which delicate interepithelial-ending nerve filaments arise.

THE URINARY PASSAGES.

The *calices and pelvis of the kidney* and the *ureter* consist of three membranes: innermost lies (1) the mucous membrane, then follows (2) the muscle membrane, which is enveloped in (3) the fibrous membrane (Fig. 254).

The constituent parts of the mucous membrane are (*a*) an epithelium that in sections exactly resembles the epithelium of a moderately contracted urinary bladder* (p. 330); (*b*) a tunica propria, which consists of delicate connective-tissue fibers, a very few elastic fibers, and many cellular elements (leucocytes also are occasionally found here), and passes without sharp boundaries into the similarly constructed, but loose submucosa.

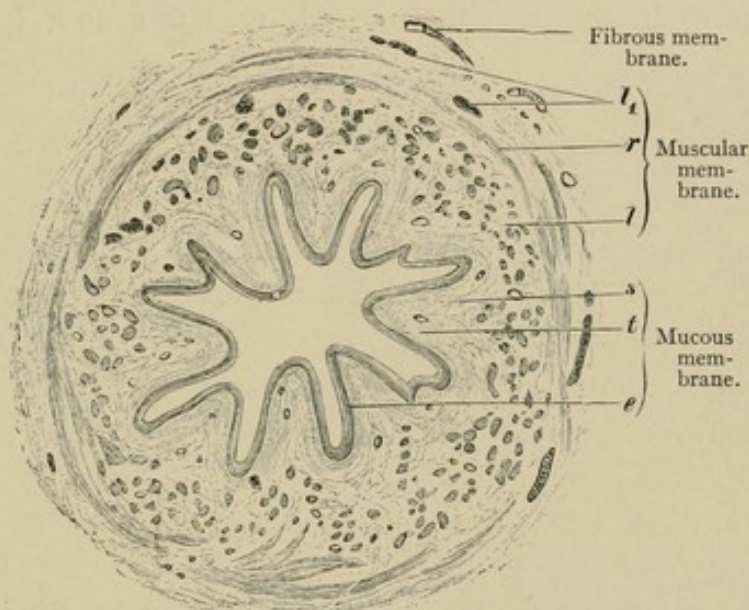


FIG. 254.—TRANSVERSE SECTION OF THE LOWER HALF OF A HUMAN URETER. $\times 15$. *e*, Epithelium; *t*, tunica propria; *s*, submucosa; *l*, inner longitudinal muscle-bundles; *r*, circular layer of muscle-bundles; *l*₁, accessory outer longitudinal muscle-bundles. Technic No. 139.

The *muscle membrane* is not, like that of the wall of the intestine, formed of a closed layer, but is frequently interrupted by connective tissue; an inner longitudinal (*l*) and an outer circular layer (*r*) of smooth muscle-fibers can be distinguished, which latter in the lower half of the ureter is covered with longitudinally disposed strands of muscle (*l*₁).† The so-called “wall-piece” of the ureter, running in the wall of the

* The isolated epithelial cells of the calices, the pelves, and the ureters also cannot be distinguished from those of the urinary bladder.

† The lowermost division of this layer, about 5 cm. long, is particularly thick and is described as the *ureteral sheath*.

urinary bladder, possesses only longitudinal muscles, that are not connected with the muscles of the latter, but end free in the tunica propria of the vesical mucous membrane. Their contraction opens the mouth of the ureters.

The fibrous membrane (tunica adventitia) consists of loose connective-tissue bundles and elastic fibers.

The mucous membrane of the renal calices continues over the surface of the renal papillæ; the circular muscle-fibers form a ring muscle round the papillæ.

The *blood-* and *lymph-vessels* are especially rich in the mucous membrane; the blood capillaries situated immediately beneath the epithelium occasionally project toward the latter and, particularly in the renal



FIG. 255.—PORTION OF A VERTICAL SECTION OF A HUMAN VESICAL MUCOUS MEMBRANE. $\times 560$. Technic No. 140.

pelves, counterfeit "intraepithelial blood-vessels." The *nerves* are partly motor—they are distributed in the muscles—and partly sensory—they form bush-like endings in the tunica propria or they terminate free between the epithelial cells.

The *urinary bladder* likewise consists of a mucous membrane, a muscle membrane, and a fibrous membrane. The epithelium of the contracted or moderately filled organ, in vertical sections (Fig. 255), resembles stratified squamous epithelium; but with this difference, that the cells of the superficial stratum are cylindric or cubical elements or thick plates. Uncertain whether to class this epithelium with the stratified squamous or the stratified cylinder epithelium, it has been named *transitional epithelium*.

It has been demonstrated by careful investigations that in reality only two strata are present, the form of which varies extraordinarily according to the content of the bladder. In the *empty*, strongly contracted organ the cells of

the superficial stratum are cubical, cylindrical, and on their under surface often provided with depressions and processes, to which the cells of the deeper stratum are attached. The latter are slender elements, expanded in the vicinity of the nucleus; the usually simple nucleus lies sometimes at the upper end, sometimes at the lower end, sometimes in the middle of the cell. This gives rise in sections to the false appearance of a stratified epithelium. In the *completely* filled bladder the superficial cells are quite flattened, the deep cells are low, cubical, and their transverse-oval nuclei lie in a *single* row. Between these two extremes many transitions exist.

With the proof that the epithelium of the bladder is in reality two-layered, the familiar fact that the terminal bars form networks, not only on the sur-

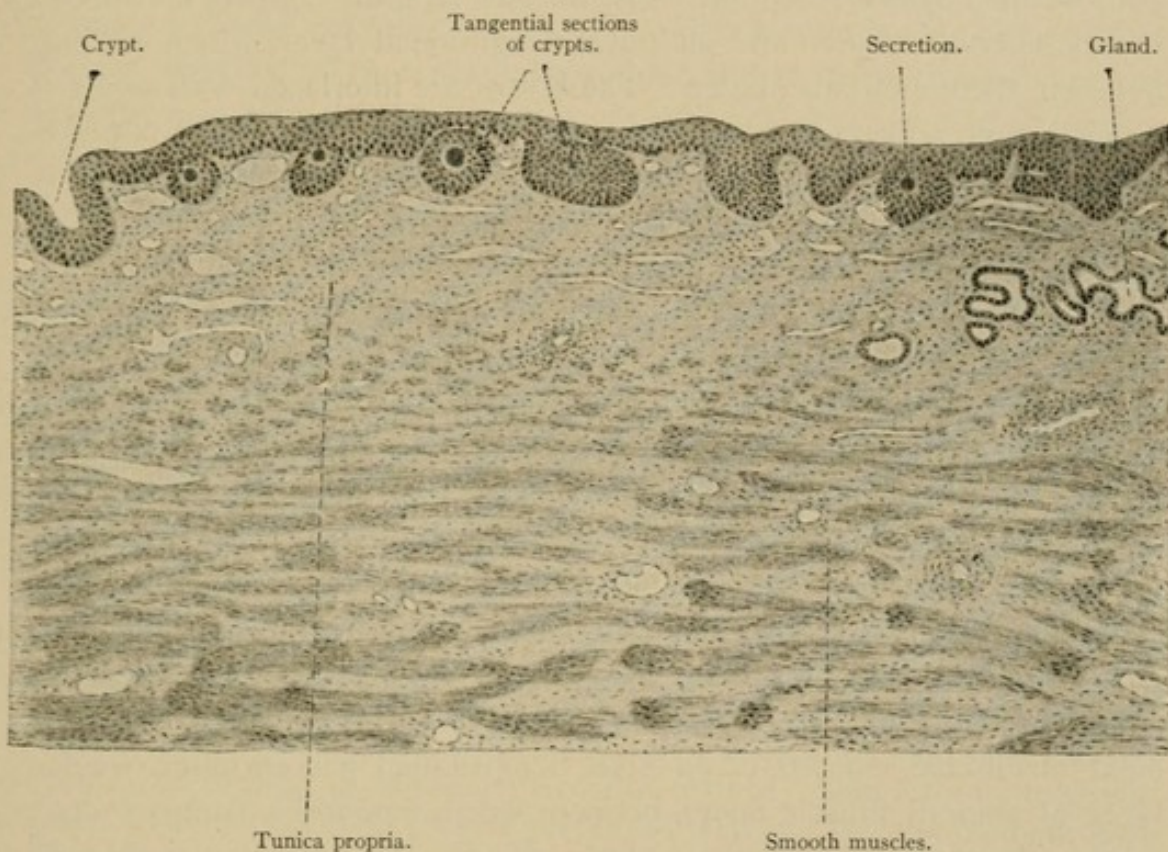


FIG. 256.—SECTION THROUGH THE FUNDUS OF THE URINARY BLADDER OF ADULT MAN. $\times 48$. Technic No. 140.

face but also in the depths, is satisfactorily explained: this occurs only in the contracted organ; in the expanded organ the network lies on the surface.

Granules can often be demonstrated in the epithelial cells, particularly in those of the superficial stratum, that possibly are precursors of secretion. These cells of the upper stratum are further distinguished by the deeper staining of their protoplasm, by the occasional presence of a cuticular border (Fig. 255), as well as by the frequent possession of several nuclei that have arisen by amitosis.

In the superficial strata of the tunica propria (also in the lower division of the renal pelvis and the upper division of the ureter) round or oblong bodies are found: sprouts of the superficial epithelium, in part

without a lumen; * little strands, partly hollow; crypts, the lumen of which contains secretion, a colloid substance. These structures are the initial stages in the development of glands, that do not until in adult life grow from the base of the crypt and develop into branched follicles clothed with cylinder epithelium.

Such true glands occur only in the urinary bladder, at the fundus, the trigonum, and the beginning of the urethra, where they exhibit all transitions to well-developed prostate glands (p. 345). The tunica propria, which blends insensibly with the submucosa, occasionally contains solitary lymph nodules. The muscle stratum consists of smooth muscle-fibers, an inner and an outer longitudinal layer, which enclose between them a circular layer. The layers are interlaced with one another in such a manner that a sharp demarcation of the same is not possible. At the base of the bladder the inner longitudinal muscle-layer is strengthened and at the beginning of the urethra the ring muscle-layer forms the not always distinct internal vesical sphincter.

The blood- and lymph-vessels,† as well as the nerves, provided with small groups of ganglion cells, behave as in the ureter.

The *female urethra* consists of a mucous membrane and a powerful muscular membrane. The tunica propria mucosæ is composed of a fine-fibered connective tissue, richly interspersed with cells, that on its surface is elevated in numerous papillæ, especially well developed on the external meatus. The epithelium varies individually, is sometimes stratified squamous epithelium, more often simple cylinder epithelium. A few branched simple tubular glands are present; they occur in small groups at the meatus and are called "periurethral" glands. The muscular membrane consists of an inner longitudinal and an outer circular layer of smooth muscle-fibers, between which extends a compact connective tissue mixed with many elastic fibers. The mucous membrane is rich in venous blood-vessels, the networks of which extend into the longitudinal layer of the muscular membrane; in this way a structure similar to the corpus cavernosum of the male urethra, the *corpus spongiosum*, is formed.

The *male urethra* (better, "male urogenital sinus") like that of the female consists of a mucous membrane and a muscular membrane, but it differs in structure in the different divisions of the canal. In the prostatic portion the epithelium resembles that of the urinary bladder; in the

* Occasionally the connection with the superficial epithelium appears to have been lost.

† Not only the muscular membrane, but also the mucous membrane of the urinary bladder contains a lymph-vessel plexus, that in the lower division of the organ is especially well developed.

membranous division it gradually passes into a stratified cylinder epithelium, which finally in the cavernous part is transformed to a simple cylinder epithelium. From the fossa navicularis on the epithelium is of the stratified squamous type. The tunica propria is rich in elastic fibers and is beset with papillæ, that are especially well developed in the fossa navicularis. Isolated, branched, alveolo-tubular simple glands, the urethral glands (*glandulæ urethrales*) (Litrii), occur throughout the entire urethra (Fig. 269). Transition forms occur between these glands and the evaginations of the mucous membrane, the "lacunæ," clothed with a simple cylinder epithelium. The muscular membrane in the prostatic division consists of an inner longitudinal and an outer circular layer of smooth muscle-fibers; both layers are still well developed in the membranous portion, but gradually cease in the cavernous portion, where the circular layer, still conspicuous in the bulbus urethræ, is the first to disappear; in the anterior part of the cavernous division only a few oblique and longitudinal bundles occur. The mucous membrane of the male urethra has a rich vascular supply (see corpus cavernosum urethræ, p. 347). The lymph-vessels lie beneath the blood-vessels. The nerves form networks intermixed with nerve-cells; the nonmedullated fibers arising from them terminate partly in free endings, partly (in the prostatic and the membranous portion) in special end apparatus (*cf.* p. 224).

TECHNIC.

No. 134.—*Isolated uriniferous tubules.*—The most suitable for this preparation are the kidneys of young animals, for example newborn kittens. Divide the kidney in halves; place one half (*a*) aside for investigation fresh; cut the other half (*b*) into pieces including cortex and medulla, and place them in 30 c.c. of pure hydrochloric acid.

(*a*) Tease a pea-sized piece in a drop of 0.75 per cent. salt solution. The red glomeruli, the convoluted and straight uriniferous tubules, can be seen with the low power. The convoluted tubules are dark and granular, the other divisions clear. With high magnification the nuclei of the clear portion of the uriniferous tubules can be distinctly perceived; the cell boundaries can best be seen in the collecting tubules. In the convoluted tubules only the fine striation of the base of the gland-cells can be seen; cell boundaries and nuclei are not visible.

(*b*) In about two hours the red pieces of kidney tissue should be transferred to a capsule containing 50 c.c. of distilled water, in which they rapidly become a dull gray, with smeary surfaces. The water is to be changed. After a few moments small pieces can be detached with needles and readily separated into tubules, in a little water on a slide. If it is desired to obtain entire uriniferous tubules transfer pieces of kidney 2 mm. square to a watch-glass in which has been placed a large cover-

glass and enough distilled water to cover the surface of the latter. The attempt should now be made to isolate the tubules with needles. If the

isolation is successful—this may be ascertained by examination with the low power—with a pipet or filter-paper carefully absorb the water from the watch-glass and then from the cover-glass, take out the latter, cleanse its free surface, and place it with the attached tubules gently on a slide on which a drop of dilute glycerol has been previously placed. The preparation may be subsequently stained under the cover-glass with picrocarmine (Fig. 257).

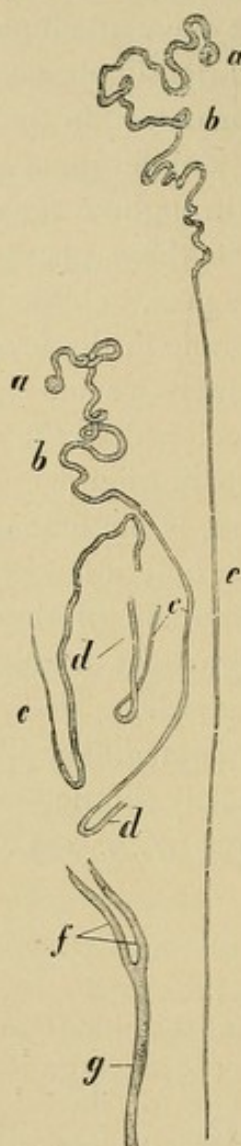


FIG. 257.—ISOLATED URINIFEROUS TUBULES OF A FOUR-WEEK-OLD RABBIT. $\times 30$. *a*, Renal corpuscle. *b*, Convoluted tubule. *c*, Henle's loop, descending limb; *d*, ascending limb. *f*, Collecting tubule. *g*, Papillary duct.

No. 135.—*The cortex and the medulla of the kidney.*—For sections the kitten's other kidney or pieces of another kidney 2 or 3 cm. square are to be fixed in 200 or 300 c.c. of Müller's fluid for four weeks (p. 33), and hardened in 100 c.c. of gradually strengthened alcohols (p. 35). Thick transverse and longitudinal sections through the cortex and similar ones through the medulla are to be examined unstained in dilute glycerol, with a low power. Thin transverse sections (*a*) through the apex of the papillæ for the papillary duct, (*b*) through the base of the papillæ (Fig. 249), and (*c*) transversely and longitudinally through the cortex are to be stained with Hansen's hematoxylin (p. 38) and mounted in xylol-balsam. The extremely delicate "brush-borders" can be seen only in very thin sections. Frequently they have fallen off.

Endeavor to cut radial sections through the cortex and the medulla, showing the boundary between the two; examine them unstained in glycerol, with the low power. Frequently the blood-vessels are still filled with blood corpuscles and can be traced for long distances.

For the study of the *glomerulus and its capsule*, also the connection of the latter with the uriniferous tubule, very thin microtome sections must be made (Figs. 247, 250).

No. 136.—*Medullary rays and Henle's loops* are especially fine in stained vertical sections of the kidneys of young animals prepared after No. 135.

No. 137.—*Blood-vessels of the kidney.*—An isolated kidney can be injected (p. 48), fixed in 300 c.c. of Müller's fluid (p. 33) for four weeks

and then hardened in 150 c.c. of gradually strengthened alcohols (p. 35). The surface aspect of the *venæ stellatæ* should be examined macroscopically. Unstained thick longitudinal and transverse sections should be studied with the low power (Fig. 252).

No. 138.—*Nerves of the kidney*.—Treat small pieces according to Golgi's method, given on p. 45; they should remain from three to six days in the osmiobichromate mixture. Result: Fig. 253.

No. 139.—*The renal pelvis and the ureters*.—Of the former pieces 1 cm. square, of the latter 1 or 2 cm. long, should be fixed in 100 c.c. of Müller's fluid (p. 33) and in fourteen days hardened in 100 c.c. of gradually strengthened alcohols (p. 35). Stain sections with Hansen's hematoxylin (p. 38) and mount in xylol-balsam.

No. 140.—Treat the *bladder* after No. 139.

No. 141.—*Epithelial cells of the renal pelvis, of the ureter, and of the bladder*.—Treat pieces of each 1 cm. square (cut open the ureter), with 30 c.c. of Ranvier's alcohol (p. 29). Isolate and stain with picrocarmine (p. 53). Mount in diluted acidulated glycerol.

No. 142. *The female urethra*.—Cut out a piece about 2 cm. long of the female urethra, together with the attached anterior vaginal wall; place it in 100 or 200 c.c. of Müller's fluid (p. 33) for fixation and in two or three weeks harden it in gradually strengthened alcohols (p. 35). Stain cross-sections in Hansen's hematoxylin (p. 38) and mount in xylol-balsam.

No. 143.—*The male urethra*.—Treat pieces 1 or 2 cm. long of the prostatic, membranous, and cavernous portions and of the fossa navicularis after No. 142. Care should be exercised not to confuse the urethral lacunæ (Morgagni), blind evaginations of the mucosa, with sections of glands.

VIII. THE REPRODUCTIVE ORGANS.

THE MALE REPRODUCTIVE ORGANS.

THE TESTES.

The testes are glands consisting of branched, pouch-like tubules, the *seminiferous tubules*, which are enveloped in a connective-tissue capsule. This capsule, the *tunica albuginea s. fibrosa* (Fig. 258), is a compact membrane, which encloses the parenchyma on all sides and on the

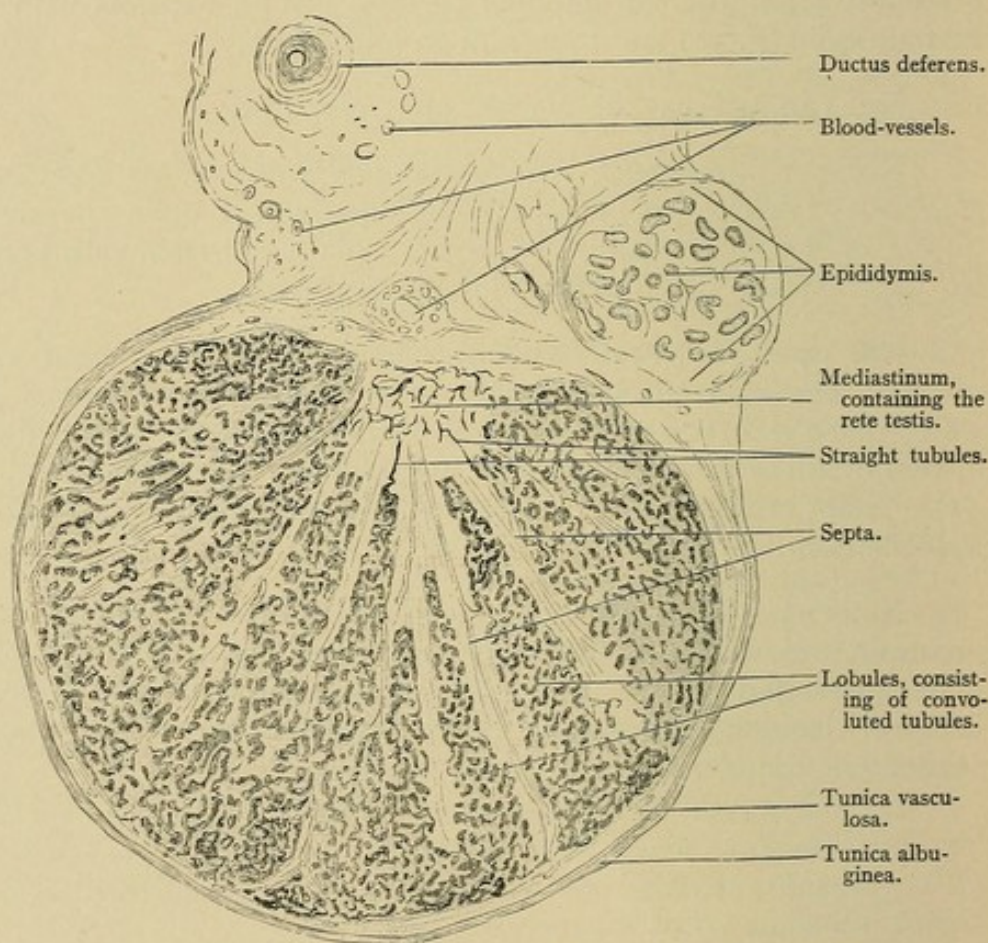


FIG. 258.—CROSS-SECTION OF THE TESTIS OF A NEWBORN CHILD. $\times 10$. Technic No. 144.

posterior upper aspect is developed into a mass, the *mediastinum testis* (*corpus Highmori*), which juts into the interior of the organ. From this a number of septa arise, the *septula testis*, which pass along divergent paths to the tunica albuginea and thus divide the parenchyma of the testis into *pyramidal lobules*, the base of which is directed toward the capsule, the apex toward the corpus Highmori. The tunica albuginea consists of dense fibrous connective tissue and numerous elastic fibers, that increase with the years; on its free surface it is covered with a

simple layer of flat epithelial cells,* on its inner surface it is in contact with a layer of loose connective tissue mixed with elastic fibers; this supports numerous vessels and is called the *tunica vasculosa*; it is connected with the interlobular septa. The mediastinum is constructed of dense connective tissue and numerous elastic fibers and encloses in its interior a network formed of freely anastomosing tubules, the *rete testis* (Halleri). The interlobular septa consist of bundles of connective tissue, which are connected with the loose connective tissue surrounding the individual seminiferous tubules. This "interstitial" connective tissue is rich in cellular elements, which are in part flat connective-tissue cells, in part spherical cells, the so-called *interstitial cells*, containing pigment or fat granules, in the sexually mature testis, also crystalloids † (Fig. 260 and 261).

The *seminiferous tubules* in their course may be divided into three

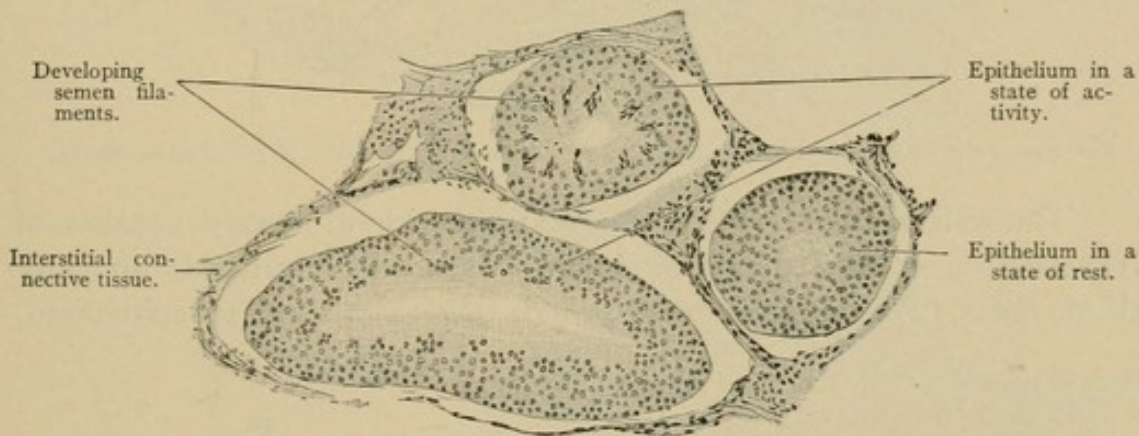


FIG. 259.—FROM A CROSS-SECTION OF THE TESTIS OF AN OX. $\times 50$. In the process of fixing and hardening the epithelium has become somewhat shrunken, so that spaces exist between it and the interstitial connective tissue. Technic No. 145.

portions: they begin as (1) the *convoluted tubules* (*tubuli contorti*), which pass into (2) the *straight tubules* (*tubuli recti*), which continue as (3) the *rete testis*. The convoluted tubules are round, serpentine canals, about $140\ \mu$ in diameter, the initial extremity of which has not yet been satisfactorily oriented; probably they are freely united with one another at the periphery, beneath the *tunica vasculosa*, and form a network, ‡ from which numerous tubules turn aside and with many windings pass toward the mediastinum. During their course the tubules diminish in number,

* This is the visceral layer of the *tunica vaginalis propria*.

† Such structures are more common in the vegetable world, but have recently been found in other animal cells, e. g. in the nuclei and the protoplasm of the nerve-cells of the porcupine and in the protoplasm of the lens epithelium.

‡ Seminiferous tubules with blind ends have been observed.

because they continually unite with one another under narrow angles. Not far from the mediastinum the convoluted tubules pass into the straight tubules (Fig. 258) which, considerably reduced in size (20 to 25 μ thick), after a short course penetrate into the mediastinum and form the rete testis, the tubules of which measure from 24 to 180 μ .

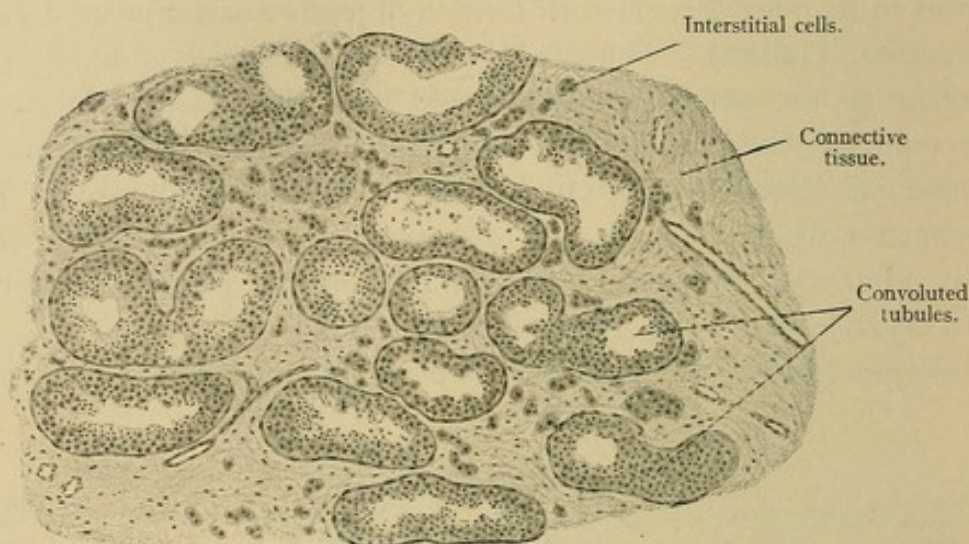


FIG. 260.—FROM A CROSS-SECTION OF THE TESTIS OF A MAN 22 YEARS OLD. $\times 50$. Technic No. 145.

The wall of the *convoluted tubules* from without inward consists of (1) several layers of flattened connective-tissue cells interlaced with many elastic fibers, (2) a thin membrana propria, and (3) a stratified epithelium,

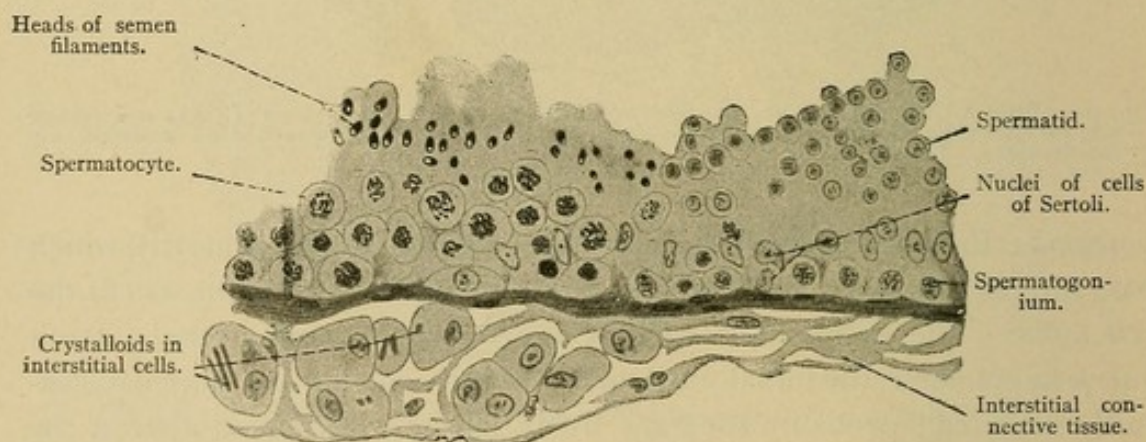


FIG. 261.—FROM A LONGITUDINAL SECTION THROUGH A CONVOLUTED TUBULE OF A HUMAN TESTIS. $\times 360$. Technic No. 145.

the appearance of which differs greatly in the separate divisions of the tubules.

When the epithelium is in a state of rest (as well in the immature testis, as in individual tubules of the sexually ripe human testis) the tubules appear to be clothed in several strata of spherical cells, the nuclei of which stain sometimes more, sometimes less intensely (Fig. 260).

When the epithelium is in a state of activity it exhibits a series of pictures that are related to the development of semen, to "spermatogenesis." The stratum, *parietal stratum*, of membraneless epithelial cells lying next to the membrana propria consists of two kinds, the *cells of Sertoli* (Fig. 262), which take no direct part in the production of the semen filaments, and the *spermatogonia* (ancestral cells), the real producers of the semen. The latter multiply by indirect division and grow to be large cells, that occupy the next layer within. These are the *spermatocytes* (mother-

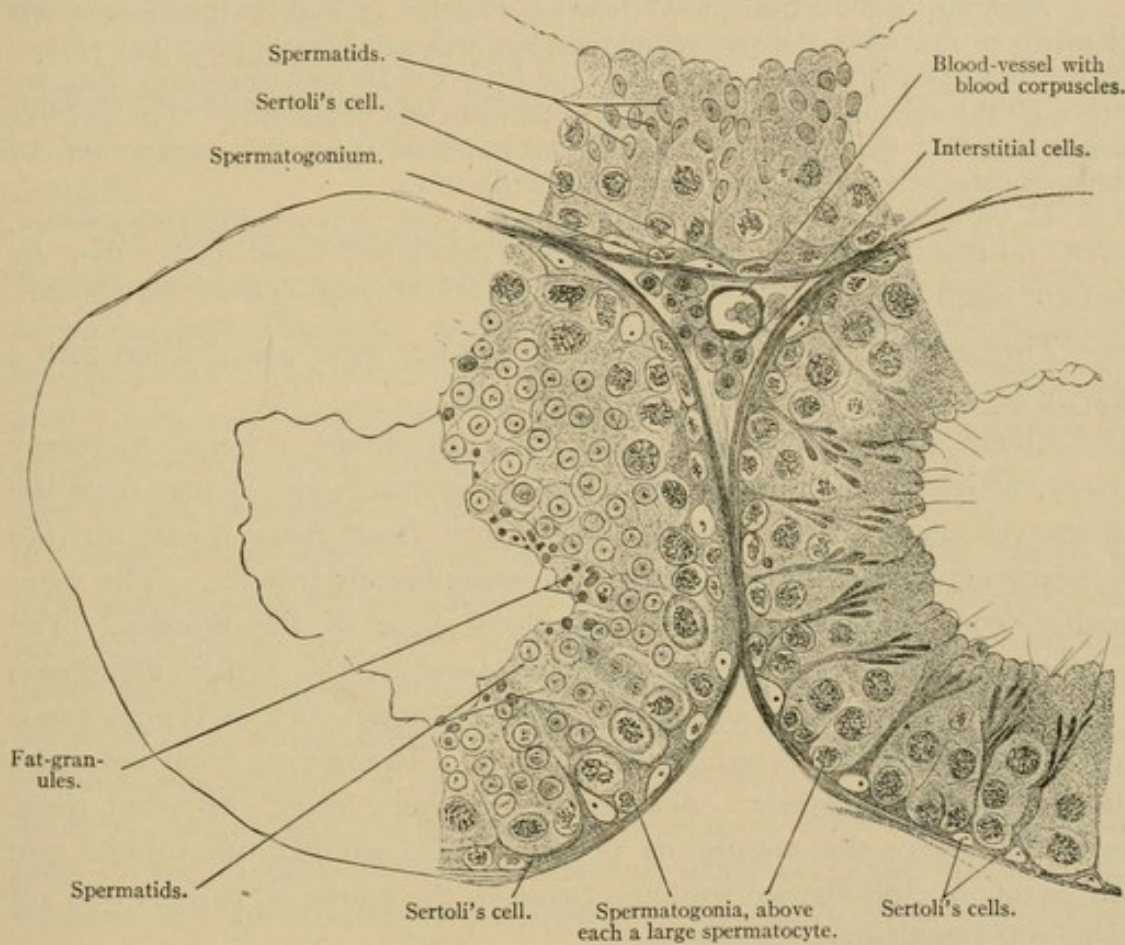


FIG. 262.—CROSS-SECTION OF SEMINIFEROUS TUBULES OF A MOUSE. $\times 360$. Observe that the nuclei of the spermatids (below on the left), at first round, become oval (above) and are transformed (below on the right) into the heads of the semen filaments. Technic No. 146.

cells), which divide twice, each giving rise to four smaller cells, the *spermatids*, lying in a zone still nearer to the center of the tubule. The latter now become *spermatozomes* (semen filaments), by the nucleus of each spermatid developing into the head, a small portion of the protoplasm forming the tail, while the axial fiber (p. 341) grows out of the distal centrosome of the diplosome (p. 66) lying beneath the surface of the cell.

The cells of Sertoli are characterized by a nucleus poor in chromatin, containing a distinct nucleolus, as well as by a protoplasm provided with

brownish fat-granules. In animals the nucleus usually lies on a level with the spermatogonia (Fig. 262), in man not seldom between the spermatocytes (Fig. 261). The rôle of the cells of Sertoli is generally assumed to be sustentative; it is supposed that during the processes just described a large number of spermatids unite with a cell of Sertoli,* that meanwhile has grown in length centrad, and that through this plasma union ("copulation") they in all probability receive nutritive material.

Another, but much-disputed theory is that the cells of Sertoli likewise are elements produced by the spermatogonia, but instead of developing they perish; their protoplasm gradually passes into the intercellular substance occurring between all the cells and there dissolves; their nucleus disappears. The tuft-like arrangement of the semen filaments is the result of the pressure exercised by the spermatogonia lying about the cells of Sertoli.

In the lumen of the seminiferous tubules (also in the epididymis) round, often multinucleated cells are found beside developed semen cells; they are spermatocytes that have not completed their development and are perishing.

The *tubuli recti* and the canals of the *rete testis* are clothed with a simple layer of cubical or flat epithelial cells.

The *arteries* of the testes are branches of the internal spermatic artery, which proceed in part from the mediastinum and in part from the tunica vasculosa to the septula testis, and from there break up into capillary networks which surround the seminiferous tubules. The *veins* arising from these networks follow the course of the arteries. The numerous *lymph-vessels* form a plexus beneath the tunica albuginea, which is in communication with the fairly close network of lymph capillaries enveloping the seminiferous tubules. The *nerves* form networks about the blood-vessels; whether single fibers branch off from these networks, pierce the membrana propria of the seminiferous tubules and terminate in club-shaped endings between the epithelial cells is not yet definitely established.

THE SEMEN.

The secretion of the testes, the *semen* (sperm), consists almost exclusively of *semen filaments* (spermatofilia, spermatozoa), pin-shaped structures about $60\ \mu$ long in which a head and a tail are distinguished (Fig. 263). In man the *head* is from 3 to $5\ \mu$ long, and from 2 to $3\ \mu$ broad, flattened, viewed from the side pyriform in shape, with the tapering end directed forward, seen from surface oval, with the anterior end rounded and containing a clear division (Fig. 263, 1). The foremost end of the head is characterized by its density, which is due to a special struc-

* Whence the "spermatoblast" of authors, see Technic No. 147, p. 369.

ture, the cap, that in man has not yet been definitely demonstrated. The *tail* when very highly magnified exhibits a fiber extending from end to end, the *axial fiber*, which is composed of delicate fibrils. Three different divisions are recognized in the tail: the round *connecting piece* (middle-piece) lying next to the head, which is $6\ \mu$ long and scarce $1\ \mu$ broad; following this is the *main-piece*, from 40 to $60\ \mu$ long, gradually tapering backwards; the tip of the tail, the *end-piece*, is about $10\ \mu$ long and consists of the projecting axial fiber.*

The spermatozoa are distinguished by their extraordinary stability (probably due to the calcareous substances which they contain).

The sinuous movements of the spermatozoa are executed by the tail alone, which propels the head before it; they seldom occur in the concentrated secretion of the testis and begin only after dilution of the semen normally effected by admixture of the secretion of the epididymis, the ampullæ, the seminal vesicles, the prostate gland, and the bulbourethral glands (Cowper). In this mixture of fluids the motions may continue for from twenty-four to forty-eight hours after death and for a still longer period in the secretions of the female genitalia. Water paralyzes the movement, which, however, may be restored by the addition of normal animal fluids of alkaline reaction and moderate concentration; normal fluids in general, also a one per cent. salt solution, exert a favorable influence on the motility of the spermatozoa, while acids and metallic salts suspend it. In motionless spermatozoa the tail is frequently looped (Fig. 263, 3).

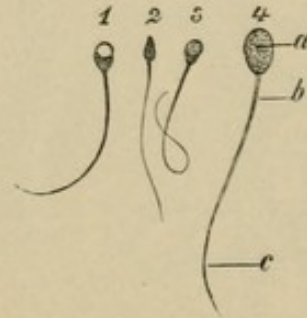


FIG. 263.—1, 2, 3. HUMAN SPERMATOTZOA. $\times 360$. 1. View from the surface. 2. View in profile. 3. Looped semen filament. 4. Spermatozoon of an ox; a, head; b, connecting-piece; c, main-piece. The end-piece and the demarcation of these parts cannot be perceived with this magnification. Technic No. 148.

THE SEMINAL PASSAGES.

The seminal passages are formed by the epididymis, the ductus deferens, the seminal vesicle, and the ductus ejaculatorius.†

From the upper end of the rete testis about fifteen *ductuli efferentes*

* The different forms of semen filaments in animals cannot be considered here. The *spiral fiber*, that is united to the axial fiber by a hyaline membrane, first discovered in birds and tailed amphibians, has also been found in mammals, *e. g.* in the rat, and appears to occur also in man.

† The tubuli recti and rete testis also belong to the seminal passages, but because of their situation in the interior of the gland they were described with it.

testis emerge, which by their progressively increasing convolutions form as many conical lobules, the *lobuli epididymidis*. The aggregate of the lobules constitutes the so-called *head* of the epididymis. By the union

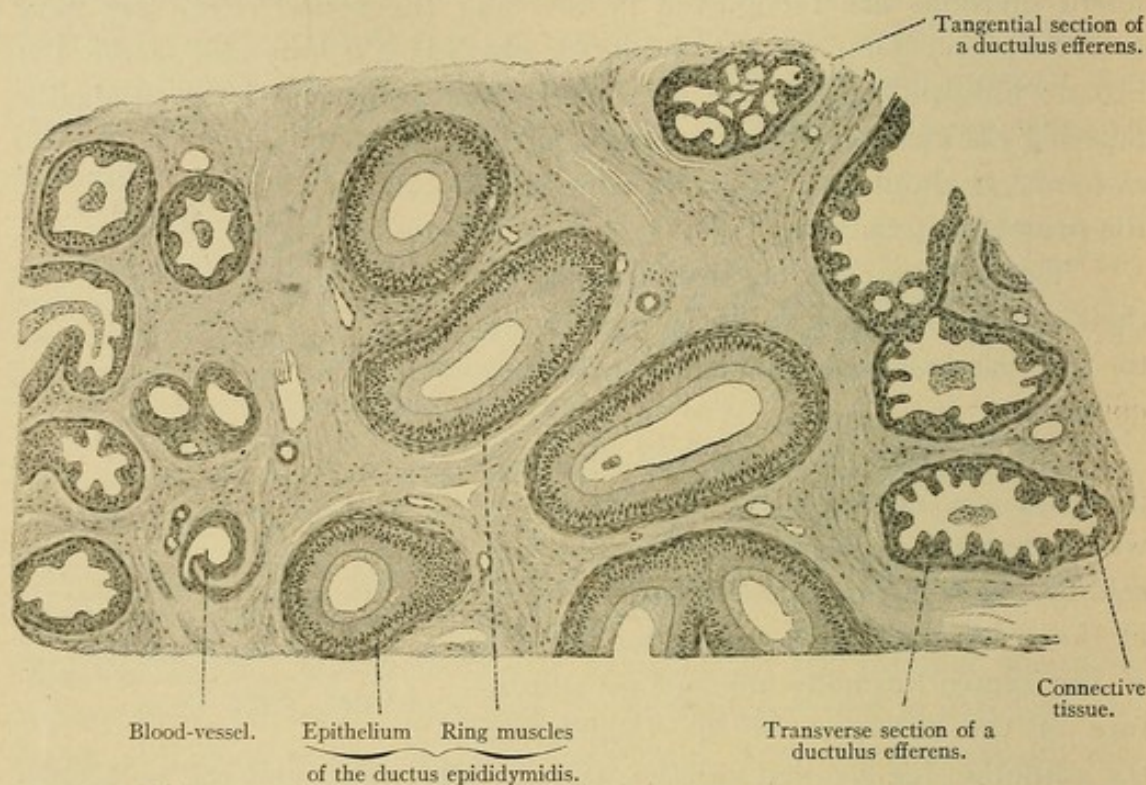


FIG. 264.—FROM A SECTION THROUGH THE HEAD OF THE EPIDIDYMIS OF MAN. $\times 50$. In the middle are transverse sections of the ductus epididymidis, on the right of the ductuli efferentes. Technic No. 151.

of the ductuli efferentes the *ductus epididymidis* arises, which with its complex convolutions forms the body and the tail of the epididymis and then continues as the *ductus deferens*.

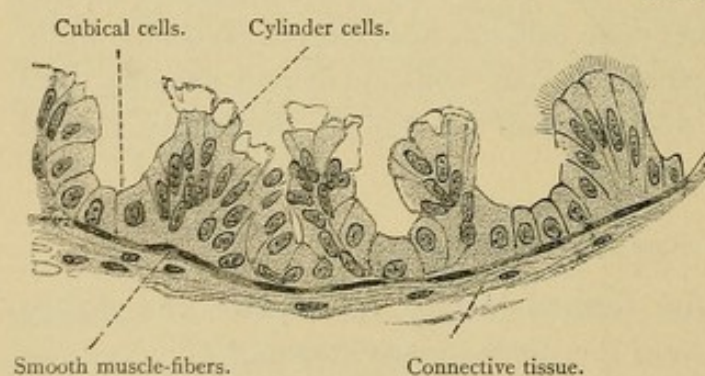


FIG. 265.—TRANSVERSE SECTION OF A DUCTULUS EFFERENS TESTIS OF AN ADULT MAN. The right-hand end of the illustration is schematic. No cilia could be seen, although those of the epithelium of the epididymis were well preserved. $\times 360$. Technic No. 151.

The *ductuli efferentes* are clothed by an epithelium consisting of totally dissimilar varieties; groups of simple ciliated cylindric elements alternate with clusters of cubical cells partly without cilia; consequently the latter have the appearance of simple alveolar glands, that do not always produce evagination of the *membrana propria** (Fig. 265).

* In a few cases instead of the alveoli there are long branched ducts, that extend out beyond the wall of the ductulus into the surrounding connective tissue.

The cells contain, besides a widely varying quantity of pigment granules, granules which indicate a secretory function; this view is also supported by the circumstance that vesicular processes resembling drops of secretion are often found on the free surface of the cells, instead of the cilia (Fig. 265). A striated tunica propria and a membrane of smooth muscle-fibers, consisting of several layers of circularly arranged elements interlaced with elastic fibers, complete the wall of the ductuli efferentes.

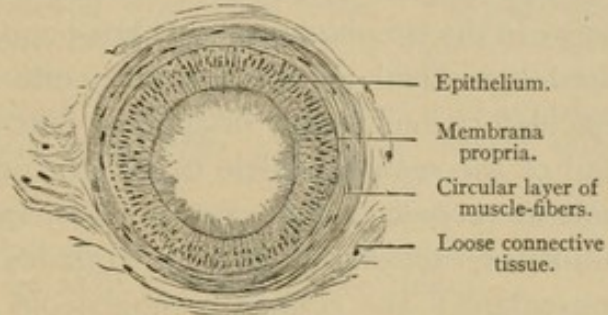


FIG. 266.—TRANSVERSE SECTION OF A HUMAN DUCTUS EPIDIDY-
MIDIS. $\times 80$. Technic No. 151.

The *ductus epididymidis* possesses a two-row epithelium (Fig. 266), the elements of which are spherical basal cells and long cylinder cells; the latter contain secretion granules and on the middle of their free surface support long hairs, that do *not* vibrate and in fixed preparations are frequently glued together in a conical process. In the epithelium tubes

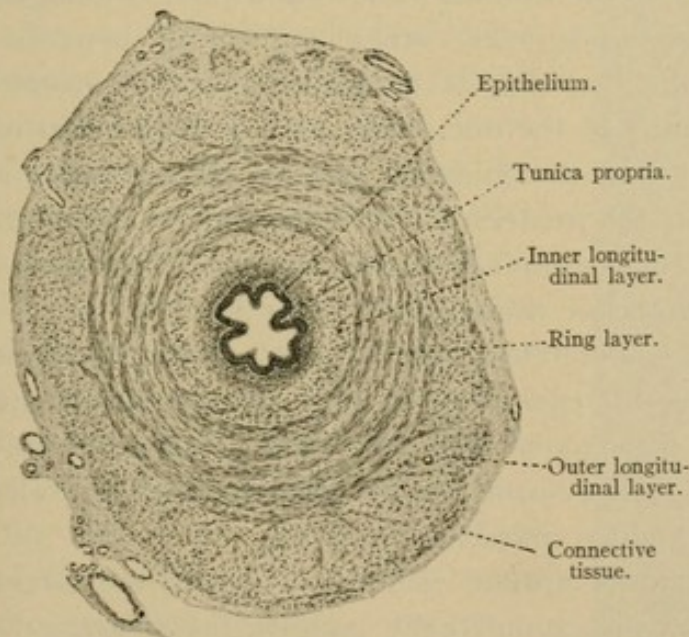


FIG. 267.—TRANSVERSE SECTION OF THE INITIAL PORTION OF THE
DUCTUS DEFERENS OF MAN. $\times 24$. The transverse sections of
the longitudinal muscles have the appearance of small circles and
dots. Technic No. 151.

or sack-like ducts, partly closed, partly opening on the free surface, are found. A delicate membrana propria and a thick layer of circular muscle complete the wall of the ductus epididymidis, the convolutions of which are held together by loose connective tissue; toward the ductus deferens the circular muscle stratum becomes thicker.

The *ductus deferens* consists either of a two-row cylinder epithelium or of a stratified squamous

epithelium (resembling transitional epithelium, p. 330); of a connective-tissue tunica propria, that outwardly is attached to a dense plexus of elastic fibers; further of an inner longitudinal, a middle circular, and an outer longitudinal layer of smooth muscle-fibers, of which the inner longitudinal layer is especially well developed in the initial portion of the duc-

tus deferens ; and finally, of a connective-tissue adventitia intermixed with elastic fibers (Fig. 267). The latter, particularly in the division between the testis and the ejaculatory duct, contains longitudinal bundles of smooth muscle-fibers.* The terminal portion of the ductus deferens expands into the *ampulla*, the musculature of which is more irregularly constructed ; between the circular muscles oblique and longitudinal strands occur, while the longitudinal muscles break up into isolated strands, that toward the ejaculatory duct wholly disappear. The mucous membrane of the ampulla and of the seminal vesicle is laid in "primary" folds, that in turn subdivide in secondary and tertiary folds ; from this develop diverticula and branched, tube-shaped processes (glands ?), that may extend deep into the muscularis † and contain homogeneous or finely granular balls of secretion. On the primary folds the epithelium is a stratified (perhaps only many-rowed), elsewhere a simple cylinder epithelium, the cells of which contain pigment granules in widely varying quantities.

The *ductus ejaculatorii* on their dorso-median side are beset with a series of appendages that do not project externally, but are wholly enclosed in the connective-tissue wall of the duct. Some of these appendages show the same structure as the seminal vesicles and therefore might be described as accessory seminal vesicles ; others are simply convolutions of alveolo-tubular glands, that may be compared with the prostate gland. The mucous membrane of the ductus ejaculatorii is like that of the seminal vesicles, except that the folds are not so complicated ; a musculature is found only on the processes, not in the wall of the duct, which is formed of inner circular strands of compacter (the "fiber membrane") and outer looser connective tissue (the "adventitia").‡

The *blood-vessels* of the epididymis, scarce in comparison with those of the testis, lie on the ductuli efferentes, in part close beneath the *membrana propria*, which occasionally they evaginate toward the epithelium. The veins of the pampiniform plexus often have thick walls containing circular and longitudinal muscles.

In addition to the networks around the blood-vessels, the *nerves* form a close plexus provided with sympathetic ganglia, the *plexus myospermaticus*, that lies partly in the muscularis of the epididymis, but chiefly in that of the ductus deferens ; delicate fibers arise from this

* They really belong to the tunica vaginalis of the spermatic cord and are known as the internal cremaster muscle.

† Sections give a very complicated picture, that can only be correctly interpreted by the reconstruction of serial sections.

‡ In the outer strata of the adventitia isolated strands of smooth muscle-fibers occur, which belong to the pelvic fascia and are in part connected with the muscle-fibers of the prostate.

plexus, which end for the greater part on smooth muscle-fibers, for the lesser part they continue into the mucous membrane.

The *paradidymis* (Giraldès), lying between the elements of the seminal cords, and the *ductulus aberrans* are atrophic remains of the embryonal mesonephros. Both consist of a tubule lined with cubical or cylindric ciliated epithelium, which is enveloped in a vascular connective tissue. The *appendix testis* (hydatid of Morgagni) is a solid lobule composed of a highly vascular connective tissue, which is covered with a ciliated cylinder epithelium; it possesses a short pedicle, which contains a little canal that is lined with cylinder epithelium. The inconstant *appendix epididymidis* is a vesicle clothed with cubical epithelial cells and contains a clear fluid. The meaning of these appendices has not yet been satisfactorily explained; it is uncertain whether they are remains of the anterior end of the embryonal Müllerian duct, that in the female becomes the oviduct, or remnants of the primitive kidney.

ACCESSORY GLANDS OF THE MALE SEXUAL ORGANS.

The *prostate* consists of gland substance, of smooth muscle-fibers, that make up about one-fourth of the bulk of the organ, and of connective tissue, intermixed with relatively few elastic fibers. The gland substance is composed of from thirty to fifty branched alveolo-tubular serous simple glands, which are characterized by their loose structure. The glands open by two large and a number of smaller excretory ducts into the urethra. The gland-cells are low cylinder elements, which in a simple layer clothe the tubules. In the larger excretory ducts the epithelium is of the transitional variety (p. 330), like that in the prostatic portion of the urethra. In elderly persons the so-called *prostatic crystals*, round, stratified masses of secretion up to 0.7 mm. in size, occur in the end-pieces. The smooth muscle-fibers found in large quantities everywhere between the gland lobules, are augmented toward the urethra and form a robust circular layer (the internal vesical sphincter muscle); numerous smooth muscle-fibers are also found on



FIG. 268.—FROM A SECTION OF THE PROSTATE OF A CONDEMNED MAN 22 YEARS OLD. $\times 53$. Technic No. 152.

the external surface of the prostate gland, where they are contiguous to the bundles of striated muscle-fibers of the musculus sphincter urethræ membranaceæ.* The prostate and the colliculus seminalis are provided with many blood-vessels. The numerous nerves form wide-meshed networks containing nerve-cells; of the nonmedullated fibers arising from these networks some approach the smooth muscle-fibers, some end in free ramifications, some (in dogs and cats) terminate in special end apparatus (p. 224), that are found in the capsule and in the interior of the organ.

The *bulbourethral glands* (glandulæ bulbourethrales, Cowper) are compound alveolo-tubular glands; the irregularly widened excretory duct sends off similarly constructed branches, that are annexed either directly or indirectly, by means of intercalated tubules, to the end-pieces. The latter have the form of tubules and of spherical vesicles or of transition forms of both. Occasionally reticular connection of the end-pieces occurs. The branches of the excretory duct are lined with a low, simple epithelium and encircled by thin rings of smooth musculature. The terminal pieces possess gland-cells resembling mucous cells and intercellular secretory capillaries. Between the gland lobules lie many smooth and striated muscle-fibers.

THE PENIS.

The penis consists of three cylindrical erectile bodies: the two corpora cavernosa penis and the corpus cavernosum urethræ, which are enveloped in fascia and skin.

Each *corpus cavernosum penis* consists of a tunica albuginea and of an erectile tissue. The *tunica albuginea* is a stout connective-tissue membrane, intermingled with many delicate elastic fibers, possessing an average thickness of one mm., in which an outer longitudinal and an inner circular layer can be distinguished. The *erectile tissue* is constructed of lamellæ and trabeculæ of connective tissue containing bundles of smooth muscle-fibers, that by means of numerous anastomoses form a network. The spaces of the net are clothed with a simple stratum of flat epithelial cells and are filled with venous blood. The thick-walled *arteries* in part pass into capillaries that form a network situated beneath the tunica albuginea, the *superficial (fine) cortical plexus*; this is connected with a many-layered net of wider venous vessels, the *deep (coarse) cortical plexus*, that lies in the superficial strata of the erectile tissue and passes into the venous spaces of the same. *Some of the arteries open directly into the deep cortical plexus. The so-called *helicine*

* Both sphincters are now included in the designation musculus prostaticus.

arteries are small branches lying within slender strands of connective tissue, which in the collapsed organ are bent in the form of a loop and in an imperfect injection appear to terminate in blind ends. The *veins* (*venæ emissariæ*) which return the blood from the corpora cavernosa penis partly arise from the deep cortical plexus, partly from the depths of the erectile tissue. They penetrate the tunica albuginea and empty into the dorsal vein of the penis.

The *corpus cavernosum urethræ* consists of two different divisions; the central portion is formed by a reticulum of the conspicuously de-

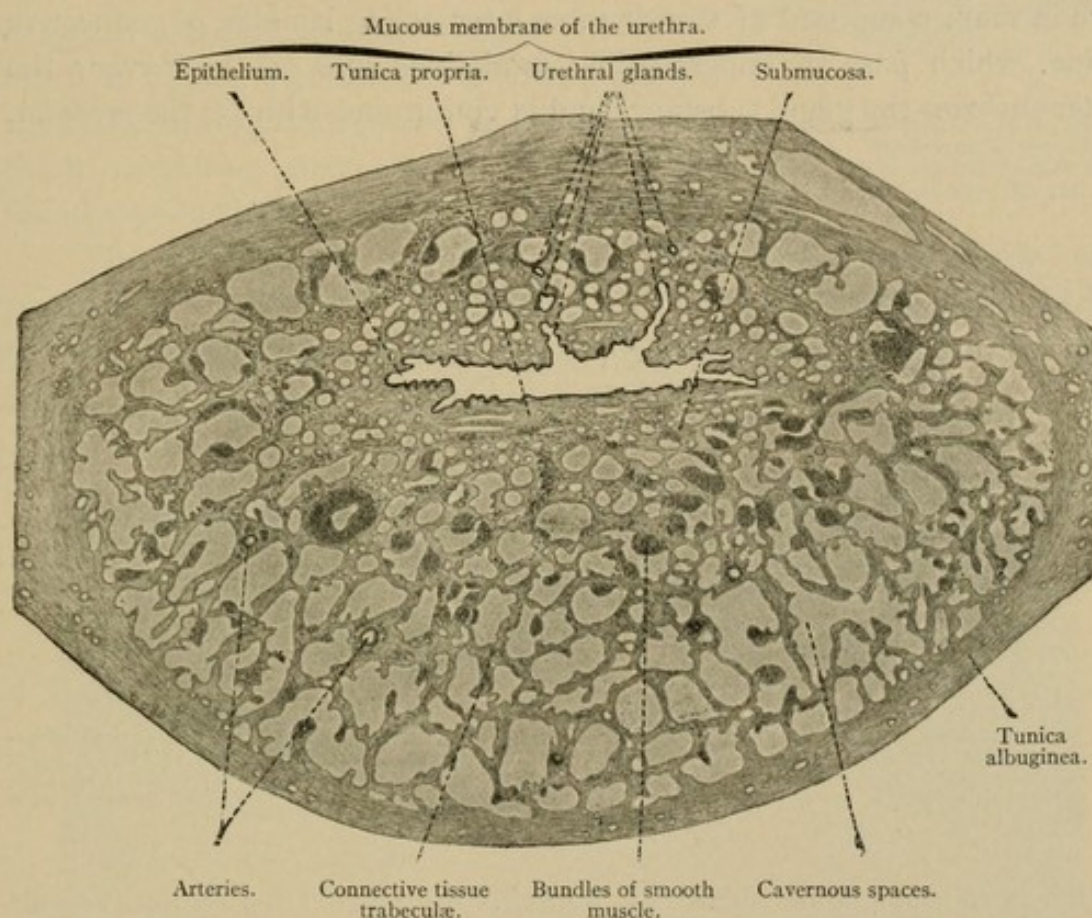


FIG. 269.—TRANSVERSE SECTION OF THE PARS CAVERNOSA URETHRÆ OF MAN. $\times 28$. Technic No. 152.

veloped veins of the submucosa of the urethral mucous membrane (p. 333); the peripheral portion in structure resembles the corpus cavernosum penis, excepting that there is no direct communication of the arteries with the venous spaces. The tunica albuginea is composed simply of a layer of circularly arranged bundles of connective tissue. The glans penis consists of greatly convoluted veins, that are held together by a conspicuously well-developed connective tissue, containing many elastic fibers and supporting the arterioles and the capillaries. (For the skin of the glans, see p. 389.)

In the tunica albuginea of the corpora cavernosa, in the glans, and also in the prepuce peculiar terminal organs of nerves are found (p. 224).

THE FEMALE REPRODUCTIVE ORGANS.

THE OVARIES.

The ovaries consist of connective tissue and of gland substance. The compact connective tissue, the *ovarian stroma*, is arranged in several strata; outermost lies (1) the *tunica albuginea* (Fig. 270), a thick structure in man, composed of two or more intersecting lamellæ of connective tissue, which pass by imperceptible gradations into (2) the *cortex*; the latter encloses the gland substance and is continuous with (3) the *medulla*,

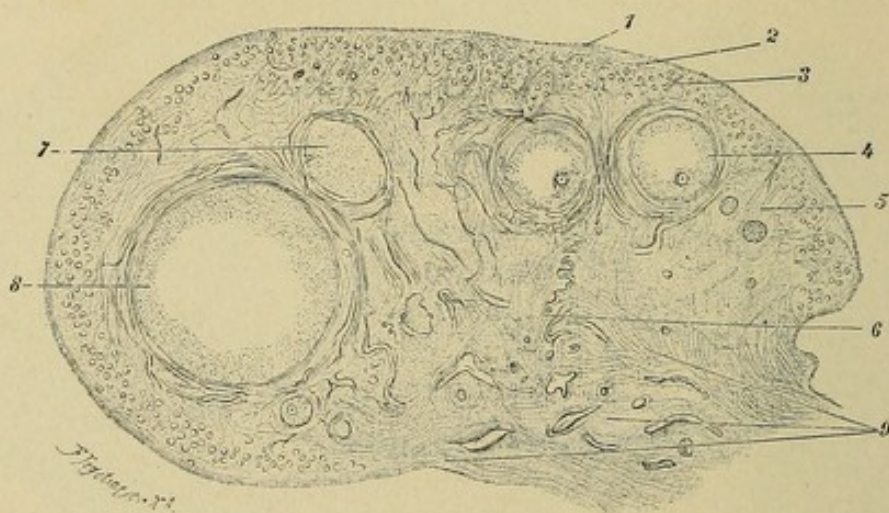


FIG. 270.—TRANSVERSE SECTION OF THE OVARY OF A CHILD EIGHT YEARS OLD. $\times 10$. 1. Germinal epithelium; 2, tunica albuginea; 3, outermost zone of the cortex containing numerous minute follicles; 4, larger follicle; 5, inner division of cortex; 6, medulla with numerous tortuous arteries; 7, follicle cut at the periphery; 8, large follicle, the cumulus oophorus not within the plane of the section; 9, hilus, containing wide veins. Technic No. 135.

which is rich in elastic fibers and contains numerous convoluted vessels accompanied by strands of smooth muscle-fibers. The *gland substance* is formed by a profusion of spherical epithelial sacs, the *egg-follicles*, each of which contains an *egg-cell*. In the human ovary there are about 36,000 follicles. The majority of the follicles are microscopically small ($40\ \mu$) and in the outer strata of the cortex form an arched zone embracing the entire organ except at the hilus, the place where the vessels enter (Fig. 270). The larger follicles occupy somewhat deeper portions of the cortex. The largest, those follicles readily perceptible by the unaided eye, when fully developed extend from the medulla to the tunica albuginea. The surface of the ovary is covered with a simple layer of very small, short cylindrical or flat cells, the *germinal epithelium*.

Only the initial stage in the development of the egg-cells takes place during embryonal life; their subsequent development, from the primordial to the fully ripened cell, may be observed in all its phases in every functionally active ovary. During the fetal period many cells of the germinal epithelium divide into two cells lying one above the other, of which the lower enlarges and becomes the *primordial egg-cell* with its large nucleus and nucleolus, while the upper cell and also its neighbor-cells become flattened and place themselves shell-like around the ovum. Such conditions are still found after birth (Fig. 271).

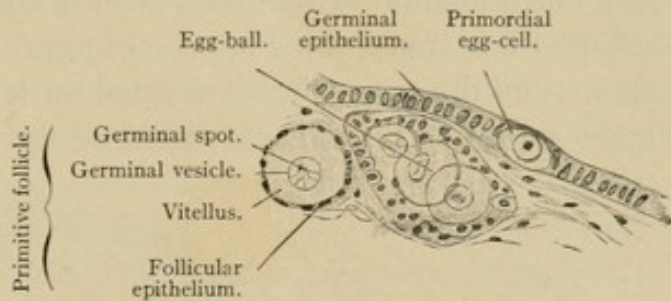


FIG. 271.—FROM A VERTICAL SECTION OF THE OVARY OF AN INFANT FOUR WEEKS OLD. $\times 240$. The primordial egg-cell has a large nucleus with a nucleolus. The egg-ball contains three egg-cells, surrounded by cylinder cells. Technic No. 153.

The egg-cell, which under circumstances may divide again, surrounded by its indifferent neighbor-cells, now moves down into the ovarian stroma, while above in the germinal epithelium new primordial eggs arise

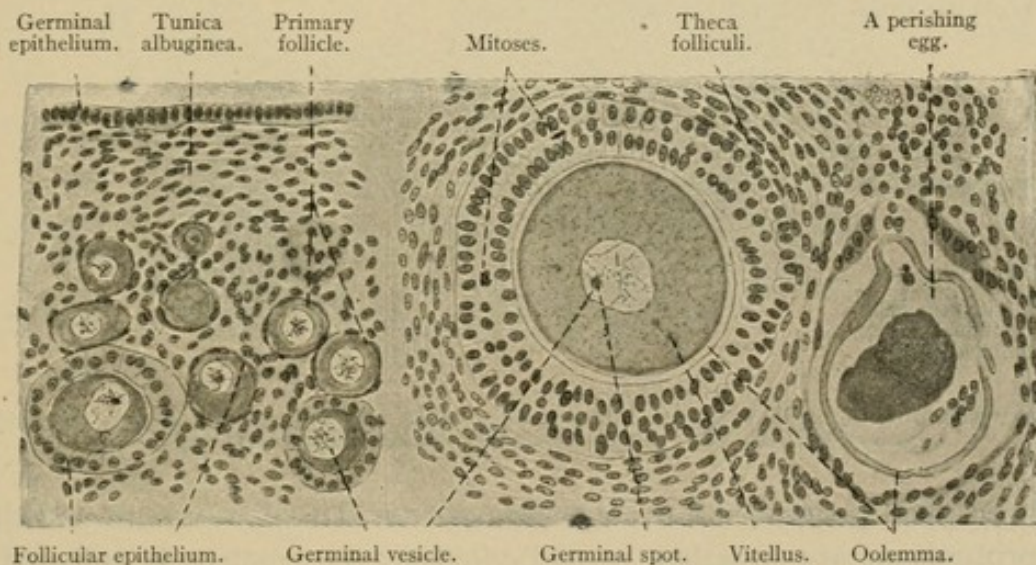


FIG. 272.—FROM A SECTION OF THE CORTEX OF A RABBIT'S OVARY. $\times 240$ Technic No. 153.

in the same way, that likewise move into the depths. Thus originate entire complexes of egg-cells and indifferent cells of the germinal epithelium, complexes which are named *egg-balls* (egg-pouches, egg-nests). Each egg-cell subsequently becomes separated from its neighbors by the rapid multiplication of the indifferent epithelial cells, as well as by proliferation of the connective tissue, and is then an isolated spherical body, the *primitive follicle*, that consists of the egg and the epithelial cells enclosing it, the so-called follicular epithelium, and of a connective-tissue sheath.

So far the processes are chiefly fetal.* The cells of the follicular epithelium now grow taller (Fig. 272, below left), then become stratified, the egg grows larger, takes up an eccentric position within the follicle, and obtains a delicate, radially striated border stratum that gradually increases in thickness, the *zona pellucida* (oölemma). With the enlargement of the egg a differentiation of its protoplasm is also accomplished; the greater portion of it is transformed into a crummy mass, the *deutoplasm*; only a zone around the eccentrically situated nucleus and a thin stratum covering the surface of the egg are distinguished by a more abundant quantity

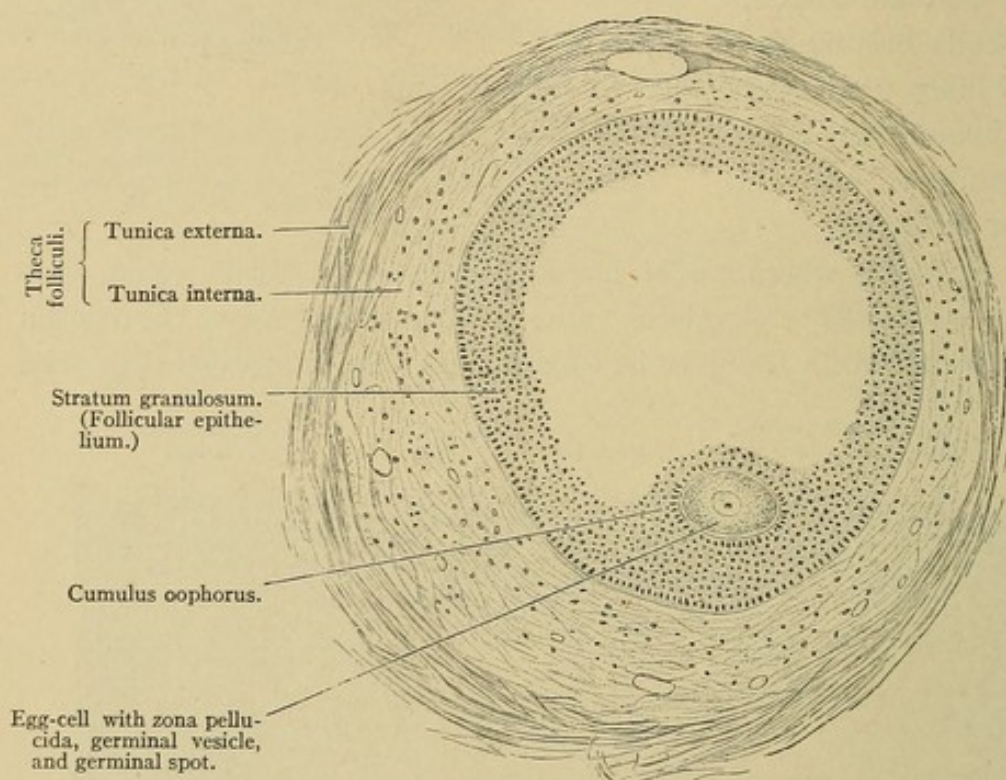


FIG. 273.—SECTION OF A LARGE VESICULAR FOLLICLE OF A CHILD EIGHT YEARS OLD. $\times 90$. The clear space within the follicle contains the liquor folliculi. Technic No. 153.

of the original protoplasm, the *egg protoplasm*. The deutoplasm and the egg protoplasm are together named *vitellus*,† the nucleus is called the *germinal vesicle* (vesicula germinativa), which contains the *germinal spot* (macula germinativa). Ameboid movements have been observed in the latter. The full-grown human egg has a diameter of about 0.3 mm.

The follicle now develops further; during continual multiplication

* In a few cases egg-balls and egg-cells with several germinal vesicles are found in sexually mature individuals, which represent elements in which the division of the cell-body has not yet taken place or perhaps they are an effect of pressure, that is two separate egg-cells were so pressed together that their dividing line disappeared.

† The *nucleus of the vitellus*, long known in animals, has recently been found in the human egg; it corresponds to the centrosome or to the archoplasm (p. 65).

of the cells of the follicular epithelium a cleft appears in their midst that becomes filled with an aqueous fluid, the *liquor folliculi*. This liquor is partly a transudate from the blood-vessels surrounding the follicle, is partly derived from the liquefaction of some of the cells of the follicular epithelium; it undergoes progressive increase in quantity and consequently the follicle soon expands to a vesicle filled with fluid, the *vesicular follicle* (Graaf), having a diameter of from 0.5 to 12 mm. Around the larger follicles the connective tissue of the stroma is arranged in circular strands forming a sheath called the *theca folliculi*, in which an outer fibrous layer, the *tunica externa*, and an inner vascular layer rich in cells, the *tunica interna*, can be distinguished (Fig. 272). Thus the follicle consists of a connective tissue sheath, the theca folliculi, and of the stratified follicular epithelium (Fig. 273), which in teasing fresh follicles be-

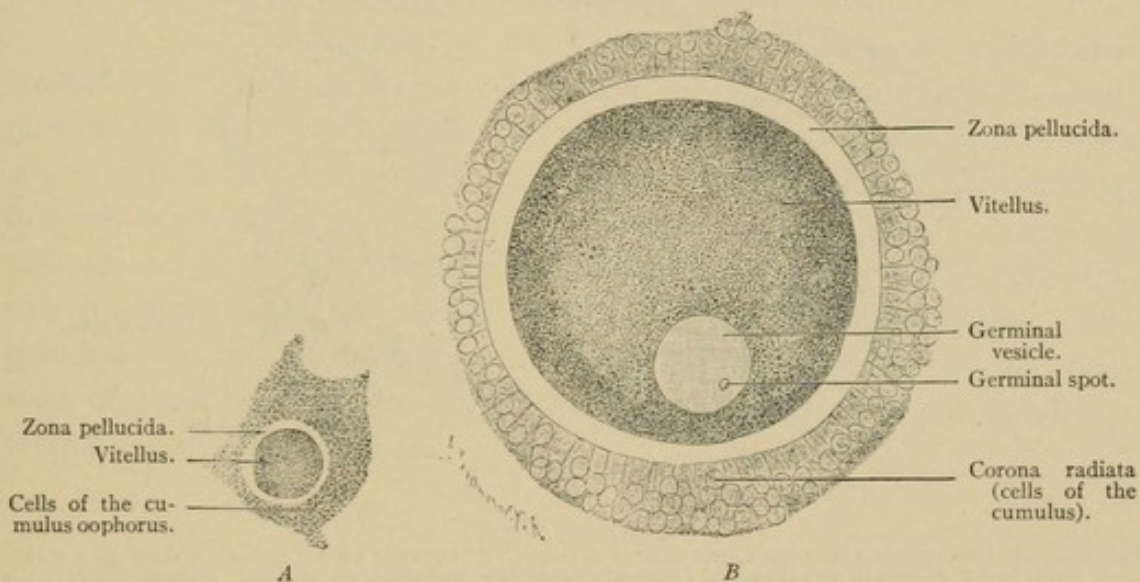


FIG. 274.—AN OVUM FROM A VESICULAR FOLLICLE OF A COW. *A* magnified 50, *B* magnified 240 times. The radial striation of the zona pellucida cannot be seen. Technic No. 154.

comes detached in large shreds, and has long been known as the *stratum (membrana) granulosa*;* at one point it presents a thickening, the *cumulus oophorus*, which encloses the egg. The cells of the cumulus which lie next to the zona pellucida are radially placed to the egg and form the *corona radiata* (Fig. 274). The greater part of the interior space of the follicle is occupied by the liquor folliculi.

When the vesicular follicle has attained its full development it bursts at the side directed toward the surface of the ovary, where its site is previously indicated by the attenuated and arched overlying

* Between the tunica interna and the follicular epithelium a delicate *membrana propria* is found in man, that in animals is often replaced by a thin ring fiber-layer.

tissue; the egg-cell escapes into the pelvic cavity, the empty follicle undergoes regressive change and is converted into the yellow body, the *corpus luteum*. When the discharged egg is not fertilized the yellow body disappears after the lapse of a few weeks; if on the other hand pregnancy occurs, the ruptured follicle develops into a large body, that possesses a diameter up to three centimeters and endures for years. At first it consists of a fibrous membrane, the former tunica externa of the theca, and of a yellow mass, that is formed of large, fatty cells, the *lutein cells*,—the multiplied and enlarged elements of the follicular epithelium—between which delicate connective-tissue septa occur, derivatives of the connective-tissue tunica interna of the theca folliculi (Fig. 275).

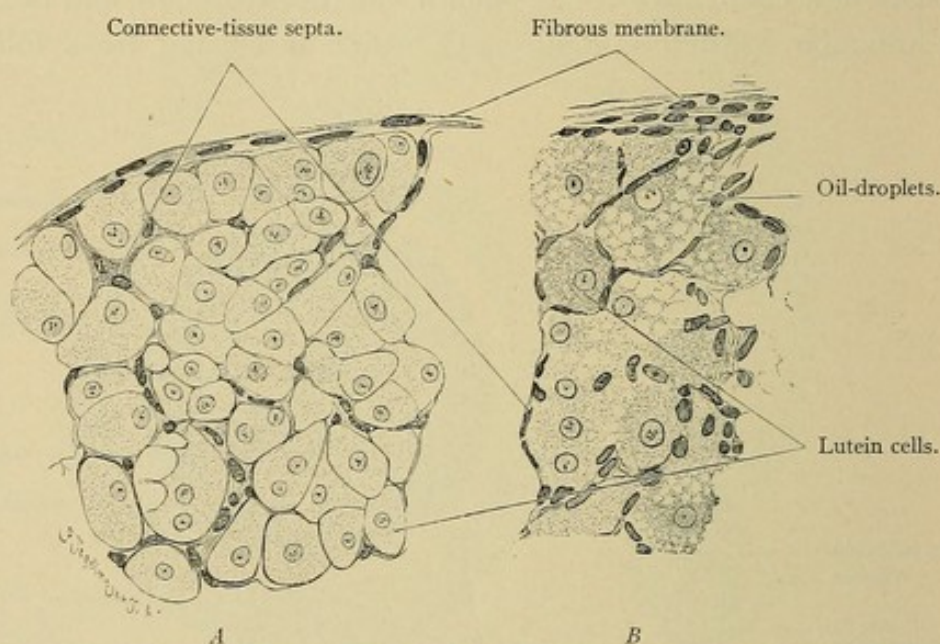


FIG. 275.—A. PORTION OF A CORPUS LUTEUM OF A RABBIT. B. PORTION OF A CORPUS LUTEUM OF A CAT
X 260. In B the lutein cells have become fatty and contain large and small oil droplets. Technic No. 153.

In the center of the corpus luteum is a gelatinous connective tissue and occasionally a cavity filled with blood. The blood is derived from the torn vessels of the tunica interna. Later the center becomes decolorized and the blood is replaced by a crummy mass, occasionally containing hematoidin crystals (see page 139).

Not all the primitive follicles attain complete development. Many undergo regressive change. Retrograde metamorphosis of larger follicles also occurs.*

* The process is effected in this wise: the tunica interna of the theca folliculi increases greatly in thickness, during which the egg dies; then cells, partly elements of the stratum granuloseum, partly leucocytes, wander into the egg and liquefy and absorb its substance. After the migratory cells have accomplished the liquefaction and resorption of the material of the vitellus they perish. Such degenerating follicles are called *atretic follicles*; they are easily recognized by the wrinkled oölemma, the part of the egg which persists the longest (Fig. 272, "A perishing egg").

The *arteries* of the ovary, branches of the ovarian and the uterine arteries, enter at the hilus, divide in the medulla, and are characterized by their tortuous course (Fig. 270). From the medulla they pass to the cortex, where they chiefly supply the capillary networks situated in the tunica interna of the follicles. The *veins* form a dense plexus at the hilus of the ovary. The numerous *lymph-vessels* can be traced to the tunica interna of the follicles. Medullated and nonmedullated *nerves* in large number enter the medulla through the hilus, in company with the blood-vessels, to the walls of which the majority of them are distributed. A few of the nerves proceed to the cortex; these form there a dense plexus of delicate, mostly nonmedullated fibers, which envelops the follicles and sends minute branches to the walls of the blood-vessels; whether nerve-fibers penetrate within the epithelium of the larger follicles is not yet definitely established.

The *epoöphoron* and the *paroöphoron* are remains of embryonal structures. The former lies within the lateral division of the mesosalpinx, at the hilus ovarii (in the cat, mouse, etc., in rare cases also in man within the hilus), and consists of a group of convoluted, blind-ending tubules, the walls of which consist of cylinder epithelium, occasionally ciliated, and of circularly arranged connective-tissue fibers. The epoöphoron is a remains of the sexual segment of the primitive kidney. The paroöphoron lies in the median division of the mesosalpinx* and consists of branched tubules lined with cylinder epithelium; it is also a remains of the mesonephros.

THE OVIDUCT.

The wall of the *oviduct* (*tuba uterina*, *Fallopia*) consists of three membranes: an inner mucous membrane, a middle muscular membrane and an outer serous envelop. The *mucous membrane* is thrown into numerous longitudinal folds, that correspond in amplitude to the size of the tube, and are highest in the ampulla, where they are united to one another by minute oblique secondary plications. The thick mucous membrane consists of (a) a simple layer of ciliated cylinder epithelium, —the ciliary wave is directed toward the uterus, and of (b) a tunica propria rich in connective-tissue cells, that lies close against the muscle membrane.†

* According to recent investigations the paroöphoron is said to correspond to structures beneath and external to the attachment of the mesovarium, that are to be sought along the free edge of the broad ligament.

† Longitudinal strands of smooth muscle-fibers lying close beneath the tunica propria in a few places, by some authors are described as *muscularis mucosæ*.

The *muscular membrane* consists of an inner thick circular and an outer, in places thin, longitudinal layer of smooth muscle-fibers. The *serous tunic* is formed by the peritoneum and by a conspicuous layer of loosely united connective-tissue bundles. Elastic fibers occur in the muscularis and in the serosa, but in children and in old women are confined chiefly to the serosa. The highly developed *blood-vessels* between the circular and the longitudinal layer of the muscularis send or receive

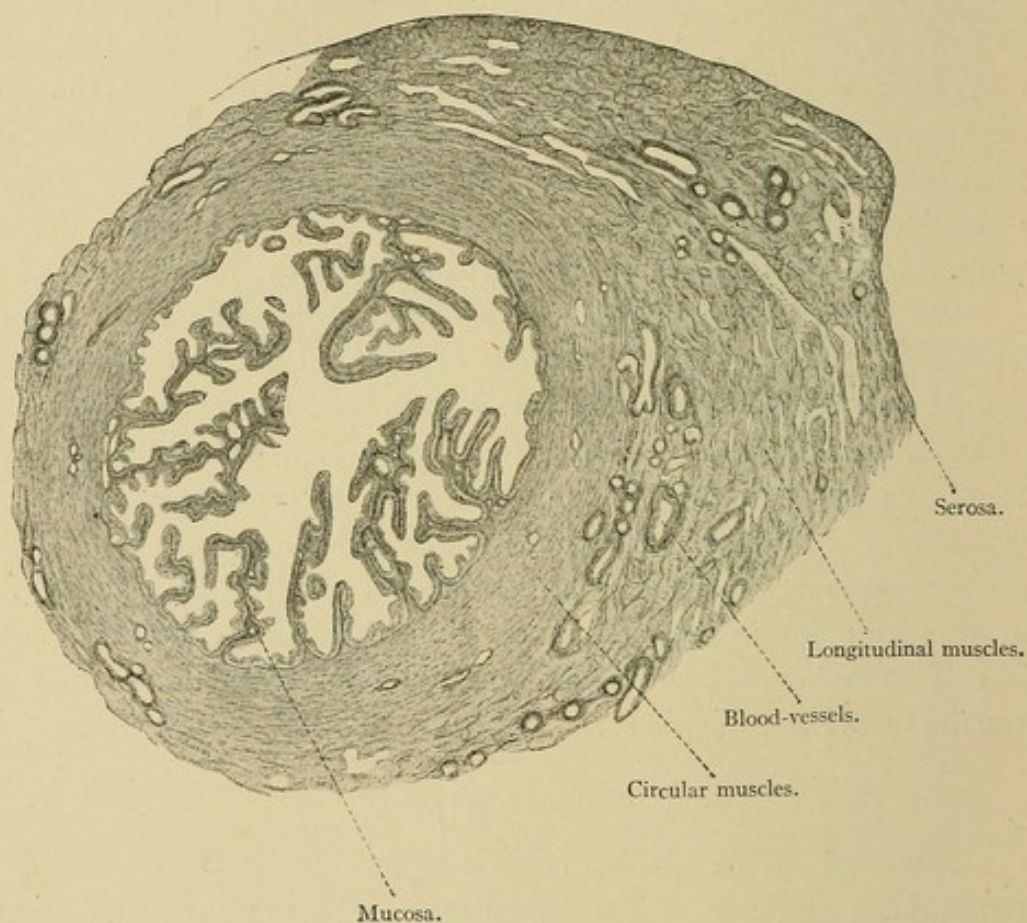


FIG. 276.—TRANSVERSE SECTION OF THE OVIDUCT OF AN ADULT WOMAN (NEAR THE AMPULLA). $\times 30$.
Technic No. 156.

branches from the mucous membrane, which is provided with a narrow-meshed capillary plexus. The larger veins run along the longitudinal folds of the mucosa. The knowledge of the exact behavior of the *lymph-vessels* is still wanting. The *nerves* (in the pig) form a rich plexus in the mucosa, from which branches ascend to the epithelium. A penetration in the epithelium has not been observed.

THE UTERUS.*

The wall of the uterus, like that of the oviduct, consists of a mucosa, a muscularis, and a serosa (Fig. 277).

* This chapter has been revised and considerably enlarged by the editor.

The *serosa* exhibits no special characteristics.

The *muscularis* consists of smooth muscle-fibers, united into bundles which interlace in all directions, so that a sharp demarcation of single layers is not possible; still in general three strata, more or less well-defined, can be distinguished: (1) an *inner*, the *stratum submucosum*, chiefly composed of bundles disposed in a longitudinal direction; (2) a *middle*, the most robust, consisting of bundles having in general a circular disposition and containing wide veins, hence the name *stratum vasculare*; (3) an *outer layer*, the *stratum supravasculare*, formed of bundles extending partly in a circular, partly in a longitudinal direction, the latter lying close beneath the *serosa*. The longitudinal bundles of

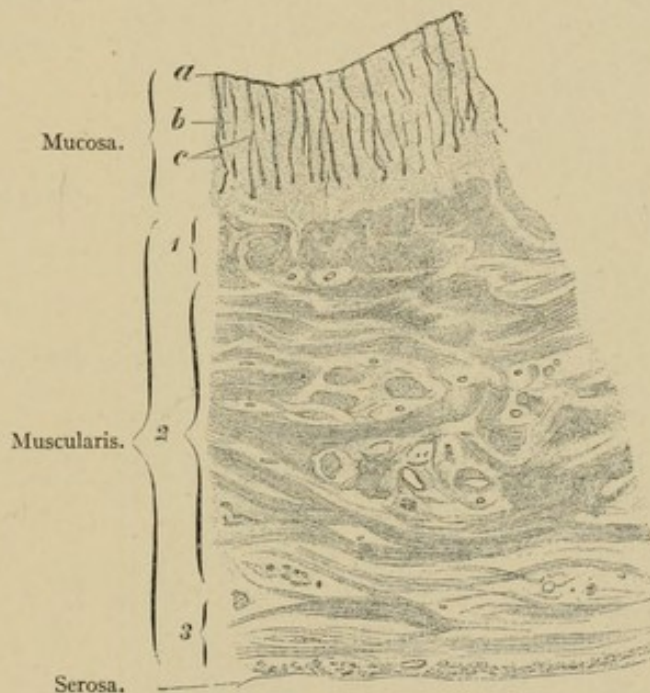


FIG. 277.—FROM A TRANSVERSE SECTION OF THE MIDDLE OF THE UTERUS OF A GIRL FIFTEEN YEARS OLD. $\times 10$. a, Epithelium; b, tunica propria; c, glands; 1, inner muscular layer (stratum submucosum); 2, middle muscular layer (stratum vasculare); 3, outer muscular layer (stratum supravasculare). Technic No. 156.

this stratum pass over into the musculature of the oviduct and into the surrounding subserous connective tissue of the folds of the peritoneum. The stratification of the muscular tissue is more pronounced in the *cervix*, where an inner and an outer longitudinal may be distinguished from a middle circular layer. The volume of the muscularis is subject to great variation, dependent on the functional state of the uterus.

The *muscle-fibers* differ somewhat from the elements of smooth muscle-tissue found in other organs. They are elongated cells, usually spindle-shaped, or are blunted and frayed at the ends. Frequently they are forked at their extremities. Their length varies greatly, in the virgin uterus from 40 to 60 μ ; during pregnancy they increase exces-

sively and at the end of the same attain a size of from 300 to 600 μ . The *nucleus* (not infrequently two or more are present in one cell) is usually oval and lies embedded in a granular substance.

The *mucosa* is sharply defined from the muscularis. It is the coat which in the different functional states of the uterus undergoes the profoundest and physiologically the most important changes. Therefore a description of the histologic structure of the mucosa of the uterus can only answer to the corresponding functional condition of the organ, and in consideration hereof will be presented in separate sections.

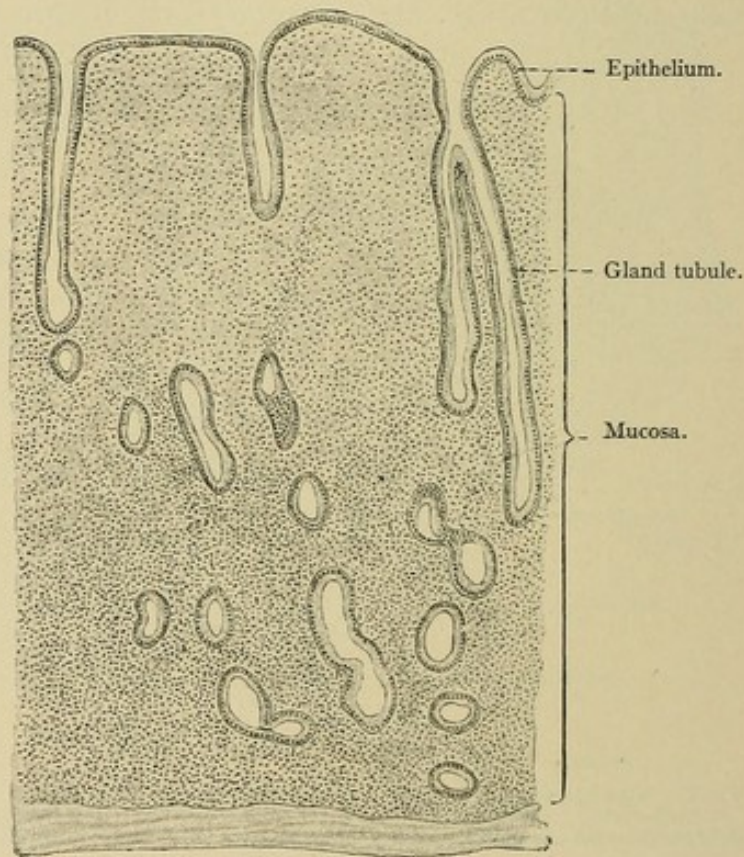


FIG. 278.—MUCOUS MEMBRANE OF THE RESTING UTERUS OF A YOUNG WOMAN. $\times 35$. (After Böhm and von Davidoff.)

It is desirable to consider:—

1. The mucosa of the virgin resting organ.
2. The mucosa of the menstruating uterus.
3. The mucosa of the gravid uterus.

The mucosa of the virgin resting uterus (Fig. 278), after the advent of puberty, has a thickness of from 1 to 2 mm. and bears on its surface a layer of simple ciliated columnar epithelium, 30 μ in height in the middle regions; the ciliary wave is directed toward the cervix. The tunica propria is formed of a fine fibrous tissue closely resembling embryonal

connective tissue; it consists of elongated cells furnished with oval nuclei, which send out in all directions branched processes that unite with those of neighboring cells and form a cellular network, the meshes of which are occupied by lymph and by numerous leucocytes.

The tunica propria supports many simple or forked gland tubules, of which the upper part pursues a more or less straight course, while the

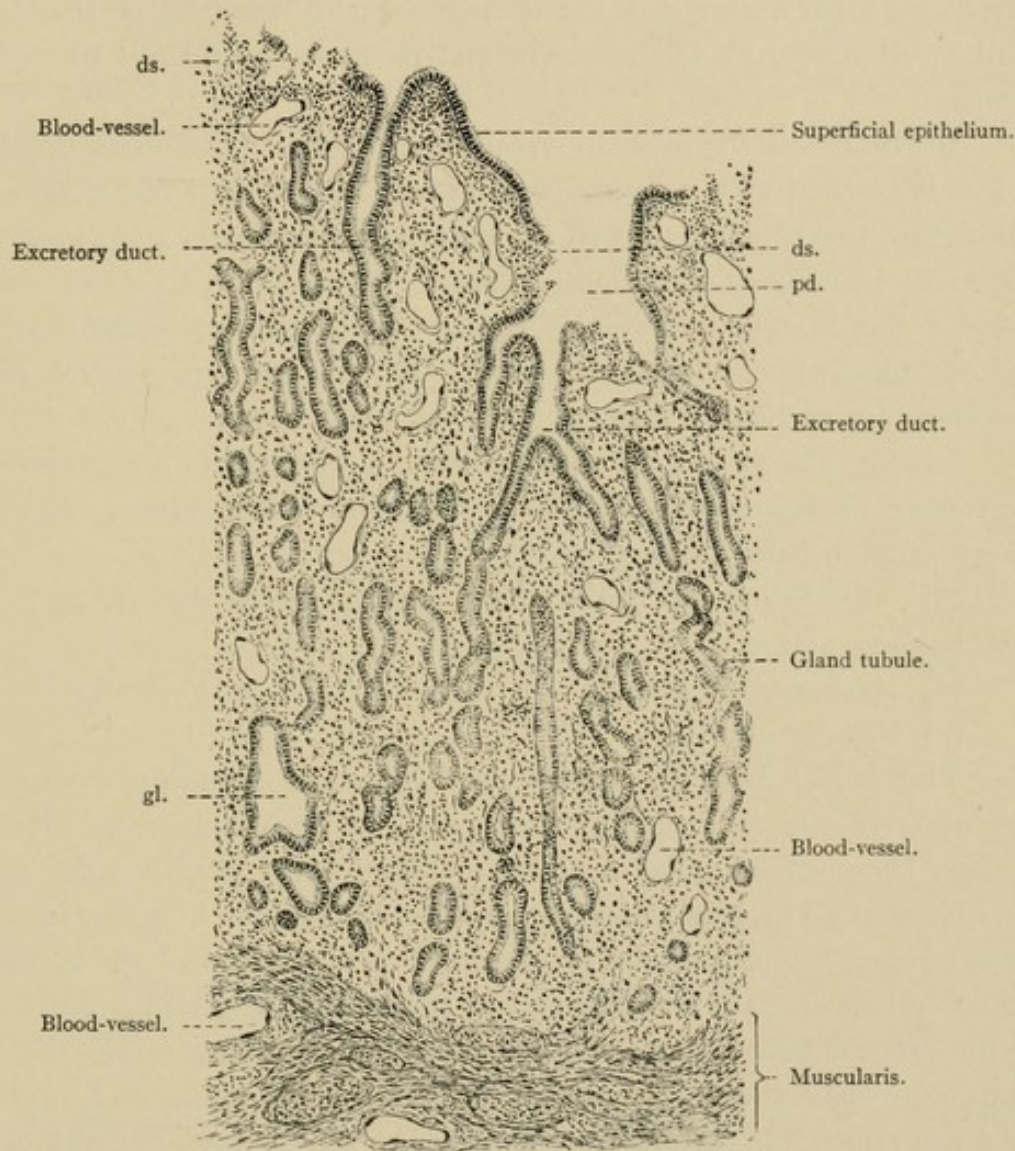


FIG. 279.—MUCOUS MEMBRANE OF A VIRGIN UTERUS DURING THE FIRST DAY OF MENSTRUATION. ds. Disintegrating surface; pd, pit-like depression of the mucous membrane; gl, gland lumen very much enlarged. $\times 30$.—(Schaper.)

lower part takes a serpentine course (Fig. 277). The glands extend close to the muscularis and here not infrequently they are bent at right angles, so that the fundus runs parallel to the muscular coat. The glands of the uterus are to be regarded as invaginations of the superficial epithelium and likewise consist of a simple layer of ciliated epithe-

lium, resting upon a delicate basement membrane composed of anastomosing connective-tissue cells.

The blood-vessels run in a winding manner from the muscularis to the surface of the mucosa and the *arteries* in particular are characterized by their extremely convoluted, corkscrew-like course. At the surface they break up into capillaries and form a close network. A similar network surrounds the gland tubules. The *veins* proceeding from the capillaries form a plexus in the deeper strata of the mucosa, that is especially well developed in the cervix and particularly around the external orifice.

In the *cervix* the mucous membrane is thicker and in its upper two-thirds is clothed with a single layer of tall ciliated cells ($60\ \mu$ high in the middle portion),* while toward the external orifice papillæ covered with a stratified squamous epithelium appear. In addition to a few scattered tubular glands, mucous follicles, the so-called *mucous crypts*, occur; they are one mm. wide, possess many evaginations, and by retention of their secretion are converted into cysts, the *ovula Nabothi*.

During the *period of menstruation* a number of progressive and regressive changes take place in the mucosa of the uterus, which may be grouped in three phases:—

(a) Thickening of the mucosa, accompanied by changes in its histologic structure.

(b) Menstruation proper.

(c) Regeneration.

The *initial phase* is characterized by a considerable increase in the thickness of the mucosa (up to 6 mm.), in consequence of which the surface becomes irregular and the orifices of the glands open in deep depressions. The thickening of the mucosa depends in a measure on an actual increase of the tissue produced by proliferation of the connective-tissue cells and the leucocytes and by growth of the gland tubules, which in the process take up an irregular course and become essentially wider. Simultaneously the blood-vessels, especially the veins and capillaries, undergo enormous distention, whereby the blood-supply of the organ is extraordinarily augmented. In this condition the mucosa is designated *decidua menstrualis*.

These changes are followed by a partial disintegration of the superficial strata of the mucosa, accompanied by an infiltration of blood into the subepithelial tissues. The molecular disintegration (associated with fatty degeneration) of the surface advances rapidly, the greatly dilated

* Transformation of these cells into goblet-cells occurs.

superficial blood-vessels become exposed, rupture, and cause hemorrhages within the uterine cavity, which flow into the vagina and give rise to the external phenomena of *menstruation*. After this discharge of blood the mucosa is rapidly reduced in thickness. The surface is now entirely devoid of epithelium and consists of connective tissue and exposed blood-vessels. This condition is immediately succeeded by the stage of *regeneration*. The hyperemia rapidly disappears, the extravasated blood is partly resorbed, partly cast off, a cellular network grows upward and restores the lost tunica propria, while from the gland-cells the epithelial covering of the mucosa is regenerated. New subepithelial capillaries are formed.

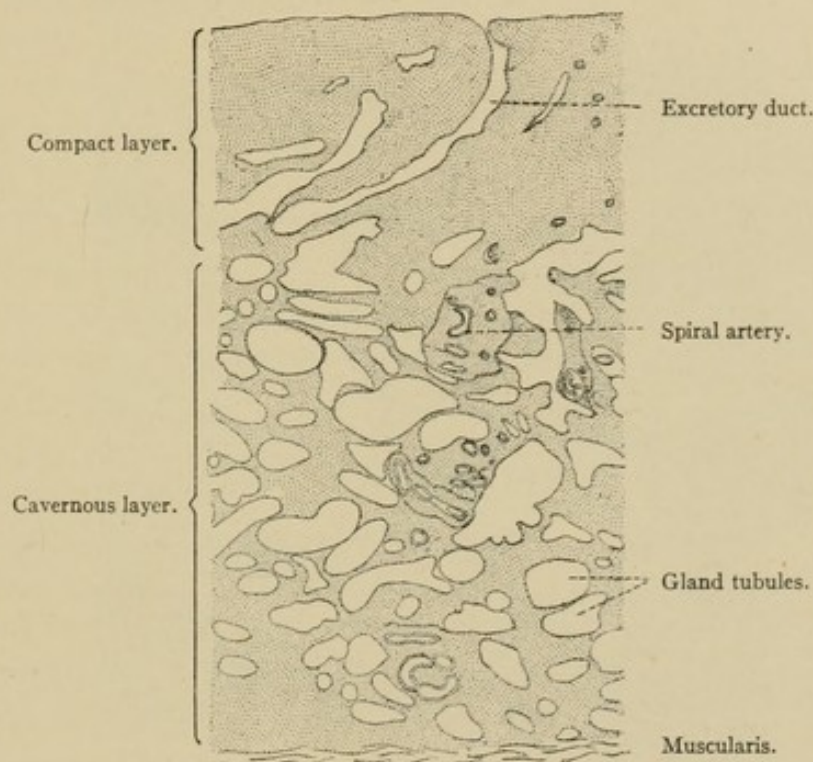


FIG. 280.—VERTICAL SECTION THROUGH THE MUCOUS MEMBRANE OF A HUMAN UTERUS ONE MONTH PREGNANT; it shows the outlines of the glands and the division of the mucosa into an upper compact and a lower cavernous layer.—(After Minot.)

The histology of the mucosa of the uterus during *pregnancy* (*decidua graviditatis*) (Fig. 280 and Fig. 281) is, on the whole, like that of the *decidua menstrualis*, with the alterations more pronounced. However, it undergoes considerable modification because of its intimate relations with the developing ovum in the uterus. These relations vary and thus in the course of development three essentially different parts of the mucosa may be distinguished:—

(a) The *decidua serotina* (*decidua basalis*), the area of the mucosa to which the ovum is attached (*placenta uterina*).

(b) The *decidua vera*, which comprises all the remaining portion of the mucosa attached to the wall of the uterus.

(c) The *decidua reflexa* (*decidua capsularis*), the portion of the mucosa which projects into the cavity of the uterus and encapsules the ovum.

The decidua serotina and the decidua vera undergo progressive development during the entire course of pregnancy and persist until its close; the decidua reflexa becomes gradually attenuated and disappears in the course of the fifth month.

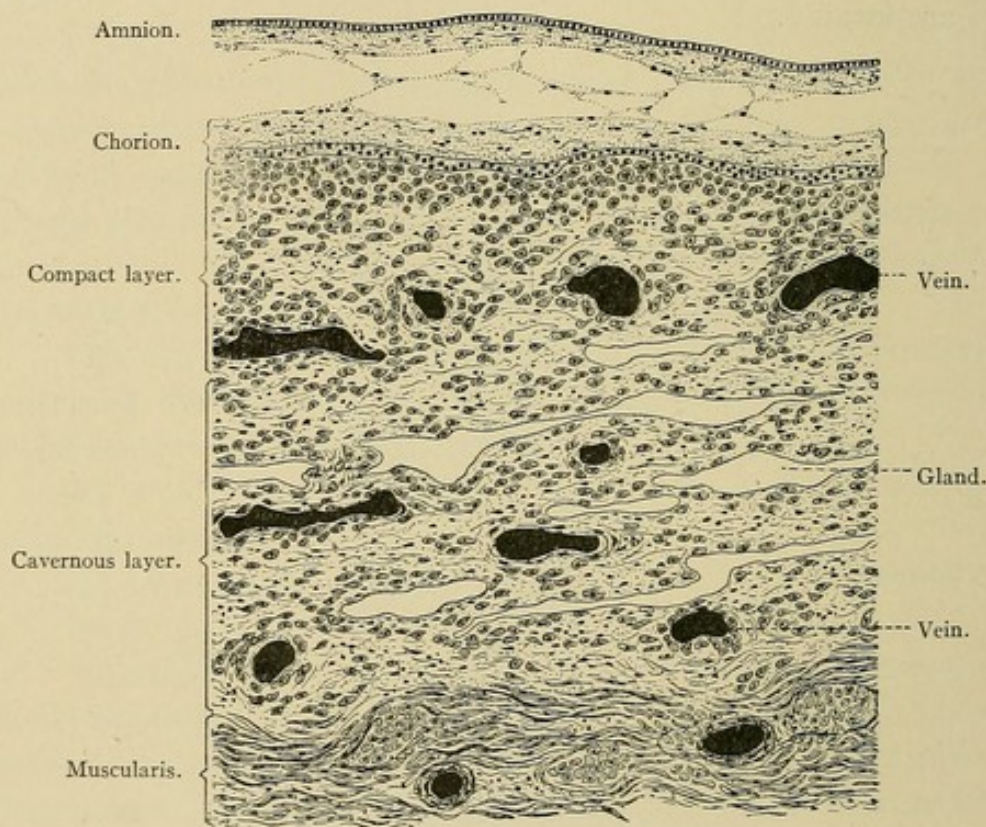


FIG. 281.—VERTICAL SECTION THROUGH THE WALL OF A UTERUS ABOUT SEVEN MONTHS PREGNANT, WITH THE FETAL MEMBRANES IN SITU. Between amnion and chorion are threads of the intermediate gelatinous connective tissue. $\times 30$.—(Schaper.)

A section of the greatly thickened mucosa (*decidua vera* and *decidua serotina*) shows the same histologic details that have been described in the menstrual decidua, but with this difference, that the progressive alterations (proliferation of the connective-tissue elements, dilation of the blood-vessels and glands) attain much greater proportions. A *superficial compact zone* and a *deep spongy zone* can always be distinguished (Fig. 280). The cavities in the latter are produced by the lower divisions of the gland tubules, which have become greatly widened and very tortuous. At a later stage of pregnancy, owing to the great

expansion of the uterus, the lumina of the glands appear compressed and straighter (parallel to the muscular coat). (Fig. 281.) Between the glands are numerous blood-vessels, spindle-cells, and multinucleated giant-cells. The epithelium of the glands early begins to loosen, and in great part the cells lie irregularly scattered in the lumen of the tubule, where they disintegrate. The orifices of the glands are gradually obliterated, since the walls after the loss of the epithelium become adherent and grow together.

The *blood-vessels* of the mucosa are all dilated, especially the superficial veins and capillaries; the latter often form distended, sinus-like cavities in the upper layer of the decidua. In the decidua serotina the arteries and the veins open on the surface of the mucosa (Fig. 283 and Fig. 284), so that here the maternal blood circulates between the chorionic villi of the placenta (see page 367). In the decidua vera the blood-vessels toward the end of pregnancy are less conspicuous.

Of especial interest are peculiar, typical cells, *decidual cells*, that appear in large numbers in the mucosa of the gravid uterus. They are flattened, spherical, oval, or branched cells of conspicuous size (0.03 to 0.1 mm.), that in the latter half of pregnancy assume a characteristic brown color. They usually possess but one nucleus, though occasionally two, three, or more are present, and in rare cases as many as

thirty or forty. The decidual cells are most numerous and most densely aggregated in the upper compact zone of the serotina (Fig. 281), which owes its typical character and brown color to these elements. Occasionally cells are found that are united with one another by means of protoplasmic processes. According to Minot, the decidual cells originate from connective-tissue elements, therefore may be regarded as a modified embryonal or so-called anastomosing connective tissue.

In a cross-section of the decidua vera in the latter half of pregnancy it will be seen that the surface of the mucosa is covered by two distinct membranes,—fetal membranes—the *chorion* and the *amnion* (Figs. 281, 283). The chorion lies next to the decidua vera and is intimately united with it. It consists of two layers, an epithelial and a connective-tissue layer, of which the former is turned toward the uterine wall, the latter



FIG. 282.—DECIDUAL CELLS FROM THE MUCOUS MEMBRANE OF A HUMAN UTERUS ABOUT SEVEN MONTHS PREGNANT. Below a "giant-cell," above to the right a cell with a karyokinetic figure. $\times 250$.—(Schaper.)

toward the amnion. Two similar layers may be distinguished in the amnion, but of these the epithelial layer, which consists of cubical cells, is turned toward the cavity of the uterus, while the connective-tissue stratum faces the chorion. The amnion and the chorion are loosely united to each other by mucous connective tissue, in which delicate fibrils may be seen extending from one membrane to the other.

The *lymph-vessels* of the uterus form in the mucosa a wide-meshed network provided with blind branches. From this small stems proceed through the muscularis and communicate with a close subserous network of larger channels.

The *nerves* of the uterus, medullated as well as nonmedullated, are very numerous. They branch—the medullated nerves after losing their medullary sheath—in the muscularis and form a dense plexus in this and in the mucosa. From the latter delicate fibrils may be traced between the epithelial cells.

THE PLACENTA.*

The placenta is an organ which from a morphologic standpoint is composed of two heterogeneous parts, of which the one is produced by the mother (placenta uterina), the other by the embryo (placenta foetalis). It is the result of the intimate union of a circumscribed area of the chorion (chorion frondosum) with that portion of the mucosa of the uterus known as the decidua serotina. The placenta serves the purpose of bringing the fetal and the maternal blood into the closest proximity, to render possible the interchange of materials between them. To subserve this function the organ possesses a peculiar histologic construction, in which the blood-vessels, especially in their arrangement and structure, take a prominent part.

In the histologic investigation of the placenta various obstacles are encountered, owing to its being an extremely soft, spongy mass, traversed by numerous wide blood-vessels. The comprehension of the minute structure will be considerably facilitated by proceeding from the previously mentioned fact that the finished organ is the product of two originally heterogeneous structures, the chorion on the one side, the decidua serotina on the other, and that their union is substantially effected in that the chorion, by means of numerous villous-like proliferations, penetrates the underlying serotina, the surface of which is peculiarly modified and further regressively altered for its reception, and as it were takes root in the same. For the investigation of these relations sections through the

* This chapter is an entirely new addition by the editor.

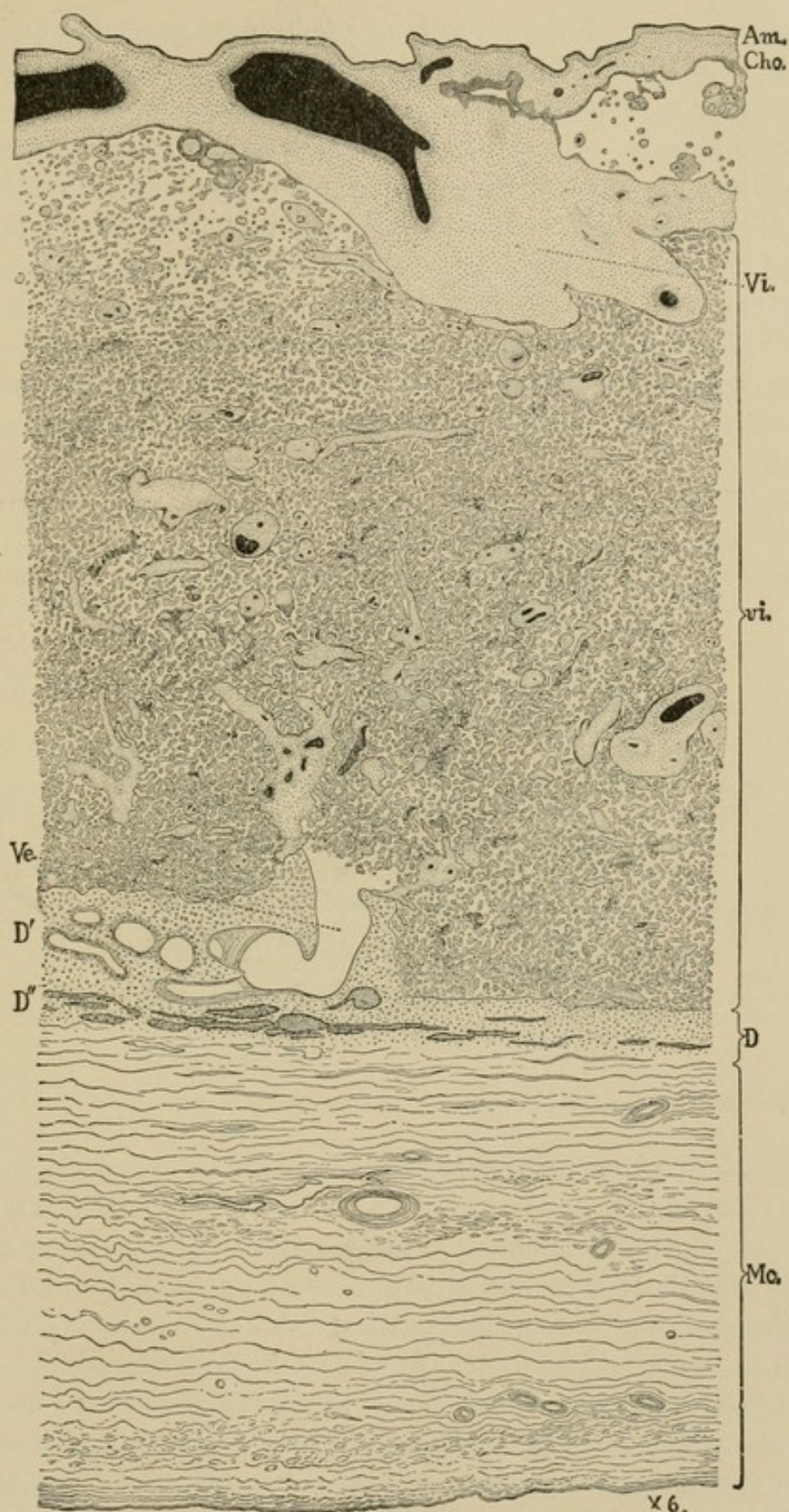


FIG. 283.—SECTION THROUGH A NORMAL HUMAN PLACENTA OF ABOUT SEVEN MONTHS, IN SITU. Am., Amnion; Cho., Chorion; Vi, trunk of a villus; vi, sections of villi in the substance of the placenta; D, decidua basalis; Mc, muscularis; D', compact layer of decidua; Ve, uterine artery opening into the placenta. The fetal blood-vessels are drawn black; the maternal blood-spaces are left white; the chorionic tissue is stippled, except the canalized fibrin, which is shaded by lines; the remnants of the gland cavities in D'' are stippled dark.—(After Minot.)

wall of the uterus with the placenta *in situ*, toward the end of pregnancy, are most instructive. In such a section two sharply defined zones may be recognized: an outer compact stratum consisting of the greatly thickened muscular coat of the uterus, covered externally by the serosa, and an inner spongy zone containing numerous inter-communicating spaces filled with blood. The latter is the placenta, that is, the united decidua serotina and chorion frondosum. The accompanying illustration (Fig. 283) shows their relations under low magnification, which will be elucidated by referring to the schematic representation in figure 284.

The surface of the placenta directed toward the cavity of the uterus is covered by a compact stratum, the *membrana chorii*, which is chiefly

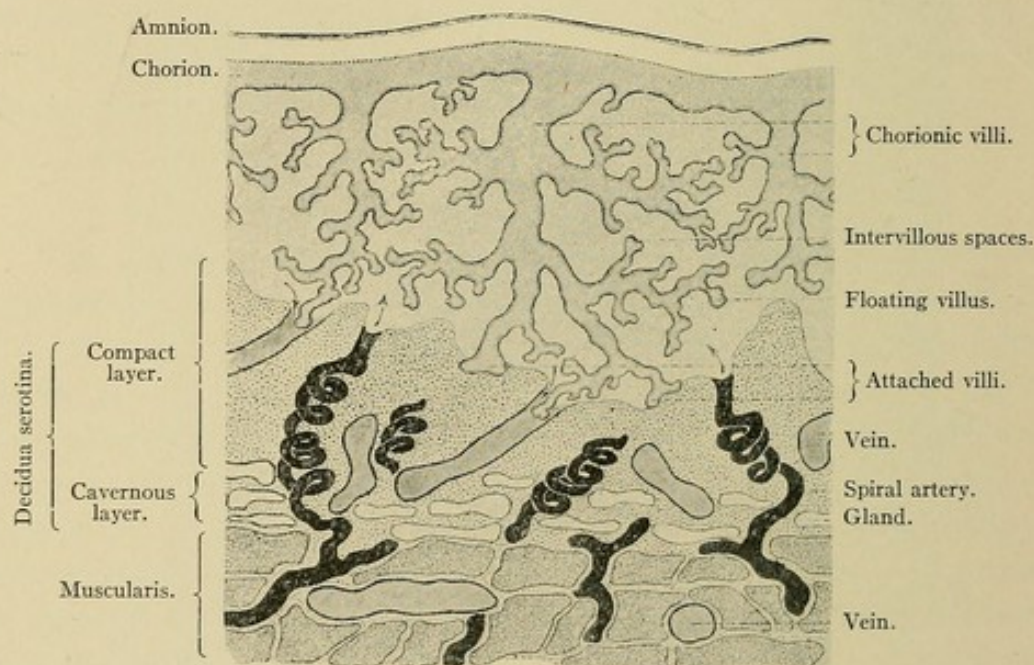


FIG. 284.—DIAGRAM OF THE HUMAN PLACENTA AT THE CLOSE OF PREGNANCY. Cf. Fig. 283.—(Schaper.)

composed of fibrillar connective tissue, in which the main branches of the umbilical blood-vessels run. The outer surface of the chorion is covered by a delicate membrane, the placental portion of the amnion, which as previously stated consists of an inner epithelial and a connective-tissue layer and is attached to the chorion by means of embryonic connective tissue. The other surface of the *membrana chorii*, that directed toward the wall of the uterus, is closely beset with innumerable villous-like structures, large and small, which in the upper part of the placenta form a dense tangle, the terminal ramifications of which are embedded in the cleft, uneven substance of the serotina. On closer study of this villous tangle it will be seen that the larger stems run a more or less direct course from the chorion to the serotina, in order to

secure a firm union with the latter, while their many much-branched lateral twigs usually establish no connection with the uterine portion of the placenta, but terminate free in the blood-spaces, the so-called *intervillous spaces*, between the chorion and the serotina. Dependent upon these relations the branches of the chorionic villi are divided into *roots of attachment*, or *main stems*, and *free processes*, or *floating villi*. From the chorion a branch of the umbilical artery enters each main stalk and within the terminal ramifications of the villus breaks up into a dense capillary network from which the umbilical veins take their origin and carry back the blood from the chorion through the umbilical cord to the fetus. Accordingly, the blood-vessel system of the fetal placenta is entirely

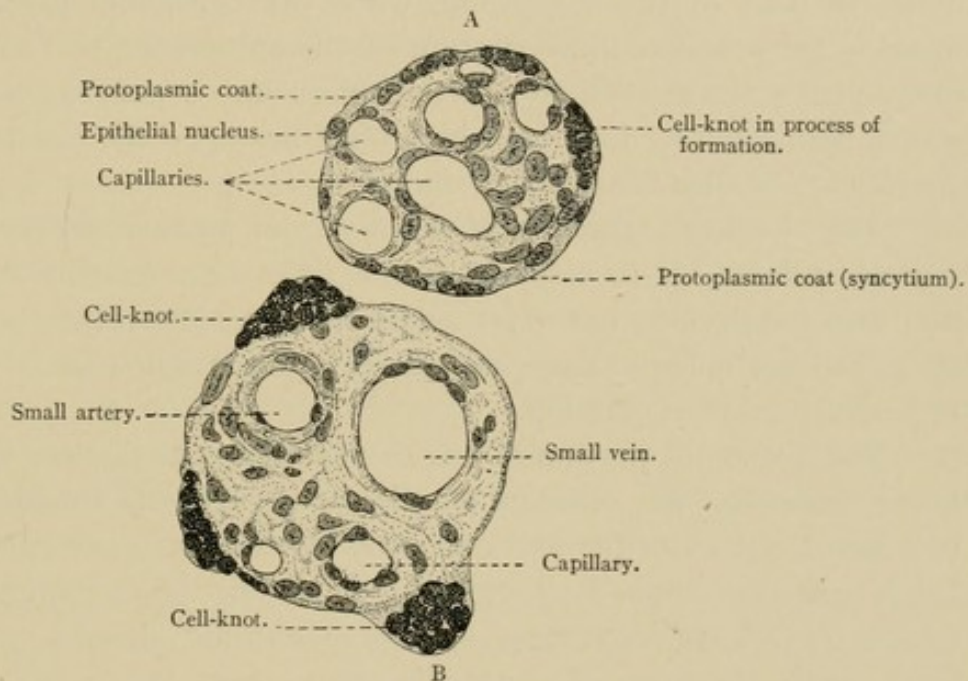


FIG. 285.—CROSS-SECTION THROUGH A SMALLER (A) AND A LARGER (B) CHORIONIC VILLUS OF A HUMAN PLACENTA AT THE END OF PREGNANCY. $\times 250$.—(Schaper.)

closed. Nowhere does a direct intermingling of the maternal and the fetal blood occur.

A cross-section of one of the smaller chorionic villi, highly magnified, shows that it is chiefly composed of mesenchymal tissue (mucous tissue), in which the blood-vessels are embedded (Fig. 285). This central supporting substance is covered by an irregular and not everywhere continuous stratum of epithelium. In the earlier months of development two distinct strata may be distinguished in the epithelium of the villi: an inner, lying immediately upon the supporting tissue, in which the cells possess large nuclei and definite contours, so that in the main they are distinctly separated from one another, and an outer layer, consisting of a continuous protoplasmic mass—*syncytium*—containing num-

erous small, irregularly scattered nuclei. Toward the end of pregnancy, however, the epithelium of the villi undergoes great alteration, as appears in the illustration (Fig. 285). On the larger villi a true epithelial investment has almost entirely disappeared and instead isolated accumulations of large round nuclei are found; they stain intensely, are embedded in a clear, homogeneous substance, and form protuberances (*Zellknoten*, *cell-knots*) on the surface of the villi. Between these cell-knots the connective-tissue of the villi frequently is covered only by a thin, homogeneous stratum, or in some cases (especially in smaller villi) this stratum still retains more or less the character of the protoplasm containing scattered nuclei. There are many indications that the latter is the remains of the syncytium, while the cell-knots probably originated in the primitive inner stratum of the epithelium of the villi.* In many places the syncytium is transformed into a peculiar hyaline substance permeated by fissures, which often lies upon the chorion in dense strata and is called *canalized fibrin*.

The histologic structure of the maternal portion of the placenta—*placenta uterina*—in its essential features has been described in connection with the decidua in the preceding chapter. But certain peculiarities, as well as the union of maternal and fetal placenta in a functional whole, require a brief consideration.

The placental portion of the decidua (Fig. 283), that forming the lower stratum of the placenta (basal plate), is greatly thinned (from 0.5 to 1 mm.), but as in the extraplacental portion an upper compact layer and a lower cavernous layer (gland lumina) may be distinguished. The decidual cells are extremely numerous and lie closely crowded. A honeycomb structure of connective-tissue septa (*septa placentaë*) arises from the surface of the serotina, directed toward the intervillous spaces, and penetrates between the villi of the chorion, separating the latter into lobes or cotyledons. Only in the peripheral regions of the placenta do these septa reach to the membrana chorii, where frequently they form on the inferior surface of the latter a thin membranous stratum, the *decidua placentalis subchorialis*. On the margin of the placenta the serotina gradually increases in thickness and passes into the vera, at which point it is closely applied to and firmly united with the chorion. Within the

* It has not been yet determined with certainty whether the epithelium of the villi of the *human* placenta is entirely derived from the epithelium of the chorion, or whether the epithelium of the serotina participates in its composition. However, recent investigations as well as comparative anatomical facts indicate that only the inner epithelial stratum of the villi comes from the chorionic epithelium, while the syncytium is derived directly from the mucosa of the uterus, the epithelium of which, on the ingrowth of the chorial villi, becomes closely applied to and blends with the epithelium of the latter.

area of the placenta, however, the chorion and the serotina are far apart and the space between them is filled with the above-described chorial villi and the blood circulating between them; it is *maternal* blood that surrounds the villi on all sides and is thus brought into the closest relation with the fetal circulation.

Of especial interest is the behavior of the blood-vessels within the placenta uterina (Fig. 283 and Fig. 284). Numerous *arteries* from the muscularis of the uterus penetrate the serotina, in which they make corkscrew-like tours during the course of which they lose their muscular coat and continue as wide tubes consisting alone of the lining epithelium. Near the surface of the decidua they usually bend sharply at right angles and then open directly into the intervillous spaces of the placenta.* *Nowhere do the arteries break up into capillaries.* The *veins* (likewise epithelial tubes, though wider than the arteries) also are in direct communication with the placental spaces; they enter the decidua usually under a very narrow angle, run more or less parallel to the surface, and unite in the deeper strata in a wide venous plexus. In accordance with the description of these conditions of the vessels the arteries and the veins within the serotina can no longer be recognized by the histologic structure of their walls, but can be distinguished only by their width and their course. In addition the arteries usually are characterized by a thin, homogeneous, enveloping stratum that stains intensely with carmine, in which a few scattered nuclei are found. This peculiar layer is probably a product of the degenerated muscular coat.

The *umbilical cord* of the fetus at term consists of the umbilical vessels, two arteries and a vein, with somewhat thinner walls, which are held together

* In regard to the relation of the decidual blood-vessels to the intervillous spaces there are two conflicting theories. According to the one the intervillous spaces are independent clefts without proper walls, that are formed in the course of development between the fetal and maternal portions of the placenta, with which the blood-vessels opening on the surface of the decidua are in direct communication. Accordingly the villi of the chorion are in direct contact with the maternal blood circulating in these spaces. The opposite view regards the blood-spaces of the placenta as the enormously widened capillaries of the decidua, which, during the mutual process of intergrowth between the placenta uterina and the placenta foetalis, the developing villi of the chorion have invaginated. According to this the blood-vessel system of the decidua is closed and the arteries and the veins communicate through a system of capillary lacunæ (the intervillous spaces). Further, the chorial villi are not directly bathed in the maternal blood, but are separated from it by a thin stratum of cells, the capillary epithelium, which lies directly upon them. Recent investigations of Keibel apparently support the latter view, since in a human placenta in an early stage of development he succeeded in tracing the epithelium of the decidual blood-vessels into the intervillous spaces and demonstrating it as a continuous stratum on the surface of the chorionic villi. It is possible that in the further development of the placenta this epithelial covering undergoes regressive change, so that in later stages it cannot as a rule be demonstrated.

by *Wharton's jelly*. The latter is a mixture of gelatinous connective tissue and connective-tissue strands, usually running longitudinally, often united net-fashion. These vessels are richly provided with transverse and longitudinal smooth muscle-fibers, between which a delicate connective tissue is found, that for the greater part is united in small, perforated membranes and forms a sack-like sheath (not a sarcolemma) around each muscle-fiber, which under the influence of hardening and staining reagents gives rise to the deceptive appearance of intercellular bridges. More or less large remnants of the allantois are found in the umbilical cord, a strand about 0.1 mm. broad, formed of epithelial cells. A simple or stratified squamous epithelium, developed from the amnion, covers the surface of the umbilical cord. Smaller blood-vessels, nerves, and lymph-vessels are wanting in the matured umbilical cord, but the jelly is penetrated by a network of juice-canals (p. 100).

THE VAGINA AND THE EXTERNAL FEMALE GENITALIA.

The *vagina* is formed of a mucous membrane, a muscle membrane, and a fibrous membrane.

The *mucous membrane* is composed of (1) a stratified squamous epithelium and (2) a tunica propria beset with papillæ, built up of small, interlacing bundles of connective tissue, and containing a few elastic fibers and a varying quantity of leucocytes. The latter occasionally exist in the form of solitary nodules; in this case numerous migrating leucocytes are found arrested in the epithelium in these localities. The deepest layer of the mucosa is formed by (3) a *submucosa*, which is composed of loosely united connective-tissue bundles and robust elastic fibers. Glands are absent in the vaginal mucous membrane.

The *muscular membrane* comprises an inner circular and an outer longitudinal layer of smooth muscle-fibers.

The outer *fibrous membrane* is a dense connective-tissue structure, rich in elastic fibers.

The *blood- and lymph-vessels* are arranged in parallel horizontal networks in the submucosa and in the tunica propria. Between the bundles of the muscular membrane lies a close network of wide veins. The *nerves* form a plexus in the outer fibrous tunic, beset with many small ganglia.

The mucous membrane of the *external female genitalia* in the vicinity of the clitoris and the urethral meatus differs from the vaginal mucosa in the possession of numerous mucous glands, from 0.5 to 3 mm. in size, and on the labia minora sebaceous glands* (without hair-follicles) from 0.2 to 2 mm. in size are found. The clitoris repeats on a diminutive scale the structure of the penis; tactile corpuscles and genital nerve corpuscles occur in the glans clitoridis.

* They are wanting at birth and are not distinct until the fifth year.

The *large glands of the vestibule* (Bartholini) are the homologues of the bulbourethral glands in the male.

The labia majora possess the same structure as the external skin.

The acid vaginal mucus contains desquamated squamous epithelial cells and leucocytes, and not infrequently an infusorium, the *trichomonas vaginalis*.

TECHNIC.

No. 144.—For a *general view of the testis* make a transverse incision * through the testis and the epididymis of a newborn child; † fix both pieces in about 50 c.c. of potassium-bichromate acetic acid (p. 32) and harden in 30 c.c. of gradually strengthened alcohols (p. 35). Stain thick transverse sections of the entire organ with Hansen's hematoxylin and dilute eosin (p. 39), and mount in xylol-balsam. Examine with very low powers (Fig. 258).

No. 145.—*Minute structure of the seminiferous tubules*.—Place small pieces (2 cm. cubes) of the fresh testis of an ox in 200 c.c. of Zenker's fluid (p. 33), and harden them in 50 c.c. of gradually strengthened alcohols (p. 35). Cut the sections as thin as possible, stain them with Hansen's hematoxylin (p. 38), and mount in xylol-balsam.

No. 146.—Still better preparations are obtained by placing the entire testis of a mouse ‡ in 10 c.c. of the platinum-acetic-osmic acid mixture (p. 34) for twenty-four hours for fixation, washing it for several hours in running water, and hardening it in 20 c.c. of gradually strengthened alcohols. Mount the unstained sections in xylol-balsam (Fig. 262).

No. 147.—*Elements of the testis*.—Place pieces about 1 c.c. in size of the fresh testis of an ox in 20 c.c. of one-third alcohol (p. 20) and in five or six hours tease the content of the tubules in a drop of the same alcohol. Stain under the cover-glass with picrocarmine (p. 53) and mount in dilute glycerol. Several preparations from different parts of the organ should be completed; then not infrequently the cells of Sertoli with attached spermatocytes, or the semen filaments produced by them, will be obtained (Fig. 286, *b*), structures that formerly were described as "spermatoblasts."



FIG. 286. — ISOLATED ELEMENTS OF THE TESTIS OF AN OX. $\times 240$. *a, c*, Mother-cells; *b*, "spermatoblast;" *d*, Immature semen filament; *e*, mature semen filament.

* In the testis of the rabbit, cat, and dog the mediastinum is not at the margin, but in the interior of the organ.

† If no incision is made into the organ it does not harden sufficiently, because the dense tunica albuginea retards the penetration of the fluids.

‡ The platinum-chlorid mixture does not fully penetrate the testes of larger animals; only the peripheral portions can be used.

No. 148.—*Elements of the semen.*—Make an incision into a fresh epididymis* and place one drop of the milk-white fluid that escapes from the cut surface on a clean slide; add one drop of salt solution, apply a cover-glass, and examine with the high power. Often the semen filaments are not active; a *gentle* warming of the slide over the flame of a spirit-lamp will quickly call forth their motility. After a time let one drop of distilled water flow under the cover-glass; the movements soon cease; the heads of the majority of the semen filaments then present their broad surface and the tail curves and forms a loop (Fig. 263, 3). Remnants of protoplasm still adhere to semen filaments not fully matured. The filaments can be preserved by diluting the semen with a drop of distilled water containing ammonia (5 c.c. distilled water + 1 drop ammonia) and allowing it to dry on the slide; apply a coverglass and fasten with cement (p. 50). In such preparations too much illumination gives rise to troublesome reflexes.

No. 149.—The stability of the semen filaments permits *investigations for forensic purposes*. For example, it may be a question as to whether spots occurring on a linen garment were caused by semen. Cut strips 2 to 10 mm. square from suspected stiff spots, soak them for from five to ten minutes in a watch-glass containing distilled water, and tease a few fibers. With a high power (500 : 1) chiefly examine the edges of the isolated linen fibers, to which the semen filaments if present are attached. Not infrequently the heads are broken off; they are recognized by their peculiar luster, their shape, and their (in man small) size.

No. 150.—*Semen filaments of the frog.*—The male frog is recognized by a well-developed wart on the ball of the thumbs. Open the abdominal cavity; the testes are a pair of oval bodies (resembling the kidneys of mammals) lying to either side of the vertebral column. Divide the organ by a transverse incision; dilute a drop of the contained fluid with a drop of salt solution. The filaments are large, the head thin and elongated, the tail so delicate that at first glance it may be overlooked. Immature filaments lie grouped in tufts.

No. 151.—*Epididymis, ductus deferens, and seminal vesicle.*—Fix pieces from 1 to 2 cm. in size in about 100 c.c. of Zenker's fluid (p. 33) and harden them in 60 c.c. of gradually strengthened alcohols. Stain the sections with Hansen's hematoxylin and mount in xylol-balsam (Fig. 264-267).

No. 152.—The *prostate* and the different divisions of the *male urethra* are to be prepared in 2 or 3 cm. cubes like No. 151 (Fig. 268, 269).

No. 153.—*The ovary.*—The ovaries of small animals may be fixed in toto, those of larger animals and of man with several incisions trans-

* For a view of the spiral fiber mentioned above (p. 341, remark *), that can be seen only with immersion lenses, I recommend the seminal filaments of the rat; they are to be examined in water.

verse to the long axis, in 100 or 200 c.c. of Zenker's fluid (p. 33) and hardened in 100 c.c. of gradually strengthened alcohols (p. 35). For a topographic view (Fig. 270) it is advisable to cut thick sections, because otherwise the contents of the follicles easily fall out. Not every section includes large follicles; it is often necessary to cut many sections in order to hit a favorable place. Human ovaries have a very thick tunica albuginea (Fig. 287) and furnish less satisfactory preparations than animal ovaries. Stain the sections with Hansen's hematoxylin (p. 38). Mount in xylol-balsam.

No. 154.—Fresh *egg-cells* may be obtained as follows. Procure the fresh ovaries of a cow. The large vesicular follicles are transparent, pea-sized vesicles, which with the scissors can be easily shelled out in toto. Transfer an isolated follicle to a slide and prick it with a needle. The

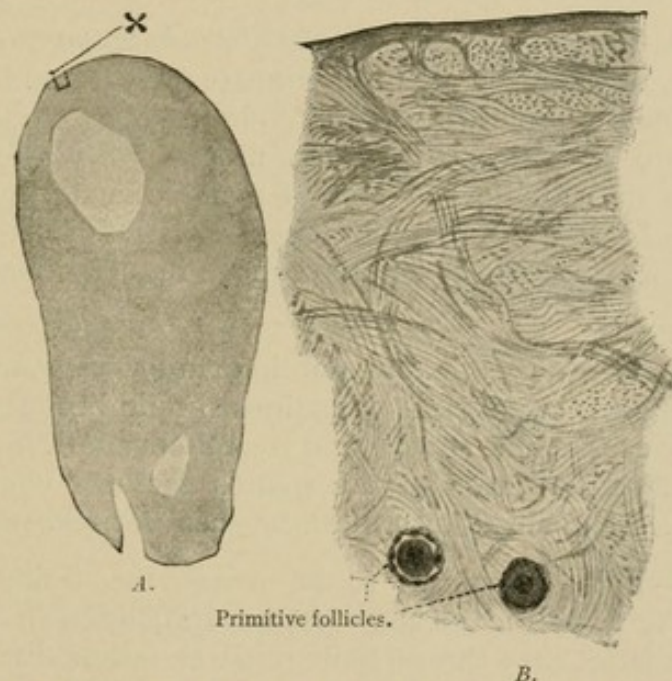


FIG. 287.—A. SECTION OF AN OVARY OF A GIRL 17 YEARS OLD. $\times 3$. X is the portion shown in B. B, tunica albuginea. $\times 120$.

needle must be carefully thrust in on the side of the follicle lying against the slide, otherwise the liquor will spurt out and carry the egg with it.

The egg, surrounded by the cells of the cumulus oophorus, escapes with the liquor folliculi and must be searched for in the uncovered preparation with a low power (Fig. 274 A). If it is desired to examine the egg with high powers place on each side of it a strip of thin paper and cover it with a cover-glass.

The beginner will sacrifice many a follicle before he succeeds in finding an egg. Often the egg does not escape when the follicle is pricked; it may then be found by teasing the follicle.

No. 155.—*Eggs of the frog*.—Place a small piece of the fresh ovary of a frog on a slide and prick all the large pigmented eggs, so that their contents escape. Place that which remains in a watch-glass with dis-

tilled water and wash by moving it to and fro with needles. Place the watch-glass on a black background; the smaller, still unpigmented follicles can then be seen. Transfer the washed object to a clean slide, apply a cover-glass, and examine it. The eggs have very large germinal vesicles; the germinal spot disappears early and usually is not to be seen. On the other hand, a dark spot occurs in the vitellus, the "nucleus of the vitellus," which corresponds to the archoplasm (p. 65). Encircling the egg is a finely striated membrane, the inner surface of which is covered with flat cells; this is the theca folliculi with the simple follicular epithelium.

No. 156.—*The oviducts*.—Fix pieces 1 or 2 cm. long in 50 c.c. of Müller's fluid and harden them in 60 c.c. of gradually strengthened alcohols (pp. 33, 35). Stain with Hansen's hematoxylin and mount in xylol-balsam. (Fig. 276.)

No. 157.—*Topographic preparations of the human uterus*.—Fix 2 cm. cubes in 100 c.c. of the Müller-formol mixture (p. 33) and harden them in 100 c.c. of gradually strengthened alcohols (p. 35). Stain in Hansen's hematoxylin and in eosin (p. 39) and mount in xylol-balsam (p. 50). (Fig. 277.) In the two-horned uteri of many animals the often greatly convoluted gland tubules can be more readily distinguished; the arrangement of the muscular strata is different, more regular than in the human organ.

No. 158.—For preparations of the human *uterine mucosa*, the fresh, living tissue obtained at surgical operations should be put into the fixing reagent. Cut pieces 1 cm. square and treat them after No. 157. Owing to the extreme tortuousness of the glands sections contain only fragments of them. The cilia can seldom be seen in fixed preparations.

No. 159.—The *placenta* is to be treated according to No. 158.* Before cutting sections the pieces must be embedded in celloidin or in paraffin; in the latter case the sections must be fastened to the slide (see Microtome Technic), in order that the innumerable branches of the villi, cut in every plane, do not fall out. The study of preparations of this kind is one of the most difficult tasks of the microscopist.

No. 160.—The *umbilical cord*. Prepare like No. 4, p. 101.

* Fixation in absolute alcohol often yields very good results.

IX. THE SKIN.

The external skin (*integumentum commune, cutis*) in its chief mass consists of connective tissue, which however is nowhere exposed, but is protected by a continuous epithelial cover. The connective-tissue portion of the skin is called *corium* or *derma*, the epithelial portion, *epidermis*. The appendages of the skin, the *nails* and the *hairs*, as well as the *glands* and the *hair-follicles* embedded within the depths of the corium, are products of the epidermis.

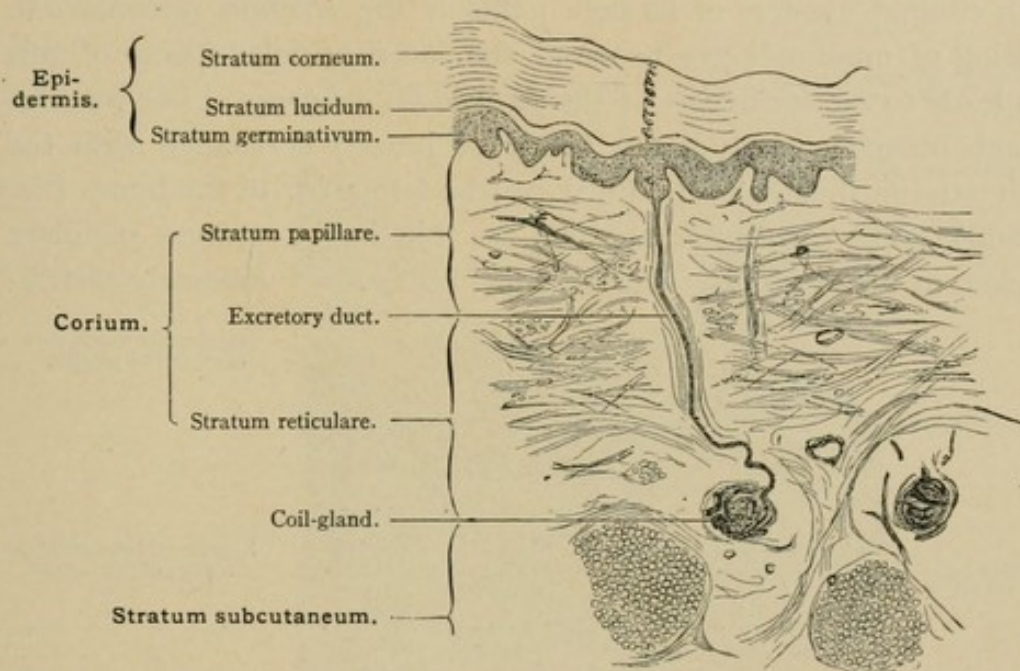


FIG. 288.—VERTICAL SECTION OF THE SKIN OF THE FINGER OF ADULT MAN. $\times 25$. With this magnification the stratum granulosum is not visible. Technic No. 161.

THE EXTERNAL SKIN.

The corium.—The surface of the *corium* is marked by many fine furrows, which intersect and bound rectangular or lozenge-shaped fields or run parallel for longer stretches between minute ridges. The lozenge-shaped fields may be seen on the surface of the greater part of the body, while the ridges are confined to the volar surface of the hand and the plantar surface of the foot. These fields and ridges are beset with numerous conical elevations, the *papillæ*, the number and size of which vary greatly in different regions of the body. The largest (up to 0.2 mm. high) and most numerous papillæ occur on the palm of the hand and on the sole of the foot; they are very slightly developed in the skin of the face.

The corium chiefly consists of netlike interlacing connective-tissue

bundles, mingled with elastic fibers, cells, and smooth muscle-fibers. In the superficial layers of the corium the *connective-tissue bundles* are delicate and are united in a closely interwoven texture; in the deeper layers they are larger and, intersecting at acute angles, form a coarse-meshed network. These differences have led to the recognition of two strata in the corium, a superficial stratum beset with papillæ, the *stratum papillare*, and a deep stratum, the *stratum reticulare*. There is no sharp demarcation between the two strata, the one gradually blending with the other (Fig. 288). The stratum reticulare is connected with an underlying network of loosely united bundles of connective tissue, the wide meshes of which contain clusters of fat-cells; this is the *stratum subcutaneum*. The storing of much adipose tissue in the interfascicular spaces of this stratum leads to the formation of the *panniculus adiposus*. The bundles of the subcutaneous stratum are firmly or loosely connected with the connective-tissue sheaths of the muscles (the *fasciæ*) or of the bones (the periosteum). The *elastic fibers*, which are thin in the stratum papillare and thicker in the stratum reticulare, form networks* uniformly distrib-

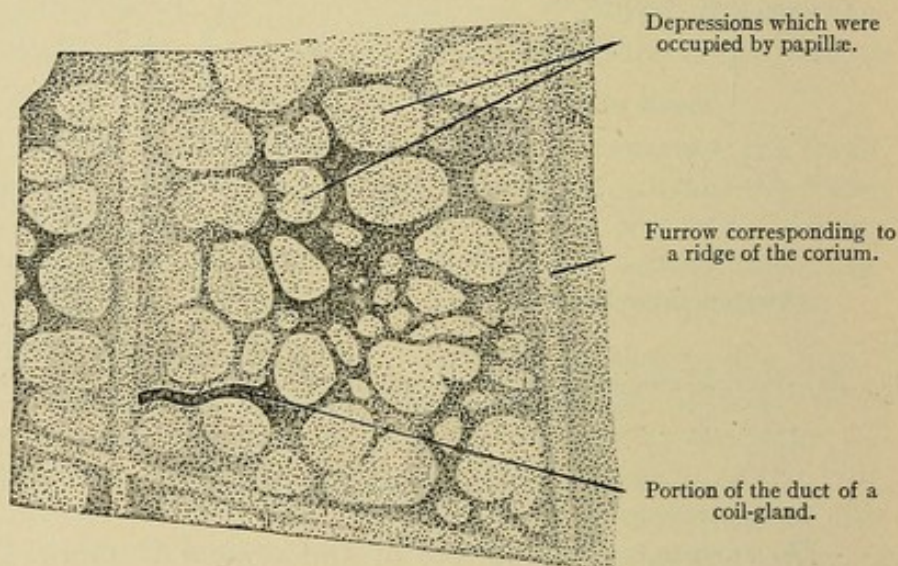


FIG. 289.—EPIDERMIS FROM THE SKIN OF THE DORSUM OF THE HUMAN FOOT, SEEN FROM THE LOWER SURFACE $\times 120$. The preparation is so to speak the cast, while the surface of the corium beset with papillæ represents the matrix. Technic No. 162.

uted throughout the corium. The easily shifted membrane of subcutaneous tissue is relatively poor in elastic fibers; particularly rich in these is the skin of the face and of the vicinity of the joints. The *cells* include spindle-shaped and plate-like connective-tissue elements, leuco-

* Recent authors distinguish four layers of elastic fibers: (1) a layer consisting of numerous thick elastic fibers, that lies immediately above the common fascia of the body; (2) a zone in the stratum reticulare, here the fibers run with the blood-vessels; (3) a dense sub-papillary plexus and (4) a sub-epithelial network. The first three layers are freely connected. In old age a notable disappearance of elastic fibers occurs.

cytes, and fat-cells. The number of the cellular elements is extremely variable. The *muscle-fibers* almost exclusively belong to the nonstriped variety and the majority are attached to the hair-follicles (p. 379); only in a few situations in the body do they occur as membranous expansions in the skin (tunica dartos,* nipple). Striated muscle-fibers occur in the skin of the face, where they radiate from the mimetic muscles.

The epidermis.—The *epidermis* consists of a stratified squamous epithelium, in which at least two sharply defined zones can be distinguished: a deep zone, the germinal stratum, *stratum germinativum* (Malpighi), which fills the depressions occurring between the papillæ of

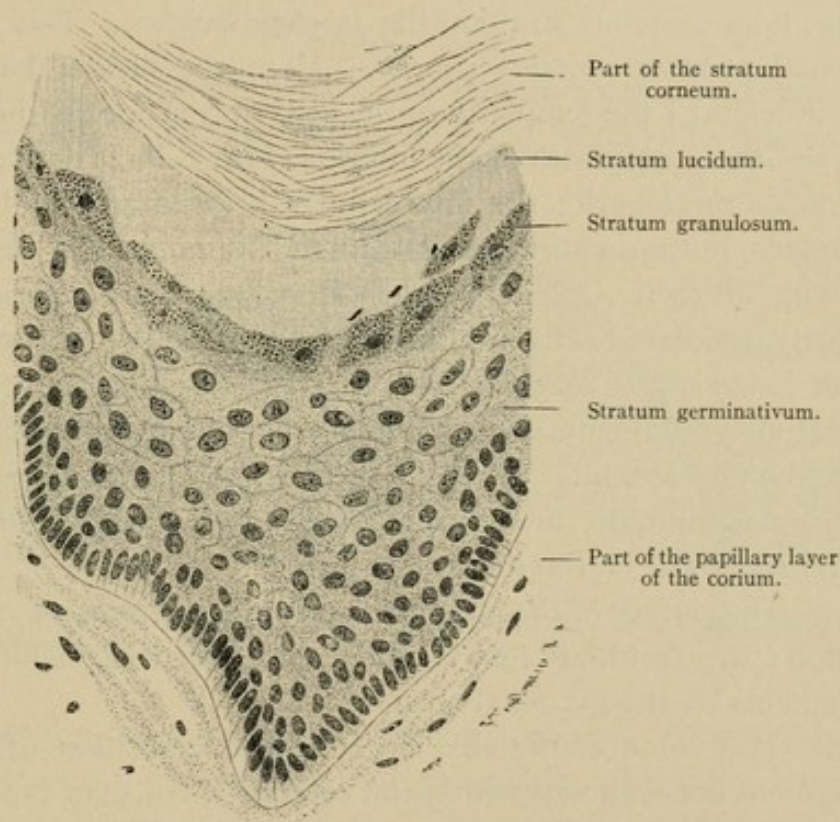


FIG. 290.—FROM A SECTION THROUGH THE SKIN OF THE SOLE OF THE FOOT OF ADULT MAN. $\times 360$. Technic No. 161.

the corium, and a superficial, firmer zone, the horny stratum, *stratum corneum*. Both strata consist exclusively of epithelial cells, which exhibit different appearances in the separate layers. In the deepest layer of the *stratum germinativum* the cells are membraneless, cylindrical, and possess oblong nuclei; these are followed by several layers of spherical cells that are beset with numerous delicate thorns and are named prickle-cells. The thorns are delicate, thread-like processes, which serve to connect

* Here the muscle-fibers are profusely interlaced with elastic fibers.

neighboring cells to one another. Therefore they are called intercellular bridges (Fig. 23, p. 79). In the stratum germinativum new cells are continually being formed by indirect nuclear division. The *stratum corneum* is not everywhere of the same structure and two different types may be distinguished: (1) In localities where the epidermis is thick, as on the palm of the hand and the sole of the foot, a stratum of cells characterized by highly refracting granules* (keratohyaline granules) lies next to the stratum germinativum. The granules are a decomposition product of the interfilar mass and the stratum is named the *stratum granulosum* (Fig. 290). These granules liquefy and form a diffuse mass, the *eleidin*, that saturates the cells and produces a glistening, homogeneous stratum, the *stratum lucidum*. This is covered by the broad *stratum corneum* proper. Here the eleidin has become firmer (pareleidin),† the exoplasm of the cells is transformed into a horny membrane, the protoplasm in the interior of the cells is desiccated and forms a delicate meshwork; the intercellular bridges are no longer connecting threads, but are short, small teeth. The nucleus is desiccated; but the cavity which it occupied persists for a long time. These partly cornified, partly desiccated cells are only slightly flattened. (2) In situations where the *epidermis is thinner* (all the remaining surface of the skin), the stratum granulosum is narrow and interrupted by gaps. The stratum lucidum is absent. The cells of the stratum corneum, enveloped in a horny membrane, are extremely flattened and are united in lamellæ. The last trace of the nucleus is lost.

The surface of the stratum corneum is subject to a constant desquamation; the resulting loss is compensated by the pushing upward of the elements of the germinal stratum.

The color of the skin is due to the deposition of fine granules of pigment between and within the cells of the deeper layers of the epidermis; only in certain localities, for example, in the vicinity of the anus, are pigmented connective-tissue cells found in the adjacent corium.

With regard to the source of the pigment of the epidermis there are two theories, of which the one attributes its origin to the connective tissue, the other to the epithelium. According to the first, hitherto widely accepted opinion, the so-called "transportation" theory, the

* These granules dissolve in a solution of potassium hydroxid, therefore do not consist of keratin, which is insoluble in this reagent.

† The pareleidin blackens like fat, but only after the prolonged action of osmic acid; therefore the black staining of the horny cells of thick epidermis cannot be attributed to the saturation of the stratum corneum with fat from the exterior, derived from the secretion of the sebaceous or coil-glands.

pigment is carried to the epithelium by pigmented connective-tissue cells, that wander from the corium into the epidermis and there disintegrate. In the human hair-bulb pigmented forms presenting great diversity in outline are actually found between the epithelial elements; some of these figures are cells, but it has not been demonstrated with certainty that they are connective-tissue cells, others are not cells, but intercellular clefts filled with pigment. The second theory is supported by the developmental history, which teaches that the pigment originates in the epithelium of the hair without the intervention of connective-tissue cells. The pigment of the retina also is certainly and exclusively of epithelial origin.

THE NAILS.

The nails are horny laminæ, which rest upon a special modification of the skin, the *nail-bed*. The nail-bed is bounded laterally by the *nail-*

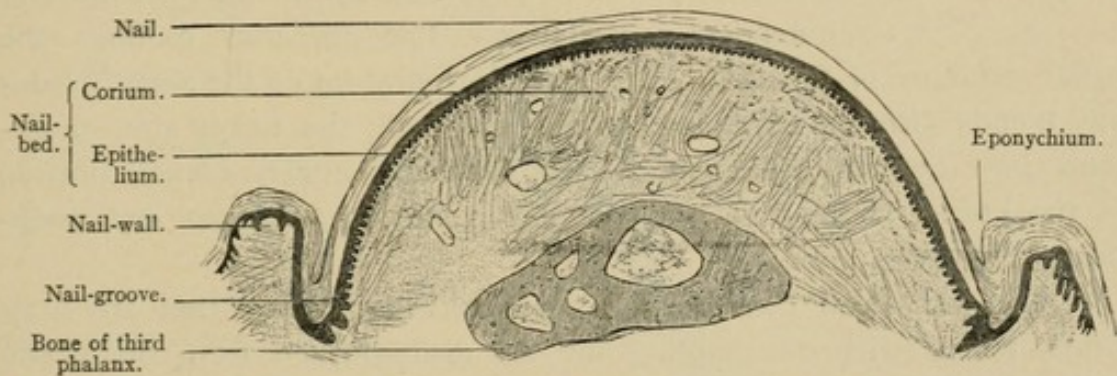


FIG. 291.—DORSAL HALF OF A CROSS-SECTION OF THE THIRD PHALANX OF A CHILD. $\times 15$. The ridges of the nail-bed in cross-section appear like papillæ. Technic No. 163.

walls, a pair of sloping folds with the descent forward. The nail-bed and nail-wall embrace a furrow, the *nail-groove*, in which the lateral border of the nail is inserted (Fig. 291). The posterior border of the nail, the *nail-root*, rests in a similar but deeper groove; here the principal growth of the nail takes place; this place is named the *matrix*.* The anterior free border of the nail projects over the *nail-ridge*, a small, seam-like prominence at the distal end of the nail-bed.

The *nail-bed* consists of corium and of epithelium. The connective-tissue bundles of the corium contain many elastic fibers and are partly disposed parallel to the long axis of the finger, partly run vertically from the periosteum of the phalanx to the surface. The surface of the corium does not possess papillæ, but minute longitudinal ridges. They begin

* Other authors name the whole nail-bed matrix, which is in a measure justified by the growth in the thickness of the nail that occurs here.

low at the matrix, increase in height toward the anterior border of the nail and terminate abruptly at the point where the latter leaves its bed. The epithelium is of the stratified squamous variety, of the same structure as that of the germinal stratum of the epidermis. It covers the ridges of the nail-bed, fills up the furrows between them, and is sharply defined from the substance of the nail. The *matrix* likewise consists of corium and of epithelium; the corium is distinguished by its tall papillæ, the

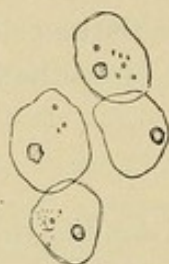


FIG. 292.—ELEMENTS OF
A HUMAN NAIL. X
240. Technic No.
164.

stratified squamous epithelium is very thick and is not sharply defined from the nail-substance, but gradually blends with it. This is the place where by continual division of the epithelial cells the material for the growth of the nail is furnished. On this account the epithelium is here called the *germ-layer* of the nail. The extent of the matrix is indicated by the *lunula*, a white, anteriorly convex field, visible to the unaided eye; it is produced by the thick, uniformly extended germ-layer. The *nail-wall* exhibits the

usual structure of the skin. The germinal stratum of the same gradually blends with the epithelium of the nail-bed; the horny stratum extends into the nail-groove and as "eponychium" covers a small portion of the border of the nail, but soon diminishes in thickness and disappears (Fig. 291).

The *nail* itself consists of horny epithelial scales, that are very firmly united with one another and are distinguished from the horny cells of the stratum corneum of the epidermis by the possession of a nucleus (Fig. 292).*

THE HAIRS AND THE HAIR-FOLLICLES.

The hairs are flexible, elastic, horny threads, which are distributed over nearly the entire surface of the body and on the integument of the cranium are united in small groups. The part of the hair which projects beyond the free surface of the skin is called the *shaft* (*scapus*); the portion obliquely embedded within the skin is named the *root* (*radix pili*); this at its lower extremity is expanded to a hollow knob, the *hair-bulb* (*bulbus pili*), which is occupied by a formation of the corium, the *hair-papilla* (Fig. 293).

Each hair-root is inserted in the *hair-follicle*, a modification of the

* The new anatomic nomenclature reckons the epithelium of the nail-bed to the nail, that according to this representation consists of two layers, the stratum corneum and the stratum germinativum.

skin in the construction of which the corium and the epidermis participate; the parts furnished by the latter are named the *epithelial root-sheaths*; the portion originating from the corium is named the *dermal* or *connective-tissue hair-follicle*. From two to five glands, the *sebaceous glands*, open laterally into the upper part of the follicle. Bundles of smooth muscle-fibers, the *arrectores pilorum*, provided with elastic tendons at the point of origin, pass obliquely down from the upper surface of the corium and attach themselves beneath a sebaceous gland to the connective-tissue hair-follicle; the point of insertion of these fibers is always on the side toward which the hair inclines and forms an acute angle with the free surface of the skin; consequently when they contract the follicle

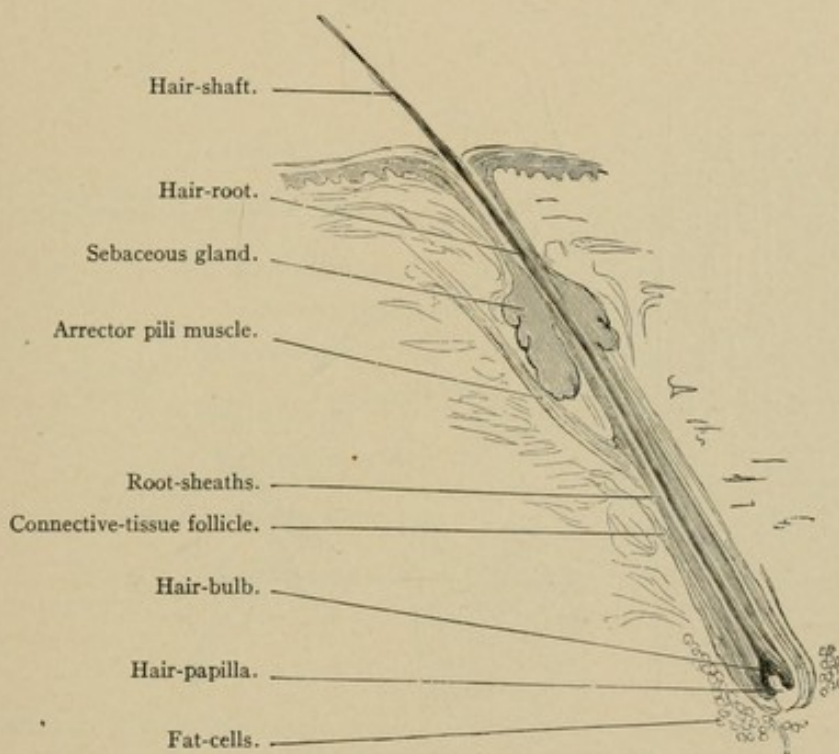


FIG. 293.—FROM A THICK SECTION OF THE HUMAN SCALP. $\times 20$. Technic No. 168.

and the shaft become erect. The arrector muscles are wanting on the lanugo hairs of the nose, the cheeks, the lips, also on the cilia and the vibrissæ.

The hair consists entirely of epithelial cells, which are arranged in three well-defined strata: (1) the *cuticle*, which covers the surface of the hair; (2) the *cortical substance*, which forms the chief bulk of the hair; (3) the *medulla*, which occupies the axis of the hair.

The *cuticle* (*cuticula pili*) consists of transparent, imbricated scales: horny, nonnucleated epithelial cells.

The *cortical substance* of the shaft consists of slender, horny epithelial cells provided with thin, linear-shaped nuclei, which are very

intimately united with one another; on the root the cells become softer and rounder, their nucleus correspondingly more spherical, the nearer they lie to the hair-bulb.

The *medulla* is absent in many hairs; when it is present (in the thicker hairs) it does not extend through the entire length of the hair. It consists of cubical epithelial cells containing keratohyaline granules (p. 375), which enclose a rudimentary nucleus and are usually disposed in twofold rows beside one another.

The colored hairs contain pigment, in solution and in the form of granules, which partly occur between and partly within the cells of the

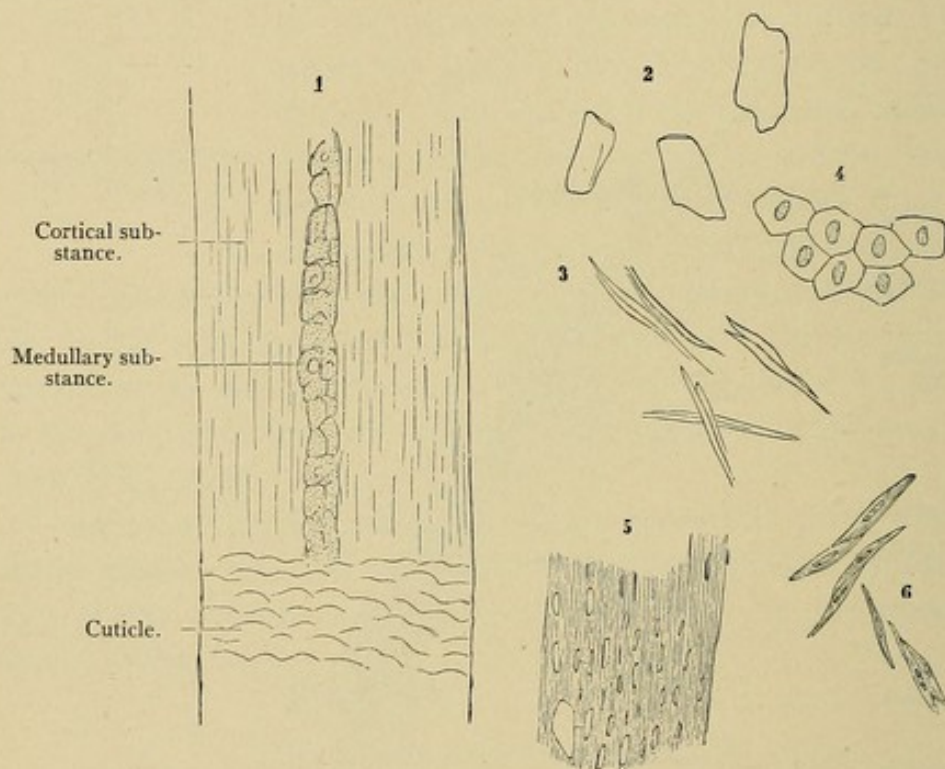


FIG. 294.—ELEMENTS OF A HUMAN HAIR AND HAIR-FOLLICLE. $\times 240$. 1. White hair; 2, scales of the cuticle; 3, cells of the cortical substance of the shaft; 4, cells of Huxley's layer; 5, cells of Henle's layer, having the appearance of a fenestrated membrane; 6, cells of the cortical substance of the root. Technics No. 166 and No. 167.

cortical substance.* In every hair which has attained its full development extremely minute *air vesicles* occur; they are found in the cortical substance as well as in the medulla, in the intercellular spaces.

The *follicle* of finer (lanugo) hairs is formed alone by the epidermal root-sheaths, but in coarser hairs the corium participates in its construction. In the follicles of the latter the following strata are distinguished: an outermost *longitudinal fiber-layer*,† formed of vascular, loose connective-

* Regarding the source of the pigment, see page 376.

† Elastic fibers occur only in the longitudinal fiber-layer; in the ring fiber-layer and in the papilla they are wanting.

tissue bundles, richly supplied with nerves ; following this is a thicker layer of circularly arranged, delicate connective-tissue bundles, the *ring fiber layer*, which is contiguous to an inner clear, transparent membrane, the *hyaline membrane*. These three strata are derived from the corium * and together are named the *connective-tissue hair-follicle* ; it is to be seen in complete development only in the lower half of the entire hair-follicle. Within the hyaline membrane lies the *outer root-sheath*, which as the continuation of the germ-layer of the epidermis consists of stratified squamous epithelium ; inward to this lie continuations of the stratum granulosum and stratum corneum, which latter extends to the point where the ducts of the sebaceous glands open into the follicle, while the former extends a little

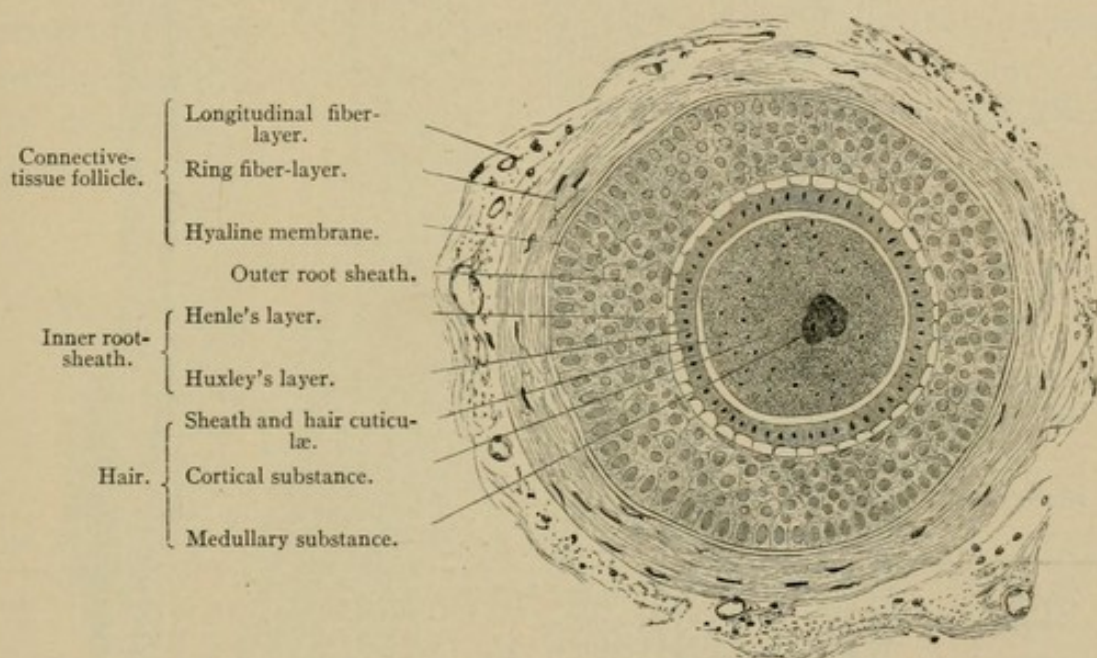


FIG. 295.—FROM A HORIZONTAL SECTION OF THE HUMAN SCALP. $\times 240$. Cross-section of a hair and hair-follicle in the lower half of the root. Technic No. 168.

farther downward ; immediately beneath (toward the papilla) the *inner root-sheath* begins *abruptly*, which in the lower portion of the follicle is differentiated into two sharply defined layers. The outer of these two, *Henle's layer*, consists of a single or double row of nonnucleated epithelial cells (here and there an atrophic nucleus is present), while the inner, *Huxley's layer*, is formed of a simple stratum of nucleated cells. The inner surface of this layer is lined with a delicate membrane, the *cuticle of the root-sheath*, which exhibits the same structure as the cuticle of the hair. Toward the base of the follicle the outer root-sheath diminishes in thickness and at the neck of the hair-papilla it disappears ; there its elements

* The inner portion of the hyaline membrane is said to be of epithelial origin.

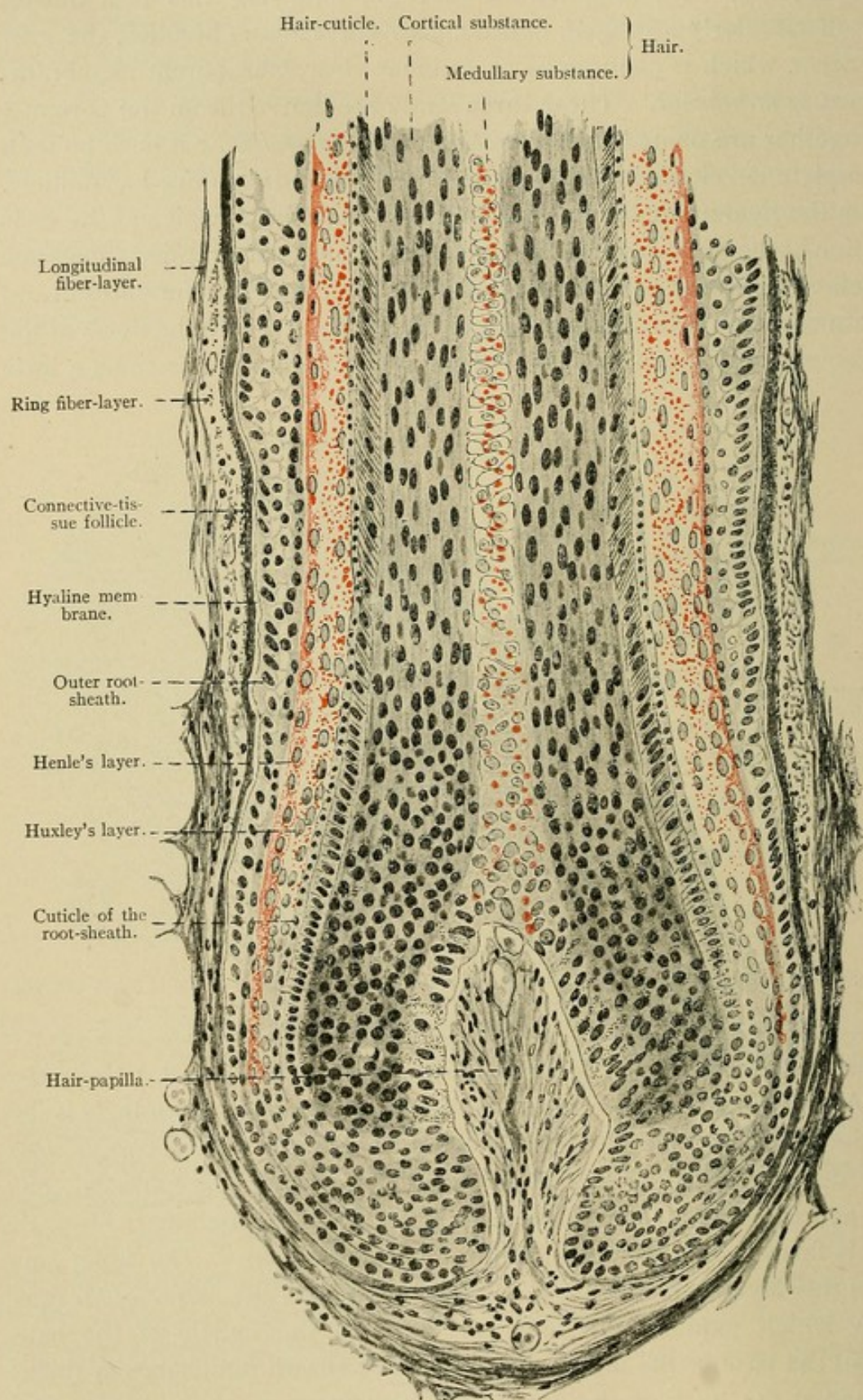


FIG. 296.—LONGITUDINAL SECTION OF THE LOWEST DIVISION OF THE ROOT OF A HAIR; the keratohyaline granules are colored red. From a vertical section of the human scalp. $\times 200$. Technic No. 168.

are drawn sharply into a transverse position and in tangential longitudinal sections of the root-sheath have the appearance of short, circular smooth muscle-fibers. The elements of the inner root-sheath and of the cuticulæ all become nucleated cells, that can be distinguished as separate layers until near the neck of the papilla; there they lose their sharp demarcation, but nevertheless can be distinguished from the cells of the hair-bulb by the pigmentation of the latter.* The hair-papilla is covered with a thin, distinctly double-contoured continuation of the hyaline membrane.

THE DEVELOPMENT OF THE HAIRS.

The first anlage of the hair appears at the end of the third embryonal month and at first in the form of a local thickening of the epidermis, which is caused by elongation of the deepest cells of the epidermis and by multiplication of the cells of the middle layers (Fig. 297). This *hair-germ* (Fig. 298) grows in length, down into the corium, and becomes a

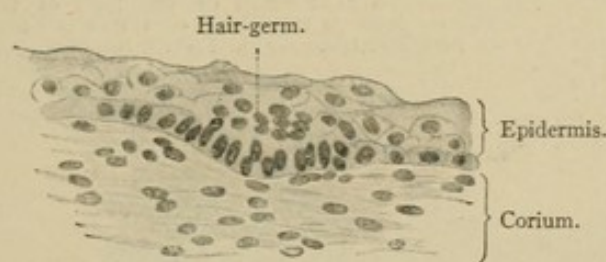


FIG. 297.—VERTICAL SECTION OF THE SKIN OF THE ABDOMEN OF A HUMAN FETUS OF FIVE MONTHS. $\times 230$. Technic No. 169.

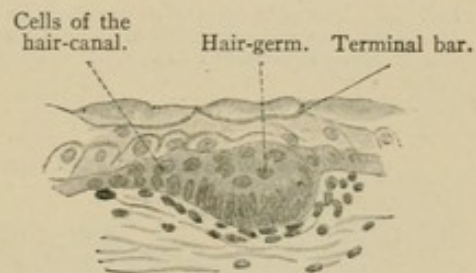


FIG. 298.—VERTICAL SECTION OF THE SKIN OF THE BACK OF A HUMAN FETUS OF FIVE MONTHS. $\times 230$. Technic No. 169.

solid epithelial peg, the *hair-peg*, at the blind end of which a thicker aggregation of connective-tissue cells, the anlage of the *hair-papilla*, has developed (Fig. 299); a second accumulation of cellular elements of the corium, appearing at the under side of the hair-peg, is the anlage of the arrector muscle (Fig. 300). The lower end of the hair-peg grows around the papilla, converting the entire anlage into the *bulb-peg*, which develops two evaginations, an upper, the future hair-follicle gland, and a lower, the future hair-matrix (Fig. 301).

The epithelial cells of the bulb-peg lying next to the papilla develop into the *hair-cone* (Fig. 302), while the remaining epithelial cells develop into the outer root-sheath. The hair-cone grows in length, its periph-

* Already at the level of the papilla keratohyaline granules appear in the cells of Henle's layer, at a somewhat higher level also in those of Huxley's layer (Fig. 296), that a little farther up disappear; from this upward the elements of the inner root-sheath are corneous.

eral cells become the inner root-sheath (Fig. 303), its axial cells become the hair, while the epithelial cells lying between these produce the cuticulæ of the sheath and the hair. In this stage the entire hair,

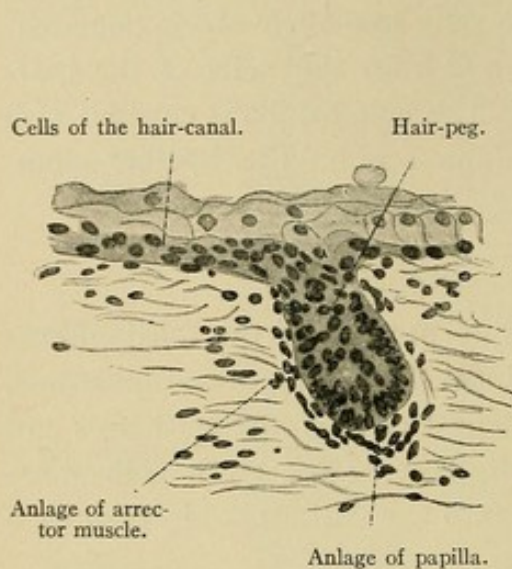


FIG. 299.—VERTICAL SECTION OF THE SKIN OF THE BACK OF A HUMAN FETUS OF FIVE MONTHS. $\times 230$. Technic No. 169.

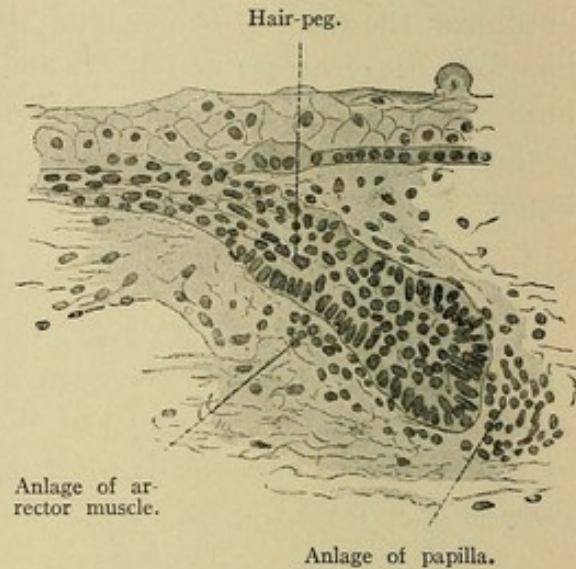


FIG. 300.—VERTICAL SECTION OF THE SKIN OF THE FLEXOR SIDE OF THE THIGH OF A HUMAN FETUS OF FIVE MONTHS. $\times 230$. Technic No. 169.

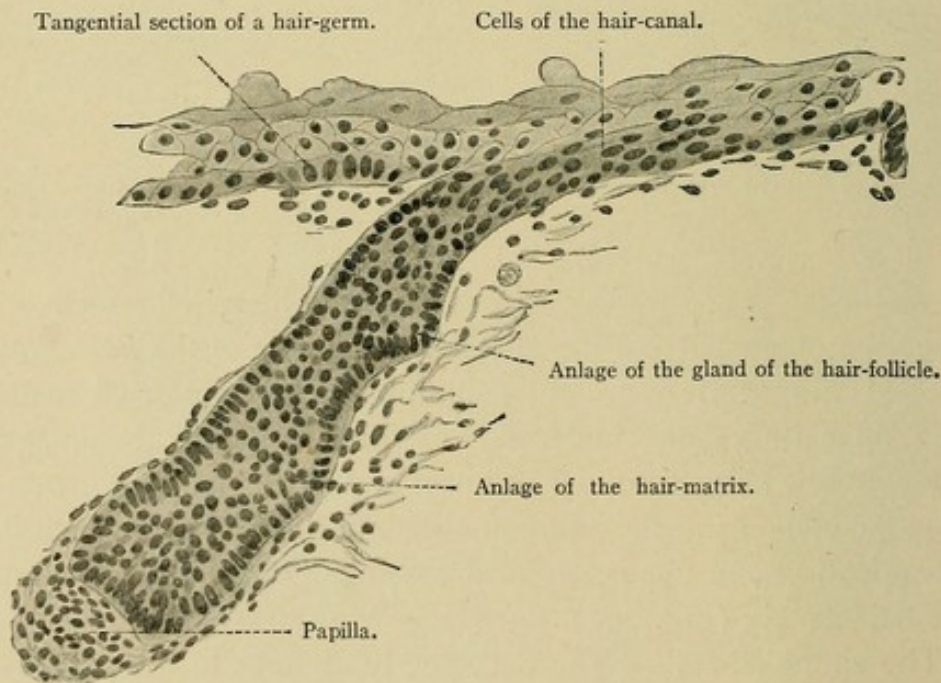


FIG. 301.—VERTICAL SECTION OF THE SKIN OF THE GLUTEAL REGION OF A HUMAN FETUS OF FIVE MONTHS. TRANSITION OF THE HAIR-PEG INTO THE BULB-PEG. $\times 230$. Technic No. 169.

including its tip, is completely enclosed in the inner root-sheath, gradually becoming horny from above downwards. This is the stage of the *sheathed hair* (Fig. 304).

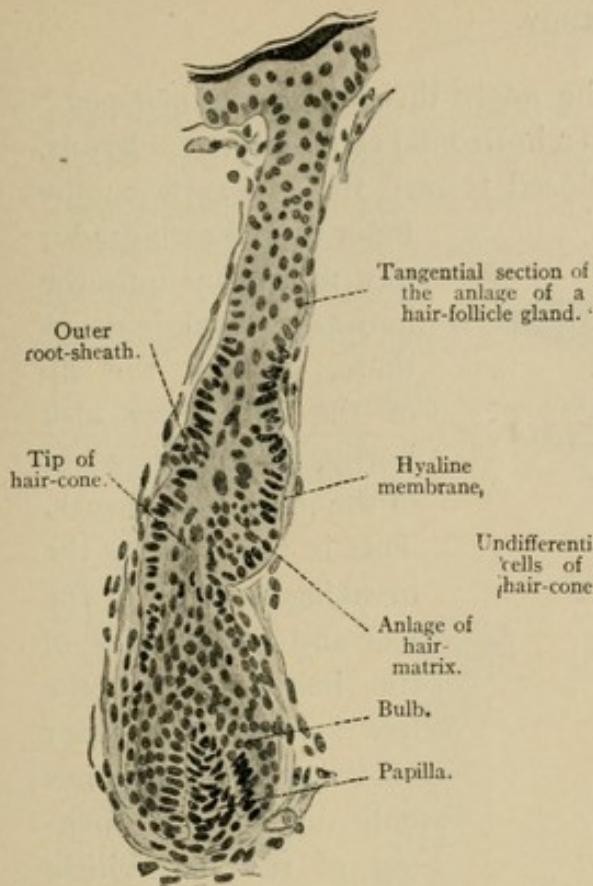


FIG. 302.—BULB-PEG. FROM A VERTICAL SECTION OF THE BRIDGE OF THE NOSE OF A HUMAN FETUS OF SEVEN AND A HALF MONTHS. $\times 230$. Technic No. 169.

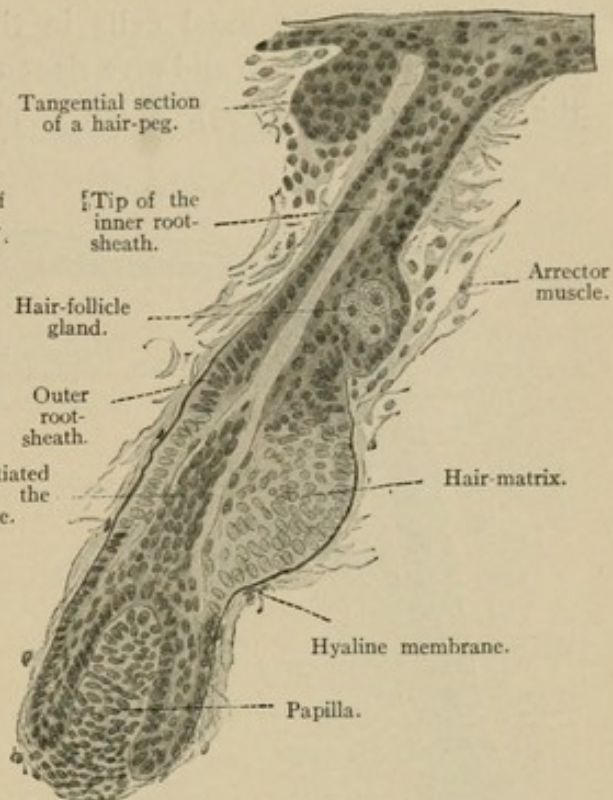


FIG. 303.—SHEATHED HAIR. VERTICAL SECTION OF THE SKIN OF THE BACK OF A HUMAN FETUS OF FIVE AND A HALF MONTHS. $\times 230$. Technic No. 169.

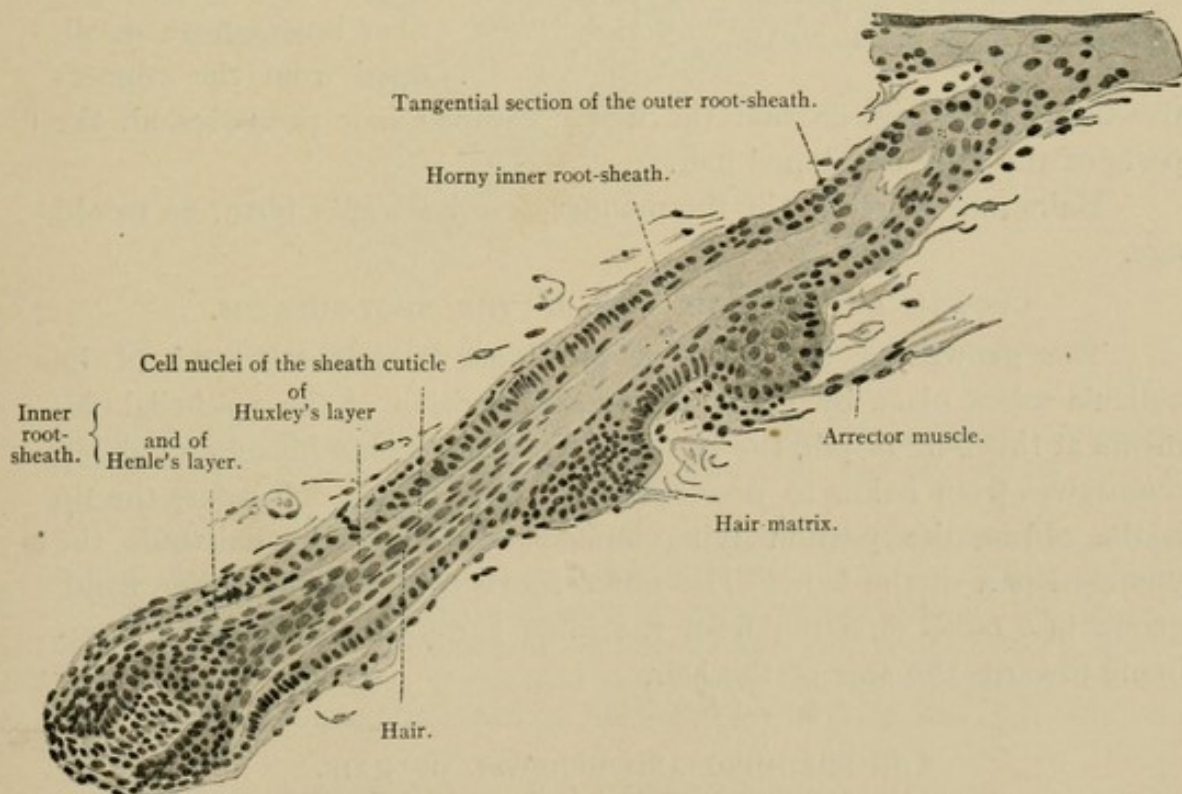


FIG. 304.—VERTICAL SECTION OF THE SKIN OF THE FOREHEAD OF A HUMAN FETUS OF FIVE MONTHS. $\times 230$. Differentiation of the sheaths of the hair. Above the point of the tangential section of the outer root-sheath the degenerating end of the inner root-sheath is seen projecting into the small portion of the hair-canal included in the section. Technic No. 169.

Meanwhile the axial cells in the *upper* division of the hair-peg* grow horny and perish and give rise to a horizontal canal in the epidermis, the *hair-canal* (Fig. 305), which is closed toward the free surface; the

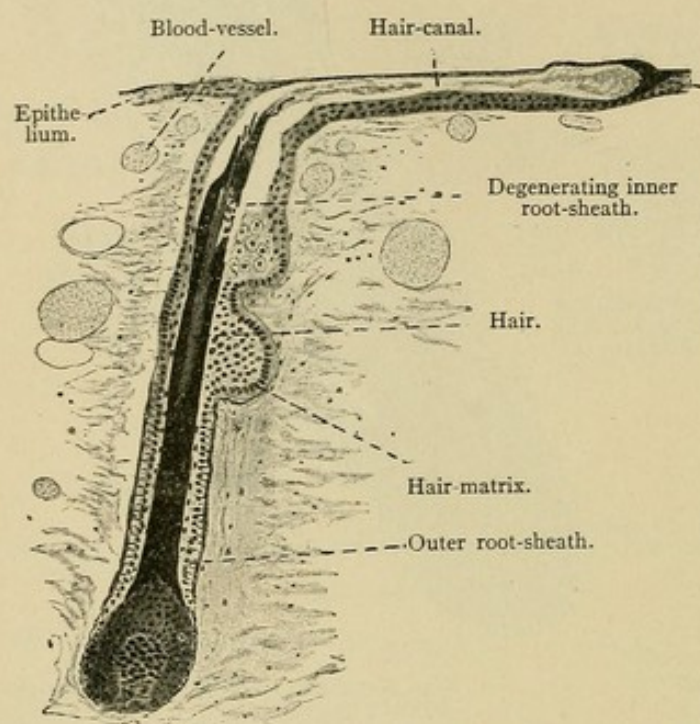


FIG. 305.—VERTICAL SECTION OF THE SKIN OF THE BACK OF A HUMAN FETUS OF FIVE AND A HALF MONTHS. $\times 120$. The staining with iron-hematoxylin has made the horny parts so black that their details are invisible. Technic No. 169.

inner root-sheath gradually moves up into the hair-canal and perishes there, so that the tip of the hair, now also horny, projects free out of the inner root-sheath. This is followed by the breaking through of the hair and the opening of the hair-canal on the free surface; the inner root-sheath now reaches only up to the opening of the hair-follicle glands. Meanwhile the hyaline membrane, the ring and longitudinal fiber layers have developed from the connec-

tive-tissue bulb-peg, so that the newly escaped hair possesses all the parts of the fully developed hair.

Hairs may originate in the manner described after birth, up to old age.

GROWTH OF THE HAIR AND OF THE ROOT-SHEATHS.

The growth, of the hair, of the inner root-sheath, and of its cuticula takes place by continual mitotic division of the epithelial elements at the bulbus pili, the *matrix cells*, that become horny and annex themselves from below to previously cornified cells. Therefore the tip is the oldest, the portion lying immediately above the hair-bulb the youngest part of the hair. The outer root-sheath, on the other hand, grows in a radial direction from the inner surface of the hyaline membrane towards the axis of the hair.

THE SHEDDING AND RENEWAL OF HAIR.

Shortly before and after birth a total exchange of hair takes place

* The differentiation of these cells of the hair-canal begins very early (*cf.* Fig. 298).

and also in adult man a constant, but not periodic renewal occurs, to replace the shed hairs of the head and beard.* In this process the hollow hair-bulb ("hollow root") is transformed into a solid, horny bulb ("solid root"), that is lifted from the papilla and crowded up under the opening of the sebaceous glands and then falls out; meanwhile a new hair originates on the papilla.

The minuter processes are as follows: the ring fiber-layer and the hyaline membrane grow thicker and the matrix cells cease to divide, those of the inner root-sheath first, while those of the hair itself continue to produce for a time

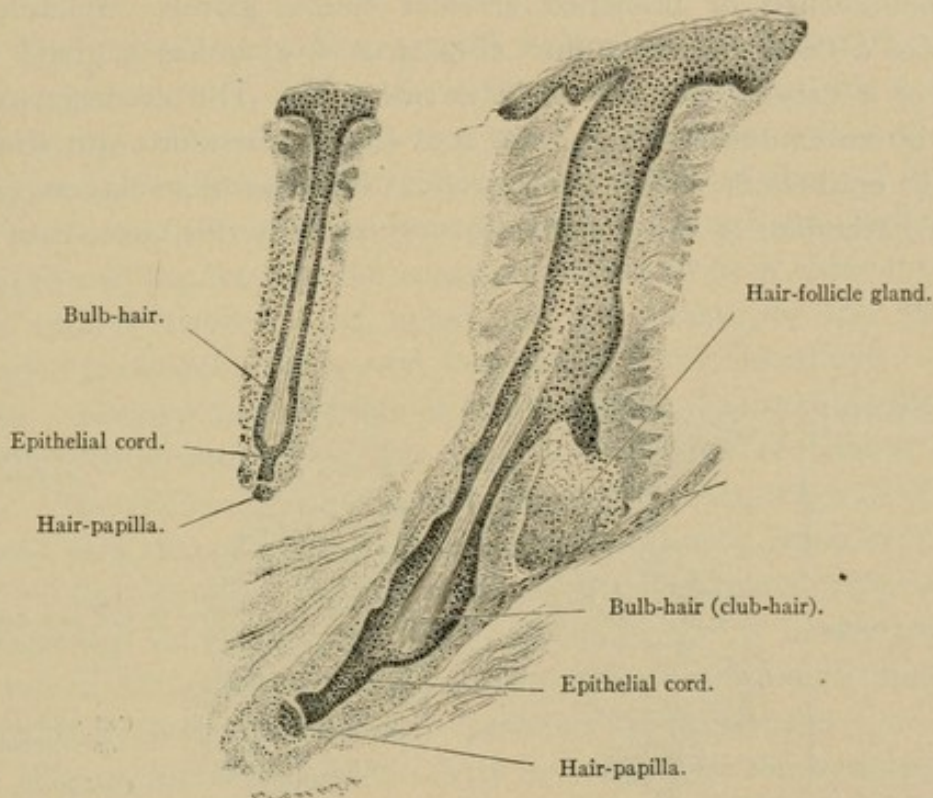


FIG. 306.—FROM A VERTICAL SECTION OF THE SCALP OF ADULT MAN. $\times 40$. Technic No. 170.

and then also cease their activity. In this way the lower end of the hair becomes transformed to a *bulb*, which gradually removes from the hair-papilla, that is still covered with a few matrix cells.

These matrix cells multiply, however, without becoming elements of the hair or sheaths, grow larger and with the aid of the thickened ring fiber-layer and the now multiplying epithelial cells of the outer root-sheath push the hair up to the level of the attachment of the arrector muscle. At this locality, of the *hair-matrix* ("bulb site"), the bulb-hair † (club-hair), having meanwhile become completely horny, remains for a time, ceases to grow, and later falls out.

* The duration of life of a scalp hair is said to be 1600 days. Regarding the shedding and renewal of the other hairs definite knowledge is wanting.

† With the ascent of the bulb-hair the inner root-sheath, that extends only up to the opening of the sebaceous glands, for the greater part is lost.

The small epithelial cord situated under the hair-matrix becomes greatly shortened and pulls up the now atrophic hair-papilla, altered in shape, while the strata of the connective-tissue hair-follicle remain and form the *hair-stalk*. After a time the elements of the epithelial cord are regenerated from the cylinder-cell layer of the hair matrix; the cord extends down to the old papilla; new matrix cells produce a young hair, in the mode previously described for the first development of hairs, that gradually moves obliquely into the original depth, and with its tip next to the bulb-hair, which later falls out, it pushes upward to the surface.

THE GLANDS OF THE SKIN.

The *hair-follicle glands* (sebaceous glands, glandulæ sebaceæ) are either unbranched or branched alveolar simple glands. Each gland consists of a short excretory duct (Fig. 307 *A*, *a*) and of a gland body formed of a varying number of little sacks (*t*). The *excretory duct* is clothed by an extension of the outer root-sheath, therefore with stratified squamous epithelium, which by a gradual decrease of its layers passes into the epithelium of the *gland body*. Externally this consists of low,

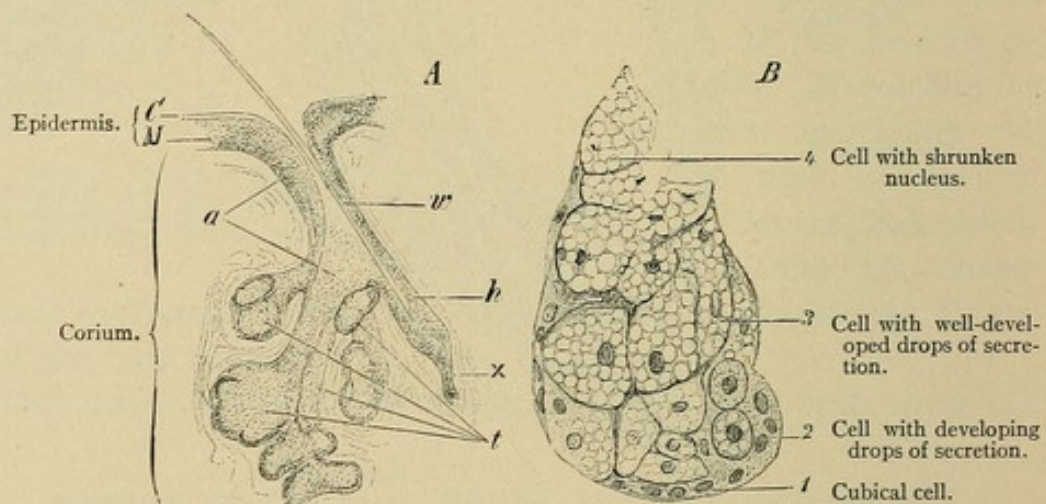


FIG. 307.—*A*. FROM A VERTICAL SECTION THROUGH THE ALA NASI OF A CHILD. $\times 40$. *C*, Stratum corneum; *M*, stratum germinativum; *t*, sebaceous gland consisting of four sacks, *a*, duct of the same; *w*, lanugo hair, about to be shed, *h*, hair-follicle of the same, at the base of which a new hair, *x*, is forming.
B. FROM A VERTICAL SECTION OF THE SKIN OF THE ALA NASI OF AN INFANT. $\times 240$. Sack of a sebaceous gland containing gland-cells in various stages of secretion. Technic No. 171.

cubical cells (*B*); within these lie spherical or polygonal cells, varying in size, which fill the entire gland-sack and exhibit all the transitional phases in the process by which the cell is converted into the secretory product of the gland. The secretion, the *sebum*, during life is a semi-fluid substance, that consists of fat and disintegrated cells. While the sebaceous glands occur as appendages of the hair-follicles of the coarser hairs (Fig. 293), in the case of the lanugo hairs reversed relations prevail, since the follicles of the latter appear as the appendages of the powerfully developed sebaceous glands (Fig. 307 *A*). The sebaceous

glands are distributed with the hairs over the entire body and are wanting only where they are absent, on the palm of the hand and on the sole of the foot. There also are sebaceous glands that are not associated with hair-follicles; for example, on the red edge of the lips, on the labia minora, on the glans, on the præputium penis; in the latter situation they are known as *glandulæ præputiales*.* The sebaceous glands are always situated in the superficial layers of the corium, in the stratum papillare. Their size varies from 0.2 to 2.2 mm.; the latter are found in the skin of the nose, where their excretory ducts are visible to the unaided eye.

The *coil-glands* (sweat glands, *glandulæ sudoriparæ*) are long, unbranched tubules, that at their lower end are rolled into a spherical coil, having a diameter of 0.3 to 7 mm. (of the latter size in the axilla). Two parts are distinguished, the *excretory duct* and the *coil* (Fig. 288). The *duct* runs a straight or sinuous course through the corium, enters between two papillæ into the epidermis, in the stratum corneum of which it is spirally twisted, and opens on the surface of the skin by a rounded lumen, the *sweat-pore*, just visible to the naked eye. The wall of the excretory duct consists of longitudinally disposed bundles of connective tissue and within this of a double layer of cubical epithelial cells. The *coil* is a much convoluted single † canal, the wall of which is formed of a simple layer of cubical cells, containing granules of pigment and of fat; external to this is a delicate *membrana propria*. Intercellular and intracellular secretory capillaries occur in the coil-glands. In well-developed glands longitudinally disposed smooth muscle-fibers occur between the *membrana propria* and the gland-cells.

The secretion usually is a fatty substance, for the purpose of lubricating the skin; only under the influence of disturbed innervation do the coil-glands discharge the watery liquid called sweat. Destruction of the gland-cells does not occur either in the one or the other mode of secretion. The coil-glands are distributed over the entire surface of the skin and are absent only on the glans penis and on the inner surface of the prepuce. They are most numerous in the skin of the palm of the hand and of the sole of the foot.

* They may be wholly wanting; to name them Tyson's glands is incorrect, because under this designation Tyson described depressions in the superficial epithelium, crypts regularly present, from $\frac{1}{2}$ to 1 cm. long, chiefly in the form of a flat pocket, occurring in the neighborhood of the frenulum præputii. Preputial glands, as well as crypts, are wanting in the glans and præputium clitoridis. In the fetus the inner surface of the præputium and the outer surface of the glans are united by a solid epithelial mass, that often does not break up until after birth, through the formation of concentrically stratified epithelial pearls.

† Branched tubules have been observed only in the axillary and circumanal coil-glands.

THE BLOOD-VESSELS, LYMPH-VESSELS, AND NERVES OF THE SKIN.

The *arteries* of the skin originate in a network lying above the fasciæ and branch as they ascend toward the surface of the skin. These branches anastomose with one another and with those of neighboring arteries and in the lower stratum of the corium form a horizontally disposed reticulum, the *cutaneous network*. The arteries supplying the skin are therefore not end-arteries.*

From this network two capillary territories are supplied; the deeper is intended for the adipose tissue (Fig. 308, *a'*), the more superficial

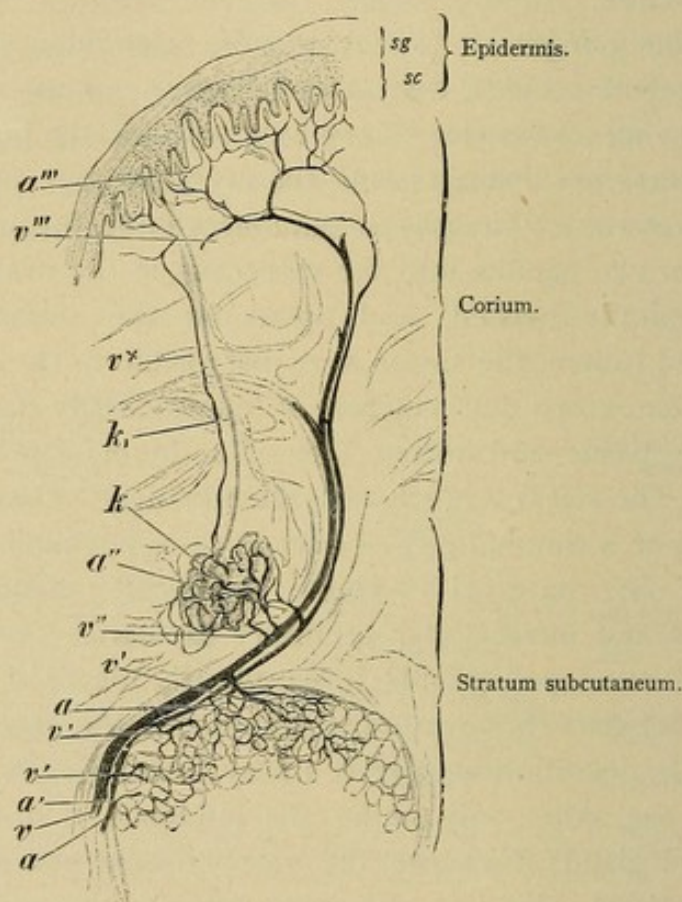


FIG. 308.—FROM A VERTICAL SECTION OF THE SKIN OF THE SOLE OF A HUMAN FOOT. $\times 50$. *sc*, Stratum corneum; *sg*, stratum germinativum; *a*, artery; *v*, vein; *a'*, *v'*, their branches to the panniculus adiposus; *a''*, *v''*, their branches to the coil-glands; *a'''*, *v'''*, their branches to the papillæ; *k*, coil-gland; *k*₁, duct of the same; *vx*, vein accompanying the duct. Technic No. 172.

appears in the form of basket-like plexuses surrounding the coil-glands (*a''*). From the cutaneous network twigs ascend that anastomose and form a second horizontal network in the upper third of the corium, the *subpapillary plexus*; from this very small twigs arise, which run for a

* "End-arteries" are those small arteries which do not anastomose with neighboring arteries, but independently supply capillary circuits of varying extent. When they become obstructed the part of the organ which they supply dies.

short distance along the rows of papillæ and send little branches into them (Fig. 308, *a'''*). These smallest twigs do not anastomose with one another, hence are end-arteries. The branches intended for the hair-follicles and the sebaceous glands also arise from the subpapillary plexus.

The blood returning from the capillary vessels of the papillæ, the hair-follicles, and the sebaceous glands is taken up by *veins* that form a horizontal plexus lying close beneath the papillæ and that occasionally are united with a second horizontal plexus lying very close below the first. From this plexus small venous trunks descend beside the arteries and lead to a third network lying in the lower half of the corium, which is not so horizontally spread out as its predecessors. This plexus takes up the veins coming from the coil-glands and then those proceeding from the lobules of adipose tissue. It should further be noted that a branch of the veins of the coil-glands passes along the excretory duct to the venous plexus of the stratum papillare (Fig. 308, *v x*) and that the hair-papilla receives an independent arterial branch. From the third venous network larger veins lead to the lower boundary of the skin, where a fourth horizontally disposed, "subcutaneous" venous network occurs, from which larger stems turn into the subcutaneous stratum and then unite with the large subcutaneous veins, some of which are provided with names.

The *lymph-vessels* form two horizontal capillary networks, of which that consisting of smaller tubules and narrower meshes lies in the papillary stratum of the corium beneath the blood vascular network; the other, wider-meshed, is situated in the subcutaneous stratum. Special networks of lymph capillaries surround the hair-follicles, the sebaceous and the coil-glands.

The *nerves* of the integument (very numerous in the palm of the hand and the sole of the foot) partly end in the subcutaneous stratum in lamellar corpuscles (p. 222); partly in terminal cylinders (p. 226), that occur in the stratum subcutaneum in the vicinity of the body of the coil-glands, also in the corium of the toe and finger pads; partly they find their ending in tactile corpuscles, in tactile cells, as free ramifications in the papillæ with vascular loops, and as intraepithelial fibers (Fig. 145). The hairs also are supplied with medullated nerve-fibers, which run up to the point where the sebaceous glands open into hair-follicles; here they divide, lose their medullary sheath, and as naked axis-cylinders, usually running longitudinally, terminate in a spoon-shaped expansion *on* the hyaline membrane (epilemmal nerve-ending); in the tactile-hairs (sensory hairs) of animals delicate twigs arise from these nerves, which pass through the hyaline membrane of the hair-follicle into the outer root-sheath and there end in tactile disks (p. 221). The hair-papilla does not

possess nerves. The nerves of the coil-glands behave similarly to those of the glands of the oral cavity (*cf.* p. 247).

THE MAMMARY GLAND.

The mammary gland, a convoluted alveolo-tubular compound gland, in children of both sexes consists chiefly of connective tissue, which encloses the branched excretory gland-ducts. These ducts have a bulbous enlargement at their termination. End-pieces are wanting. The mammary gland of the adult male exhibits the same structure.

In the adult female the mammary gland up to the occurrence of

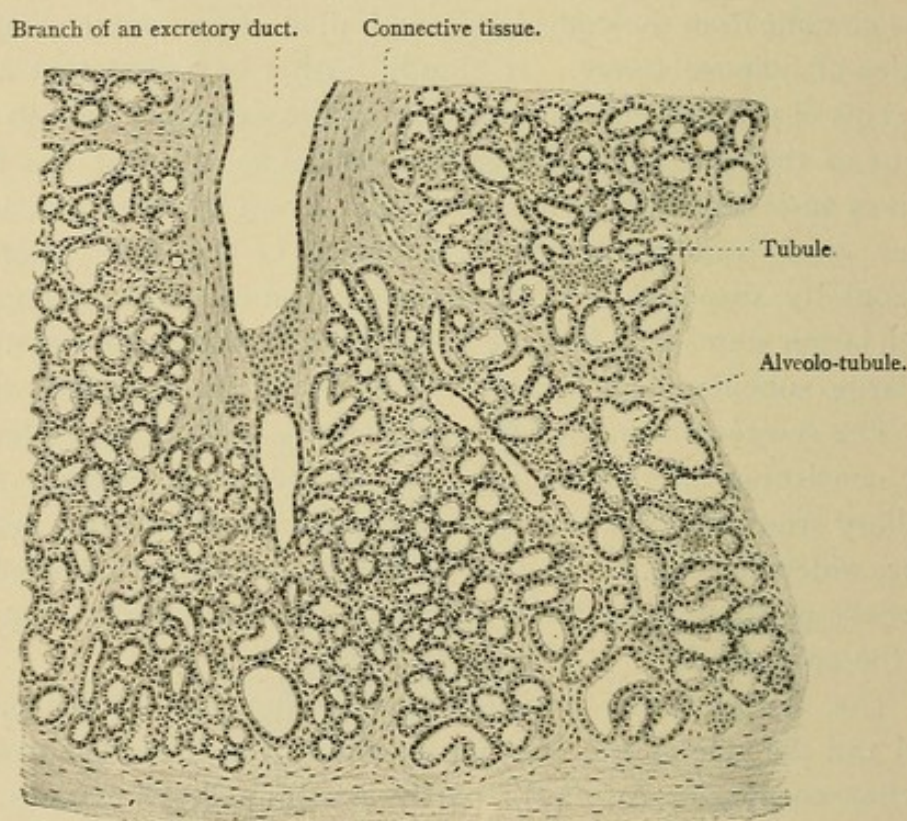


FIG. 309.—SECTION OF A HUMAN MAMMARY GLAND AT THE PERIOD OF LACTATION. $\times 50$. Technic No. 174.

pregnancy is a disk-shaped body, that consists chiefly of connective tissue and of the excretory gland-ducts. End-pieces are present only in limited number at the ends of the smallest branches of the excretory ducts.

At the time of pregnancy and of lactation the mammary gland consists of from fifteen to twenty alveolo-tubular compound glands, which are held together and united in a common body by loose connective tissue containing fat-cells. Each of these glands has its own excretory duct opening on the nipple, that shortly before its termination is provided with a conspicuous spindle-shaped expansion, the *milk-sack* or *ampulla*

(*sinus lactiferus*) and by means of dichotomous ramifications is connected with the end-pieces. The latter lie close beside one another and are bound together by connective tissue into small lobules.

Touching the microscopic structure, the excretory ducts consist of a cylinder epithelium,* followed outside by a *membrana propria* and connective-tissue bundles chiefly having a circular arrangement.

The end-pieces differ in structure during the period of gestation and during the period of lactation. During gestation the end-pieces are clothed with a simple cubical or somewhat flattened epithelium; their lumen contains leucocytes that have wandered in from the underlying interstitial connective-tissue through the epithelium. Some of these leucocytes perish (their nucleus is ragged, often divided into several pieces), the others take up fat drops furnished by the gland-cells and grow to be conspicuous bodies, the *colostrum corpuscles* (Fig. 312). Basket-cells (remark *, p. 86) and a delicate *membrana propria* separate the end-pieces from the interstitial connective tissue, that is not only rich in uninuclear leucocytes, but also contains many oxyphile cells (p. 138).

After parturition the gland-cells are larger, filled with stainable granules (precursors of the secretion?) and with fat drops, which latter usually lie on the side of the cell directed toward the gland-lumen and often exceed in size the nucleus of the cell (Fig. 310).

When lactation has been established for a couple of days some of the gland-cells appear flat (empty of secretion), some as tall cylinders, that with a ragged top extend toward the lumen; both forms are united with each other by transitional forms and contain (the tall cells more often) two nuclei. Both forms contain fat drops; these are not as in the sebaceous glands the product of a fatty *degeneration* of the cell, but the product of an act of secretion, that the cell repeats many times and of which it does not perish.† Besides the fat drops ("milk globules"), the gland-lumen contains free nuclei, that appear to be extruded from the gland-cells. These nuclei perish by liquefying (*cf.* p. 72) and regulate the

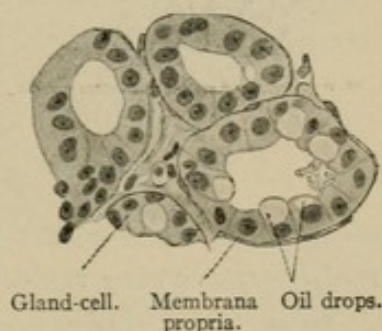


FIG. 310.—FROM A SECTION OF THE MAMMARY GLAND OF A NURSING WOMAN. $\times 250$. Technic No. 174.

* Not seldom a stratified squamous epithelium, instead of cylinder epithelium, is found in the trunks of the excretory ducts.

† This statement is not vitiated by the well-established fact that some gland-cells die. The death of these cells is due to the intensity of their secretory activity, which may result in the rapid ageing and finally in the demise of individual cells.

nuclein constituent of the milk.* Colostrum corpuscles and leucocytes are now wanting, and the greatly reduced interstitial connective tissue contains extremely few leucocytes and eosinophile cells.

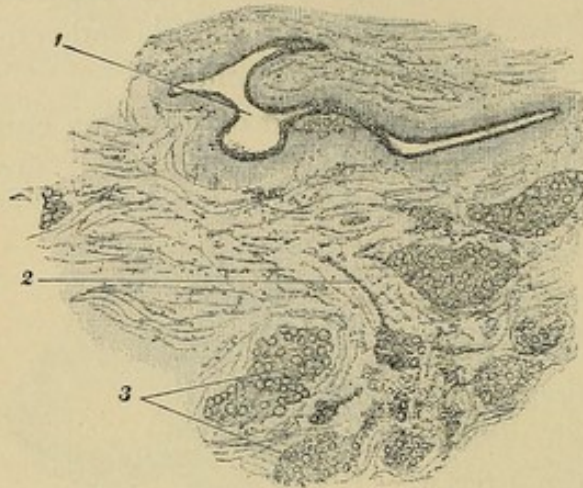


FIG. 311.—FROM A THICK SECTION OF THE MAMMARY GLAND OF A WOMAN LAST PREGNANT TWO YEARS BEFORE. $\times 50$. 1. Large excretory duct; 2, small excretory duct; 3, gland lobules, separated from one another by connective tissue. Technic No. 173.

the deepest strata of the epidermis, by tall papillæ, and by the presence of smooth muscle-fibers, which latter are partly circularly arranged around the orifices of the ducts, partly ascend vertically to the apex of the nipple. In the integument of the areola accessory mammary glands, the *areolar glands* (Montgomery), occur during pregnancy and lactation.

The *blood-vessels* approach the mammæ from all sides and form capillary networks embracing the gland-tubules. The *lymph-vessels* form capillary plexuses lying within and between the gland-lobules. Lymph-vessel networks also occur in the vicinity of the ampullæ and the areolæ.

The *nerves* are in part vascular nerves, in part behave like those of the glands of the oral cavity (p. 247).

When lactation is ended a gradual regressive metamorphosis takes place, that is soon manifested by the abundant development of the interlobular connective tissue† (Fig. 311). The lobules become smaller, the end-pieces begin to atrophy. In elderly persons all the end-pieces and the lobules have disappeared and only the excretory ducts remain.

The skin of the *nipple* and of the *areola* is characterized by deep pigmentation, due to the presence of pigment granules in

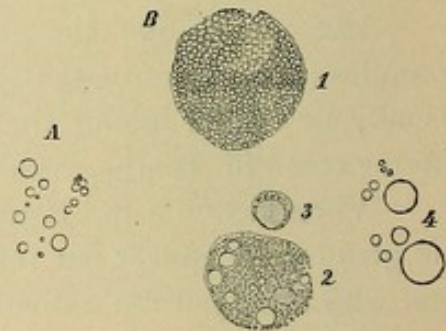


FIG. 312.—A. MILK GLOBULES FROM HUMAN MILK. $\times 560$. Technic No. 175. B. ELEMENTS OF THE COLOSTRUM OF A PREGNANT WOMAN. $\times 560$. 1. Cell containing uncolored fat globules; 2, cell containing minute colored fat globules; 3, leucocyte; 4, milk globules. Technic No. 176.

* Nuclear divisions by mitosis do not occur in the functioning mammary gland; it is assumed that the nuclei here are produced by amitosis (remark *, p. 69).

† Leucocytes also reappear, that behave in exactly the same manner as during the period of gestation and become colostrum corpuscles, etc. Therefore the leucocytes invariably are present when retention occurs.

Microscopically *milk* consists of a clear fluid, the *milk plasma*, in which large fat-drops from 2 to 5 μ in size, the *milk globules*, are suspended. In addition isolated cells enclosing fat globules (leucocytes) are found in milk.

The elements of milk secreted before and in the first few days after parturition appear somewhat different. Beside the milk globules the *colostrum corpuscles* occur, leucocytes enclosing a spherical nucleus, some of which contain small, yellowish and larger, uncolored fat globules, others only uncolored fat-drops.

The *witches' milk*, which can be pressed out of the hollowing gland-ducts of the newborn, is a fluid resembling the colostrum.

TECHNIC.

No. 161.—*Strata of the skin ; coil-glands*.—Cut from the pad of the finger, the palm of the hand, or the sole of the foot pieces of skin, as fresh as possible, from 1 to 2 cm. square, together with a thin stratum of the subjacent fat and place them in 30 c.c. of absolute alcohol. To prevent curling of the pieces pin them on small cork plates with the epidermis turned toward the cork, and place the whole in absolute alcohol. On the following day remove the pieces from the cork plates and place them for from three to four weeks in 50 c.c. of 90 per cent. alcohol. Cut thin and thick sections. The latter are indispensable in order to obtain the excretory ducts of the coil-glands in their entire length. The most suitable for this purpose is the skin of children, from the sole of the foot, because the still short ducts of the coil-glands here run vertically (Fig. 288). Stain with alum carmine for ten minutes (p. 39); the red coils can be seen with the unaided eye; mount in xylol-balsam. Examine with the low power. In thick sections the papillæ often are indistinct, because they are encircled by the red-colored stratum germinativum; the screw-like ends of the excretory ducts are most distinctly seen when the object is faintly illuminated or with oblique illumination (see p. 55, remark *).

To render the stratum granulosum visible bulk-staining with borax-carminé for two or three days (p. 40) is recommended. The granules of this stratum are then stained an intense red (Fig. 290).

No. 162.—Pretty preparations of the under surface of the *epidermis* are obtained by fixation of shreds of the epidermis of the dorsum of the foot, that often can be detached from injected cadavers,* in 30 c.c. of absolute alcohol. Stain for two minutes in Hansen's hematoxylin and mount in xylol-balsam (Fig. 289).

* The epidermis can also be detached from the corium by macerating a little piece of skin in 0.33 per cent. acetic acid.

No. 163.—For preparations of the *nails* fix the distal finger joint of a child from eight to twelve years of age (of adults, that of the little finger, if possible of women), two or four weeks in 100 or 200 c.c. of Müller's fluid (p. 33) and harden in about 100 c.c. of gradually strengthened alcohols (p. 35); decalcify (p. 36); harden again and stain thick cross-sections ten minutes in alum-carmines (p. 39); mount in xylol-balsam. (Fig. 291.) In cutting sections place the knife on the *volar side* (not on the nail side) of the finger joint. The substance of the nail frequently shows differently colored strata. In the nails of old cadavers the germ-layer often becomes loosened from the ridges.

No. 164.—*Elements of the nails*.—Place pieces of nail 1 or 2 mm. broad, in a test-tube containing 5 c.c. of concentrated potash-lye and heat it over a flame until it boils up once. Transfer the nail with a drop of the lye to a slide and scrape off some of the softened surface; apply a cover-glass. On examination with a high power cells will be found like those in Fig. 292. For comparison investigate the horny cells of the stratum corneum, which may be obtained by lightly scraping the pad of the finger with a scalpel. Examine the polygonal scales in a drop of distilled water, with a high power.

No. 165.—*Hairs*.—Place a hair in a drop of salt solution on a slide and examine it with the low and the high power; the most suitable for study are white hairs and the hairs of the beard. The hair cuticle of man is very delicate and the transverse markings produced by the imbrication of the cells are often very indistinct; usually only fine wavy lines are visible. The hairs of many animals, on the other hand, show the cuticula very well, for example, sheep's wool.

No. 166.—For the demonstration of the *elements of the hairs*, place a piece of hair 1 or 2 cm. long in a drop of pure sulfuric acid on a slide and apply a cover-glass; press lightly on the cover-glass with a needle and the cortical substance will split into fibers, which consist of adherent cortical cells. Slightly warm the slide, press again with a needle, so that the cover-glass becomes slightly displaced; numerous free elements, superficial scales and cortical cells, will then be seen (Fig. 294).

No. 167.—For the exhibition of the *elements of the hair-follicles* (and the *hairs*) cut from a mustachioed human upper lip a piece 2 cm. square and place it in dilute acetic acid (5 c.c. of acetic acid to 100 c.c. of distilled water). In two days the individual hairs with their sheaths can be easily withdrawn and their elements separated by teasing in a drop of distilled water (Fig. 294). The cells of Henle's sheath float in small complexes in the preparation and closely resemble fenestrated membranes (Fig. 294, 5). The fenestra are spaces normally occurring between Henle's cells, through which processes of the cells of Huxley's stratum extend to the outer root-sheath. Not infrequently a hair-follicle is obtained in which shedding is taking place (similar to Fig. 306).

No. 168.—For the study of *hairs and hair-follicles* place pieces 2 or 3 cm. square of the quite fresh skin of the scalp in about 200 c.c. of a 3

per cent. solution of potassium bichromate (p. 21, No. 10 *a*) for from four to eight weeks; wash them for from one to three hours in running water and harden in the dark in about 100 c.c. of gradually strengthened alcohol. Longitudinal sections of sufficient thinness, which include the entire length of the follicle, are very difficult to cut. Macroscopic orientation as to the direction of the hair is first necessary. To obtain preparations like that in Fig. 293 thick unstained sections are to be mounted in glycerol. Thin sections usually include only a portion of the hair-follicle. It is much easier to cut thin cross-sections, but care must be taken to make the cut *vertical to the longitudinal direction of the hair*, not parallel to the surface of the skin. In this way hairs and hair-follicles at different levels are obtained in a single section. These sections are to be stained with Hansen's hematoxylin and with eosin* (p. 39) and mounted in xylol-balsam. Especially fine are the sections of the hair-follicles near to the hair-bulb (Fig. 295).

No. 169.—*For the development of hair* cut pieces about 2 cm. square of the skin of the forehead (not of the hairy scalp) of a five- or six-months'-old human embryo; span them on cork (see No. 161); place them for fourteen days in 100 or 200 c.c. of Müller's fluid (p. 33) and harden in about 100 c.c. of gradually strengthened alcohols (p. 35). Staining the objects in bulk in borax-carmin (p. 40) is advised; or the sections may be stained in Hansen's hematoxylin (p. 38). Embed the tissue in liver; endeavor to cut sections exactly in the direction of the hair-follicle, which is much more easily done than in the hairy scalp of the adult. Mount in xylol-balsam. The sections exhibit all stages of development. The preparations of figures 302, 303, and 305 were fixed in Zenker's fluid. The details pictured in figures 297 to 306 can be seen only in thin microtome sections.

No. 170.—*Shedding and renewal of hair*.—The eyelids of newborn children are most suitable. Treat like No. 191. Cut sagittal sections. Vertical sections of the hairy scalp often yield good results.

No. 171.—*The sebaceous glands*.—Fix and harden the alæ nasi of newborn children in 100 c.c. of a 3 per cent. solution of potassium bichromate (like No. 168). Cut thick (Fig. 307 *A*) and thin (Fig. 307 *B*) sections; stain them with dilute carmin (p. 39), and with Hansen's hematoxylin (p. 38), and mount in xylol-balsam. Sections lengthwise of the nose often show sebaceous glands and hair-follicles, but they must be exactly vertical. The alæ of the nose of adults, on account of the very large sebaceous glands with their wide excretory ducts, do not furnish good microscopic specimens. Small sebaceous glands with hair-follicles can be seen with the unaided eye in stripping off the macerated epidermis of old cadavers.

No. 172.—*The blood-vessels of the skin*.—Inject with Berlin blue the entire hand of a child through the ulnar artery or a foot through the pos-

* *Slow* staining with eosin (3 *a*, p. 39) stains the granules of keratohyaline an intense red (Fig. 296).

terior tibial artery (p. 48) and place it in from 1 to 2 liters of Müller's fluid (p. 33); after several days cut pieces 2 or 3 cm. square of the palm of the hand or of the sole of the foot, place them for from two to four weeks in 100 or 200 c.c. of Müller's fluid and harden them in 100 c.c. of gradually strengthened alcohols (p. 35). Cut thick sections and mount them unstained in xylol-balsam (Fig. 308). The papillæ in such sections often can only be recognized by the capillary loops. To the beginner it appears as if the loops extend into the stratum germinativum.

No. 173.—*For a general view of the mammary gland* place the nipple and a portion of the gland (3 or 4 cm. square) in from 60 to 100 c.c. of absolute alcohol. If possible obtain the glands of an individual that was pregnant not too long a time before, also the glands of virgins, etc. Make vertical sections through the nipple and in any direction through the gland-substance, and stain them with Hansen's hematoxylin (p. 38); mount in xylol-balsam. (Fig. 311.)

No. 174.—*For the minute structure of the mammary glands* place the warm, living tissue (3 to 5 mm. cubes) of a mammal during gestation or lactation in 5 c.c. of Flemming's mixture (p. 34), and harden after one or two days in 30 c.c. of gradually strengthened alcohols. Cut *very thin* sections, stain them with safranin (p. 41), and mount in xylol-balsam. The structure is often difficult to understand on account of the small size of the gland-cells (in the rabbit). (Fig. 309, 310.)

No. 175.—*The elements of milk.*—Put a drop of salt solution on a clean slide and add to it a drop of milk. The milk is to be obtained by placing the cover-glass upon the nipple of a nursing woman and then pressing out a drop. Examine with a high power (Fig. 312 A).

No. 176.—*The elements of colostrum.*—Obtain the colostrum from a pregnant woman shortly before parturition. Proceed as in No. 175. Be careful to avoid pressure on the cover-glass. The nuclei of the colostrum corpuscles can rarely be distinctly seen without further treatment; on the addition of a drop of picrocarmine they appear as simple, round, dull-red spots (Fig. 312 B).

X. THE ORGAN OF VISION.

The organ of vision consists of the *eyeball* (*bulbus oculi*), the *optic nerve*, the *eyelids*, and the *lacrimal apparatus*.

THE EYEBALL.

The eyeball is a hollow globe, which encloses partly formed, partly fluid contents. The wall of the globe is composed of three membranes: (1) the *tunica externa*, a connective-tissue membrane, in which an anterior transparent division, the *cornea*, may be distinguished from the remaining opaque portion, the *sclera*; (2) the *tunica media*, rich in ves-

sels, which includes three divisions, the *choroid*, the *ciliary body*, and the *iris*; (3) the *tunica interna*, the *retina*, which contains the terminal apparatus of the optic nerve. The formed contents within the eyeball are the *lens* and the *vitreous body*.

The first anlage of the eyeball, the *primary optic vesicle*, is a hollow epithelial sphere, which is connected with the brain by a stalk. By invagination in front and below the primary vesicle is converted into the *secondary vesicle*, a two-layered cup that becomes the retina (the outer layer develops into the pigmented epithelium (p. 412), the inner layer into the retina proper); while the stalk is transformed into the optic nerve. From the border of the invagination, where the inner and outer layers of the secondary vesicle meet and blend, smooth muscle fibers develop, the sphincter and beside it the dilatator pupillæ muscles.* While the vitreous body and the lens come to lie within the cavity of the optic cup, the connective tissue surrounding the cup separates into two layers, an outer, which furnishes the *tunica externa*, and an inner, which furnishes the *tunica media* of the eyeball.

THE TUNICA EXTERNA.

The *cornea* consists of five strata, which named from without inward form the following (Fig. 313): (1) the corneal epithelium, (2) the anterior basal membrane, (3) the substance proper, (4) the posterior basal membrane, (5) the corneal "endothelium."

The *corneal epithelium* is a stratified squamous epithelium and consists of a lowermost layer of sharply contoured cylindric cells, which is followed by three or four (more in animals) layers of spherical cells, that in turn are covered by several strata of flattened elements, still possessing nuclei. The thickness of the epithelium in man is 0.03 mm. At the rim of the cornea the epithelium is continuous with that of the conjunctival sclera.

The *anterior basal membrane* (Bowman's membrane), in man is a distinctly visible stratum, measuring up to 0.01 mm. in thickness, and is almost homogeneous in appearance. Its upper surface is provided with minute serrations and ridges for the attachment of the cylinder cells of the corneal epithelium; at its under surface it gradually passes into the substantia propria of the cornea, of which it passes as a special modification. The name "anterior elastic lamina" is not recommended, because the membrane does not consist of elastic substance.

The *substance proper* (substantia propria corneæ) constitutes the chief bulk of the cornea. It consists of delicate connective-tissue fibrillæ, running a straight course, which are united by a (fluid?) interfibrillar cement-substance into bundles of nearly uniform thickness; the bundles

* According to this the muscles are of ectodermal derivation, contrary to the majority of other smooth muscles, that originate from the mesoderm.

in turn are united by an interfascicular cement-substance into flat lamellæ, which lie in many superposed strata and are held together by an interlamellar cement-substance. The lamellæ are arranged parallel to the surface of the cornea and run in the direction of every meridian. A number of bundles running obliquely, the so-called arcuate fibers, unite each lamella with its neighbor above or below; especially well-developed arcuate fibers occur in the anterior strata of the substantia propria.

Buried in the cement-substance is a system of freely branched canals, the *corneal canaliculi*, juice canaliculi, which at many places are expanded to broad, oval lacunæ, the *corneal spaces*, juice-spaces (Fig.

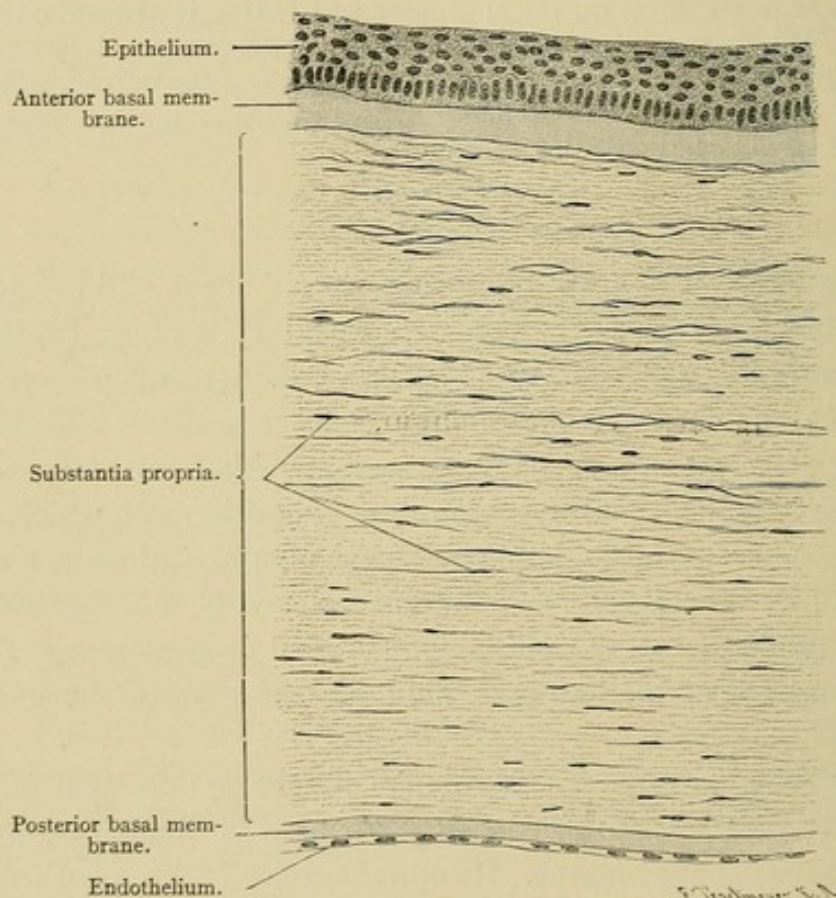


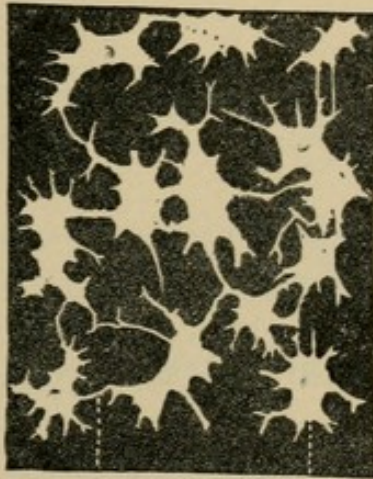
FIG. 313.—VERTICAL SECTION OF A HUMAN CORNEA. $\times 100$. Technic No. 177 b.

314). The latter lie between the lamellæ, while the canaliculi also penetrate between the bundles. In some animals, *e. g.* the frog, the canals branch at right angles. The lacunæ and canaliculi contain a serous fluid and cells, "fixed" *corneal cells* and *wandering cells* (leucocytes). The corneal cells are stellate, flattened connective-substance cells, possessing large, very irregularly shaped nuclei (Fig. 315). According to one view these cells lie against one wall of the canal system, according to another opinion they completely fill the spaces and canals.

The *posterior basal membrane* (membrane of Descemet, posterior elastic lamina) is a transparent elastic layer, only 0.006 mm. thick. In adult man the posterior surface, at the periphery of the cornea, is beset with hemispherical elevations, the so-called warts.

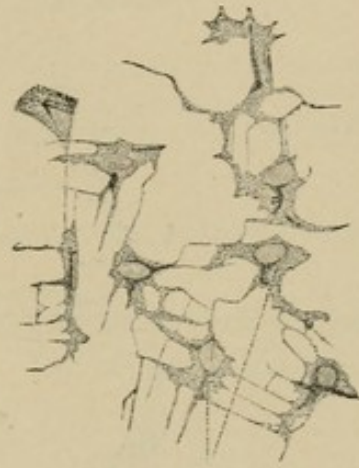
The *corneal endothelium* is composed of a single layer of flat, polygonal cells with spherical nuclei (in animals the nuclei have the shape of a kidney or a horse-shoe).*

The *sclera* consists chiefly of connective-tissue bundles, which interlace in different directions, principally meridional and equatorial, and of many elastic fibers† running parallel with the bundles, as well as of flat, connective-substance cells, which like the fixed corneal cells lie in juice-



Corneal canaliculi. Corneal spaces.

FIG. 314.—HORIZONTAL SECTION OF THE CORNEA OF AN OX. Silver-preparation; negative picture; the canalicular system is light upon a dark ground. $\times$ about 240. Technic No. 182.



Corneal cells.

FIG. 315.—HORIZONTAL SECTION OF THE CORNEA OF A RABBIT. Positive picture of the corneal canaliculi. $\times$ about 240. Technic No. 184.

spaces, that are more irregularly shaped in the sclera than those in the cornea. The thickness of the sclera is greater at the back (1 mm.) and gradually diminishes toward the front.

Between the sclera and the choroid is a layer of loose tissue, rich in elastic fibers and branched pigment-cells and flattened elements free from pigment ("endothelial" cells), which on separating the sclera from the choroid adheres partly to the former and partly to the latter; the portion on the sclera is called the *lamina fusca sclerae*, that on the choroid, *lamina suprachorioidea*.

* These forms are conditioned by the centrosome surrounded by a large court and further especially distinguished by the possession of net-like cords (*cf.* p. 65).

† They are especially profuse at the points of insertion of the eye-muscles.

THE TUNICA MEDIA.

The *choroid* (chorioidea) is distinguished by the great abundance of its blood-vessels, which are arranged in two layers. The superficial layer, lying to the inner side of the lamina suprachorioidea, the *lamina vasculosa* (layer of larger vessels) (Fig. 316) contains the ramifications of the arterial and venous vessels, that are enveloped in lymph sheaths and embedded in a ground-substance (stroma) consisting of delicate elastic fiber-nets and numerous branched pigment-cells. In addition the stroma contains the elements accompanying the larger arteries; namely, fibrillar connective tissue, smooth muscle-fibers, and flat, nonpigmented cells,

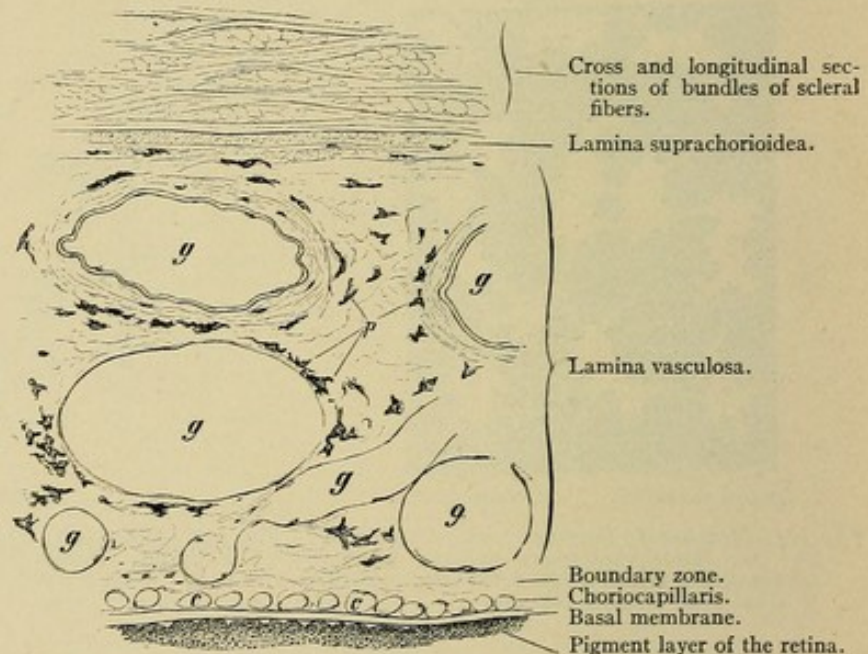


FIG. 316.—VERTICAL SECTION THROUGH A PART OF THE HUMAN SCLERA AND THE ENTIRE CHOROID. $\times 100$.
g. Larger vessels; p, pigment cells; c, cross-sections of capillaries. Technic No. 177 c.

that are united in delicate "endothelial" membranes. The deeper layer, the *lamina choriocapillaris*, or layer of capillary networks, is composed of a narrow-meshed net of wide capillaries, between which no formed elements are found. Between the two vascular laminæ lies the *boundary zone of the ground-substance*, consisting of fine networks of elastic fibers and almost devoid of pigment. In ruminants and horses this zone consists of wavy bundles of connective tissue, to which is due the metallic reflex seen in the eyes of these animals. This shining membrane is known as the *tapetum fibrosum*. The similar iridescent *tapetum cellulosum* of carnivora is composed of several strata of plate-like cells containing numerous minute crystals.

Attached to the lamina choriocapillaris is a close net of fine elastic

fibers and then follows the *lamina basalis* or vitreous membrane, a structureless lamella, up to $2\ \mu$ thick, which on its outer surface is provided with delicate, lattice-like markings. The polygonal areas noticeable on

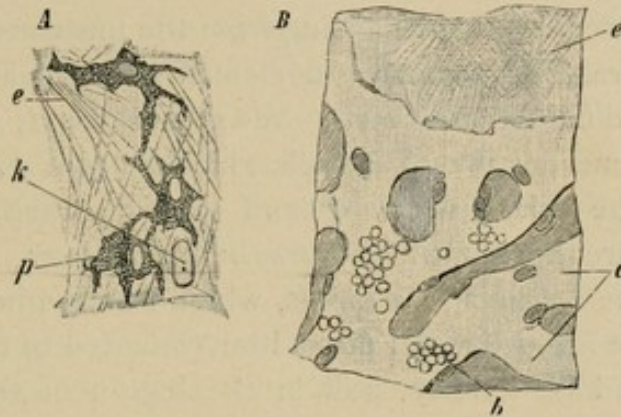


FIG. 317.—A. FROM A TEASED PREPARATION OF A HUMAN CHOROID. $\times 240$. *p*. Pigment cells; *e*, elastic fibers; *k*, nucleus of a flat nonpigmented cell; the cell-body is invisible. B. PORTION OF A HUMAN CHORIOCAPILLARIS AND THE ADHERENT LAMINA BASALIS. $\times 240$. *c*. Wide capillaries, some of which contain (*h*) blood corpuscles; *e*, lamina basalis, showing a fine "lattice-work." Technic No. 178 a.

its inner surface are imprints of the retinal pigment. The vitreous membrane is related to the elastic membranes.

The *ciliary body* is formed by the ciliary processes and the muscular ring lying upon them, the ciliary muscle. The *ciliary processes* are from

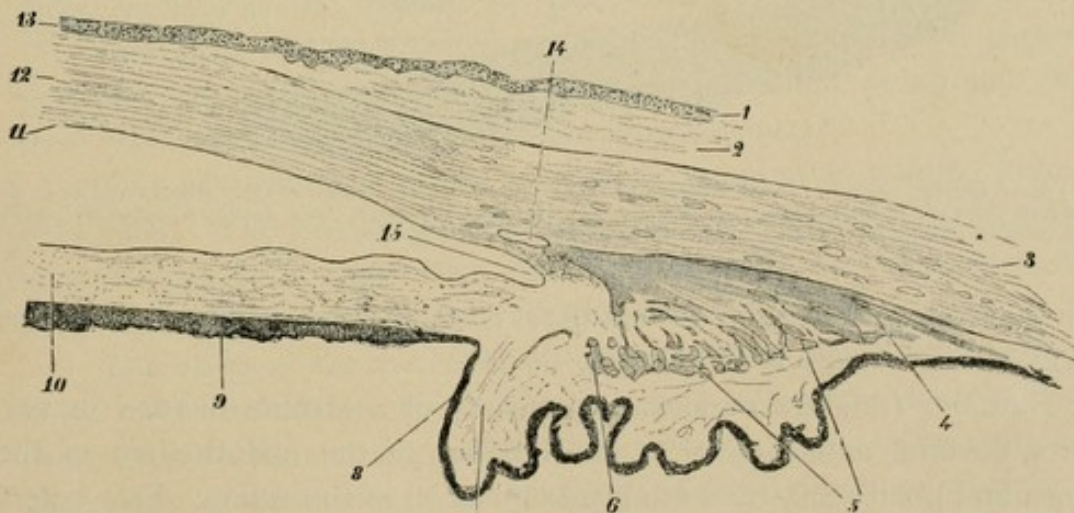


FIG. 318.—MERIDIONAL SECTION THROUGH THE IRIDAL ANGLE OF MAN. $\times 30$. 1. Epithelium, 2, connective tissue of the conjunctiva. 3. Sclera. 4, 5, 6, 7, and 8. Ciliary body; 4, meridional, 5, radial, 6, circular fibers of the ciliary muscle; 7, ciliary process; 8, ciliary portion of the retina. 9. Iridal portion of the retina. 10. Stroma of the iris. 11, 12, and 13. Cornea; 11, posterior elastic lamina; 12, substantia propria; 13, epithelium. 14. Venous sinus of the sclera. 15. Angle of iris. Technic No. 177 a.

seventy to eighty meridionally placed folds, which begin low at the ora serrata (p. 414), gradually attain a height of one millimeter, and terminate with an abrupt descent near the edge of the lens. Each ciliary process consists of fibrillar connective tissue, that contains elastic fibers

and numerous blood-vessels and inwards is bounded by a continuation of the vitreous membrane, that here is distinguished by minute intersecting folds. The blood-vessels of the ciliary processes supply the intraocular fluid.* The *ciliary muscle* is an annular band about 3 mm. broad, anteriorly 0.8 mm. thick, arising from the inner wall of the venous sinus of the sclera. The nonstriped elements of which it is composed extend in three different directions. We distinguish 1, *meridional fibers* (Fig. 318, 4), numerous muscle-bundles, intermingled with elastic fibers, lying next to the sclera, which extend to the smooth portion of the choroid; they are known as the *tensor chorioideæ*; 2, *radial fibers*, lying next to the meridional bundles, which from without inward progressively assume a more radial disposition (oriented to the center of the bulbus oculi) and posteriorly, still in the region of the ciliary body,

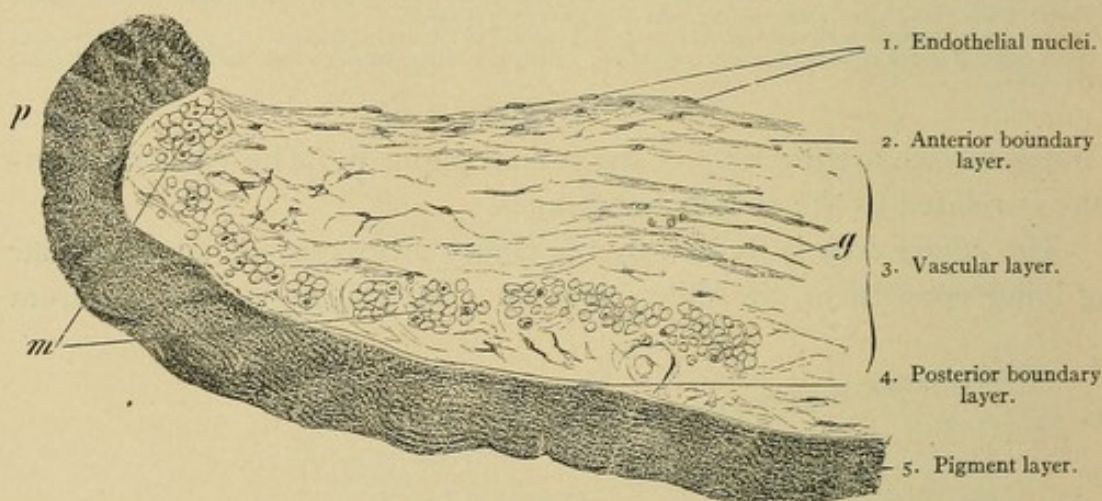


FIG. 319.—VERTICAL SECTION OF THE PUPILLARY PORTION OF A HUMAN IRIS. $\times 100$. About one-fifth of the entire width of the iris is shown. *g*, Blood-vessel, with thick connective-tissue sheath; *m*, sphincter pupillae muscle cut transversely; *p*, pupillary border of the iris. Technic No. 178 c.

turn and follow a circular course (5); 3, *circular (equatorial) fibers*, the so-called *ring-muscle of Müller* (6).

The *iris* (rainbow membrane) consists of a stroma divided in two layers, covered anteriorly by a continuation of the endothelium of the cornea and posteriorly by a modified extension of the retina. Five layers are distinguished in the iris: (1) the "endothelium," (2) the anterior boundary layer, (3) the vascular layer, (4) the posterior boundary layer, (5) the pigment layer.

The *endothelium* covers the anterior surface of the iris and, like that of the cornea, consists of a single layer of flattened, polygonal cells.

* The ciliary processes perhaps serve to regulate the intraocular pressure, that despite the action of the ciliary muscle is not increased; the regulation is effected by compression of the ciliary processes.

The *anterior boundary layer* (reticular layer) comprises three or four strata of networks, which are formed by stellate connective-substance cells. This network resembles the reticulum of adenoid tissue and on its posterior surface gradually passes into the vascular layer.

The *vascular layer of the iris* contains numerous vessels radially disposed (to the pupil), in a stroma consisting of slender, loosely united bundles of connective tissue. There are smooth muscle-fibers in the vascular layer, arranged in (a) circular fiber-bundles at the pupillary margin of the iris, the *sphincter pupillæ muscle*, up to 1 mm. broad, and (b) in animals (the rabbit) a few fibers spreading in a radial direction from this, which do not form a continuous stratum and are lost periphery-ward between the fibers of the dilatator muscle; in man only traces of these fibers are present. In the anterior boundary layer and in the vascular layer pigmented cells occur in greatly varying numbers; in blue eyes they are absent.

The *dilatator muscle* of the pupil extends from the ciliary margin of the iris to near the pupillary margin and unites with the connective tissue occurring here between the sphincter bundles, there between the bundles of the ciliary muscle. It consists of a continuous stratum of spindle-shaped smooth muscle-fibers, of which each exhibits an anterior, non-nucleated, contractile division and a posterior, nucleated, pigmented division; the anterior division in particular can be distinctly seen in radial sections of the iris and has long been known under the name of:

The *posterior boundary layer* (Bruch). The posterior pigmented portion forms with the adjacent, likewise pigmented, polygonal cells of the "pars iridica retinæ" a common pigment-mass:

The *pigment layer* of the iris. The pigment is wanting here only in albinos. The posterior surface of the pigment layer is covered by a very delicate, little membrane, the *limitans iridis*, a continuation of the vitreous membrane of the pars ciliaris retinæ (p. 414).

The *angle of the iris* (corneal furrow). The place where the transition of the sclera into the cornea occurs is of especial interest, because there the iris, the cornea, and the ciliary body meet. The transition of the sclera into the cornea is absolutely direct; the more wavy bundles of the sclera without interruption in continuity pass over into the slender fibril-bundles of the cornea, the system of juice canaliculi of the sclera communicates with that of the cornea. The line of transition, microscopically not sharply defined, is oblique, because the transformation of the sclera into the tissue of the cornea takes place sooner in the posterior than in the anterior portion of the tunica externa. The posterior stratum of the substantia propria corneæ and the posterior basal mem-

brane meet at the periphery with the ciliary border of the iris ; this place is called the *angle of the iris* (Fig. 318, 15). Here the iris sends toward the posterior surface of the posterior basal membrane connective-tissue processes, the *iridal processes*, that in animals (cattle, horses) are powerfully developed and constitute the so-called *ligamentum iridis pectinatum*. In man these processes are scarcely developed at all. The posterior basal membrane at its entire periphery splits into fibers, which blend with the iridal processes ; these fibers receive reinforcements from the elastic tendons and the intermuscular connective-tissue of the ciliary muscle and accessions in a lesser degree from the sclera. Accordingly the tissues that participate in the construction of the fibers occupying the angle of the iris are contributed by all the structures that meet one another there : the cornea, sclera, iris, and ciliary muscle. The endothelium of the posterior surface of the posterior basal membrane, continued on to the surface of the iris, forms a cover for these fibers. The spaces occurring between these fibers, that stand in open communication with the anterior chamber of the eye and contain the same fluid, are called the *spaces of Fontana*. In man they are scarcely developed.

THE TUNICA INTERNA.

The transparent *retina*, in a perfectly fresh condition colored red by the visual purple, extends from the entrance of the optic nerve to the pupillary margin of the iris and in this tract three zones can be distinguished : (1) the *pars optica retinae*, the actual territorial expanse of the optic nerve ; this portion of the retina, alone sensitive to light, clothes the entire posterior segment of the eyeball, to within a short distance of the ciliary body, where it terminates in a sharp, macroscopically perceptible, serrated line, the *ora serrata* ; (2) the *pars ciliaris retinae*, extending from the ora serrata to the ciliary margin of the iris ; (3) the *pars iridica retinae*, which covers the posterior surface of the iris from the ciliary to the pupillary margin. The ciliary and the iridal portion of the retina are together named *pars cæca*.

The *pars optica retinae* falls into two divisions, an outer, the layer of the visual cells (neuro-epithelial division), and an inner layer, the cerebral division ; in each of these divisions several layers can be distinguished, four in the neuro-epithelial, five in the cerebral ; if the pigment layer (pigment-epithelium) lying close beneath the choroid, that genetically belongs to the retina, is added, there are ten layers, that counted from without inward are arranged in the order given in figure 320.

The elements of these layers are only in part nervous or epithelial in their nature ; the other part is formed of *supporting substance*, that how-

ever is not of the nature of connective tissue (see the neuroglia of the spinal cord, p. 195). The most conspicuous elements of the supporting tissue are the *radial fibers* (Müller's supporting fibers), slender cells, which

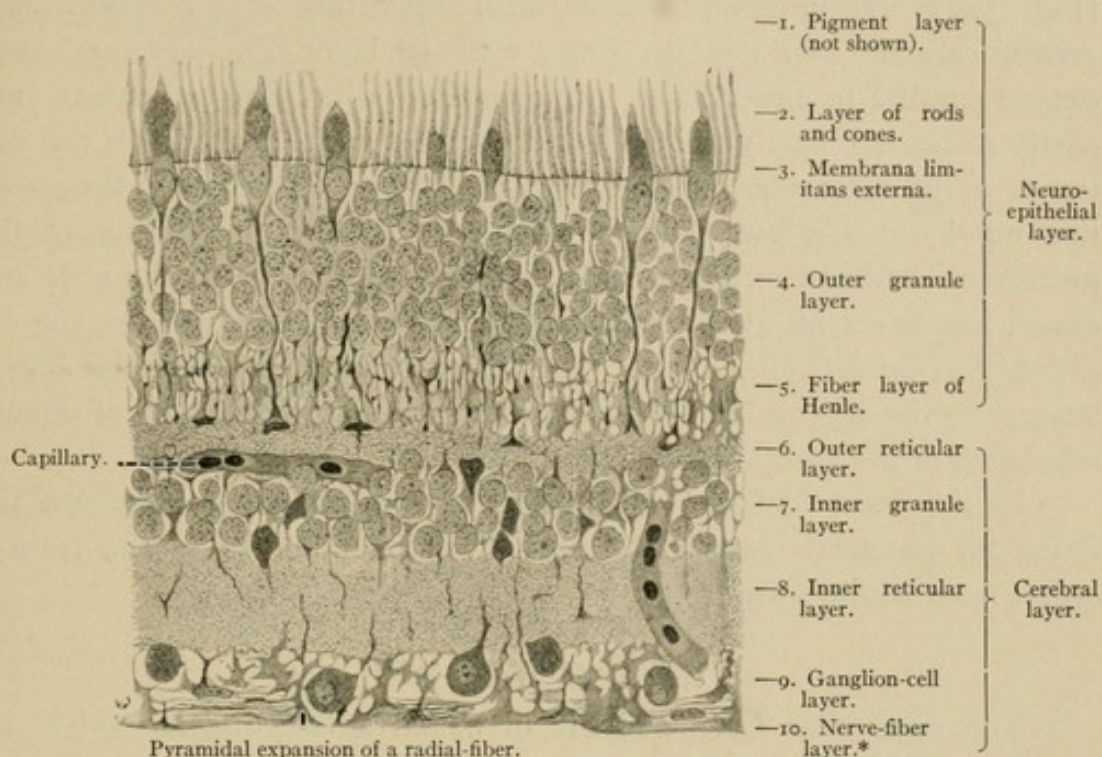


FIG. 320.—VERTICAL SECTION OF A HUMAN RETINA, FROM THE POSTERIOR PORTION OF THE EYEBALL. $\times 400$.
 —(Schaper.)

extend from the inner surface of the retina through all the layers to the rods and cones. The inner end of the fibers is characterized by an expanded base, the *radial-fiber pyramid* (Fig. 321, *k*); the bases of these pyramids are so closely placed beside one another that they apparently form a continuous membrane on the inner surface of the retina, the so-called *membrana limitans interna* (Fig. 321, *l*). From the apex of the pyramids the radial-fibers, with progressive decrease in thickness, proceed through the inner reticular layer to the inner granule layer, where they are provided with a nucleus (Fig. 321, *n*); from here they pass through the outer reticular and outer granule layers to the external limiting

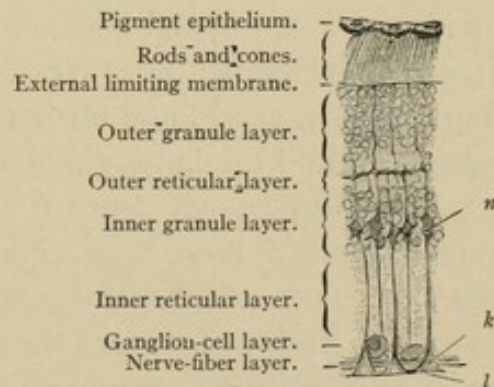


FIG. 321.—VERTICAL SECTION OF THE RETINA OF A RABBIT. $\times 240$. *k*, Expanded base of radial fibers; *n*, nucleated portion of the same; *l*, "membrana limitans interna." Technic No. 178 *e*.

*To these the *membrana limitans interna* is added as an eleventh layer, but it does not represent an independent structure (see the radial fibers, p. 407).

membrane, with which they unite. Throughout their entire course the radial fibers give off lateral processes and lamellæ, especially profuse in the outer granule layer, for the support of the nervous elements (Fig. 322). In addition to these radial supporting cells, *concentric supporting cells* are found in the outer reticular layer (Fig. 322, oo); they extend parallel to the surface, are provided with long processes, are partly nucleated, partly nonnucleated; in the neighborhood of the entrance of the optic nerve, in the nerve-fiber layer, as well as in the ganglion nervi optici, a few glia-cells are found. From the surface of the membrana limitans externa delicate processes extend to the rods and cones, the bases of which they embrace in crib-like structures, the so-called *fiber-baskets* (Fig. 322). A portion of both the reticular layers belongs to the supporting substance, as also the small quantity of cement substance in the ganglion-cell layer.

In the more detailed description of the individual layers of the retina for practical reasons the series will be taken up in the reverse order, from within outward.

THE CEREBRAL LAYER.

The *nerve-fiber layer* consists of naked axis-cylinders, which, arranged in bundles, are united plexus-like. At the entrance of the optic nerve, where the layer is thickest, the fibers expand in a radial direction to the ora serrata. The radial arrangement of the fibers is disturbed in the territory of the macula lutea (p. 412). The majority of the axis-cylinders are centripetal fibers, which originate from the ganglion cells situated in the retina; the others are the axis-cylinder processes of cerebral ganglion cells, centrifugal fibers (Fig. 322), which terminate in free ramifications around the large ganglion cells of the inner granule layer.

The *ganglion-cell layer* ("ganglion nervi optici") consists of a simple layer of large* multipolar ganglion cells, containing Nissl's bodies (p. 115), which send one usually† *unbranched* process (nerve-process) centralward, toward the nerve-fiber layer, one or more *branched* processes (dendrites) peripheryward, toward the inner reticular layer; there the processes divide and form delicate ramifications spread out parallel to the surface, in different planes, which construct a dense tangle with the processes of other ganglion cells (Fig. 322).

* A few of these cells are marked by their large size; such giant ganglion cells occur at tolerably regular intervals; "twin ganglion cells," united with each other by a short bridge, have been found in this layer; only one of the twin cells possesses a nerve-process.

† Recently collaterals have been found on a few nerve-processes, that turn back and envelop neighboring ganglion cells in their ramifications (Fig. 322).

The *inner reticular layer* ("neurospongium, granular layer") consists of a very delicate network of the supporting substance, which supports a dense nervous tangle formed of the processes of all the ganglion cells of the retina.

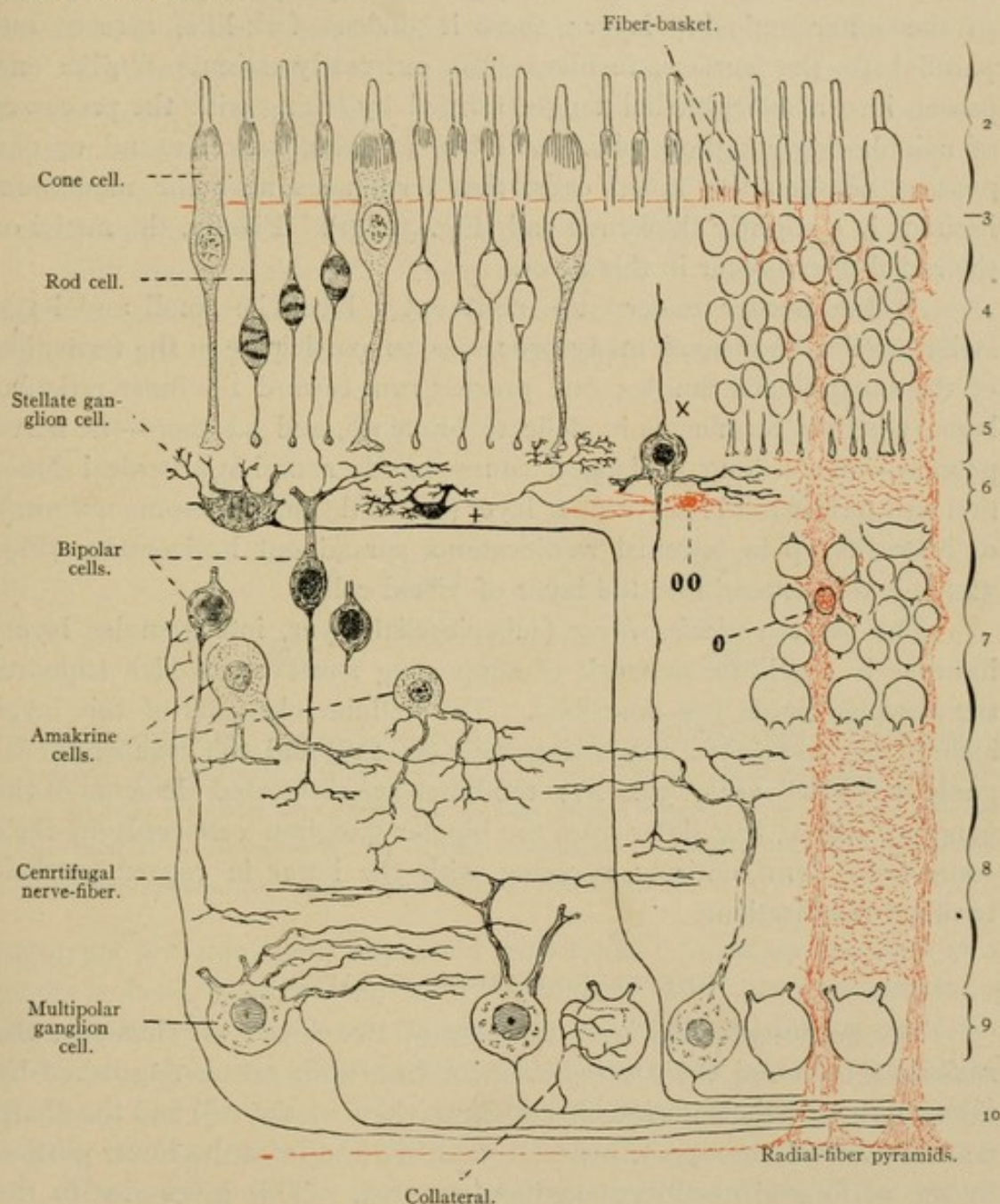


FIG. 322.—SCHEME OF THE HUMAN RETINA. Supporting substance red. O. Nucleated portion of radial fibers. Compare 2-10 with Figure 320.

The *inner granule layer* includes elements named "granules," that differ greatly in their nature. The innermost stratum consists of large ganglion cells, amacrine * cells, which send branched processes into the

* That is, without a long process; they were formerly called spongioblasts, because they were erroneously regarded as the producers of the "neurospongium."

inner reticular layer. The remaining strata, for the greater part, are composed of small bipolar ganglion cells ("ganglion retinae"), the central process of which extends into the inner reticular layer and there breaks up into delicate varicose branches, while the peripheral process passes to the outer reticular layer; there it divides fork-like, spreads out parallel to the surface, resolves into extremely minute fibrillae and passes into a subepithelial tangle formed by felting with the processes of neighboring ganglion cells.* All bipolar ganglion cells send up one process between the visual cells, that terminates near the membrana limitans in a slightly thickened end (Fig. 322 x). Finally, the nuclei of the radial-fibers occur in this layer.

At the border toward the next outer layer lie small and large stellate cells; they send many processes to participate in the formation of the subepithelial tangle; one process runs toward the inner reticular layer, where it terminates in delicate branches, and another—the nerve process—after a long horizontal course bends round in a vertical direction and passes to the nerve-fiber layer (this is disputed by some authors), or it breaks up in terminal ramifications spread out horizontally (Fig. 322 +), that extend into the layer of visual cells.

The *outer reticular layer* (subepithelial layer, intergranular layer) likewise is a delicate network of supporting substance, which supports the nervous tangle just described. The cellular elements of this layer include the concentric supporting cells (p. 408) and the "subepithelial ganglion cells" (Fig. 322 x); the latter are dislocated elements of the ganglion retinae, that differ from the bipolar ganglion cells only in their compressed form, entirely agreeing with the latter in regard to their terminal ramifications.

THE NEURO-EPITHELIAL LAYER.

The neuro-epithelial layer consists of two kinds of elements, the *rod-visual cells* and the *cone-visual cells*, that both are distinguished by the situation of their nucleus in the lower half of the cell and the sharp demarcation of the upper, nonnucleated division from the lower portion by the perforated membrana limitans externa. This gives rise to the appearance of different layers; the inner, nucleated portion of the visual cells being known as the outer granule layer, the outer nonnucleated division as the layer of rods and cones. Between these two lies the limiting membrane.

* Latterly it has been attempted to distinguish two forms of ganglion cells, of which the one stands in relation to the rod-visual cells, the other to the cone-visual cells; the differences are very insignificant.

The rod-visual cells. The outer halves of these elements are the *rods*, slender cylinders ($60\ \mu$ long, $2\ \mu$ thick), which consist of a homogeneous outer segment and a finely granular inner segment. The outer segment is the exclusive seat of the *visual purple*. The inner segment possesses in its outer end an ellipsoidal, fibrillated body, the *fiber-apparatus*. The inner halves of the rod-visual cells are named *rod-fibers*; they are exceedingly delicate filaments, which are provided with a nucleated expansion, the *rod-granule*. The nucleus is marked by from one to three clear, transverse bands. The basal end of the cell terminates in a minute process-free, club-shaped expansion (Fig. 322).

The cone-visual cells. The outer halves of these cells, the *cones*, likewise consist of an outer segment and an inner segment. The outer segments are conical and shorter than those of the rods. The inner

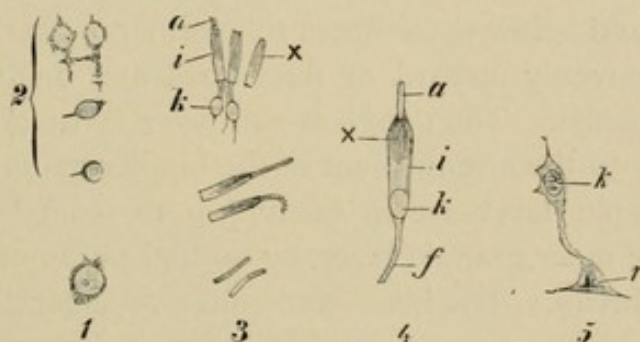


FIG. 323.—ISOLATED ELEMENTS OF THE RETINA OF AN APE. $\times 240$. 1. Mutilated ganglion cell of the ganglion of the optic nerve. 2. Elements of the inner granule layer. 3. Rod-visual cells and fragments of the same; below, two outer segments, one of which exhibits transverse striation, the beginning of a disintegration into transverse platelets; above are two rods, the outer segment of the lower one falling apart. Uppermost are more complete rod-cells; *a*, outer segment; *i*, inner segment; *k*, rod-granule; *x*, fiber-apparatus. 4. Cone-visual cell: *a*, outer segment; *i*, inner segment; *k*, cone-granule; *f*, cone-fiber, torn at lower end; *x*, fiber-apparatus. 5. Radial fiber; *k*, nucleus of the same; *r*, pyramidal base. Technic No. 181.

segments are thick and expanded pouch-like; therefore the cone as a whole is flask-shaped. The inner segment of the cones also contains a fiber-apparatus. The inner halves of the cone-visual cells are the *cone-fibers*; these are broad and rest with an expanded pyramidal foot on the outer reticular layer. The nucleated enlargement, the *cone-granule*, usually lies immediately to the inner side of the *membrana limitans*.

The number of the rods is much greater than that of the cones. The latter occur at regular intervals, so that three or four rods always lie between two cones (Fig. 320).

The basal portions of the visual cells, resting upon the outer reticular layer, usually are distinctly recognized as a special, radially striated layer (Fig. 320), *Henle's fiber-layer*; in the territory of the macula lutea (see below), this fiber-layer is particularly broad and gradually diminishes—often very unsymmetrically—toward the ora serrata.

The *pigmented epithelium* consists of a simple layer of hexagonal cells, which on their outer surface, that directed toward the choroid, where the nucleus lies, are free from pigment (Fig. 321), while their inner division contains numerous rod-shaped, brown pigment-granules ("fuscin"), from 1 to 5 μ long. From this inner division numerous delicate processes extend between the rods and cones. In albinos and on the tapetum (p. 402) the epithelium is free from pigment.

In the macula lutea and fovea centralis, also in the ora serrata, the structure of the retina above described presents noteworthy modifications.

The macula lutea and fovea centralis. In the territory of the macula the layers of the retina exhibit the following variations: Delicate fibers of the optic nerve (the so-called papillo-macular bundle) run from the optic entrance directly to the adjacent median portion of the macula; above and below these fibers thicker nerve-fibers run from the optic entrance convexly upward or downward and unite at the lateral margin of the macula. The ganglion-cell layer is greatly increased in thickness, owing to the arrangement of the bipolar ganglion cells, which instead of a simple layer are in many (up to nine) layers over one another; also the inner granule layer, by multiplication of its elements, is almost twice as broad. The inner and outer reticular layers suffer no essential change. The neuro-epithelial layer is composed of the here somewhat smaller cone-visual cells alone. Already at the margin of the macula the rod-visual cells diminish in number and within the macula they are wanting altogether; as a result the cone-fibers are visible in a wide extent; here they alone form the fiber-layer of Henle. The cone-granules, on account of their large number, lie in several rows one above the other. The radial-fibers no longer stand vertically to the thickness of the retina, but obliquely toward the fovea.

Toward the *fovea centralis*, situated in the middle of the macula, the layers of the retina become gradually thinner and are in part totally suspended. With the exception of a few delicate fibers, the nerve-fiber layer first disappears; then the cerebral layers blend with one another in a thin layer. In the center of the fovea (*fundus foveæ*) the neuro-epithelial layer (cone-cells) alone is present. The decrease in the layers differs individually, so that the form of the fovea is sometimes flat, sometimes deep with steep borders.*

A diffuse yellow pigment, soluble in alcohol, saturates the macula and the fovea.

* The latter form is shown by the fovea represented in Fig. 324.

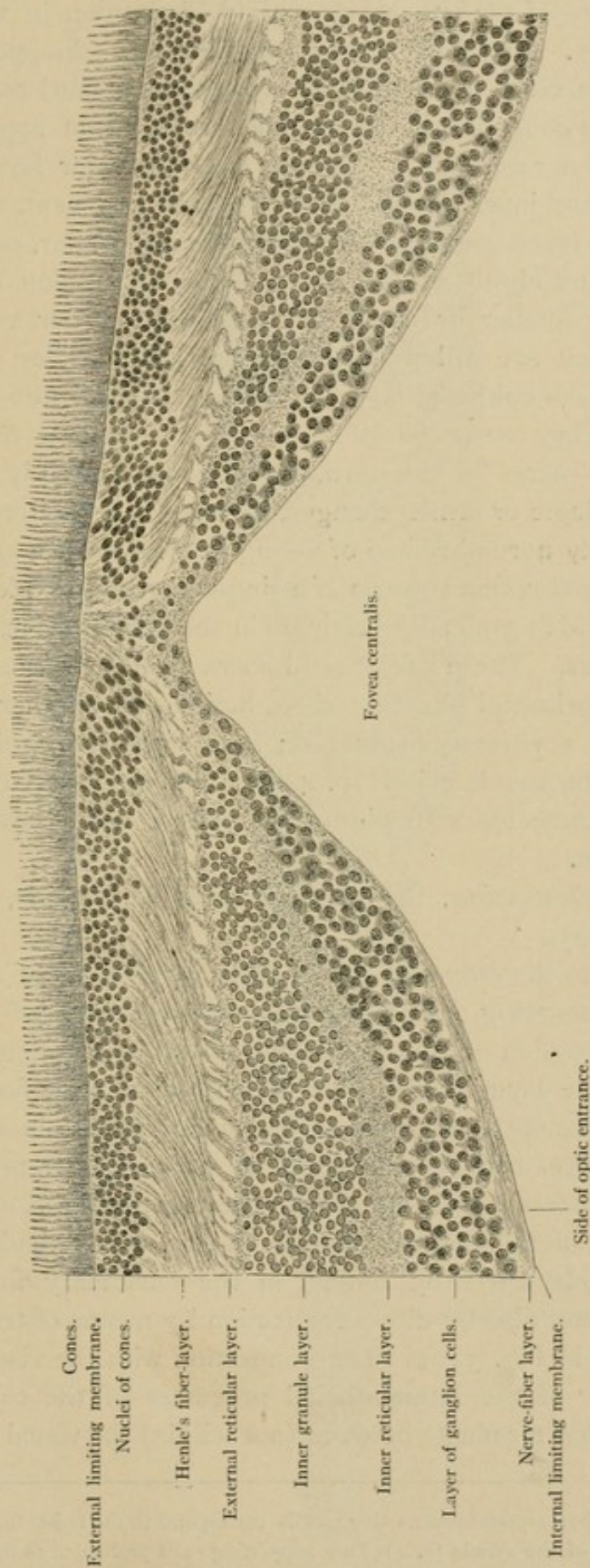


FIG. 324.—HORIZONTAL SECTION THROUGH THE MACULA AND THE FOVEA OF A MAN SIXTY YEARS OLD.—(After Schaffer.) $\times 135$. The nerve-fiber layer, like all the layers, is thicker on the side toward the entrance of the optic nerve than on the opposite side; in the latter situation the nerve-fibers are seen in transverse section as minute dots. The section is not through the exact center of the fovea, for there are only cone-visual cells and no remnants of the confluence of the inner granule and ganglion-cell layers are present.

In the territory of the *ora serrata* a rapid diminution in the retinal layers takes place. Optic fibers and ganglion cells disappear before reaching the *ora serrata*. Of the visual cells the rod-visual cells are the first to vanish; the cone-visual cells are still retained but appear to be deprived of their outer segment. Then the outer reticular layer is lost, so that the outer and inner granule layers become confluent, and finally the inner reticular layer ceases. The radial fibers of Müller, on the contrary, persist and are highly developed. [Within the region of the *ora serrata* commonly smaller or larger clefts or even rather voluminous spaces occur, which are called *vacuoles* (Fig. 325). They are either confined to the neuro-epithelial layer or extend centrally into the inner reticular layer. They are probably filled with a lymphatic fluid. The meaning of these spaces is unknown, but they are certainly not to be regarded as pathologic or senile changes, because they are rather common in the perfectly normal retinae of young individuals.—EDITOR.]

The *pars ciliaris retinae* consists of a simple layer of slender cylinder cells (Fig. 325), which gradually originate in the blended visual-cell and inner granule layers. These cells * send fibers from their inner surface, that extend in a horizontal direction close beside one another and have the appearance of a vitreous membrane; farther front toward the lens these fibers form the zonula ciliaris (p. 419). The outer surface of these cylinder cells is connected with pigmented cells, a continuation of the pigment epithelium.

The *pars iridica retinae*, the pigment layer of the iris, has been described (*cf.* p. 405).

With regard to the *connections of the nervous elements of the retina*, according to the foregoing description the nerve-processes of the ganglion cells of the ganglion of the optic nerve, as well as a few stellate cells of the inner granule layer (?) furnish the centripetal optic fibers, while the centrifugal nerve-fibers terminate in free endings in the inner granule layer. The ganglion cells of the ganglion retinae apparently do not possess a nerve-process; their union with the other nervous elements is effected by means of the nervous tangles in the two reticular layers, and not only as elsewhere by contact in the customary manner (*cf.* remark†, p. 118), but also by direct connection by means of true anastomoses (not shown in Fig. 322). The connection with the visual cells is effected by means of the intraepithelial processes of the cells of the ganglion retinae, that terminate between (not within) the visual elements.

* According to other representations these cells correspond to the radial fibers (p. 407), in which case the fibers of the zonula (p. 419) are supporting cells prolonged to the lens.

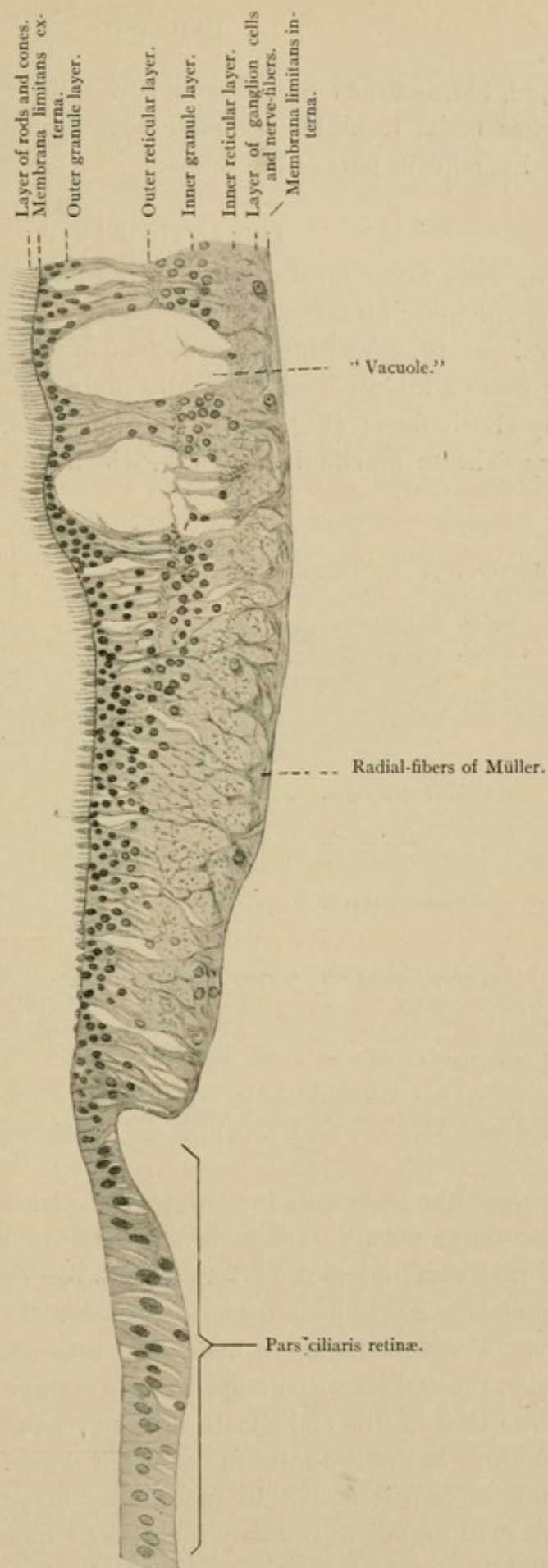


FIG. 325.—MERIDIONAL SECTION OF THE ORA SERRATA AND THE ADJACENT PORTION OF THE PARS CILIARIS RETINÆ OF A MAN THIRTY-SEVEN YEARS OF AGE. $\times 180$.—(Schaper.)

(Fig. 322, left, between the second and third rod-fiber.) Physiologic researches make it highly probable that the visual cells constitute the essential percipient part of the retina.

THE OPTIC NERVE.

The *optic nerve* in its entire intraorbital course is enveloped in sheaths which are processes of the cerebral membranes. Outmost is the dural sheath, consisting of firm connective-tissue bundles, externally having more a longitudinal, internally more a circular arrangement, and of many elastic fibers (Fig. 326); following this, toward the interior, is the very delicate arachnoidal sheath, which sends numerous, branched

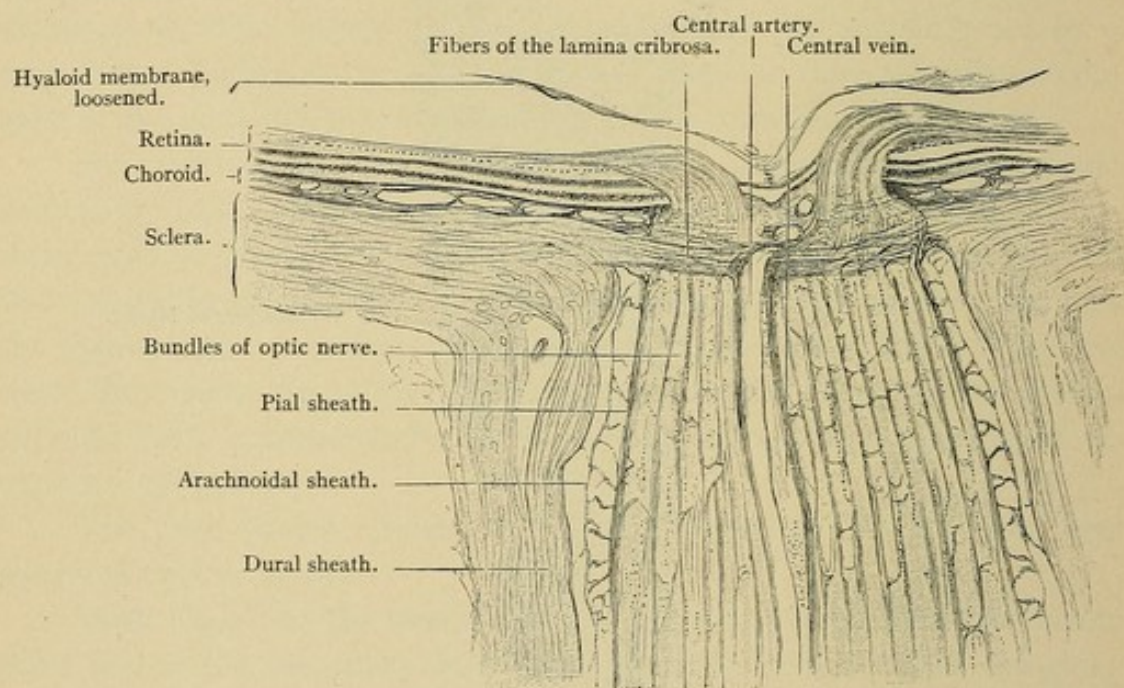


FIG. 326.—LONGITUDINAL SECTION OF THE OPTIC ENTRANCE OF A HUMAN EYE. $\times 15$. Above the lamina cribrosa the narrowing of the optic nerve is visible. The central artery and vein have been for the most part cut longitudinally, but above at several points transversely. Technic No. 177 d.

connective-tissue trabeculæ inward to the pial sheath, while the union with the dural sheath is established by a few tight fibers. Innermost lies the pial sheath, which closely invests the optic nerve and sends off numerous lamellæ, which form sheaths for the individual nerve-fiber bundles. These lamellæ are connected with one another by transverse trabeculæ, the resultant structure being a transverse lattice-work.

The tissue of the pial sheath does not penetrate within the nerve-fiber bundles, but only forms an outer envelope for them. The nerve-fiber bundles consist of delicate medullated fibers without a neurilemma; they are held together by many long-rayed neuroglia cells. The same are found in greatest number on the surface of the optic nerve, are

numerous in the periphery of the fiber-bundles, and their processes spin themselves around each individual nerve-fiber. This creates a conspicuous distinction between these and the peripheral nerves, in which glia-cells are wanting. At the entrance of the optic-nerve into the eyeball the dural sheath passes into the sclera; the arachnoidal sheath at its anterior end resolves into fibers, so that the subdural space lying on its outer side communicates with the subarachnoid space on its inner side. The pial sheath blends with the sclera, which here is pierced with numerous holes for the nerve-fibers passing through it; this portion is very rich in elastic fibers and is called *lamina cribrosa*. The choroid also participates, though in a slight degree, in the formation of the lamina cribrosa. The nerve-fibers lose their medullary sheath at the optic entrance, consequently the entire nerve is considerably reduced in size, and bending round spread out radially on the inner surface of the retina. At the turning point the fibers form a ring-like wall around the blood-vessels entering from the axis of the optic nerve (*papilla nervi optici*), that gradually levels towards the periphery (Fig. 326). The funnel-shaped depression encircled by the papilla varies greatly in size and is called the physiologic excavation of the optic nerve.

The central artery and central vein of the retina lie in the axis of the distal half of the optic nerve; the connective tissue enveloping these vessels is freely connected with the pial sheath, as well as with the lamina cribrosa.

THE LENS.

The more intricate structure of the lens can be understood only by considering its developmental history. The lens arises by constriction from the outer germ-layer and then represents a hollow vesicle formed of a simple layer of cylinder epithelial cells. The cells of the anterior wall of this vesicle, by a trifling alteration in their form, become the *lens epithelium*; the cells of the posterior wall grow out to long *lens-fibers*, the number of which undergoes considerable augmentation by repeated division of the cells situated at the equator of the lens-vesicle. Eventually the lens-fibers fill the entire cavity, so that the lens then represents a solid body, in its chief bulk consisting of lens-fibers—they are collectively designated the *substantia lentis*—and only on its anterior surface covered by the lens epithelium, which at the equator, by gradual elongation of its elements, is transformed into the lens-fibers. The whole is enveloped in the *lens capsule*, which perhaps is exclusively developed from the epithelial lens-vesicle.

In the *substantia lentis* a soft cortical substance and a firm core may be distinguished. It consists entirely of epithelial cells, greatly extended in length, named the *lens-fibers*. They have the form mostly of hexagonal, prismatic bands, that at one or both ends possess a bulbous enlargement. Three varieties are distinguished: central fibers, transition fibers,

and chief fibers. The central fibers are nonnuclear, possess waved or dentated edges, and are centered toward the axis of the lens. The transition fibers have also lost their nucleus. These two varieties form the core of the lens. The chief fibers form the major part of the substantia lentis and are distinguished by smooth edges and an oval nucleus lying in the neighborhood of the equator. All the fibers are bound to one another by a small quantity of cement substance, that is more abundant at the anterior and posterior poles of the lens and in maceration experiments leads to the formation of the so-called anterior and posterior lens-stars. All lens-fibers run in a meridional direction, beginning at the anterior lens-star, to the posterior lens-star; but no lens-fiber embraces the entire half of the lens: the nearer the anterior pole a fiber arises,

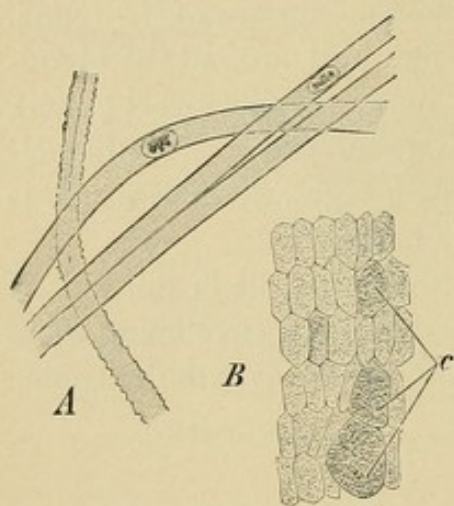


FIG. 327.—LENS-FIBERS OF A NEWBORN INFANT. A. Isolated lens-fibers, three with smooth, one with dentated borders. $\times 240$. Technic No. 187. B. Human lens-fibers cut transversely; c, section through club-shaped ends. $\times 560$. Technic No. 188.

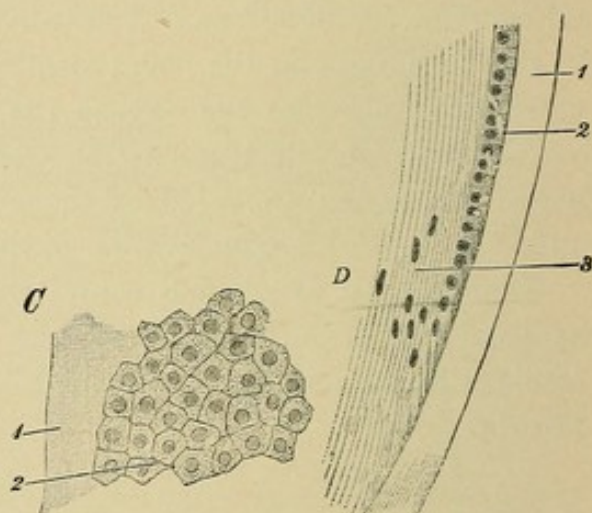


FIG. 328.—CAPSULE AND EPITHELIUM OF A LENS OF ADULT MAN. C. Inner aspect. $\times 240$. Technic No. 189 a. D. Lateral aspect, from a meridional section through the equator of the lens; 1, capsule; 2, epithelium; 3, lens-fibers. $\times 240$. Technic No. 189 b.

the more remote from the posterior pole it finds its terminus. The chief fibers are arranged in radial lamellæ,* the number of which in adult man exceeds 2000.

The *lens epithelium* is composed of a simple layer of cubical cells, low at the anterior lens-pole (2.5μ), becoming gradually higher toward

* In the lower vertebrates and in rodents among the mammals (e. g., in the squirrel) the lamellæ are of great regularity; in apes and man, on the other hand, very irregular. Also the transverse section of the lens-fibers in the latter are marked by their great irregularity. In this we detect the expression of a greater elasticity and adaptability of the whole lens, that thereby is peculiarly fitted to respond to the demands of accommodation. It is well known that the amplitude of accommodation in man and apes is very much greater than in the other mammals. Regarding the erroneous account of the construction of the lens out of concentric lamellæ, see Technic No. 188.

the equator ($-10\ \mu$), which extend over the anterior surface of the lens up to the equator; back of the equator the epithelial cells are arranged in meridional rows,* which is owing to the cell divisions and cell displacements that take place at the equator. At the posterior ends of these rows the epithelial cells gradually elongate and form the lens-fibers.

The *lens capsule* in man is a crystal clear, elastic membrane, from 6.5 to $25\ \mu$ thick anteriorly, from 2 to $7\ \mu$ thick posteriorly.

THE VITREOUS BODY.

The ectodermal vitreous body, ectodermal because probably originating from the anlage of the retina, consists of a fluid substance, the *vitreous humor*, and of fibers, which extend through the fluid in all directions.† The surface of the vitreous body is enveloped in a very resistant, structureless membrane, the *hyaloid membrane*, that anteriorly continues as the vitreous membrane of the pars ciliaris retinæ (p. 414) and in certain localities contains scanty fibrils, as well as a few cells. Of the latter two forms may be distinguished: (1) round cells, resembling leucocytes; (2) stellate and fusiform cells. Cells containing clear vacuoles probably are forms undergoing degeneration.

Regarding the hyaloid canal, see p. 420.

THE ZONULA CILIARIS.

In a zone situated immediately anterior to the ora serrata delicate, homogeneous fibers arise‡ from the inner surface of the hyaloid membrane, as well as from the cells of the pars ciliaris retinæ, which pass into the depressions between the ciliary processes and toward the lens, where they find their attachment, anteriorly, posteriorly, and at the equator, on the lens capsule. These fibers in their totality form a membrane, nowhere of perfect continuity, the *zonula ciliaris*, the *radial girdle*, the anchoring structure of the lens. The communicating spaces occurring between the posterior zonula fibers and the anterior surface of the vitreous body are named *spatia zonularia* (canalis Petiti).§ The spatia are not entirely closed toward the posterior chamber of the eye.

* In man the rows are short and not so regular as, for example, in the cow and the hog.

† A regular arrangement of the tissues of the vitreous body is very difficult to demonstrate; but recent pathologic-anatomic discoveries give strong evidence that the fibers are extended somewhat after the manner of the septa of an orange.

‡ A few zonula fibers originating or ending in the vitreous body support the genetic homogeneity with the vitreous body.

§ Other authors name the triangular space occurring between the zonula fibers going to the anterior and posterior surfaces of the lens capsule the canal of Petit, which is incorrect, be-

THE BLOOD-VESSELS OF THE EYEBALL.

The blood-vessels of the eyeball are separated in two sharply defined territories, which are in communication only at the entrance of the optic nerve.

I. *Territory of the vasa centralia retinae* (Fig. 329).—The *central artery of the retina* (*a*), at a distance of from 15 to 20 millimeters from the eyeball, enters the axis of the optic nerve and runs within it to the surface of the optic entrance. Here it divides into two main branches, of which the one is directed upward, the other downward, each of which, subdividing, supplies the entire pars optica retinae to the ora serrata. During its course in the optic nerve the artery gives off numerous small branches, which enclosed in the processes of the pial sheath run between the nerve-fiber bundles and anastomose with small arteries (*b*) that have entered the sheaths of the nerve from the surrounding adipose tissue and also with twigs of the short ciliary arteries (at *c*). In the retina itself the artery breaks up into capillaries, which extend into the outer reticular layer.* The veins proceeding from the capillaries run parallel with the branches of the arteries and finally unite in the *vena centralis retinae*, likewise enclosed in the axis of the optic nerve (Fig. 329, *a'*).

In the embryo a twig from the central artery of the retina, the *hyaloid artery*, passes through the vitreous body to the posterior surface of the lens. The artery atrophies before birth, but the canal which transmitted it may still be found in the vitreous body of the adult; it is called the *hyaloid canal*, or *Cloquet's canal*.

II. *Territory of the vasa ciliaria*.—This territory is characterized by the complementary veins taking a course entirely different from that of the arteries.

Of the *arteries*, the short ciliary arteries (Fig. 329, Roman numerals) supply the smooth portion of the choroid, while the long ciliary arteries (Fig. 329, Arabic numerals) and the anterior ciliary arteries (Fig. 329, Greek letters) are primarily destined for the ciliary body and the iris.

The branches, about twenty, of the *short ciliary arteries* (*arteriæ ciliares posticæ breves*) penetrate the sclera in the vicinity of the optic entrance (I); after giving off twigs (II) which supply the posterior half

cause the zonula fibers approaching the anterior and posterior surfaces of the lens form no membranes; the fibers interlace in such manner that one portion of the fibers approaching the anterior surface of the lens come from behind, while fibers proceeding to the posterior surface come from in front.

* Only the cerebral layer of the retina is vascular; in the fundus foveæ centralis the cerebral layer is absent and with it the vessels.

of the surface of the sclera, the arteries break up into a narrow-meshed capillary network, the *lamina choriocapillaris* (III). At the optic entrance

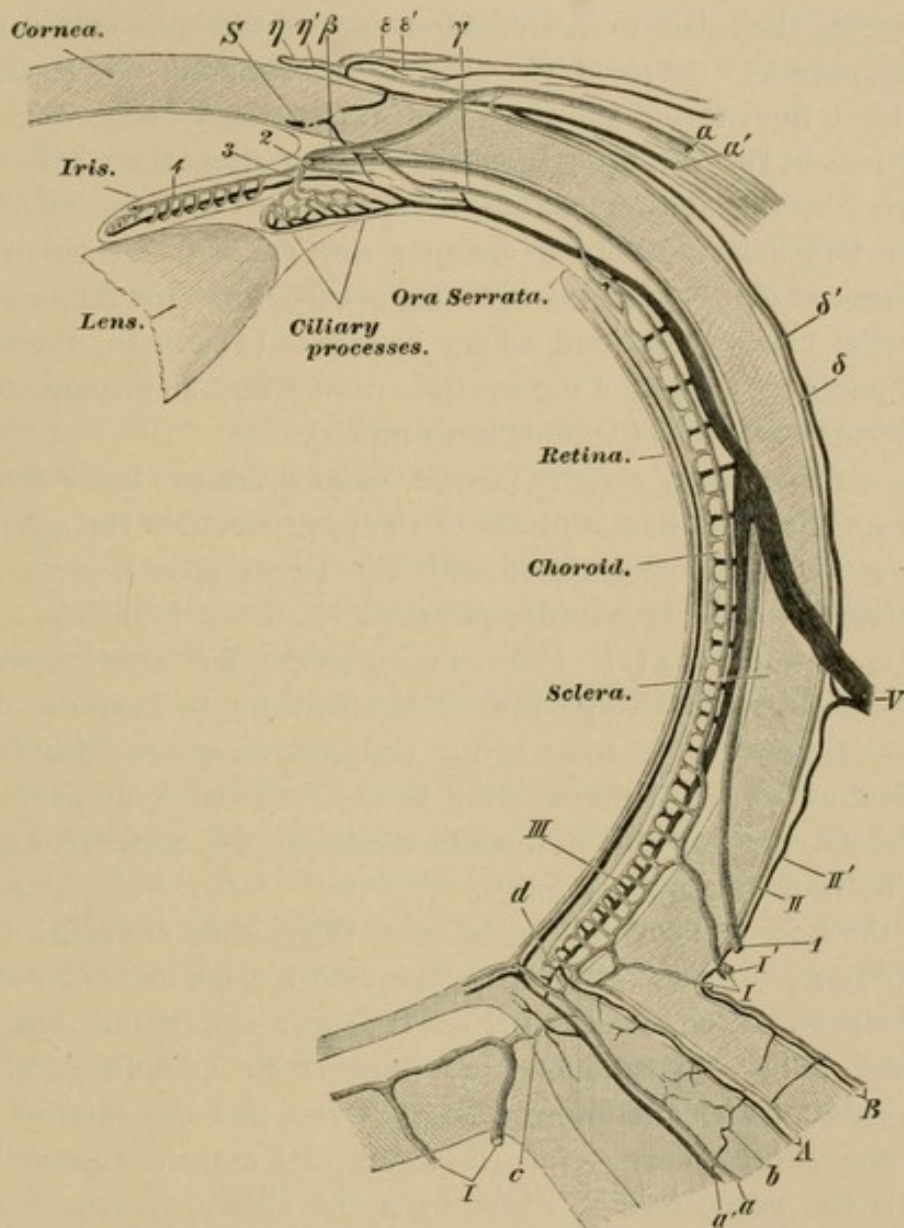


FIG. 329.—SCHEME OF THE VESSELS OF THE EYE, ACCORDING TO LEBER. External tunic stippled, middle tunic white, internal tunic and optic nerve stippled crisscross. Arteries light. Veins dark.
Territory of the central vessels of the retina (small Italic letters): *a*, artery, *a'*, vein, central of retina; *b*, anastomosis with vessels of the sheath; *c*, anastomosis with branches of the posterior short ciliary arteries; *d*, anastomosis with choroidal vessels.
Territory of the vessels of the sheath (large Italic letters): *A*, inner, *B*, outer vessels of the sheath.
Territory of the posterior short ciliary vessels (Roman numerals): *I*, artery, *I'*, vein (short posterior ciliary); *II*, episcleral arterial, *II'*, episcleral venous branches of the same; *III*, capillaries of the choriocapillaris.
Territory of the posterior long ciliary vessels (Arabic numerals): 1, posterior long ciliary artery; 2, circulus iridis major cut transversely; 3, branches to the ciliary body; 4, branches to the iris.
Territory of the anterior ciliary vessels (Greek letters): *α*, artery, *α'*, vein (anterior ciliary); *β*, connection with the circulus iridis major; *γ*, connection with the choriocapillaris; *δ*, arterial, *δ'*, venous episcleral branches; *ε*, arterial, *ε'*, venous branches to the scleral conjunctiva; *η*, arterial, *η'*, venous branches to the corneal limbus.
V, vena vorticiosa. *S*, cross-section of the venous sinus of the sclera.

the arteries anastomose with branches of the arteria centralis retinae (Fig. 329, *c*) and in this way form the *circular artery of the optic nerve* (circulus arteriosus nervi optici); at the ora serrata they anastomose with recurrent

twigs of the long ciliary and of the anterior ciliary arteries (for the latter anastomosis see Fig. 329, r).

The two *long ciliary arteries* (arteriæ ciliares posticæ longæ) (1) likewise penetrate the sclera in the neighborhood of the optic entrance; the one artery passes to the nasal, the other to the temporal side of the eyeball, between the choroid and the sclera to the ciliary body, where each artery divides in two diverging branches running along the ciliary margin of the iris; by the anastomoses of these branches with the branches of the other long ciliary artery a vascular ring (2) is formed, the *larger arterial circle of the iris* (circulus iridis major), from which numerous twigs arise for the ciliary body and ciliary processes (3) and for the iris (4). Near the pupillary margin of the iris the arteries form an incomplete ring, the *smaller arterial circle* (circulus iridis minor).

The *anterior ciliary arteries* (arteriæ ciliares anticæ) come from the arteries supplying the recti muscles of the eye, penetrate the sclera near the corneal margin, communicate with the larger arterial circle of the iris (β), supply the ciliary muscle, and send recurrent branches to unite with the choriocapillaris (γ). Before the anterior ciliary arteries penetrate the sclera they give off twigs toward the *back* for the anterior half of the sclera (δ), toward the *front* to the conjunctival sclera (ϵ) and to the corneal limbus (η). The cornea itself is non-vascular, only at the margin, in the anterior lamellæ of the substantia propria, is there a circumferential network of capillary loops.

All the *veins* run toward the equator, where they converge to four (more rarely five or six) small stems, the whorl veins or *venæ vorticosæ*, which forthwith pierce the sclera (Fig. 329) and empty into one of the ophthalmic veins. In addition there are small complemental veins that run parallel with the short ciliary arteries and the anterior ciliary arteries, the short ciliary veins (Fig. 329, I') and the anterior ciliary veins (a'); the latter receive twigs from the ciliary muscle, from the episcleral vascular network (Fig. 329, δ'), from the conjunctival sclera (ϵ'), and from the circumferential capillary loops of the cornea (η'). The episcleral veins also communicate with the *venæ vorticosæ*, at the equator (at V). Finally the anterior ciliary veins communicate with the *sinus venosus scleræ* (Schlemm) (S). This is a *venous wreath* encircling the cornea, that, lying within the sclera, still possesses completely closed walls.* It takes up small veins from the capillary network of the ciliary muscle.

* The communication with the anterior chamber of the eye formerly described is factitious; the assertion that such communication existed was based on the fact that colored fluids injected into the anterior chamber pass over into the venous wreath by filtration.

THE LYMPH PATHS OF THE EYEBALL.

The eye possesses no proper lymph-vessels, but a series of intercommunicating lymph spaces. Two complexes of such spaces can be distinguished in the eye, occupying an anterior and a posterior territory. The anterior territory comprises :—

1. The *juice canaliculi* of the *cornea* and the *sclera*.
2. The *anterior chamber* of the eye, which, by means of the capillary cleft between the iris and the lens, communicates with—
3. The *posterior chamber* of the eye. The latter is in open connection with—
4. The *spatia zonularia*.

The last three spaces stand in close relation to one another and may be injected from the anterior chamber.

The posterior territory includes :—

1. The *hyaloid canal* (*canalis hyaloideus*) (p. 420); also
2. The *intervaginal lymph space*, that is, the subdural and the subarachnoid space of the sheath of the optic nerve.
3. The narrow cleft between the choroid and the sclera: the *perichoroidal space*.
4. The *spatium interfasciale* (Tenon) which extends on the dural sheath of the optic nerve, as the supradural space, to the optic foramen.

These spaces may be injected from the subarachnoid space of the brain. The content of these spaces is a filtrate from the vessels, which also saturates the vitreous body. The quantity of fluid in the perichoroidal space, also in the interfascial space, normally is exceedingly minimal. Both these spaces serve to facilitate the movements of the choroid and of the eyeball and may be regarded as synovial spaces.

THE NERVES OF THE EYEBALL.

The nerves of the eyeball penetrate the sclera in the circumference of the entrance of the optic nerve and run forward between the sclera and the choroid; after giving twigs provided with ganglion cells to all the choroidal vessels, they form upon the ciliary body a ring plexus intermingled with ganglion cells, the *ciliary ganglionic plexus* (*plexus gangliosus ciliaris*), from which branches arise for the ciliary body, the iris, and the cornea. The *nerves of the ciliary body* terminate in delicate, pointed ends on the blood-vessels and on the ciliary muscle, partly between the muscle-bundles of the ciliary body in the form of branched terminal trees, which perhaps subserve the muscular sense, and partly on the scleral surface of the ciliary body in the form of a delicate plexus.

The medullated *nerves of the iris* form networks and lose their medullary sheath as they pass to the pupillary margin; their terminal ramifications are in part distributed to the sphincter and dilatator muscles and to the vascular walls, while another portion forms a sensory plexus lying close beneath the anterior iridal surface. Ganglion cells are wanting in the iris of man and of mammals.

The *nerves of the cornea* first enter the sclera and form a circular plexus, the *plexus annularis*, surrounding the corneal margin, from which branches arise for the conjunctiva and for the cornea. In man the twigs in the conjunctiva terminate in spherical end-bulbs (p. 222), lying close under the epithelium; they are also found in the substance proper of the cornea, for a distance of from one to two millimeters within the corneal limbus. The corneal nerves lose their medullary sheath after entrance in the substantia propria, and as naked axis-cylinders penetrate the entire cornea. They form networks, which according to the plane they

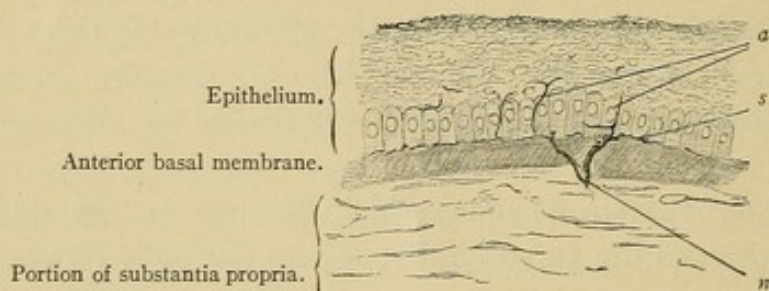


FIG. 330.—FROM A VERTICAL SECTION THROUGH THE HUMAN CORNEA. $\times 240$. *n*. A dividing nerve penetrating the anterior basal membrane; *s*, subepithelial plexus beneath the cylindrical cells; *a*, fibers of the intraepithelial plexus ascending between the epithelial cells. Technic No. 186.

occupy are described as the *stroma-plexus*, which lies in the deeper strata of the cornea; the *sub-basal plexus*, which is situated beneath the anterior basal membrane; the *subepithelial plexus*, which lies close under the epithelium. From the latter plexus exquisitely delicate nerve-fibrillæ ascend into the epithelium between its elements and form the exceedingly fine *intraepithelial plexus*, the ramifications of which terminate in free ends between the epithelial cells. The nerves found in the sclera form a plexus on the blood-vessels and in the lymph spaces, on which latter endings occur in the form of thickly branched structures. In addition free nerve-endings, like those in the dura, are found.

THE EYELIDS.

The eyelids, *palpebræ*, are folds of the external skin, which enclose muscles, loose and compact connective tissue, and glands. The outer leaf of the eyelid retains the usual character of the external skin; the inner leaf, that toward the eyeball, is considerably modified and is called

the *palpebral conjunctiva*. The external skin of the eyelid extends over the anterior free margin of the lid and does not pass into the palpebral conjunctiva until it reaches the posterior border, the *palpebral border*.

The construction of the eyelid is best studied in sagittal sections (Fig. 331). Counting from before backward the following strata are found:

1. The *external skin*, which is thin and beset with fine lanugo hairs,

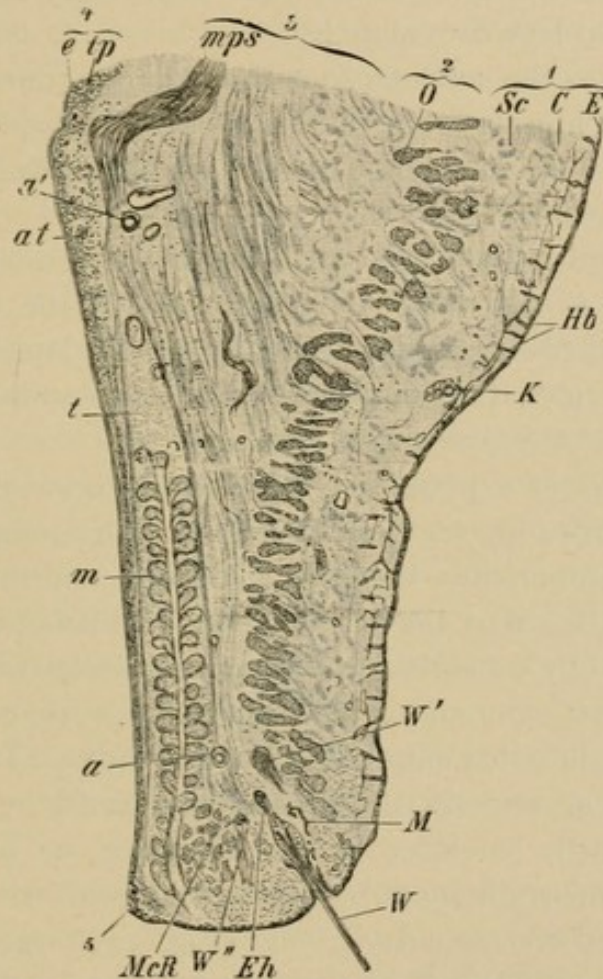


FIG. 331.—SAGITTAL SECTION OF THE UPPER EYELID OF A CHILD SIX MONTHS OLD. $\times 10$. 1. Integument: *E*, epidermis; *C*, corium; *Sc*, subcutaneous tissue; *Hb*, hair-follicles of lanugo hairs; *K*, coil-gland; *W*, eyelash, with the anlage of a new hair (*Eh*); *W'*, *W''*, portions of follicles of eyelashes; *M*, portion of a ciliary gland. 2. Territory of the orbicularis palpebrarum muscle: *O*, bundles of this muscle cut transversely; *McR*, tarsal muscle. 3. Expanded tendon of the levator palpebrarum superior; *mps*, superior palpebrarum muscle. 4. Conjunctival portion: *e*, conjunctival epithelium; *tp*, tunica propria; *at*, accessory tear-gland; *t*, tarsus; *m*, tarsal glands, the mouth of the excretory duct is not shown; *a*, transverse section of the arcus tarsus; *a'*, transverse section of the arcus tarsus externus. 5. Margin of the eyelid. Technic No. 191.

the follicles of which it encloses; in the corium small coil-glands are found, also pigmented connective-substance cells, that as is well known are of rare occurrence in the corium elsewhere. The subcutaneous tissue is very loose, rich in fine elastic fibers, poor in fat-cells, that may be entirely wanting. Near the border of the lid the corium is more compact and beset with more conspicuous papillæ. In the anterior edge of the margin of the lid two or three rows of robust hairs, the *cilia* (*W*),

are obliquely implanted, the follicles of which extend far into the corium. The cilia undergo rapid shedding; their length of life is said to be about from one hundred to one hundred and fifty days; consequently new hairs in all stages of development are frequently found among the eyelashes (*cf.* p. 383). The hair-follicles of the cilia are provided with small sebaceous glands, in addition to which they take up the excretory ducts of the *ciliary glands* (Moll) (M) which in their minute structure resemble the coil-glands, from which they differ only in having their lower end less convoluted.

2. Posterior to the subcutaneous tissue lie the transverse bundles of the cross-striated muscle-fibers of the *orbicularis palpebrarum muscle*; the portion of the muscle lying behind the cilia (McR) is named the tarsal muscle (Riolan).

3. Behind the muscle the expansion of the tendon of the levator palpebræ muscle is met, which is partly lost in the connective tissue present, the so-called fascia palpebralis, and partly attached to the upper margin of the tarsus*; the latter portion contains smooth muscle-fibers (*mps*), the *superior palpebral muscle* (Müller).

4. The *tarsus* is a plate of dense-fibered connective tissue, which gives firmness and support to the eyelid. It lies immediately in front of the palpebral conjunctiva, to which it belongs, and occupies the lower two-thirds of the height of the entire eyelid. In its substance the *tarsal glands* (Meibom) (*m*) are embedded, elongated bodies which consist of a wide excretory duct, opening on the palpebral border, and of little vesicles with short stalks, that empty into it on all sides. In their histology the tarsal glands agree with the sebaceous glands. At the upper end of the tarsus, partly enclosed in its substance, lie branched tubular glands, which in their minute structure coincide with the tear-glands and therefore are called *accessory tear-glands* (Fig. 331, *at*); they principally occur in the inner (nasal) half of the eyelid.

Behind the tarsus lies the *conjunctiva* proper, which consists of an epithelium (*e*) and a tunica propria (*tp*). The former is a stratified cylinder epithelium, with several strata of spherical cells in the depths and a stratum of mainly short cylindrical cells on the surface. The latter possess a narrow hyaline cuticular border. Goblet-cells also occur in varying number. At the posterior palpebral border the epithelium gradually passes into the stratified squamous variety, that occasionally extends far over on the palpebral conjunctiva. The lower portion of the

* In the lower eyelid the expansion of the inferior rectus muscle likewise contains smooth muscle-fibers, the *inferior palpebral muscle*.

palpebral conjunctiva is smooth. In the upper portion, on the contrary, the epithelium forms irregular pocket-like depressions, the "conjunctival recesses," that differ greatly in individual development and in sections, when highly developed, may resemble glands. The tunica propria of the conjunctiva consists of connective tissue, of lymphoid cells and plasma-cells in varying number. In animals, especially in ruminants, the latter form true nodules, the so-called *trachoma glands*, from the summit of which leucocytes wander through the epithelium to the surface; in man the migration of leucocytes also occurs but in a slighter degree. In the region of the conjunctival recesses the tunica propria is divided into papillæ by the above described depressions of the epithelium, hence the name "papillary body."

The palpebral conjunctiva passes from above (on the lower lid from below) over to the eyeball, the anterior surface of which it covers. At the turning point, the *fornix conjunctivæ*, a loose sub-conjunctival tissue consisting of connective-tissue bundles occurs under the tunica propria. The epithelium is the same as that on the palpebral conjunctiva; the tunica propria contains fewer leucocytes, but also in man normally possesses up to twenty small lymph nodules and a few mucous glands. The scleral conjunctiva is modified in so far that its stratified cylinder epithelium within a certain distance of the cornea is transformed into the stratified squamous variety, which continues in that of the cornea (*cf.* also Fig. 318).

The rudimentary *third eyelid* (*plica semilunaris*) consists of connective tissue and stratified squamous epithelium. The *caruncula lacrimalis* resembles the external skin in minute structure, only the stratum corneum is absent, and contains fine hairs, sebaceous and accessory tear-glands.

The *blood-vessels* of the eyelids proceed from branches approaching from the outer and inner angles of the eye, that form an arch, the *arcus tarseus* (Fig. 331, *a*), at the margin of the lid, and a second arch, the *arcus tarseus externus* (*a'*), at the upper end of the tarsus. Branches from these arches ramify in the skin, surround the tarsal glands, and penetrate the tarsus to supply a capillary network lying beneath the conjunctival epithelium; they also supply the fornix conjunctivæ, the scleral conjunctiva, and anastomose with the anterior ciliary arteries.

The *lymph-vessels* form a very dense network in the tarsal conjunctiva, a very thin network on the anterior surface of the tarsus. According to some authors the lymph-vessels of the scleral conjunctiva are closed at the corneal limbus; according to others they send minute canaliculi into the tissue of the cornea and through these are in communication with the juice-canal system.

The *nerves* form a very dense plexus in the tarsus and in the palpebral conjunctiva, which is characterized by a peculiar, coil-like, twisted arrangement of its fibers. One portion of the tarsal plexus surrounds the tarsal glands* and here consists of many nonmedullated and few medullated nerve-fibers; another portion terminates in the walls of the blood-vessels. From the "conjunctival plexus" medullated nerve-fibers arise, that run obliquely toward the margin of the lid and the palpebral conjunctiva, lose their medullary sheath, in part penetrate directly into the epithelium, where they ramify and terminate in free endings, also in end-bulbs (p. 222) lying close under the epithelium. These end-bulbs are found in large numbers not only in the papillæ of the margin of the lid and in the palpebral conjunctiva, but also in the ocular conjunctiva and in the margin of the cornea (*cf.* p. 424).

THE LACRIMAL ORGAN.

The *lacrimal gland* is a compound tubular gland provided with several excretory ducts. The excretory ducts (Fig. 332, *B*) are clothed

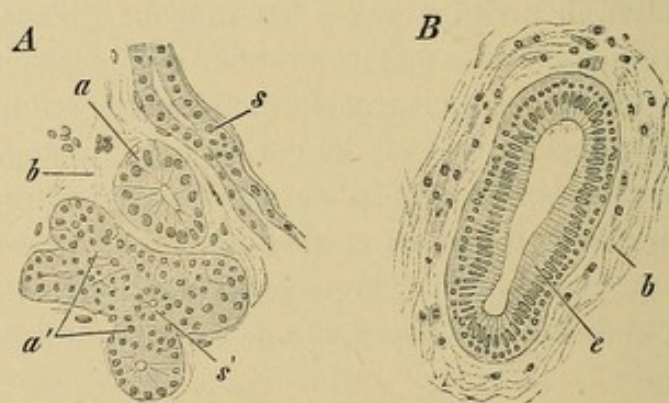


FIG. 332.—FROM A THIN SECTION OF A HUMAN LACRIMAL GLAND. $\times 240$. *A*. Gland-body; *a*, tubule cut transversely; *a'*, group of tubules, mostly cut obliquely, the lumen of only one tubule visible, below; *s*, intercalated tubule with cubical (above to the right) and flat (below to the left), epithelial cells; *s'*, intercalated tubule in cross-section, lined with moderately high cylindrical cells; *b*, connective tissue. *B*. Cross-section of an excretory duct; *c*, two-rowed cylindrical epithelium; *b*, connective tissue. Technic No. 192.

with a two-row cylinder epithelium and gradually pass into long intercalated divisions, narrow tubes clothed with low epithelium (Fig. 332 *A*, *s s'*). These finally continue in tubules that are clothed with two forms of cells and enveloped in a membrana propria. The gland cells of the one form in the replete state are tall, but when empty of secretion are considerably shorter. The secretion collecting center lies in the lumen half of the cell. The cells of the other form are low; the

* Whether nerve-fibers penetrate between the gland-cells has not yet been distinguished with certainty; probably the nerves of the tarsal glands behave like those of the glands of the mouth cavity (p. 247).

secretion balled together in large globular masses occupies the entire cell, except a small zone at the cell-base. Intercellular secretory capillaries, as well as secretion granules (the latter in the cat), have been demonstrated. Between the gland-cells and the *membrana propria* lie a few flat cells, extensions of the deep stratum of the epithelium of the excretory duct.

Blood-vessels and nerves behave as in the glands of the oral cavity, but the terminal ramifications of the latter are said to form an interepithelial net.

The walls of the *lacrimal canaliculi* consist of a stratified squamous epithelium, of a *tunica propria* rich in elastic fibers, beneath the epithelium also rich in cellular elements, and of cross-striped muscle-fibers, for the greater part running longitudinally.

The lacrimal sac and the naso-lacrimal duct consist of a two-rowed cylinder epithelium and of a *tunica propria* which is chiefly adenoid in character and separated from the underlying periosteum by a dense plexus of veins.

TECHNIC.

No. 177.—Carefully cut the fresh *eyeball* out of the optic cavity and secure as much as possible of the optic nerve; then with the scissors remove the attached fat and muscle and with a *sharp* razor make an incision at the equator, about 1 cm. long, through all the membranes of the eye. Then place the eyeball in 150 c.c. of potassium bichromate acetic acid solution (p. 32); after from twelve to twenty hours, beginning at the incision already made, divide the eyeball with the scissors completely into an anterior and a posterior half and change the fluid. After another twelve or twenty hours wash the pieces and harden them in 100 c.c. of gradually strengthened alcohols (p. 35).

(a) Carefully remove the lens from the anterior half of the eyeball and treat it further like No. 188; then cut out a quadrant and with the attached ciliary body and iris embed it in liver and cut sections through the *iris angle*. The thick sections are to be stained with Hansen's hematoxylin (p. 38) and mounted in xylol-balsam (Fig. 318).

(b) From the remaining three-fourths of the anterior half of the eyeball cut out a piece of the cornea, 5 or 10 mm. square, embed it in liver and make sections through the *strata of the cornea* (Fig. 313). The alternating lamellæ of the *substantia propria* can only be well seen in unstained sections mounted in dilute glycerol.

(c) From the posterior half of the eyeball cut pieces including the three tunics, 5 or 10 mm. square, and cut sections, not too thin, for the study of the *strata of the sclera and choroid* (Fig. 316). Stain them with Hansen's hematoxylin and mount in xylol-balsam. In sectioning, the retina usually becomes loosened.

(d) For preparations showing the *entrance of the optic nerve* cut around the point of entrance at a distance of about 5 mm. from the same

through all the tunics of the eye; embed this portion with about one centimeter of the optic nerve in liver and cut sections (not too thin). Place the knife so that it strikes the retina first, then the choroid and sclera, and passes through the optic nerve longitudinally; stain with dilute carmine (p. 39) and with Hansen's hematoxylin (p. 38), and mount in xylol-balsam. Examine with very low magnification (Fig. 326).

No. 178.—Remove a fresh eyeball according to the method given in No. 177, make an incision* at the equator and place it in from 100 to 200 c.c. of Müller's fluid (p. 33). In from twelve to twenty hours divide it with the scissors into an anterior and a posterior half. In two or three weeks carefully wash both halves in slowly running water for from one to two hours. Then cut out pieces including all the tunics, about 8 mm. on a side, and use for them the following preparations:—

(a) *Teased preparation of the choroid*.—Tease and mount a fragment in a drop of dilute glycerol; it exhibits large blood-vessels, capillaries of the choriocapillaris, branched pigment-cells, elastic fibers, sometimes also the lamina basalis; the "lattice-work" of the latter is often indistinct. Isolated membranes may be stained with Hansen's hematoxylin and mounted in xylol-balsam, but the more delicate structures are thus rendered indistinct (Fig. 317).

(b) *Elements of the retina*.—Carefully tease a small piece of the retina in a drop of Müller's fluid. Along with many fragments of the elements a few more or less well-preserved parts will be found. Human eyes have very large, beautiful cone-visual cells, while those of many mammals are very small; wholly unsuitable in this respect are the eyes of the rabbit; unfortunately, human eyes are usually no longer in a sufficiently fresh condition when the investigation is made. The outer segments of the cones, also of the rods, are extremely delicate and rapidly disintegrate after death, falling into transverse plates and at the same time curving like a shepherd's crook. Later they disappear entirely. In order to see beautiful cone-visual cells examine according to the method just given the eyes of fishes. (See further, No. 180 and No. 181.)

(c) The remaining parts of the eyeball are to be transferred from the water to 80 c.c. of gradually strengthened alcohols (p. 35) for hardening; when the hardening is completed cut out the iris, embed it in liver, and make meridional and equatorial sections; stain them in Hansen's hematoxylin (p. 38) and mount in xylol-balsam (Fig. 319).

(d) Cut out a portion 1 cm. long of the retina, including the *ora serrata*, which is macroscopically visible as a wavy line, embed it in liver, and make meridional sections; stain them in hematoxylin (p. 38) and mount in xylol-balsam (Fig. 325). The pictures are often very intricate.

(e) Treat in the same manner a piece of the *retina*, which is best taken from the posterior portion of the eye, where the optic-fiber stratum is thickest. The radial-fibers of Müller can be seen in their entire length only in accurate vertical sections (Fig. 320 and Fig. 321).

*The unopened eyeball can be put in the Müller's fluid for 2 or 3 weeks and, after washing, then divided and put into the alcohol.

(f) In the same manner treat meridional sections through the *macula* and *fovea*.^{*} It is not difficult to cut sections of the macula, but on the other hand very difficult to obtain satisfactory sections of the extremely delicate fovea. The retina should not be loosened from the choroid, but the two should be sectioned together.

No. 179.—*The retina, after Golgi*.—For this purpose *thick* retinæ are most suitable, therefore select the eyes of large animals. Divide the eye into an anterior and a posterior half, remove the vitreous body, and with forceps and scissors carefully dissect a piece of the retina from the choroid. Cautiously roll this piece into a cylindrical or spherical clump and dip it for one second in thin celloidin solution; expose it for a few seconds to the air, until the envelope of celloidin is somewhat stiffened, and then place the piece in the Golgi mixture (p. 21). (The object of this rolling in of the retina is to prevent the formation of precipitates on its surface.) Let the object remain in the Golgi mixture for from twelve to seventy-two hours, then transfer it for twenty-four hours to the silver solution (p. 46). Then repeat the procedure (p. 47). The impregnation occurs first, after twelve hours, in the rods and cones; after another twelve hours in the bipolar cells and the amakrines, later in the cells of the ganglion nervi optici and in the nerve-fibers, last in the supporting cells.

Potassium-bichromate-formol furnishes good results (p. 33). Fixation of rods and cones, also radial fibers, for 2 days and of nerve-cells for from 3 to 6 days in pure solution of potassium bichromate is better. Still better results are obtained by the vital methylene-blue staining (p. 42) but this requires great skill for correct orientation.

No. 180.—*Fresh elements of the retina*.—Select the warm eyes of animals just killed. Divide the eyeball at the equator and carefully remove the vitreous body from the posterior half; cut small pieces about 3 mm. square from the wholly transparent retina and tease gently in a drop of the vitreous humor; place two thin strips of paper one on either side of the preparation (p. 53), and apply a cover-glass. Isolated elements will be found only occasionally here and there; on the other hand, very good surface views are not infrequently obtained in which the rods and cones are perceptible in optical cross-section, the former as small, the latter as large circles. If at the same time a little piece of the pigmented epithelium has been transferred to the slide, the regular hexagonal cells of the same can be plainly seen with the low power. The light spots in these cells are their nuclei (Fig. 16). These cells are very unstable and soon lose their sharp contours; molecular motion of the pigment granules may be very frequently observed.

No. 181.—The best method for isolating the *elements of the retina*

^{*} Among mammals only the ape possesses a yellow macula and a central fovea; but the majority of mammals—insectivora and certain rodents excepted—have an “area centralis,” without yellow pigmentation, but similar in structure to the macula. A simple or multiple fovea is always present in birds and reptiles; a fovea has also been found in bony fishes.

is the following : Place the unopened eye,* freed from fat and muscle, in 1 per cent. osmium solution. In twenty-four hours cut the eye open at the equator and for maceration place it for two or three days in distilled water; then with scissors cut out a piece of the retina about 2 mm. square and tease it in a drop of water; the preparation may be stained under the cover-glass with picrocarmine (p. 53) and mounted in dilute glycerol. With the high power, in addition to many fragments the source of which is not always to be determined with certainty, elements like those pictured in Fig. 323 may be found.

No. 182.—*Corneal spaces and canaliculi*.—Select an eye as fresh as possible; of the eyes of animals, that of the ox is the most suitable; with a scalpel vertically applied scrape away the epithelium of the cornea; spray the denuded surface with distilled water; cut through the eye in front of the insertion of the ocular muscles and place the anterior segment containing the entire cornea down on the epithelial side; then with forceps and scalpel remove the ciliary body, the lens, and the iris, so that only the anterior portion of the sclera and the cornea remain, which are to be placed in 40 c.c. of a 1 per cent. solution of silver nitrate. The whole is then stood in the dark for from three to six hours, after which the object is transferred to 50 c.c. of distilled water and exposed to sunlight (see further, p. 45). Harden the object in 50 c.c. of gradually strengthened alcohols and cut horizontal sections, which are most easily obtained if the cornea is held over the left index-finger. It is best to take the sections from the posterior surface of the cornea, since the spaces and canaliculi are more regular there. The sections may be stained in Hansen's hematoxylin (p. 38) and mounted in xylol-balsam. The pictures are negative, the spaces and canaliculi white on a brown or brown-yellow ground (Fig. 314). Carefully examine the usually somewhat thinner margins of the section; in sections stained with hematoxylin the large nuclei of the fixed corneal corpuscles are a dull blue; the contours of the cells can seldom be perceived.

No. 183.—*Fixed corneal corpuscles by the gold method*.—The method described on page 47 is to be somewhat modified, as follows: Express the juice from a fresh lemon; filter it through flannel. Kill the animal,† cut out the cornea and place it for five minutes in the lemon juice, in which it becomes transparent; wash it in 5 c.c. of distilled water for one minute; transfer it to 10 c.c. of gold-chlorid (p. 23) solution and place it in the dark for fifteen minutes. Then with glass rods transfer the cornea to 10 c.c. of distilled water for one minute, then to 50 c.c. of dis-

* It is advisable to select the eyes of small animals—*e. g.*, a small water salamander (*triton tæniatus*)—in which the sclera is thin and allows the osmium solution to penetrate easily. For such an eye 1 or 2 c.c. of the solution will be sufficient. The form of the rods is quite different from those of mammals; they are thick and are provided with long outer segments; the cones are small.

† Frogs are especially recommended; their corneal canaliculi are very regular and their posterior corneal lamellæ easily detached.

tilled water, to which 2 drops of acetic acid have been added, and expose it to daylight; in from twenty-four to forty-eight hours the reduction is completed (*cf.* p. 48). The object is then placed in 10. c.c. of 70 per cent. alcohol (in the dark); on the following day cut out a little piece of the cornea, and with needle and scalpel placed at the edge separate thin lamellæ from the posterior surface; with a little attention this can be successfully done without much trouble. The lamellæ mounted in xylol-balsam furnish very beautiful pictures (Fig. 315).

No. 184.—Very beautiful preparations of the *corneal canaliculi* are obtained by the method of *Drasch*. The objects are not to be taken from the animal recently killed, but twelve or twenty-four hours after death, during which time the cadaver must be kept in a cool place. Small pieces of the cornea are to be cut out, about 6 mm. square, placed in 5 c.c. of 1 per cent. gold-chlorid solution plus 5 c.c. of distilled water, and stood in the dark for one hour; during this time frequently stir the fluid with a glass rod. With glass rods transfer the pieces to 30 c.c. of distilled water, in which they should remain (in the dark) for from eight to sixteen hours. They are then to be transferred to 25 c.c. of distilled water plus 5 c.c. of formic acid and exposed to daylight. When the reduction is completed (p. 47) the dark-violet pieces are to be hardened in gradually strengthened alcohols and in about six days thin sections parallel to the surface can be cut and mounted in xylol-balsam.

No. 185.—*Nerves and blood-vessels of the fresh cornea*.—Select the eye of an ox and cut out the cornea with the adjoining portion of the sclera, extending from the limbus to the insertion of the ocular muscles; with scalpel and forceps remove the ciliary body, iris, and lens, immediately cut out a quadrant of the cornea, place it with the epithelial side up on a slide and apply a cover-glass; add a few drops of the vitreous humor. The very thick preparation must be examined with a low power. When the superficial strata of the cornea are in focus the loop-shaped blood-vessels can be seen at the scleral margin; the majority still contain blood corpuscles. Medullated nerve-fibers are found here, as well as in the deeper strata; they are arranged in big bundles and can be traced only for a short distance within the cornea. The elongated pigment streaks found in the eye of the ox have no relation to the nerves.

This method is not serviceable for the exhibition of the finer distribution of the nerves.

No. 186.—*Nerves of the cornea*.—(a) *Gold method*.—Cut out the cornea twelve or twenty-four hours after death, detach the ciliary body and the iris, and treat it according to the method given in No. 184. When the hardening is completed cut horizontal sections, which contain the epithelium and the uppermost lamellæ of the cornea, and vertical sections through the thickness of the cornea. Mount in xylol-balsam (Fig. 330).

(b) *Methylene-blue staining*.—Kill a rabbit; remove the entire eye-

ball, free it from the attached remnants of ocular muscles and connective-tissue, place it in a watch-glass and with a sharp scalpel make a deep incision at the equator through all the coats of the eye; the escaping vitreous humor is caught in the watch-glass. Then, beginning at the incision made, cut out the entire cornea, place it on a slide with the concave surface upward and with the handle of the scalpel scrape off the ciliary body, iris, and lens, which is easily done; transfer the cornea thus cleansed to a second watch-glass containing from 3 to 10 drops of the vitreous humor and from 3 to 4 drops of a 0.06 per cent. methylene-blue solution (p. 42). The concave surface of the cornea should be uppermost and covered by the staining fluid.

The time required for staining cannot be given with certainty; therefore it is advisable after several hours to place the cornea with the *convex* surface up on a clean slide and, without a cover-glass, to examine it with the low power; if it is not sufficiently stained return it to the watch-glass and examine it again in about ten minutes.

So soon as the nerves can be distinctly seen the cornea is to be transferred for from eighteen to twenty hours to 20 c.c. of the ammonia solution (p. 26); then cut out a quadrant and mount it in dilute glycerol, to which a drop of the ammonia solution has been added; after being kept in the dark for twenty-four hours the preparation is sufficiently transparent and can be investigated with the high power.

No. 187.—*Lens-fibers*.—Cut open the eyeball back of the equator; remove the vitreous body and lens; the pigment covering the ciliary processes remains attached to the margin of the lens. Loosen the lens from the vitreous body and place it in 50 c.c. of Ranvier's alcohol (p. 20). In about two hours thrust needles into the anterior and the posterior surface of the lens and strip the capsule from a small area; this is easily done; if lens-fibers are attached to the capsule it does not matter. On pricking the lens a turbid white fluid escapes; shake the alcohol and let the lens remain in it for from ten to forty hours. At the expiration of this time the lens can be easily separated into shell-like pieces. Tease a small strip of one of these pieces in a small drop of distilled water on a slide. Apply a cover-glass, taking care to avoid pressure; if it is desired to preserve the fibers, stain with picrocarmine (p. 53), staining usually occurs in a few minutes, and mount in dilute acidulated glycerol (Fig. 327 A).

No. 188.—*Lens-fibers in transverse section*.—Place a lens in 50 c.c. of 0.05 per cent. chromic acid (p. 32). A wad of cotton must be placed on the bottom of the bottle or the lens will adhere to the glass and burst. This may also be prevented by frequently shaking the bottle. In from twenty-four to forty-eight hours, with a needle break the lens into shell-like pieces;* transfer them after ten or fifteen hours to 30 c.c. of 70 per

* The shell-like fragments obtained in maceration experiments are the source of the false doctrine of the concentric stratification of the lens; they are simulated also in meridional sections; that which is here visible are individual *fibers*, not *lamellæ*. Equatorial sections through

cent. alcohol, which is to be replaced on the following day by an equal quantity of 90 per cent. alcohol. With the scissors cut the pieces through in the region of the equator, and so embed them in liver that the first sections will pass through the zone lying next to the equator. If the sections, which need not be very thin, have passed through the fibers transversely they will appear as sharply defined hexagons; if, on the contrary, the sections are oblique the single fibers will appear to be separated from one another by irregular zigzag lines; they may even be cut partially lengthwise. The sections are to be transferred directly from the blade to the slide and mounted in dilute glycerol (Fig. 327 *B*).

No. 189.—*The lens-capsule and the lens epithelium*.—Place the eyeball free from muscle and fat in 100 or 200 c.c. of Müller's fluid (p. 33). Treat it further as follows:

(a) *Surface view of the lens-capsule and epithelium*.—After two or three days cut the eye open, take out the lens, remove as far as possible the zonula fibers, and with small forceps strip off a piece of the anterior lens-capsule; place it for about five minutes in a watch-glass with distilled water, which is to be changed once, then stain it in Hansen's hematoxylin (p. 38); mount in xylol-balsam. The capsule is stained a homogeneous light blue; the nuclei and contours of the epithelial cells are very sharp (Fig. 328 *C*). If it is desired to obtain the lens-capsule alone strip off a portion of the posterior lens-capsule.

(b) *Sections of the capsule and epithelium*.—Let the eyeball remain in Müller's fluid for two weeks; remove the lens, wash it for one hour in running water and harden it in 50 c.c. of gradually strengthened alcohols (p. 35). Embedding in celloidin is advisable. Cut meridional sections through the anterior surface and through the equator of the lens, which are to be stained with Hansen's hematoxylin and mounted in xylol-balsam (Fig. 328 *D*), and equatorial sections that begin at the posterior pole. The sections through the anterior surface show beautifully the attachment of the lens-fibers to the rays of the lens-star. Since the firm core is very difficult to cut, it is advisable when this part of the lens is reached to loosen and extract the core by means of a small knife and to fill the cavity with celloidin.

No. 190.—*The blood-vessels of the eye*.—For this purpose surface preparations are especially suitable. On opening a fresh eye at the equator the course of the central artery of the retina is macroscopically perceptible. For the exhibition of the blood-vessels of the choroid place an eyeball completely freed from attached muscle and fat on a small glass funnel, which has been thrust into a low glass bottle, and with scissors and forceps, beginning at the equator, carefully dissect off the sclera. With a little practice the entire sclera can be removed from a little behind the ora serrata up to the optic entrance without injury to the choroid; care

the lens exhibit the image, not of an onion, but of an orange,—of *radial lamellæ*. The falling apart in shells is owing to the fact that lens-fibers of approximately the same age also possess like consistence, like physical and chemic nature.

must be taken not to tear it. (Beginners should be content to remove only one quadrant of the sclera.) All the firmer cords of attachment (the *venæ vorticosæ*) between the sclera and the choroid must be cut through. Then by careful brushing with a camel's-hair pencil moistened in water remove the attached portions of the lamina suprachorioidea from the choroid; by this manipulation the course of the larger blood-vessels is brought to view. So far the investigation may be pursued on the uninjected eye (compare with 178 a). For the study of the blood-vessels of the ciliary body and the iris it is necessary to use an injected eye, divided anterior to the equator, fixed in Müller's fluid and hardened in alcohol. The iris and the ciliary body can be easily stripped from the sclera; remove the lens and then mount in xylol-balsam. Examine at first with a very low power.

No. 191.—*The eyelid*.—Fix the *upper eyelid* of a child in ca. 60 c.c. of potassium-bichromate-acetic-acid (p. 32) for from 1 to 3 days and after washing for 3 hours in running water harden in ca. 50 c.c. of alcohols of ascending degrees of strength (p. 35). For topographic preparations (Fig. 331) cut thick sections; for the finer details (Fig. 37 C, p. 92) cut thin sections. Stain with Hansen's hematoxylin (p. 38) and mount in xylol-balsam.

No. 192.—*The lacrimal glands*.—The lower *tear-gland* in man can be easily removed, without visible external injury, from the fornix of the conjunctiva. In the rabbit this gland is very small and when fresh resembles pale muscle tissue. It must not be confused with Harder's gland lying in the median angle of the eye. Treat like No. 118 (p. 304). Small pieces 1 mm. square can be used. The excretory duct and the tubules are easily seen; difficult, on the other hand, it is to see the intercalated tubules, the epithelium of which varies greatly in height and occasionally is so low that care must be taken not to confuse them with blood capillaries (Fig. 332).

XI. THE ORGAN OF HEARING.

The organ of hearing consists of three divisions; the innermost, the *internal ear*, encloses the end apparatus of the auditory nerve; the other divisions, the *middle ear* and the *external ear*, are only accessory apparatus.

THE INTERNAL EAR.

The internal ear consists of two membranous saccules lying within the bony vestibule (vestibulum), that communicate with each other by means of a minute canal, the *ductus utriculo-saccularis*. The one saccule, the *utricle* (utriculus), is in connection with membranous tubules, the *semi-circular canals* (ductus semicirculares), each of which at the point where it opens in the utricle possesses a dilatation, the *ampulla*. The other sac-

cule, the *sacculus*, connects by means of the *ductus reuniens* with a long, spirally wound, membranous tube, the *cochlea* (ductus cochlearis).

The sacculus and the utriculus, the semicircular canals and the cochlea are called the *membranous labyrinth*. This is enclosed within the petrous bone in cavities having similar outlines, the *bony labyrinth*, which it does not completely fill. The unfilled space is occupied by a watery fluid, the *perilymph*. A similar fluid, the *endolymph*, is contained in the interior of the membranous labyrinth.

The saccules and the semicircular canals agree in structure, but the cochlea is so essentially different that it requires a separate description.

THE SACCULE, THE UTRICLE, AND THE SEMICIRCULAR CANALS.

The walls of these structures consist of three layers. The outermost is a connective-tissue layer rich in elastic fibers; this is followed within by a delicate basal membrane beset with minute excrescences, which on its inner surface is covered by a simple squamous epithelium. This simple structure undergoes alteration at the places where the filaments of the auditory nerve are distributed, which in the saccule and the utricle are named the *maculae*, in the ampullae of the semicircular canals the *cristae acusticae*. The connective tissue and basal membrane here become thicker; the squamous epithelium already at the periphery of the maculae and cristae becomes transformed into a cylinder epithelium with a cuticular border, and this passes into the neuro-epithelium of the maculae and cristae. The neuro-epithelium likewise is a simple layer and consists of two kinds of cells: (1) *fiber-cells*, tall, slender elements occupying the entire height of the epithelium, slightly expanded at the upper as well as at the lower end, which contain an oval nucleus; they are the supporting elements; (2) *hair-cells*, cylindrical elements occupying only the upper half of the epithelium, which in their lower, rounded division contain a large, spherical nucleus and on their free surface bear a bundle of long, delicate, agglutinated filaments, the "auditory hair." The hair-cells are the terminal apparatus of the auditory nerve. The nerve-fibers of the *ramus vestibularis nervi acustici* are in connection with the hair-cells in this way: on entering the epithelium the nerve-fibers lose their medullary sheath, divide, and as naked axis-cylinders ascend to the base of the hair-cells; there each fiber divides into three or four varicose twigs, that run horizontally beneath several hair-cells, parallel to the surface of the epithelium, and finally turn upward and terminate in contact with the lateral surface of a hair-cell in a free pointed end.* During their horizontal course they send upward a few

* The horizontal branches interlace and form a small but close "lattice-work," that also

twigs, that in the same manner end in contact with the hair-cells. These ends do not reach to the surface of the epithelium. The free surface of the neuro-epithelium is covered by a continuation of the cuticular border,



FIG. 333.—OTOLITHS
FROM THE SACCULUS
OF AN INFANT. $\times$
560. Technic No.
193.

a "limitans," which is perforated by the auditory hairs. The maculae acusticae are covered by a soft substance (a cuticula?), in which innumerable prismatic crystals of calcium carbonate, the *otoliths*, from 1 to 15 μ in size, are embedded; together they form the "otoconia," the auditory sand. On the cristae acusticae the so-called *cupula* occurs, in fresh preparations an invisible jelly, that on the application of fixation fluids coagulates and thus becomes visible.

By means of strands of connective tissue (*ligamenta sacculorum et ductuum*) the saccules and the semicircular canals are secured to the the bony labyrinth, the inner surface of which is covered with a thin periosteum and flattened connective-tissue cells.

THE COCHLEA.

The membranous cochlea, the *ductus cochlearis*, does not entirely fill the space within the bony cochlea. It lies with one wall in contact with the outer wall* of the bony cochlea (Fig. 334); the upper or vestibular wall, the *vestibular membrane* (Reissner), bounds the scala vestibuli; the lower or tympanic wall, the *membranous spiral lamina*, is directed toward the scala tympani. The angle in which the vestibular and the tympanic wall meet lies on the free end of the osseous spiral lamina. There the periosteum and the connective tissue of the ductus cochlearis are especially well developed and form an eminence, the *limbus spiralis*, which rests with a broad base on the bony spiral lamina, slopes upward and terminates in a sharp edge. This edge is called the *labium vestibulare*, the free margin of the bony spiral lamina is called the *labium tympanicum*,† between the two runs the *sulcus spiralis* (Fig. 341). The inner surfaces of the ductus cochlearis are covered by an epithelium of very different nature in the different localities; the outer surfaces, toward the scala vestibuli and the scala tympani, are covered by a delicate continuation of

by the application of other methods than that of Golgi appears to consist of a peculiar layer of strongly refracting granules. The granules are the varicosities and the optical cross-sections of the horizontal fibers.

*I here follow the customary description, in which the cochlea is placed in such a manner that the base is directed downward, the summit upward; accordingly "inner" is toward the axis of the cochlea, "outer" toward the periphery.

†The names were bestowed at the time in which the *limbus spiralis* was accounted as part of the *lamina spiralis ossea*.

the periosteum which clothes both scalæ. On the outer wall of the cochlea the periosteum becomes greatly thickened and in cross-section appears as a huge crescentic mass, the *ligamentum spirale*, that extends above and below the attached surface of the ductus cochlearis (Fig. 335).

The *minute structure* of the outer and the vestibular wall of the membranous cochlea is comparatively simple, of the tympanic wall, on the other hand, is extremely complicated.

The *outer wall* and the *spiral ligament* together consist of epithelium and connective tissue. The latter, next to the bone, is a dense fibrous tissue (the periosteum) and this passes into a loose connective tissue which contributes the chief bulk of the spiral ligament. The epi-

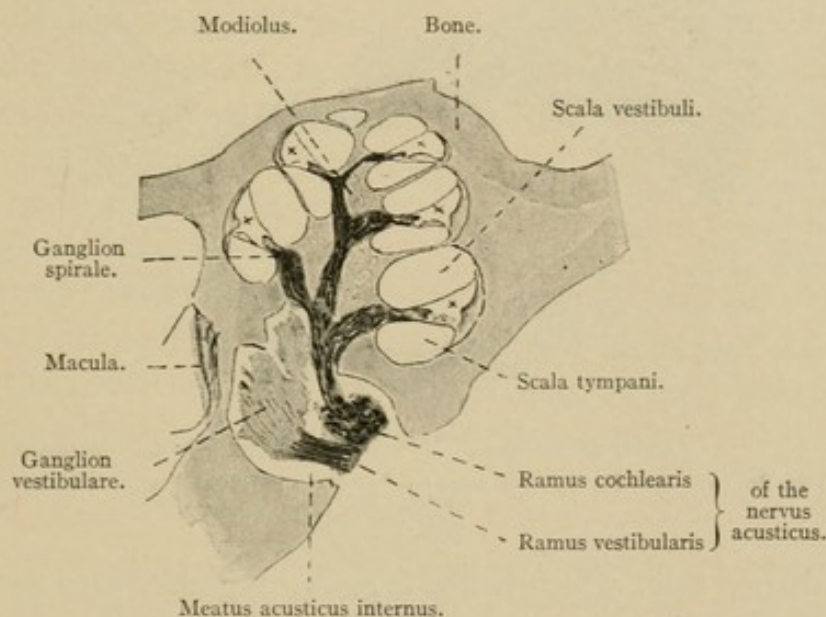


FIG. 334.—HORIZONTAL SECTION THROUGH THE ANTERIOR PORTION OF THE PETROUS BONE OF A KITTEN. $\times 8$. The ductus cochlearis, x, fell within the plane of the section five times. The variable color of the bone is owing to the incomplete penetration of the fixation medium. Technic No. 195.

thelium consists of a layer of cubical epithelial cells. A dense network of blood-vessels, the *stria vascularis*, occupies three-fourths of the height of the outer cochlear wall, and downwards is bounded by a vein that projects farther into the lumen of the cochlea, the *vas prominens* (Fig. 335). The capillaries of the stria vascularis lie close beneath the epithelium (Fig. 341); they are the source of the endolymph.

The *vestibular wall*, *membrana vestibularis* (Fig. 335), consists of a process of the periosteum of the scala vestibuli, that is, of delicate fibrous connective tissue and flattened cells, which on the surface turned toward the ductus cochlearis is clothed with a simple layer of polygonal epithelial cells.

The *tympanic wall* consists of two divisions (1), the *limbus spiralis*

with the free margin of the osseous spiral lamina, and (2) the lamina spiralis membranacea.

The *limbus spiralis* consists of a compact connective tissue, rich in

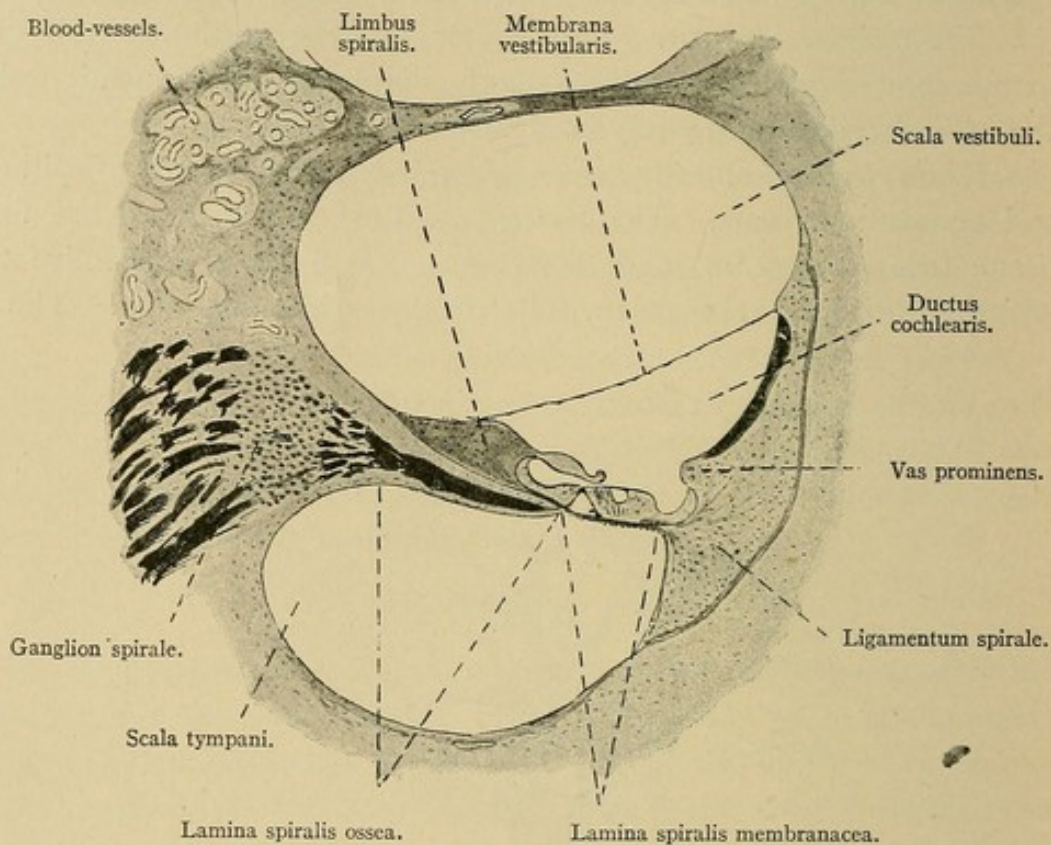


FIG. 335.—THE PORTION OF FIGURE 334 MARKED "SCALA VESTIBULI" AND "SCALA TYMPANI." $\times 50$.
Technic No. 195.

spindle-shaped cells, which below is grown together with the periosteum of the lamina spiralis ossea, on its free surface is beset with peculiarly shaped papillæ. They have the form of irregular hemispheres; toward the labium vestibulare they develop into small, elongated plates,

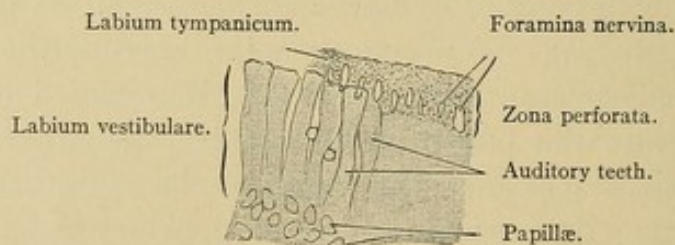


FIG. 336.—A SURFACE VIEW OF THE LAMINA SPIRALIS OF A CAT. $\times 240$. The vestibular lamina is seen from above; between the auditory teeth two nuclei of the epithelial cells are visible. On the left of the picture the plane of the auditory teeth is in focus, on the right, the plane of the zona perforata. Technic No. 194.

Huschke's auditory teeth (Fig. 336 and Fig. 339), that lie in a simple row beside one another. The surface of the limbus is covered by a simple layer of much flattened epithelial cells, which at the edge of the labium

vestibulare passes into the cubical epithelium of the sulcus spiralis (Fig. 339, *A*).

The upper surface of the free margin of the osseous spiral lamina is perforated by a single row of slit-like openings, the *foramina nervina* (Fig. 336) through which the nerves enclosed in the bony lamina emerge, to penetrate within the epithelium of the lamina spiralis membranacea. Therefore this zone of the osseous spiral lamina is called *zona perforata* (Habenula).

The *membranous spiral lamina* (lamina spiralis membranacea) consists of (1) the *membrana basilaris*, an extension of the limbus spiralis and of the periosteum of the osseous spiral lamina, (2) the *tympanal lamella*, a process of the periosteum of the scala tympani, which clothes the lower surface of the basilar membrane, and (3) the *epithelium of the ductus cochlearis*, which rests upon the upper surface of the basilar membrane.

The *membrana basilaris* consists of a structureless lamella, which contains rigid, perfectly straight fibers, extending from the labium tympanicum to the spiral ligament, and also oblong nuclei. This gives to the membrane a finely striated appearance (Fig. 337, *f*).

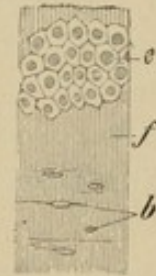


FIG. 337.—SURFACE VIEW OF THE LAMINA SPIRALIS MEMBRANACEA OF A CAT. $\times 240$. Strata of the zona pectinata drawn with change of focus. *e*, Indifferent epithelium (cells of Claudius) of the ductus cochlearis in focus; *f*, the fibers of the membrana basilaris in focus; *b*, the nuclei of the tympanal lamella in focus. Technic No. 194.

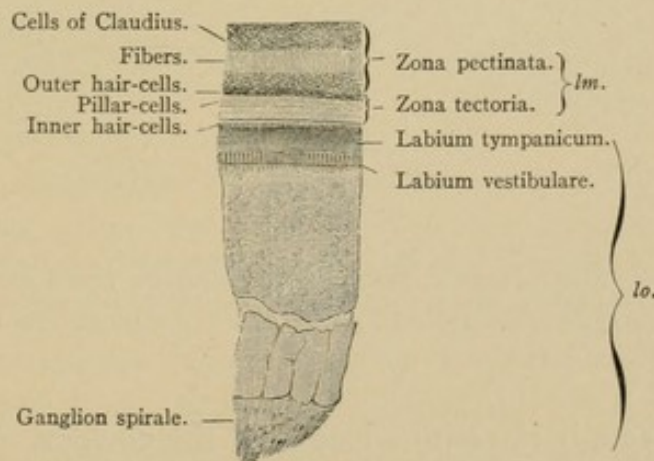


FIG. 338.—LAMINA SPIRALIS OF A CAT, SEEN FROM THE VESTIBULAR SURFACE. The membrana tectoria has been removed. $\times 50$. *lo*, Lamina spiralis ossea, the inner half fractured and broken at several places; at the posterior border of the same cells of the spiral ganglion project forth. *lm*, Lamina spiralis membranacea. The cells of Claudius have partly fallen off, so that the fibers of the membrana basilaris are visible as a delicate striation. Technic No. 194.

The *tympanic lamella* consists of a delicate connective tissue containing spindle-cells, the fibers of which are disposed vertically to the fibers of the basilar membrane (Fig. 337, *b*).

The *epithelium* of that half of the membranous spiral lamina toward

the axis of the cochlea is differentiated into the neuro-epithelium of the *spiral organ* (organon spirale, Corti), while that occupying the outer half, toward the spiral ligament, consists of indifferent epithelial elements. Therefore the membranous spiral lamina is divided into two zones: an inner, occupied by the spiral organ, *zona tecta*, and an outer, *zona pectinata*, so named because of the striations of the basilar membrane shimmering through it.

The most remarkable elements of the spiral organ are the *pillar-cells*, peculiarly shaped, for the greater part rigid structures, arranged in

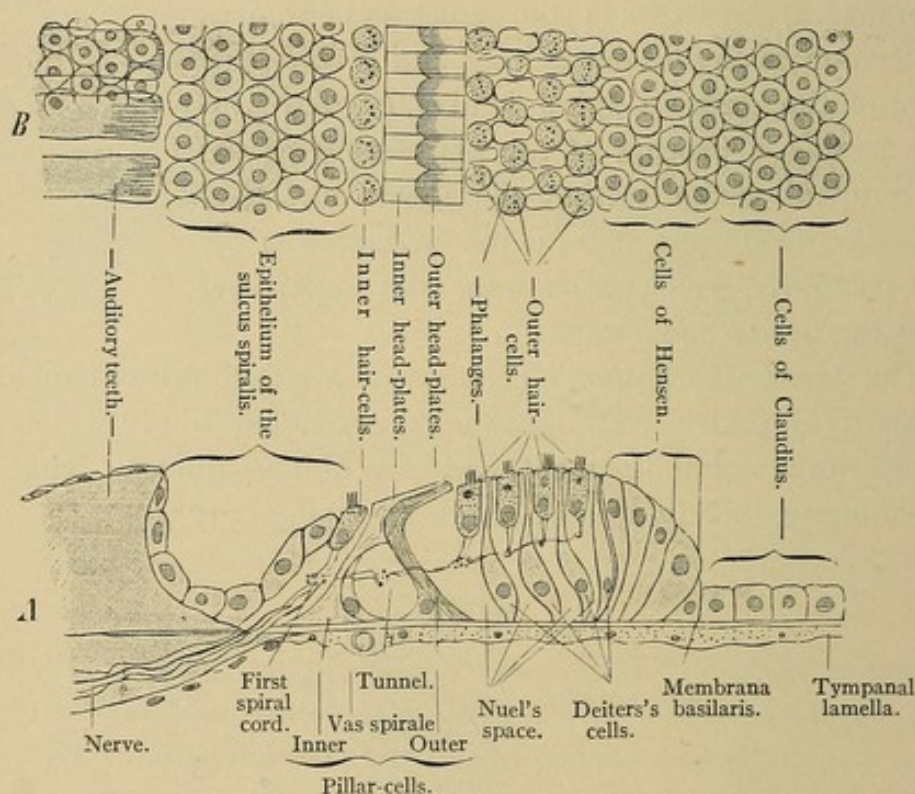


FIG. 339.—SCHEME OF THE STRUCTURE OF THE TYMPANIC WALL OF THE DUCT OF THE COCHLEA. A. View from the side. B. View from the surface. In the latter the free surface is in focus. It is evident that the epithelium of the sulcus spiralis, lying in another plane, as well as the cells of Claudius, can only be distinctly shown by depressing the tube. The membrana tectoria is not drawn. The spiral nerve-bundles are indicated by dots.

two rows through the entire length of the cochlea. The inner row of pillar-cells form the *inner pillars*, the outer row, the *outer pillars* (Fig. 339). The two rows of pillars are obliquely inclined toward one another and form an arch, the *arcus spiralis*, which spans a triangular space, the *tunnel*, the base of which is directed toward the basilar membrane. The tunnel is nothing else than a very large intercellular space, that is filled with a soft mass, with intercellular substance. Regarding the histology of the pillar-cells the following details are to be considered: The *inner pillar-cells* are rigid bands, in which a *three-sided, expanded foot*, a *slender*

body, and a *concave head*, with the concavity directed outward, are distinguished. The head is furnished with a small process, the "head-plate" (Fig. 339). The body and foot of the cell are surrounded by a scant amount of protoplasm, that only to the outer side of the foot in the vicinity of the nucleus is present in somewhat larger amount. The *outer pillar-cells* exhibit the same details, excepting that the portion containing the nucleus lies to the inner side of the foot; the rounded articular head rests in the concave facet of the head of the inner pillars, the broader head-plate is covered for the greater part by the head-plate of the inner pillars.* To the inner side of the inner pillars lies a *simple* row of cells, the *inner hair-cells*, short cylindrical elements that do not extend to the basilar membrane; they possess a rounded base and about forty long, stiff hairs on their free surface. To the inner side of the inner hair-cells lies the cubical epithelium of the sulcus spiralis internus. On the

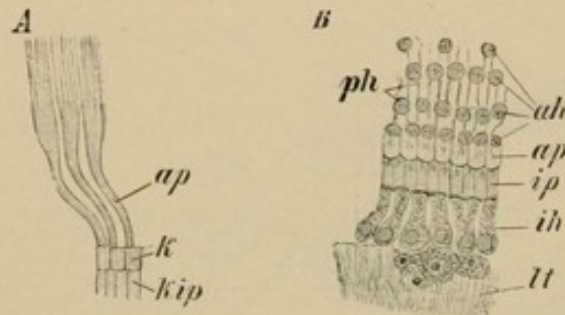


FIG. 340.—FROM THE LAMINA SPIRALIS MEMBRANACEA OF A CAT. $\times 240$. A. Outer pillar-cells; *k*, head-plates of the same, upper surface in focus; *ap*, body and lower end of the same sketched under gradual depression of the tube; *kip*, portions of the head-plates of inner pillar-cells. B. *lt*, Labium tympanicum, partly covered by the epithelium of the sulcus spiralis; *ih*, inner, *ah*, outer hair-cells, between these the phalanges, *ph*, forming the membrana reticularis; *ap*, head-plates of the outer, *ip*, of the inner pillar-cells. Technic No. 194.

outer side of the outer pillars lie the *outer hair-cells*; they resemble the inner hair-cells, but possess hairs that are one-third shorter and are characterized by a dark body situated in the upper half of the cell, the *spiral body* (Hensen).† The outer hair-cells are arranged not in *one*, but in several (usually four) rows; they do not lie in contact with one another, but are held apart by *Deiters's cells*; these are slender cells, each of which contains a rigid filament and at its upper end supports a *cuticular* process, that has the shape of a digital phalanx. The free spaces between the "phalanges" are occupied by the upper ends of the outer hair-cells ‡

* The nucleus-like inclusion found in the heads of the inner and the outer pillars, also that in the feet of the latter, has no relation to a nucleus, but probably is of a horny nature.

† In the scheme (Fig. 339 A) this body is indicated by a dark spot close beneath the auditory hairs.

‡ The inner hair-cells are held apart from one another by short processes of the inner pillar-cells. These processes are not shown in Fig. 339.

(Fig. 340). The cells of Deiters are supporting elements, that exhibit much in common with the pillar-cells; like these they consist of a rigid filament and a protoplasmic portion, like these they have a head-plate (named phalanx). The difference consists only in this, that the transformation into rigid parts is not so far advanced in the cells of Deiters. The phalanges are joined to one another and form a beautiful netted membrane, the *membrana reticularis*.

The outer hair-cells do not extend down to the basilar membrane, but occupy only the upper half of the spaces between the cells of Deiters; the lower divisions of these spaces remain unoccupied and are called

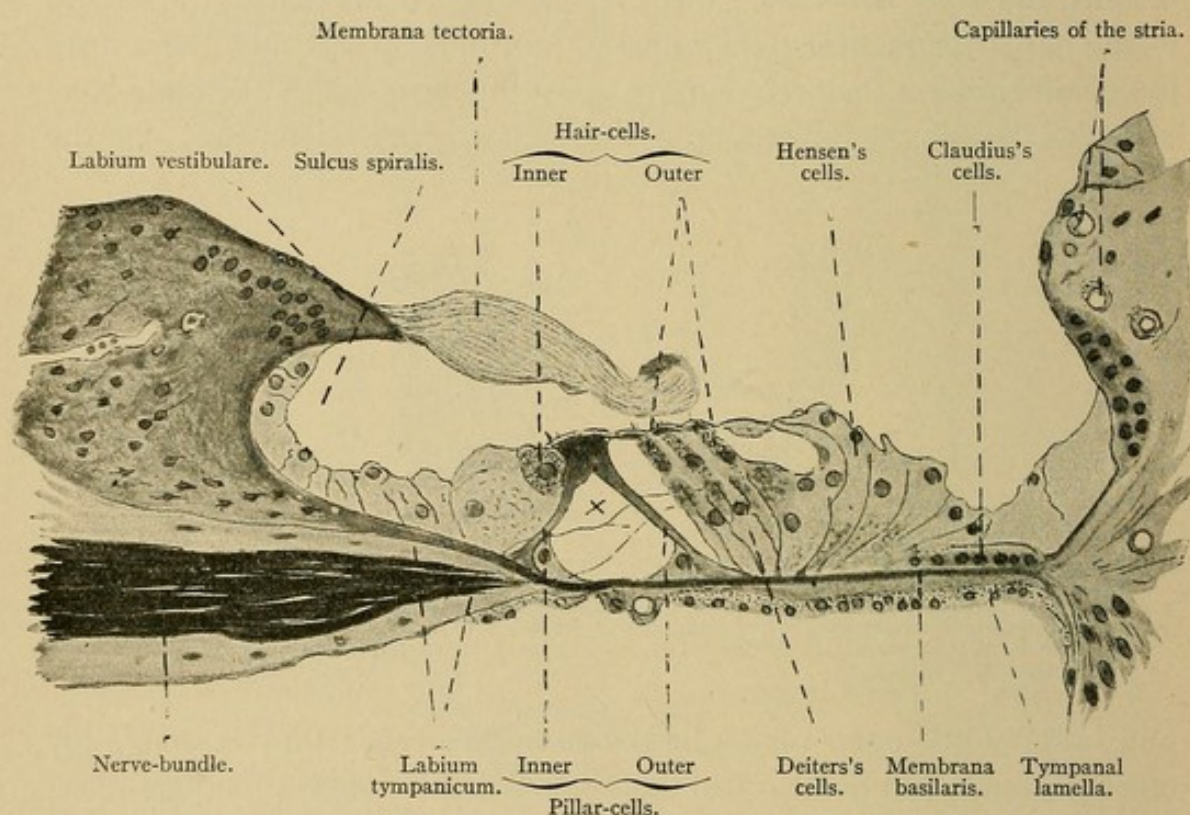


FIG. 341.—PORTION OF FIGURE 335. Magnified 240 times. *x*. Tunnel traversed by nerve-fibers.

Nuel's spaces, or, since they communicate with one another, the space of Nuel (Fig. 339, *A*). The latter also has the significance of an intercellular space and communicates with the tunnel.

External to the last row of Deiters's cells lie the *cells of Hensen*, slender cylinders, that gradually decrease in height and pass into the indifferent epithelium of the cochlear duct, the elements of which, so far as they cover the basilar membrane, are called the *cells of Claudius*. These two varieties of cells, as well as the epithelial elements of the sulcus spiralis, also contain a rigid fiber, that however is less developed than in the cells of Deiters. The centrosomes of all the epithelial cells of the spiral organ lie near the free surface.

A soft, elastic cuticular formation, the *membrana tectoria*, lies above the sulcus spiralis and the spiral organ (Fig. 341). It is attached to the vestibular lip of the sulcus and extends to the outermost row of hair-cells.

The cochlear branch (*ramus cochlearis*) of the *auditory nerve* penetrates into the axis of the cochlea and in its spiral uninterrupted course gives off branches which pass toward the root of the osseous spiral lamina; here each medullated nerve-fiber loses its medullated sheath and passes into a nerve-cell, that like those of the spinal ganglia possesses a connective-tissue capsule; these nerve-cells collectively form the *ganglion spirale*,* which winds around the entire periphery of the axis of the cochlea (Fig. 334); from the opposite pole of each cell springs a second nerve-fiber,† that soon acquires a medullated sheath and unites with neighboring fibers in a wide-meshed plexus enclosed in the osseous spiral lamina; this plexus extends near to the labium tympanicum, where the fibers lose their medullated sheath, pass through the foramina nervina (p. 441) and end in the epithelium. This occurs in such a manner that they bend in the direction of the circumvolution of the cochlea and run in spiral cords, of which the first passes to the inner side of the inner pillar-cells (Fig. 339 A), the second into the tunnel, the third between the outer pillar-cells and the first row of the cells of Deiters, while the remaining three run between the cells of Deiters. From these cords delicate fibers proceed to the hair-cells, on which (not within) they terminate.

The arteries of the labyrinth. The auditory artery gives only a small twig to the membranous labyrinth and another small twig to the osseous labyrinth; the majority of its branches pass to the exit of the fifth, seventh, eighth, ninth, and tenth cranial nerves and to the under surface of the cerebellum. The artery for the membranous labyrinth divides into two branches: 1. The *arteria vestibularis* (Fig. 342) sends twigs to the vestibular nerve and to the lateral-upper half of the sacculus and of the utricle, as well as to the corresponding portions of the upper and lateral semicircular canals, which supply a capillary plexus that in general is wide-meshed, but at the terminal points of the vestibular nerve, the *cristæ* and *maculæ*, is narrow-meshed. 2. The *arteria cochlearis communis* subdivides in two branches. The one branch, the *arteria vestibulo-*

* The *ganglion spirale* possesses the same structure as a spinal ganglion, with a single difference: the *ganglion* cells here are not unipolar, but bipolar, as in the embryonal ganglia (p. 113). The *ganglion vestibulare* in the internal meatus also possesses bipolar ganglion cells.

† In early developmental stages this fiber exhibits the character of a dendrite and only gradually becomes a slender fiber (*cf.* remark*, p. 116).

cochlearis, supplies one twig to the median-posterior half of the sacculus, utriculus, and semicircular canals and in its minute ramifications behaves like the vestibular artery; another twig ramifies in the initial third of the first turn of the cochlea. The other branch, the *arteria cochlearis propria*, supplies the remaining district of the cochlea; on entering the axis of the cochlea it divides into three or four branches, which in their

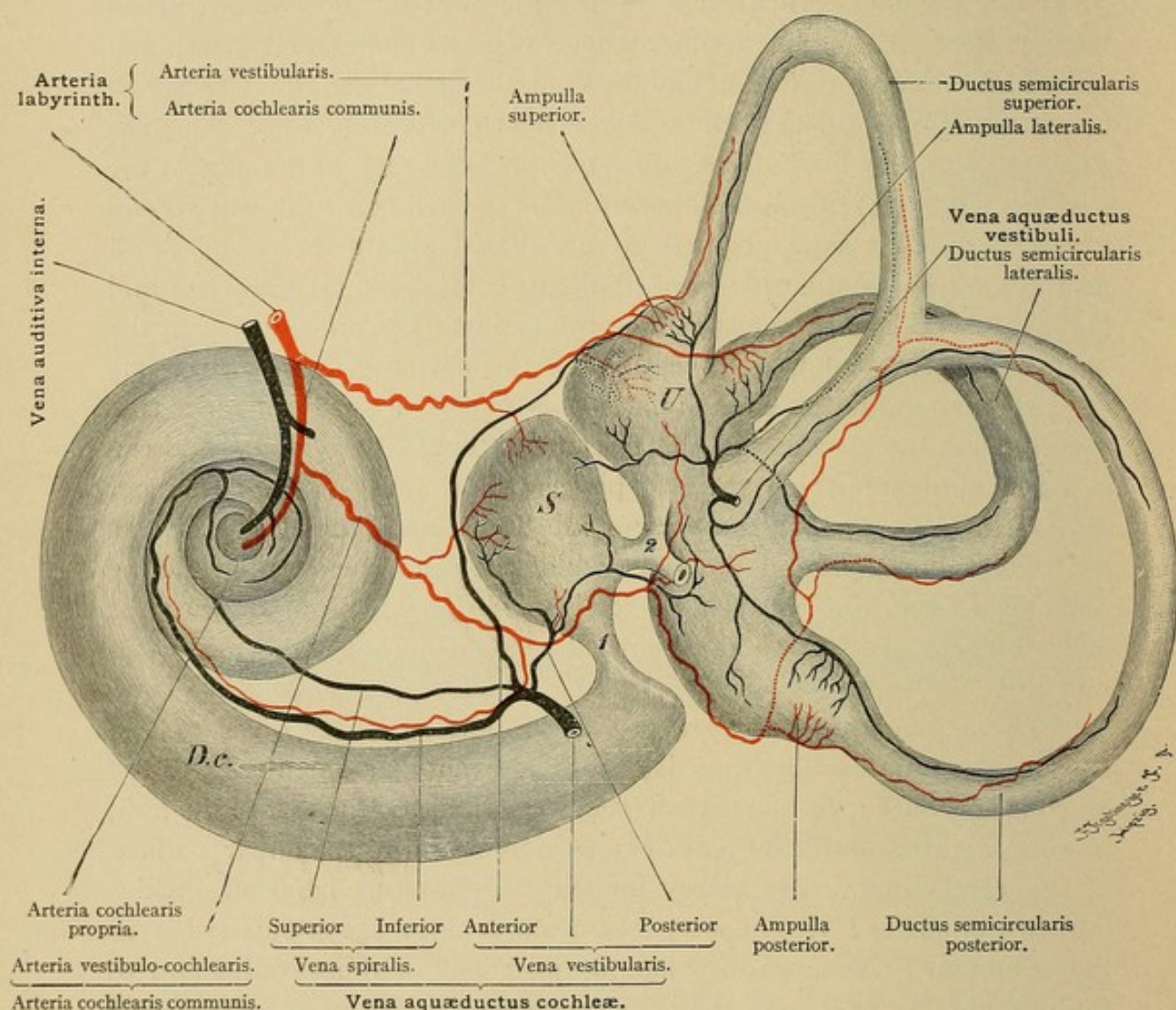


FIG. 342.—SCHEME. BLOOD-VESSELS OF THE RIGHT HUMAN LABYRINTH. MEDIAN AND POSTERIOR ASPECT. D.c. Ductus cochlearis. S. Sacculus. U. Utriculus. 1. Ductus reuniens. 2. Ductus utriculo-saccularis. The saccus endolymphaticus is cut off.

spiral ascent form the tractus arteriosus spiralis. From this about 30 or 35 radial twigs arise, which supply three separate capillary territories: (1) the canal in which the ganglion spirale is enclosed, (2) the lamina spiralis, (3) the intermediate and outer walls of the scalæ (Fig. 343, 1, 2, 3).

The *veins* of the labyrinth follow three separate paths:

1. The *vena aquæductus vestibuli* runs through the aquæductus

vestibuli; it collects the blood from the semicircular canals and from one portion of the utriculus; it opens in the sinus petrosus superior (Fig. 342).

2. The *vena aquæductus cochleæ* runs through the aquæductus cochleæ; it collects the blood from one portion of the utriculus, from the sacculus and from the cochlea. The venous radicles in the cochlea behave in the following manner: The veins uniting in the *vas prominens* and in the *vas spirale* (Fig. 343, *a*, *b*) pass in the wall of the scala

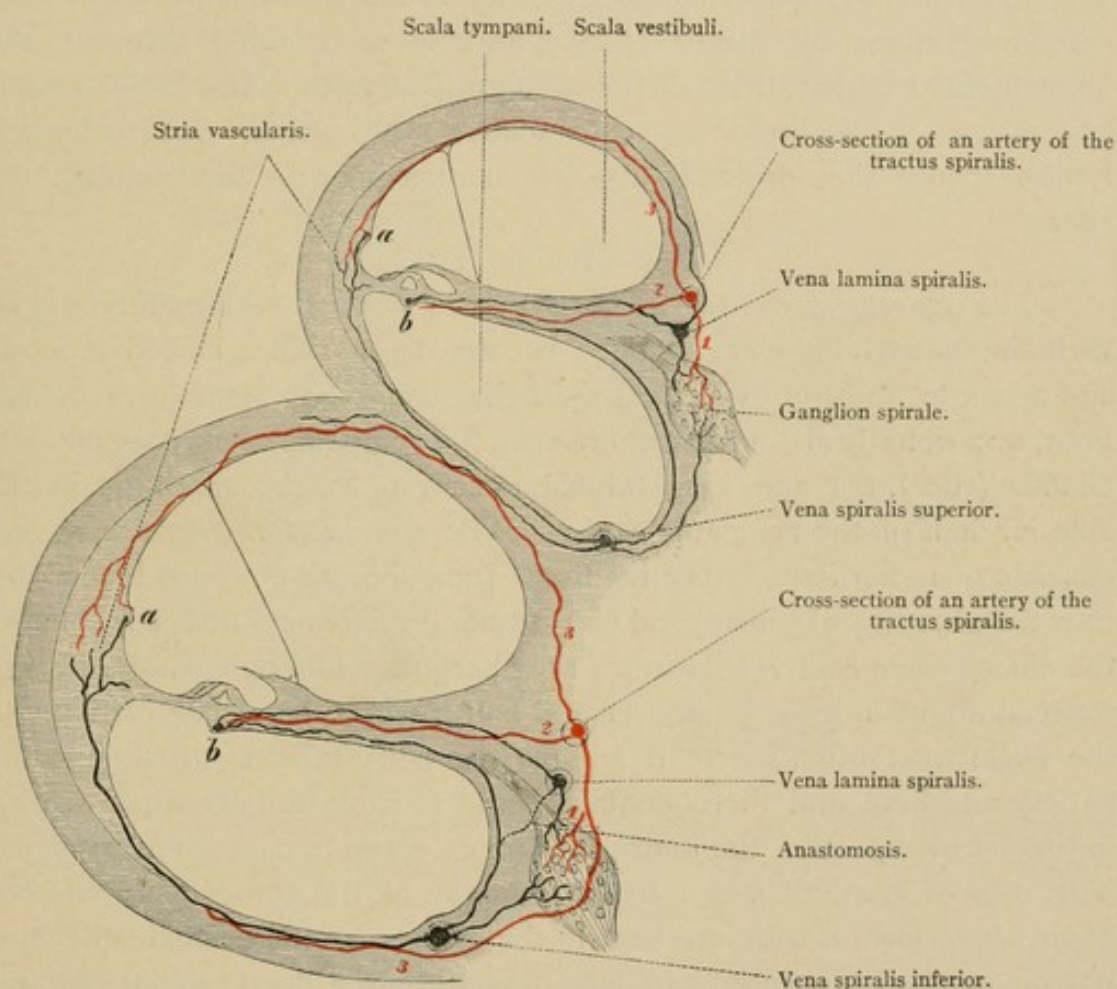


FIG. 343.—SCHEME. VERTICAL SECTION OF THE RIGHT HALF OF THE FIRST (BASAL) AND SECOND TURNS OF THE COCHLEA. *a*. Vas prominens. *b*. Vas spirale.

tympani to the spirally running *vena spiralis*, lying beneath the spiral ganglion; this originates from the confluence of two veins, of which the lower receives the blood from the first (basal) and a portion of the second turn of the cochlea, while the upper spiral vein collects the blood from the remaining cochlear turns. The spiral vein also takes up one set of the capillaries in the canal of the spiral ganglion and is united by anastomosis with a vein lying above this canal, the *vena lamina spiralis* (Fig. 343). This receives the blood from the other set of capillaries of

the spiral ganglion, as well as from the lamina spiralis,* and opens in the central vein of the cochlea.

3. The *central vein of the cochlea* is the main radicle of the internal auditory vein. The latter takes up veins from the auditory nerve and from the bone, and in all probability opens in the vena spinalis anterior.

The lymph paths. The endolymph in the interior of the membranous labyrinth communicates with the subdural lymph spaces by means of minute tubules passing from the saccus endolymphaticus. The perilymphatic spaces (*cf.* p. 437) are in connection with the subarachnoid space by means of a lymph-vessel running through the aquæductus cochleæ, the "ductus perilymphaticus." The blood-vessels and nerves are encircled by conspicuous perivascular and perineural lymph spaces, that probably also are connected with the subarachnoid space.

THE MIDDLE EAR.

The *mucous membrane of the tympanic cavity* is intimately united with the underlying periosteum. It consists of thin connective tissue and a single stratum of cubical epithelial cells, that sometimes on the floor, occasionally also in larger areas of the tympanic cavity, are ciliated. Glands (short, 0.1 mm. long follicles) occur only and sparingly in the anterior half of the tympanic cavity. The *mucosa of the eustachian tube* consists of a fibrillar connective tissue (containing numerous leucocytes near the pharyngeal orifice) and of a stratified ciliated cylinder epithelium; the ciliary wave is directed toward the pharynx. Mucous glands occur in especial abundance in the pharyngeal half of the tube. The cartilage of the eustachian tube, where it adjoins the bony tube, is of the hyaline variety and here and there contains rigid (not elastic) fibers (*cf.* p. 97); in the anterior portion the matrix of the cartilage is penetrated by dense networks of elastic fibers. In the mucosa of the tympanic cavity the *blood-vessels* form a wide-meshed, in the mucosa of the eustachian tube a narrow-meshed superficial capillary network and a deep capillary plexus enveloping the glands. The *lymph-vessels* run in the periosteum of the tympanic cavity. With regard to the terminations of the nerves exact information is still wanting.

THE EXTERNAL EAR.

The *tympanum* consists of a lamina of connective tissue, *lamina propria*, in which the fiber-bundles on the surface facing lateralward are

*The vestibular membrane is nonvascular in the adult. The arrangement of the blood-vessels in the cochlea is such that the scala vestibuli is chiefly encircled by arteries, the scala tympani mainly by veins. The portion of the scala tympani adjacent to the lamina spiralis membranacea is thus removed from the influence of arterial pulsation.

radially arranged and connected with the periosteum of the sulcus tympanicus, while on the surface toward the tympanic cavity the fiber-bundles are circularly arranged. On its inner surface the tympanum is covered by the mucous membrane of the tympanic cavity, on its outer surface by the skin clothing the external auditory canal. Both coverings are very firmly attached to the lamina propria, are smooth, and are without papillæ. Where the malleus lies against the tympanum it is provided with a cover of hyaline cartilage.

The *external auditory meatus*, so far as it is cartilaginous and on the whole length of its upper wall, is clothed with a thick extension of the skin, which is characterized by its great abundance of peculiar coil-glands,

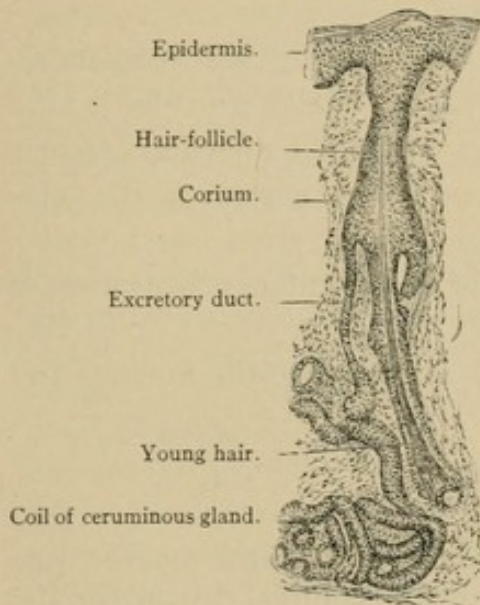


FIG. 344.—FROM A VERTICAL SECTION THROUGH THE SKIN OF THE EXTERNAL AUDITORY MEATUS OF AN INFANT. $\times 50$. The excretory duct opens into the hair-follicle. Technic No. 198.

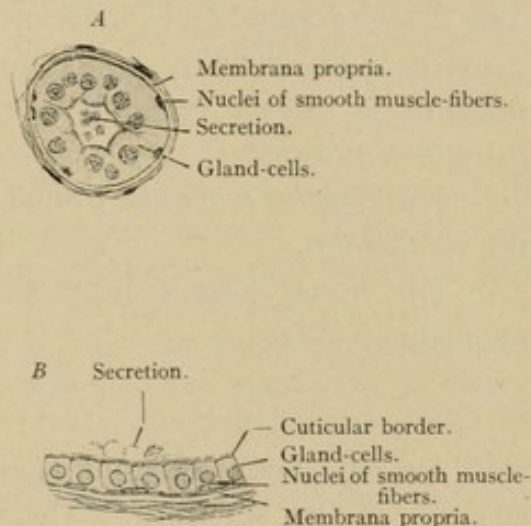


FIG. 345.—A. CROSS-SECTION OF A COIL-TUBULE OF THE SKIN OF THE EXTERNAL AUDITORY MEATUS OF AN INFANT. B. LONGITUDINAL SECTION OF A COIL-TUBULE FROM THE EXTERNAL AUDITORY MEATUS OF A TWELVE-YEAR-OLD BOY. $\times 240$. Technic No. 198.

the *ceruminous glands* (*glandulæ ceruminosæ*). In some respects these glands correspond with the ordinary larger coil-glands (sweat-glands) of the skin; like these they possess an excretory duct clothed in several layers of epithelial cells, and the canals of the coil contain a simple layer of usually cubical gland-cells, which rest on smooth muscle-fibers and a conspicuous basement membrane (Fig. 344); they are distinguished from the coil-glands by the very wide lumen of the coiled tubules, that particularly in adults is greatly dilated, and by numerous pigment granules and fat droplets within the gland-cells, which frequently exhibit a distinct cuticular border. The excretory ducts are narrow and in children open in the hair follicles, in adults close beside the hair-follicles on

the free surface. The secretion, the *cerumen* (ear-wax), consists of pigment granules, fat globules, and cells filled with fat; the latter probably come from the glands of the hair-follicles. In the (remaining) region of the bony external auditory meatus the skin is thin and without ceruminous glands.

The *cartilage* of the external auditory canal and of the pinna is of the elastic variety.

The *vessels* and *nerves* are distributed as elsewhere in the skin; only on the tympanum do they exhibit special peculiarities. Close behind the handle of the malleus an *artery* descends, which breaks up into radially disposed branches; the blood is returned by two paths: (1) by a *venous plexus* extending along the handle of the malleus and (2) by a *venous plexus* lying on the margin of the tympanum.

These vessels lie in the external skin covering the tympanum. The mucous membrane covering the tympanum is also provided with a dense capillary network, which anastomoses with the cutaneous vascular network by means of perforating branches at the margin of the tympanum.

The *lymph-vessels* are principally found in the cutaneous stratum of the tympanum.

The *nerves* form delicate networks lying beneath the mucous and cutaneous covers.

TECHNIC.

A fundamental condition is an exact knowledge of the macroscopic anatomy of the labyrinth. The difficulties, the failures, depend in the main on inaccurate knowledge of the anatomy of the bony labyrinth. As a preliminary all parts lying lateral to the promontory (os tympanicum and ossicles of the ear) must be removed, so that this is distinctly visible.

No. 193.—*Otoliths*.—Chisel out the promontory, beginning at the upper margin of the fenestra vestibuli, to the lower margin of the fenestra cochleæ. Then, especially if the bone is placed in water, the white spots (maculæ) in the sacculus and utriculus can be seen. With delicate forceps lift out the saccules and spread a small piece in *diluted* glycerol on a slide. The otoliths are present in large numbers, but are very small, so that their shape can only be distinctly seen with the high power (240 diameters). The glycerol must not be too thick, or the otoliths will become completely invisible (Fig. 333).

In taking out the saccules portions of the *semicircular canals* are not infrequently also removed; stain these with picrocarmine and mount them in dilute glycerol. Only the epithelium and here and there in optical cross-sections the delicate hyaline membrane can be seen. The connective tissue is very scanty.

No. 194.—*Surface preparations of the membranous cochlea.*—The base of the cochlea lies in the bottom of the internal auditory meatus, the apex is directed toward the eustachian tube, therefore the axis of the cochlea is horizontal and transverse to the long axis of the petrous bone.

Chisel open the free portion of the cochlea, that is, remove the promontory close to the fenestra cochleæ, open the apex of the cochlea, and having removed the superfluous osseous mass as far as practicable place the preparation in 20 c.c. of 0.5 per cent. osmic acid (5 c.c. of 2 per cent. osmic acid to 15 c.c. of distilled water). In from twelve to twenty hours wash the preparation for about one hour and then place it in 200 c.c. of Müller's fluid. In from three to twenty days (or later) open the cochlea fully and examine it under water. The osseous spiral lamina can be seen as a delicate lamella, the membranous spiral lamina as a delicate membrane, attached to the axis of the cochlea; with fine forceps break off a little piece of the osseous spiral lamina; do not lift it with the forceps, but carefully with needle and section-lifter remove it from the fluid and transfer it to a drop of dilute glycerol on a slide. It is advisable to break off the axial portion of the bony spiral lamina on the slide with needles, because the relatively thick osseous process renders it difficult to apply a cover-glass. The vestibular surface of the lamina must be directed upward; it can be recognized by the auditory teeth, which are visible when the upper surface is in focus (Fig. 336), while the other portions are not distinct until the tube is depressed and the lower planes are focused. With the low power only the interstices of the auditory teeth are at first visible as dark streaks (Fig. 338, labium vestibulare); the papillæ likewise cannot be seen immediately, even with the high power, but become distinct after the second or third day. The chief difficulty lies not in the finishing, but in the proper examination of the object; the picture alters with the slightest change in focus. In Fig. 339 *B* the membranous spiral lamina is drawn schematically, as seen with the upper surface in focus, therefore only the free surface of the structure, drawn as seen from the side in *A*, is visible. It is clear that in lowering the tube the head-plates of the pillar-cells are no longer visible, but their bodies (as circles in optical cross-section); the reticular membrane likewise disappears, it can be seen only when the surface is in focus. The preparation may be stained with picrocarmine and preserved in dilute glycerol. The foregoing directions are intended to apply to the human ear and that of the cat. The labyrinths of children are recommended.

No. 195.—*Sections of the bony and membranous cochlea.*—Remove the cochlea of a child* from the labyrinth. The compact osseous substance of the cochlea is surrounded by spongy bone so soft that it can be removed with a stout penknife. Having done this, with a chisel make small openings in the cochlea at two or three places, about 1 mm. square, in order to facilitate the penetration of the fixation fluid. Then

* Among animals the cochlea of the guinea-pig or the bat is recommended; it is not embedded in spongy bone and without further chiseling and puncturing can at once be placed in the fixing fluid. The cochlea of kittens is also recommended.

place the cochlea in 30 c.c. of Hermann's solution (p. 22). After 48 hours remove the object, wash it for a few seconds in methyl alcohol, transfer it to crude pyroligneous acid for from 12 to 24 hours, and then harden it in about 60 c.c. of gradually strengthened alcohols (p. 35). When the hardening is completed the cochlea is decalcified in concentrated aqueous (or better alcoholic) solution of picric acid. When the object is decalcified harden it again, first in 50 per cent., then in 70 per cent. alcohol, and after about a week embed it in liver or in celloidin and cut sections parallel with the long axis of the cochlea. Mount them in xylol-balsam.

It is not very difficult to obtain preparations affording a general view. The vestibular membrane is often torn, so that the ductus cochlearis and scala vestibuli appear as a common space. The spiral organ leaves most to be desired; only very thin sections which pass through the organ vertically furnish wholly intelligible pictures; usually a section contains several inner and outer pillar-cells, in part only fragments of them; the cells of Hensen appear puffed and swollen (Fig. 341), so that orientation presents many difficulties to the beginner.

No. 196.—*The nerves of the maculae, cristae, and cochlea.*—For this purpose the ear of the newborn, up to ten-day-old mouse is recommended, treated according to the method given on page 45, No. 17. The base of the cranium, after removal of the vertex, brain, and lower jaw, is placed for from three to four days in the osmio-bichromate mixture and for two days in the silver solution. As a rule it is necessary to employ the "double" method (p. 47). Cut horizontal and frontal sections through the undecalcified cranium. The former are the more readily made.

No. 197.—*The eustachian tube.*—To obtain transverse sections (including cartilage and mucosa) the oblique direction of the tube downward, forward, and inward must be ascertained. Cut out the entire pharyngeal division of the tube together with the surrounding muscles and fix it in 200 or 300 c.c. of Müller's fluid (p. 33). In from three to six weeks wash it in running water and harden it in 100 c.c. of gradually strengthened alcohols (p. 35). The sections may be stained in Hansen's hematoxylin (p. 38) and mounted in xylol-balsam. For a general view examine with the low power.

No. 198.—*The ceruminous glands.*—Cut off the ear and the cartilaginous auditory meatus close to the bony auditory meatus. From the cartilaginous portion cut a piece 1 cm. square and place it in 30 c.c. of absolute alcohol. The tissue may be sectioned on the following day. If it is desired to see the coil and the excretory duct the sections must be tolerably thick (—0.5 mm.) (Fig. 344). Nuclear staining with Hansen's hematoxylin (p. 38) may be employed. Examine thin unstained sections in diluted glycerol; in these the fat- and the pigment-granules can be seen. The organs of infants are especially recommended for these preparations. In adults the tubules are widely dilated and do

not furnish satisfactory general views. On the other hand, the cuticular border of the gland-cells is distinct in older children and in adults, which in the newborn I miss (*cf.* Fig. 345).

XII. THE OLFACTORY ORGAN.

In this chapter the structure of the entire nasal mucous membrane will be described. The olfactory mucous membrane proper in man is confined to the middle of the superior turbinal bone and to the corresponding portion of the nasal septum; the remaining portions of the nasal fossæ (the accessory nasal spaces included) are covered with respiratory mucous membrane. In addition there is another division in the region of the movable nose (vestibulum nasi) which is clothed by a continuation of the external skin.* Accordingly three divisions of the nasal mucous membrane differing in structure are to be distinguished.

THE VESTIBULAR REGION.

The mucous membrane of the vestibular region consists of a stratified squamous epithelium and a tunica propria supporting papillæ, in which numerous sebaceous glands and the hair-follicles of the stiff nasal hairs (vibrissæ) are embedded.

THE RESPIRATORY REGION.

The respiratory division of the nasal mucous membrane consists of a many-rowed ciliated cylinder epithelium (Fig. 20), that sometimes contains many, sometimes few goblet-cells, and of a conspicuous tunica propria, on the inferior turbinal bone up to four millimeters thick, which is built of fibrillar connective tissue and of a large, variable number of leucocytes, and toward the epithelial border is condensed to a homogeneous membrana propria provided with minute perforations. These leucocytes are occasionally balled together in solitary nodules and often wander in large numbers through the epithelium into the nasal fossæ (*cf.* p. 260).

The tunica propria in man contains branched alveolo-tubular mixed glands (*cf.* p. 242); the serous divisions are provided with intercellular secretory capillaries, serous and mucous gland-cells with a trophospongium (p. 64). Not infrequently they open in funnel-shaped depressions, which are lined by an extension of the surface epithelium and on the in-

* The boundaries are very variable; stratified squamous epithelium is frequently found on the middle, less often on the inferior turbinal.

ferior turbinal are perceptible to the unaided eye. In the accessory nasal spaces the epithelium and tunica propria are considerably thinner (— 0.02

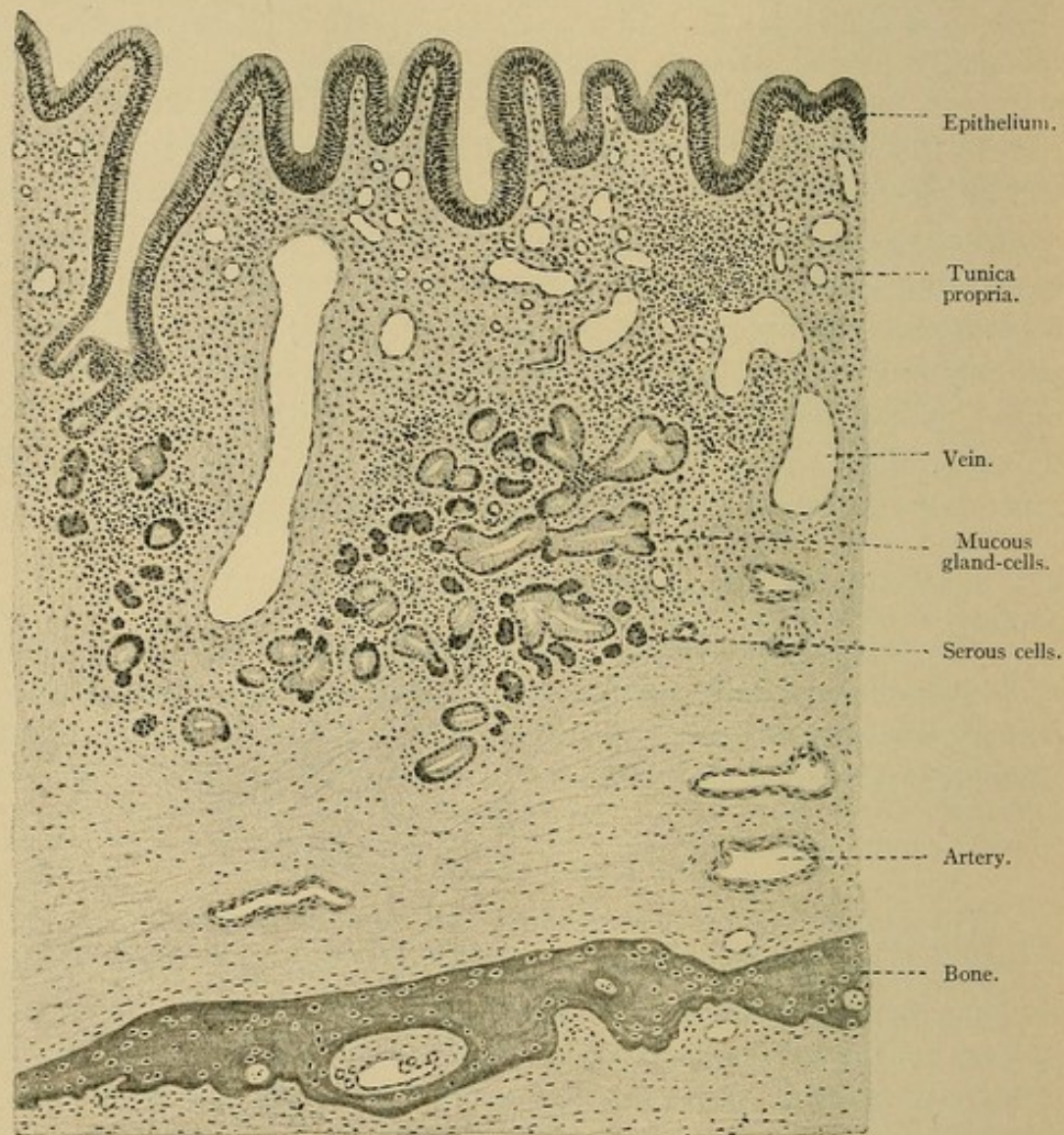


FIG. 346.—VERTICAL SECTION THROUGH THE MUCOSA OF THE INFERIOR TURBINAL OF MAN. $\times 48$. On the left is a funnel-shaped depression taking up a portion of an excretory duct; nearby on the right is the section of a large vein. Technic No. 200.

mm.), but otherwise of the same structure; the glands are small and few in number.

THE OLFACTORY REGION:

The mucous membrane of this region by its yellowish-brown color can be macroscopically distinguished from the rosy mucosa of the respiratory division. It consists of an epithelium, the olfactory epithelium, and of a tunica propria. In the olfactory epithelium two forms of cells occur. The one form (Fig. 347, *st*) is cylindrical in its upper half and here contains a yellowish pigment and minute granules, often arranged

in longitudinal rows. The lower half is slenderer, the edges are serrated and invaginated, the inferior end is forked and is said to unite with the forked ends of neighboring cells in a protoplasmic network. These elements are called *supporting cells*. Their usually oval nuclei lie at the *same* level and in vertical sections occupy a narrow belt, the *zone of the oval nuclei* (Fig. 350). The second form (Fig. 347, *r*, and Fig. 348) possesses a usually spherical nucleus and only in the vicinity of the latter an appreciable amount of protoplasm; from this a slender cylindrical process, bearing minute hairs, extends upward, while below from the opposite side a very delicate process continues directly into the axis-cylinder of a nerve-fiber. These cells, the *olfactory cells*, are ganglion cells and their lower process is a centripetal nerve-fiber. Their round nucleolated nuclei lie at different levels and occupy a broad belt, the *zone of the round nuclei* (Fig. 350, *sr*).^{*} In addition to these two forms of cells there are intermediate forms, that sometimes resemble the olfactory elements, sometimes the supporting cells. At the border of the epithelium, toward the connective tissue, there is a protoplasmic network furnished with nuclei, the so-called *basal cells* (Fig. 350, *b*). The surface of the epithelium is covered with an extremely delicate, homogeneous membrane, the *membrana limitans olfactoria*; it is pierced by the "ciliated" ends of the olfactory cells and is itself covered with a peculiar mass, regarded by some authors as a cuticular formation similar to that of the intestinal epithelium, by others as delicate cilia, by still others interpreted as minute drops of discharged mucus (Fig. 337, *s*).

The *tunica propria* consists of a loose feltwork woven of rigid connective-tissue fibers, intermingled with delicate elastic fibers, which in some animals (for example, in the cat) toward the epithelium is condensed to a structureless membrane. Numerous glands, the *olfactory glands* (glandulæ olfactoriæ, Bowman), are embedded in the tunica propria; they are either simple or (for example, in man) branched follicles, in which an excretory duct (*a*) situated in the epithelium, a body (*k*),

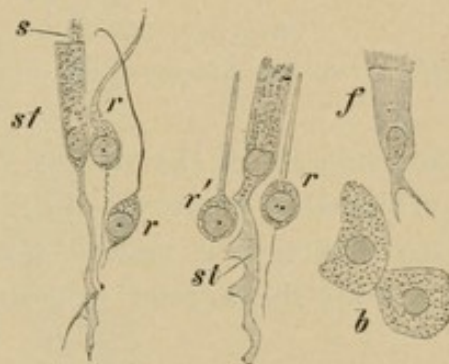


FIG. 347.—ISOLATED CELLS OF THE OLFACTORY MUCOSA OF A RABBIT. $\times 560$. *st*, Supporting cells; *s*, extruded mucus resembling cilia; *r*, olfactory cells, at *r'* the lower process has been torn off; *f*, ciliated cell; *b*, cells of olfactory glands. Technic No. 199.

^{*} Occasionally in the non-nucleated epithelial territory round nuclei, varying in quantity, are found above the zone of oval nuclei; they either belong to dislocated olfactory cells (Fig. 350) or are the nuclei of wandering, often pigmented leucocytes.

and a fundus (*g*) can be distinguished* (Fig. 349). The cells of the body of the glands are pigmented. The olfactory glands (also those of man) until recently were regarded as serous glands; latterly they have been interpreted as mucous glands. The tunica propria also carries the

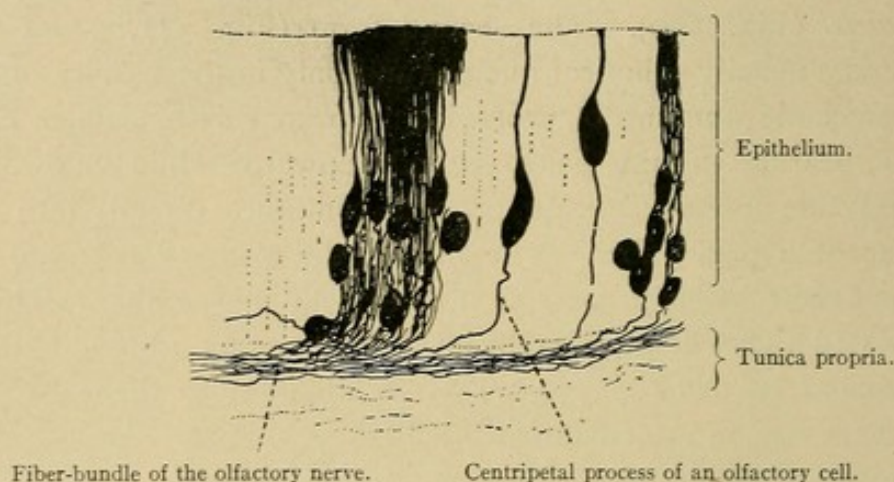


FIG. 348.—VERTICAL SECTION THROUGH THE OLFACTORY REGION OF A YOUNG RAT. $\times 480$. Technic No. 202.

ramifications of the nerves. The branches of the olfactory nerve are enveloped in processes of the dura and consist exclusively of nonmedullated fibers, that readily separate into their component fibrillæ; the fibers are the inferior processes of the olfactory cells united in bundles,

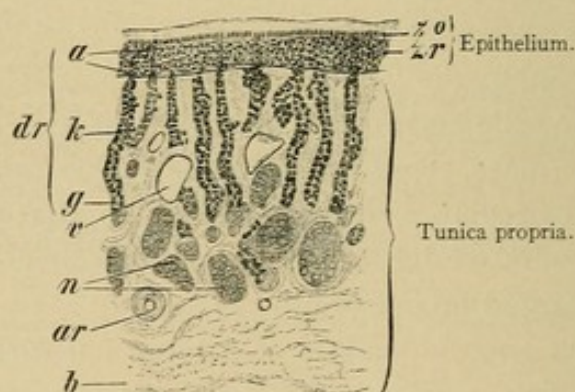


FIG. 349.—VERTICAL SECTION OF THE OLFACTORY MUCOSA OF A RABBIT. $\times 50$. *zo*, Zone of oval, *zr*, zone of round nuclei. *dr*, Olfactory glands; *a*, excretory duct, *k*, body, *g*, fundus. *n*, Branches of olfactory nerve cut transversely. *v*, Vein; *ar*, artery. *b*, Bundles of connective tissue in cross-section. Technic No. 201.

which pass in shallow curves from the epithelium and descend into the tunica propria and by union with neighboring bundles form the branches of the olfactory nerve. The terminal ramifications of the fifth nerve lie

* The olfactory glands frequently overstep the territory of the olfactory mucous membrane and are found in the adjoining divisions of the respiratory mucous membrane.

within the tunica propria; delicate fibers that ascend into the epithelium and there terminate in free ends possibly belong to the fifth nerve.*

Of the *blood-vessels* of the nasal mucosa the arterial stems run in the deeper strata of the tunica propria (Fig. 346, Fig. 349); they supply a capillary network that reaches close beneath the epithelium. The *veins* are remarkable for their conspicuous development (Fig. 346); over the posterior end of the inferior turbinal they form so dense a network that the tunica propria resembles cavernous tissue.

The *lymph-vessels* form coarse-meshed nets lying in the deeper strata of the tunica propria. The lymph-vessels of the olfactory mucosa can be injected from the subarachnoid space, through the perineural sheaths

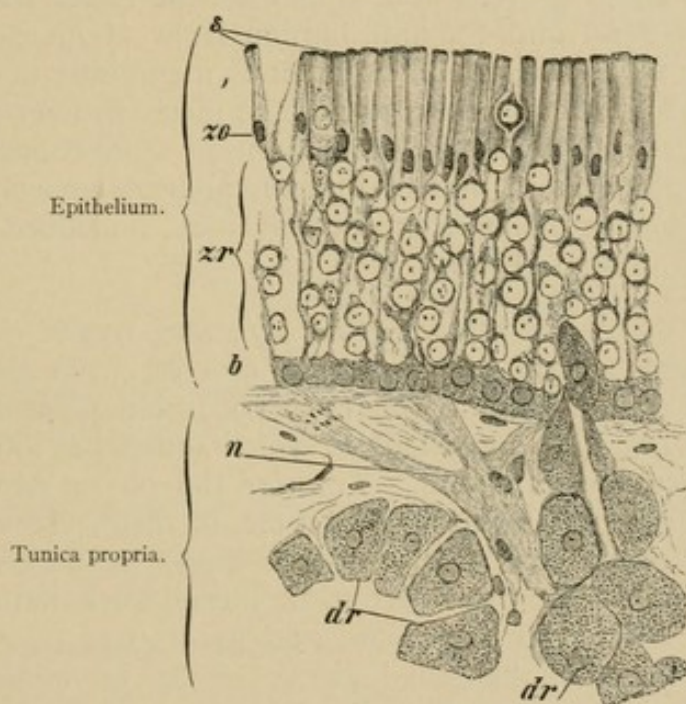


FIG. 350.—VERTICAL SECTION THROUGH THE OLFACTORY MUCOSA OF A RABBIT. $\times 560$. *s*, Cuticular border; *zo*, zone of oval, *zr*, zone of round nuclei; *b*, basal cells; *dr*, portions of olfactory glands, on the right the lower portion of the excretory duct is shown; *n*, branch of the olfactory nerve. Technic No. 202.

of the branches of the olfactory nerve obtained from the cerebral membranes on passing through the cribriform plate.

Medullated twigs of the fifth nerve can be found in the respiratory as well as in the olfactory mucosa.

TECHNIC.

No. 199.—*Olfactory cells*.—Saw open in the median line the head of a rabbit just killed. The olfactory mucosa is easily recognized by its

* Different authors have described structures in the nasal mucous membrane resembling the taste-buds. However, it is not certain but that folds of the nasal mucous membrane may have been mistaken for these "olfactory buds."

brown color. With fine scissors carefully cut out a piece of the mucosa, about 5 mm. square, together with the corresponding portion of the turbinal bone, and place it in 20 c.c. of one-third alcohol (p. 20). In from five to seven hours transfer the same to 5 c.c. of picrocarmine and on the following day to 10 c.c. of distilled water. In about ten minutes remove the piece and *lightly* toss it against a slide on which a drop of diluted glycerol has been placed; stirring with the needle is to be avoided. Carefully apply a cover-glass. In addition to many fragments of cells many well-preserved supporting elements will be seen. Very frequently the exceedingly delicate central process of the olfactory cells is wanting (Fig. 347).

No. 200.—*The mucous membrane of the respiratory region.*—Incise a piece of the mucosa about 5 or 10 mm. square on the lower half of the nasal septum; strip it off and fix and harden it in about 20 c.c. of absolute alcohol (p. 20). Use the nasal mucous membrane of the rabbit's head (No. 199) for thin sections; embed the pieces in liver and stain the sections with Hansen's hematoxylin; mount in xylol-balsam. For general views the mucous membrane of human cadavers answers, which is to be treated in the same manner, except that thick, unstained sections are to be mounted in diluted glycerol.

No. 201.—*The mucous membrane of the olfactory region.*—Remove pieces from 3 to 6 mm. square of the brown mucosa from the upper portion of the nasal septum of a rabbit (No. 199), and place them for three hours in 20 c.c. of Ranvier's alcohol, which somewhat loosens the elements of the olfactory epithelium. Transfer the pieces carefully to 3 c.c. of 2 per cent. osmium solution plus 3 c.c. of distilled water, and place the whole for from fifteen to twenty-four hours in the dark. At the expiration of this time the pieces are to be placed for a half-hour in 20 c.c. of distilled water and then hardened in 30 c.c. of gradually strengthened alcohols. The hardened pieces are to be embedded in liver and sectioned; the sections are to be stained from twenty to thirty seconds in Hansen's hematoxylin and mounted in xylol-balsam.

In order to obtain good views of the *glands* make thick sections *transverse* to the course of the *nerve-fibers* (Fig. 349). For the exhibition of the *nerve-fibers* and the epithelium thin sections *parallel* to the course of the fibers are suitable (Fig. 350).

No. 202.—*The nerve-processes of the olfactory cells* may be obtained in preparations made according to No. 196, p. 452. Often the duct system of the olfactory glands is blackened.

XIII. THE GUSTATORY ORGAN.

The gustatory organ, the *taste-buds*, are usually ellipsoidal bodies, about 80 μ long and 40 μ broad, occasionally they are more nearly spherical, which are completely embedded in the epithelium of the oral mucous

membrane; their base rests upon the tunica propria, their upper end reaches near to the surface of the epithelium, which here exhibits a small, often funnel-shaped depression, the *taste-canal*, the outer end of which is named the *outer taste-pore*, the inner end the *inner taste-pore*. Each taste-bud consists of two kinds of slender epithelial cells; the one either

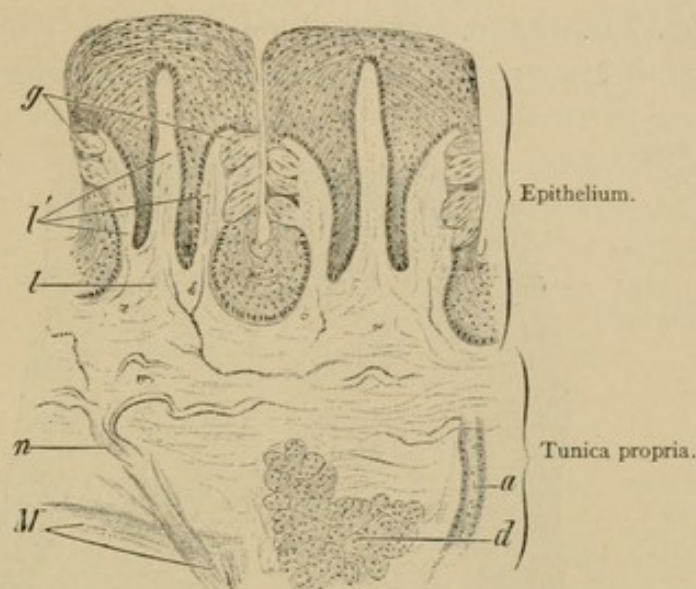


FIG. 351.—VERTICAL SECTION OF TWO RIDGES OF THE PAPILLA FOLIATA OF A RABBIT. $\times 80$. Each ridge, *l*, bears three secondary ridges, *l'*; *g*, taste-buds; *n*, medullated nerves; *d*, serous gland; *a*, portion of an excretory duct of a serous gland; *M*, muscle-fibers of the tongue. Technic No. 204.

are everywhere of the same diameter or taper at the basal end, which occasionally is forked, while the upper end is prolonged to a fine point; their protoplasm is clear. These cells constitute the bulk of the taste-bud, are principally situated at the periphery of the bud, and are called *tegmental cells* (cover-cells). They serve as support and sheath for the



FIG. 352.—FROM A VERTICAL SECTION OF THE PAPILLA FOLIATA OF A RABBIT. $\times 560$. Technic No. 204.

gustatory cells (taste-cells), which are the real sensory epithelial elements. The gustatory cells are small and only slightly thickened where the nucleus is situated, which is sometimes nearer the lower end, sometimes in the middle, rarely at the upper end of the cell. The upper division of the cell is cylindrical or—more frequently—conical, and bears

on its free end a refractile process, a cuticular formation, that reaches to the inner taste-pore (Fig. 352); the lower division is sometimes slender, sometimes thick, and terminates in a blunted end or in a triangular foot, without however extending into the connective tissue of the mucosa. Their protoplasm is granular.

The taste-buds chiefly occur in the lateral walls of the vallate papillæ (*cf.* Fig. 185, p. 258) and on the ridges of the foliate papillæ (Fig. 351), (*cf.* p. 259), in lesser number on the anterior and posterior-

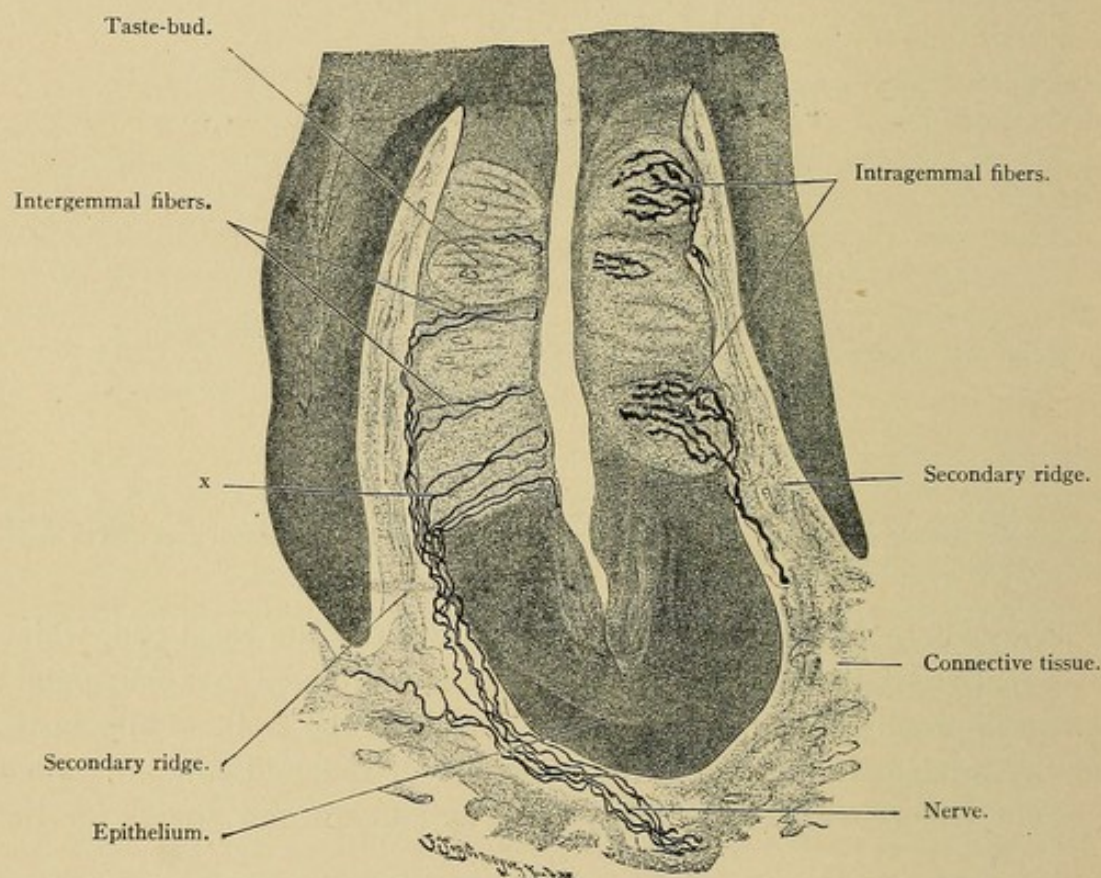


FIG. 353.—FROM A VERTICAL SECTION OF THE FOLIATE PAPILLA OF A RABBIT. $\times 220$. At x the intergemmal fibers lie upon a taste-bud. For orientation compare with Fig. 351. Technic No. 205.

lateral fungiform papillæ and on the posterior surface of the epiglottis. In the human fetus of from 5 to 7 months they are more numerous * than in the adult.

The conjecture that the terminal ramifications of the glossopharyngeal nerve have the same anatomic relation to the gustatory cells that the olfactory nerve-fibers have to the olfactory cells has been shown to be erroneous. The terminal branches of the glossopharyngeal nerve

* They are found on many filiform papillæ, as well as on the vallate papillæ. Later they atrophy and their remains are taken away by leucocytes that have wandered in. Not infrequently in adults leucocytes—often in large quantities—are found in the interior of the taste-buds.

consist of medullated and nonmedullated nerve-fibers beset with microscopic (sympathetic) ganglia,* which form a dense plexus in the tunica propria, from which numerous branches arise. Some of the latter, perhaps, terminate in the connective tissue in end-bulbs, but the majority of the (nonmedullated) fibers penetrate into the epithelium. Here two kinds of fibers can be distinguished. The one kind, the "intragemmal" † fibers, enter the taste-buds (Fig. 353), divide and form a plexus beset with numerous conspicuous varicosities, that extends up to the taste-pore; the intragemmal ramifications of the nerve-fibers do not anastomose with one another, nor do they unite with the gustatory cells, but all terminate in free ends. The other kind, the smoother "intergemmal" fibers, penetrate the epithelial areas between the taste-buds and without dividing usually extend into the uppermost strata of the epithelium.

TECHNIC.

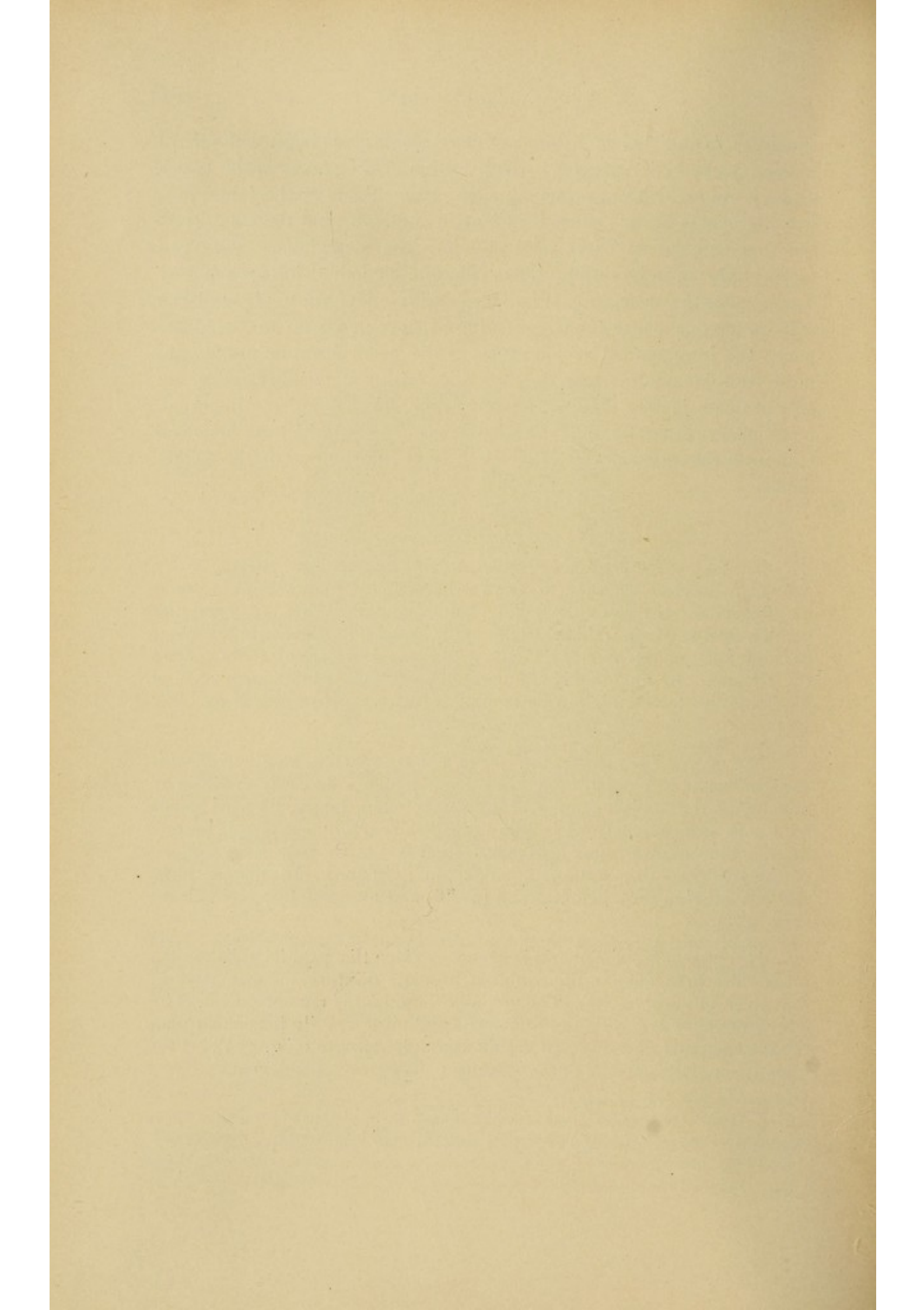
No. 203.—*For orientation regarding the number and position of the taste-buds* proceed according to the method in No. 102 (p. 298). Suitable objects are the vallate papillæ of any animal (*cf.* Fig. 185) and the papillæ foliatæ of the rabbit. The latter consist of elevated groups of parallel folds of the mucosa, situated one on either edge of the root of the tongue. In moderately thin sections vertical to the long axis of the folds the taste-buds can be recognized with the low power as clear spots.

No. 204.—*The minute structure of the taste-buds.*—Dissect off with straight scissors a papilla foliata of a rabbit just killed, with as little as possible of the subjacent muscle substance. Pin the papilla with spines on a cork stopper, the muscle side toward the cork, and expose it for one hour to the vapor of osmic acid (see further p. 34, No. 10). Thin sections of the hardened preparation embedded in liver are to be stained thirty seconds in Hansen's hematoxylin (p. 38) and mounted in xylol-balsam (Fig. 351).

No. 205.—*Exhibition of the nerves.*—Place the papillæ foliatæ of a rabbit for three days in the osmio-bichromate mixture, for two days in the silver solution (p. 45). The "double" method is recommended. The intergemmal fibers are more numerous and more readily blackened than the intragemmal fibers, which are exceedingly delicate (Fig. 353). Frequently single tegmental and gustatory cells become blackened.

* Whether the so-called "taste granules" beneath the epithelium of the papillæ foliatæ are multipolar nerve-cells is very questionable; a nerve-process has not yet been demonstrated.

† From gemma, the bud.



APPENDIX.

MICROTOME TECHNIC.

THE MICROTOME.

The commonly used microtomes are constructed according to two different principles.

The principle of the one kind consists therein, that the object to be sectioned is elevated by the shifting of the object-holder up an inclined plane.

In the other form the object is elevated in a vertical direction by a micrometer-screw.

Both kinds are excellent instruments.*

All parts of the microtome should be kept as clean as possible. When not in use it should be protected from dust by covering it with a light wooden case. The slideway in which the knife moves must be kept scrupulously clean. It should be occasionally cleansed with a cloth moistened in benzin and then should be freely lubricated with vaselin, so that the sliding-block will pass evenly throughout the entire slideway at the lightest touch.† Especial care must be bestowed upon the knife. Only with a very sharp knife can series of very thin sections be made.

* The workmanship of the sliding microtomes of Thoma, made by Jung in Heidelberg, is excellent, as I know from my own experience. The size No. IV is especially recommended. The microtomes constructed on the same principle by G. Miehe, in Hildesheim, are also highly recommended.

† The slideway of the Thoma microtome should be less freely oiled, to prevent the object-holder from recoiling backward.

Editor's remark: The *automatic microtome of Minot* is widely used, particularly in American laboratories. This instrument is distinguished from those above described by the great rapidity with which it can be worked. Therefore it is to be highly recommended, especially for the preparation of long series of paraffin sections attached one to the other in the form of a ribbon ("ribbon-cutting"). In exactness of action it is hardly surpassed by the German models, from which it altogether differs in construction. The object is moved by the rotation of a wheel in a vertical direction up and down across the edge of a knife and previous to every cut is advanced toward the knife a certain distance, which is regulated by an automatic micrometer-screw.

It is difficult to recommend in particular any of the microtomes mentioned. Each has its advantages and disadvantages, and to be successfully used demands a certain amount of experience and practice, which determines the individual preference for a certain instrument.

The Minot microtome is made by E. Zimmermann, Leipzig, Germany, and in the United States by the Bausch & Lomb Optical Co., New York. The latter also make a very satisfactory sliding microtome, on the principle of the Schanze microtome.

A really sharp knife should easily cut a thin hair held at one end between the fingers.

EMBEDDING.

THE PARAFFIN METHOD.

The following materials and apparatus are required :—

1. *Paraffin*: two kinds, a soft (melting-point 45° Celsius) and a hard (melting-point 52° Celsius). Of this prepare a mixture which melts at 50° Celsius. Much depends on the proper proportions of the two sorts of paraffin in the mixture; many a failure is due to an unsatisfactory mixture. The precise proportions cannot be given, because the consistence of the paraffin depends in a great measure on the outer temperature. Hard objects, as well as the cutting of very thin sections, require a harder mixture than usual. For winter, at a room-temperature of 20° Celsius, a mixture of 30 grams of soft and 25 grams of hard paraffin* answers for most purposes.

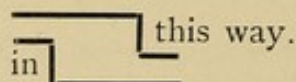
2. *Chloroform*: 20 c.c.

3. *Paraffin-chloroform*: a saturated solution (5 grams of the paraffin mixture and 25 c.c. of chloroform). This solution is liquid at room-temperature.

4. *An embedding oven* of block-tin, with double walls between which is a space to be filled with water.† A small gas-burner is to be placed beneath the oven. On top there are three openings; two lead into the space between the walls, into one a Reichert thermo-regulator‡ is to be inserted, into the other a thermometer; the third opening leads into the air space of the oven and into this a second thermometer is to be inserted. The oven should be 25 cm. long, 15 cm. high, and 15 cm. wide.

The embedding oven with its accessories is indispensable if much embedding in paraffin is to be done; but the paraffin can be melted on a water-bath and kept liquid with a small spirit flame.

5. *An embedding frame*. This consists of two adjustable bent metal plates, placed together



Instead of this frame little paper trays made of stiff paper or cardboard can be used.

The objects to be embedded must be absolutely free from water and to this end should have lain three days in absolute alcohol which has been changed several times; they are then transferred to a bottle containing 20 c.c. of chloroform, in which they should remain until the

* To be obtained from Dr. Grüber, of Leipzig.

† Made by R. Jung, Heidelberg, Germany, and in the United States by the Bausch & Lomb Optical Co., New York.

‡ To be obtained of the Bausch & Lomb Optical Co., New York.

following day.* From this the objects should be carried to the solution of paraffin in chloroform and in from two to eight hours, according to their size, transferred to a capsule containing melted, but *not too hot* paraffin.† In about a half hour the objects are to be transferred to a second capsule with melted paraffin, where, according to their size, they are to remain from one to five hours.‡ The paraffin should not be heated more than two or three degrees above its melting point; for the mixture advised the air in the oven should have a temperature of from 50° to 53° C.

When the objects have been in the paraffin bath the required length of time, place a slide in a broad dish and on this the embedding frame, into which paraffin and object now are to be poured. While the paraffin is still fluid with a heated needle place the object in the desired position; so soon as this is done carefully pour cold water into the dish until it reaches the upper margin of the frame; the paraffin will at once begin to solidify, whereupon more water may be added until the entire frame is submerged. By this manipulation the paraffin hardens into a homogeneous mass, whereas otherwise it is apt to crystallize and is then difficult to cut and also has an injurious influence on the structure of the embedded tissues. In about ten minutes the metal frames can be removed; the paraffin block should be allowed to remain in the water on the slide until it is completely solid.

The embedded object may be sectioned in a half hour. In case it is to be used later mark it with a needle. In the paraffin the object can be kept for an indefinite period.

THE CELLOIDIN METHOD.

The celloidin in plates (Grübler) has a soft consistence. The plates are to be cut in small pieces and dried in the air, in a place free from dust; they become yellow and as hard as stone. Then 16 grams of this dry celloidin are dissolved in 100 c.c. of absolute alcohol plus 100 c.c. of ether. The half of this 8 per cent. solution is diluted with 50 c.c. of absolute alcohol plus 50 c.c. of ether. The half of this 4 per cent. solution is diluted with 25 c.c. of absolute alcohol plus 25 c.c. of ether.

All three solutions should be preserved in well-stoppered, wide-necked bottles, containing from 15 to 20 grams of copper sulfate that has been heated to white heat (p. 20, No. 3), and if they become too thick can be diluted by adding equal parts of alcohol and ether.§

* This is sufficient for all cases; for small objects from one to two hours will be enough.

† If the paraffin has been melted on a water-bath, place the flame at such a distance that the surface remains covered by a thin film of solid paraffin.

‡ This is done in order to remove all the chloroform from the object. It is self-evident that the same capsule must be used for the transfer from the paraffin-chloroform. If after frequent use the capsule contains much chloroform, it can be driven off by stronger heating of the paraffin. So long as the paraffin contains any chloroform bubbles will rise on dipping in a heated needle.

§ After a time the solutions become turbid and milky; it is better then to let them dry completely and to redissolve the pieces in the alcohol-ether mixture.

The tissues to be embedded must be completely free from water and must have lain one or two days in absolute alcohol which has been changed several times. From this the objects are transferred for twenty-four hours into a mixture of equal parts of alcohol and ether and then into the 2 per cent., the 4 per cent., and the 8 per cent. celloidin solution and remain in each for twenty-four hours. The objects can remain longer in the celloidin solutions, but usually are thoroughly saturated in the course of twenty-four hours; but large objects enclosing many spaces must remain in the thick solution about eight days. Each object then should be quickly placed on a cork stopper and some celloidin poured over it. In doing this care must be taken not to press the object against the cork, or it will easily become detached. There should be a stratum of celloidin one or two millimeters thick between the cork and the object.* Now the whole is to be placed under a bell-glass, for from one-half to four hours, to slowly dry; delicate objects dry in a half hour; the bell-glass should not be air-tight, and to prevent this should be supported on one side on a needle or something similar.

When dry the objects are placed in a jar containing about 30 c.c. of 80 per cent. alcohol and to keep them submerged glue the under surface of the cork stopper by means of celloidin to the inner surface of the lid of the jar. On the following day the alcohol should be replaced by 70 per cent. alcohol, in which the objects may remain an indefinite length of time.

The celloidin objects need not be immediately mounted. In this case take the objects from the 8 per cent. celloidin solution and let it stand for several hours in a tightly covered capsule, until the air-bubbles in the celloidin have disappeared. Then remove the cover and let the capsule with the objects stand under a bell-glass for from 6 to 12 hours, or until a membrane has formed over the surface. Then place the capsule with its contents in 70 per cent. alcohol; after 24 hours cut out of the solid celloidin a block containing the objects and preserve it in 70 per cent. alcohol.

In order to cut very thin sections the celloidin must be *hardened*; for this purpose take the objects embedded in celloidin from the 70 per cent. alcohol and put them for two days or more into an alcohol-glycerol mixture (80 per cent. alcohol one part, pure, concentrated glycerol from six to ten parts). The larger the proportion of glycerol to alcohol, the harder the celloidin becomes; an extreme limit is one part of alcohol to 30 parts of glycerol. Still greater difference in the proportions produces strong curling of the sections. In order to prevent the buckling of the elastic celloidin block dry it carefully with filter-paper when it is removed from the alcohol-glycerol mixture, make a pair of lateral incisions and dip it into liquid paraffin; such blocks cannot be preserved dry, they must be returned to the alcohol-glycerol mixture.

Preparations fixed by Golgi's method require special treatment, since the absolute alcohol has an injurious influence if the object remains in it

* This stratum must not be thicker; even well-hardened celloidin is elastic and a thick stratum of such elastic material would cause the object to give in sectioning.

beyond one hour. When the tissue is taken from the silver solution it is hardened in 30 c.c. of 95 per cent. alcohol for fifteen or twenty minutes, then in 30 c.c. of absolute alcohol for fifteen minutes, then placed in the thin celloidin solution for five minutes. Meanwhile, in the previously smoothed lateral surface of a broad piece of elder-pith make an excavation just large enough to take in the *whole* preparation; insert it, cover it with celloidin solution, fit a second piece of elder-pith on the first, pour on more celloidin, and place the whole for five minutes under a bell-glass to dry; then transfer it to 80 per cent. alcohol for five minutes and cut sections with a knife flooded with 80 per cent. alcohol. The microtome is altogether unnecessary; satisfactory sections can easily be cut free-hand. If the microtome is used, the thickness of the sections should vary from 40 to 120 μ . The elder-pith should be trimmed off so that only a small shell (1 mm.) encircles the celloidin.

SECTIONING.

PARAFFIN OBJECTS.

The paraffin block containing the object is to be secured in a hollow cylinder coated with hard paraffin (in the Thoma microtome) or (in the microtome of Miehé) to a little plate* instead of the clamp. The plate is simply warmed and the paraffin block glued to it by pressure. In the case of the cylinder, warm it and also the base of the paraffin block; press the latter lightly into the cylinder and by means of a heated needle inserted between them establish a firm union. In order quickly to solidify the paraffin place the cylinder or the plate for five minutes in cold water. The projecting portion of the paraffin block containing the object should then be trimmed to a four-sided column, the base of which is right-angled square. The column should not be taller than one centimeter and the object should be encircled by a layer of paraffin not over one or two millimeters broad.

The cylinder (or the plate) with the object should now be placed in the microtome. Sections are to be cut with the blade of the knife dry. The position of the knife depends on the nature of the object.

Sectioning with the knife placed obliquely.† If the object is large and of unequal resistance the knife should be so clamped that it forms a very acute angle with the long axis of the microtome. The paraffin block should so stand that the knife strikes it first on one *corner* of the column. The knife should be moved *slowly* and pressure upon it should be carefully avoided.

Sectioning with the knife placed transversely. Screw the knife down perpendicular to the long axis of the microtome, turn the paraffin column so that the blade will strike it first on a *surface*. The knife should

* Instead of the plate I use cylindric pieces of soft wood, about 3 cm. high and 1.5 cm. in diameter, which are screwed in the object clamp.

† In Miehé's microtome the carrier of the clamp must be changed to the middle of the instrument, when the knife is to be placed obliquely.

be *rapidly* moved with a planing movement and then the sections will adhere to one another at their edges and form long ribbons. When the paraffin is of the right consistence the first section lies smoothly on the blade and is shoved by the second section in the direction of the back of the knife. If however the first sections show an inclination to curl and fall over the edge, they must then be carefully held with a delicate sable brush and led back to the right position. Ribbon-cutting is most successful when the sections have a thickness of 0.01 of a millimeter; thicker sections easily curl and do not readily adhere to one another at their edges.

OBSTACLES IN SECTIONING AND THEIR REMEDY.

Every one who has worked with paraffin is probably able to explain different unsuccessful attempts.

1. The knife glides over the object and cuts a partial section or none. The reason for this may lie in the microtome; the slideway may not be clean; examine the vertical portion of the slideway. Or the knife is not sharp enough, or the under surface has paraffin adhering to it; in the latter case remove the knife and with a cloth wetted with turpentine carefully cleanse it. Knives with thin backs buckle if the distal end of the blade is used; thus it happens that when the knife is obliquely placed the blade cuts the object only at the edge where it first touches and glides over the rest without cutting it. In microtomes of earlier construction the cause of this often lies in the unsatisfactory manner in which the block of paraffin is secured.

Secondly, the trouble may be found in the object; it may be too hard, or of very unequal resistance, or poorly embedded; in the latter case there are two possibilities. Either the preparation was not thoroughly dehydrated, in which case it exhibits opaque spots, or it contains chloroform; in this case it is soft, and light pressure with needle on the surface leaves a mark or even presses out fluid. In both cases the procedure of embedding must be repeated, reversing the series of processes to the absolute alcohol (in the latter case to the paraffin bath).

Finally, the consistence of the paraffin may be at fault.

2. The sections curl. This can be prevented by holding a small sable brush or bent needle lightly against the sections as they are cut.* The cause of this curling lies in the hardness of the paraffin, which is also responsible for—

3. The sections break. The usefulness of the paraffin depends in a high degree on the outer temperature. If the paraffin is too hard do not endeavor to reduce its consistence by the admixture of soft paraffin, —this is the last resource—but employ simpler measures. Cut the sections near a stove or near a lamp; often *slight* warming of the knife is sufficient. Even very good paraffin crumbles when cut with a cold knife.

* A "section-smoother" for microtomes in which the object is elevated vertically is made by Kleinert of Breslau. See further, Born, "Zeitschr. f. wissensch. Mikroskopie," Bd. x, p. 157.

4. The sections fold and become pressed together. As a result of this the sectioned objects acquire a false outline. The reason for this lies in a too soft paraffin. This difficulty may be overcome by frequently placing the block in cold water or by cutting the sections in a cold room (in summer, in the morning hours).

CELLOIDIN OBJECTS.

Trim the embedded object until the enveloping stratum of celloidin is only one or two millimeters thick; clamp the knife obliquely, so that it makes a very acute angle with the long axis of the microtome. Moisten the blade with 70 per cent. alcohol by means of a sable brush; this must be done after every second or third section is cut. The sections should be removed with a brush and transferred to a dish containing 70 per cent. alcohol. Very thin sections (less than 0.02 mm.) cannot be cut unless the celloidin has been hardened (p. 465).

PRESERVATION OF SECTIONS.

PARAFFIN OBJECTS.

If the sections are not very thin and are not in ribbons they may be placed in a capsule with 5 c.c. of carbol-xylol and when the paraffin is dissolved transferred to a second capsule with carbol-xylol. From this the sections, if the tissue has been stained in bulk, are carried to a slide and mounted according to the directions given on page 48. If the sections are unstained transfer them from the carbol-xylol to 5 c.c. of ninety-five per cent. alcohol, which is to be changed in two minutes. In another two minutes the sections may be stained.

In the case of serial sections and very thin sections, it is necessary first to glue the dry sections to the slide.

For this purpose the slide must be *absolutely clean*; wash it with alcohol and dry it with a clean, *not oily*, cloth or place it for a half hour in cold soapsuds. On the well-dried slide arrange the sections (or portion of the "ribbon"), and at the edge of the same place a drop of distilled water by means of a delicate sable brush. Another section (or portion of the ribbon) is now placed on the slide, another drop of water added, and so on until the slide is covered. It does not matter if the sections float. Pass the slide through a spirit-flame or place it for from one to three minutes in the oven; * on being slightly warmed, the sections spread out flat and smooth. Then arrange them with a needle and by slightly inclining the slide let the water flow off or absorb it with a strip of filter-paper and, protected from dust, let the whole dry. On the following day pour carbol-xylol over the slide and if the sections are already stained mount them in xylol-balsam. In case the sections are not stained the carbol-xylol is to be wiped off and the slide placed in ninety-five per cent. alcohol. †

* The paraffin must not be allowed to melt; the resulting mixture of melted paraffin and water is not soluble in carbol-xylol.

† The carbol-xylol, also the alcohol, must be quickly wiped off, because the sections are worthless if they are allowed to become dry. Care must also be exercised in placing the stain-

After five minutes take the slide from the alcohol, which is to be quickly wiped off around the sections, and either place it in the stain or cover it with a few drops of the solution, *e. g.* hematoxylin. Then slowly transfer the slide to a dish with distilled water and either preserve it in dilute glycerol (p. 49), or after the customary preliminary treatment with ninety-five per cent. alcohol and carbol-xylol (p. 50), mount it in xylol-balsam.

CELLOIDIN OBJECTS.

Place the sections in a dish containing 20 c.c. of 90 per cent. alcohol. If the object has not been previously stained in bulk, staining in bulk is advisable, the sections may be subsequently stained; but anilin colors cannot be used, since they also stain the celloidin; even hematoxylin imparts a light-blue tint to the celloidin. The sections must not be placed in absolute alcohol, since this dissolves the celloidin; they are to be taken from the 95 per cent. alcohol and placed in carbol-xylol; when the clearing is completed mount them in xylol-balsam.

Serial sections of celloidin objects are used only for special purposes, for example, for the central nervous system. See the articles by Weigert, in the "Zeitschrift für wissenschaftliche Mikroskopie," Bd. ii., p. 490, Bd. iii., p. 480, Bd. iv., p. 209, and by Obregia, "Neurologisches Centralblatt," Leipzig, Jahrg. 9, 1890, p. 195. The negative varnish recommended by the former is to be obtained of Dr. Grüber.

ing fluid on the sections, to see that it really covers them. Loosening of the sections occurs only when there was not enough water between the section and the slide—the water must be evenly diffused between the two. The sections can also be fastened to the cover-glass and this method permits the use of smaller quantities of the staining solution, alcohol, and other reagents.

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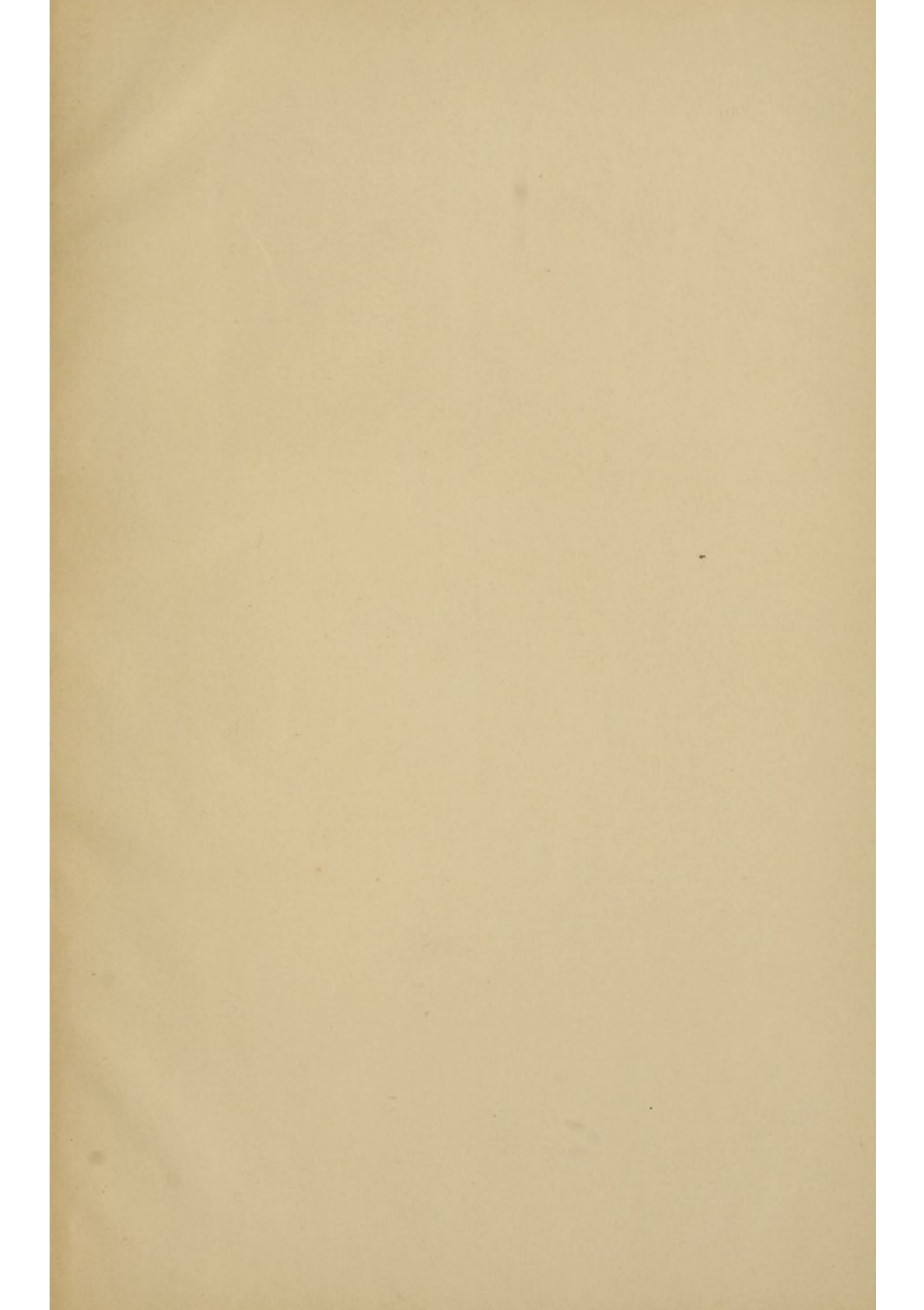
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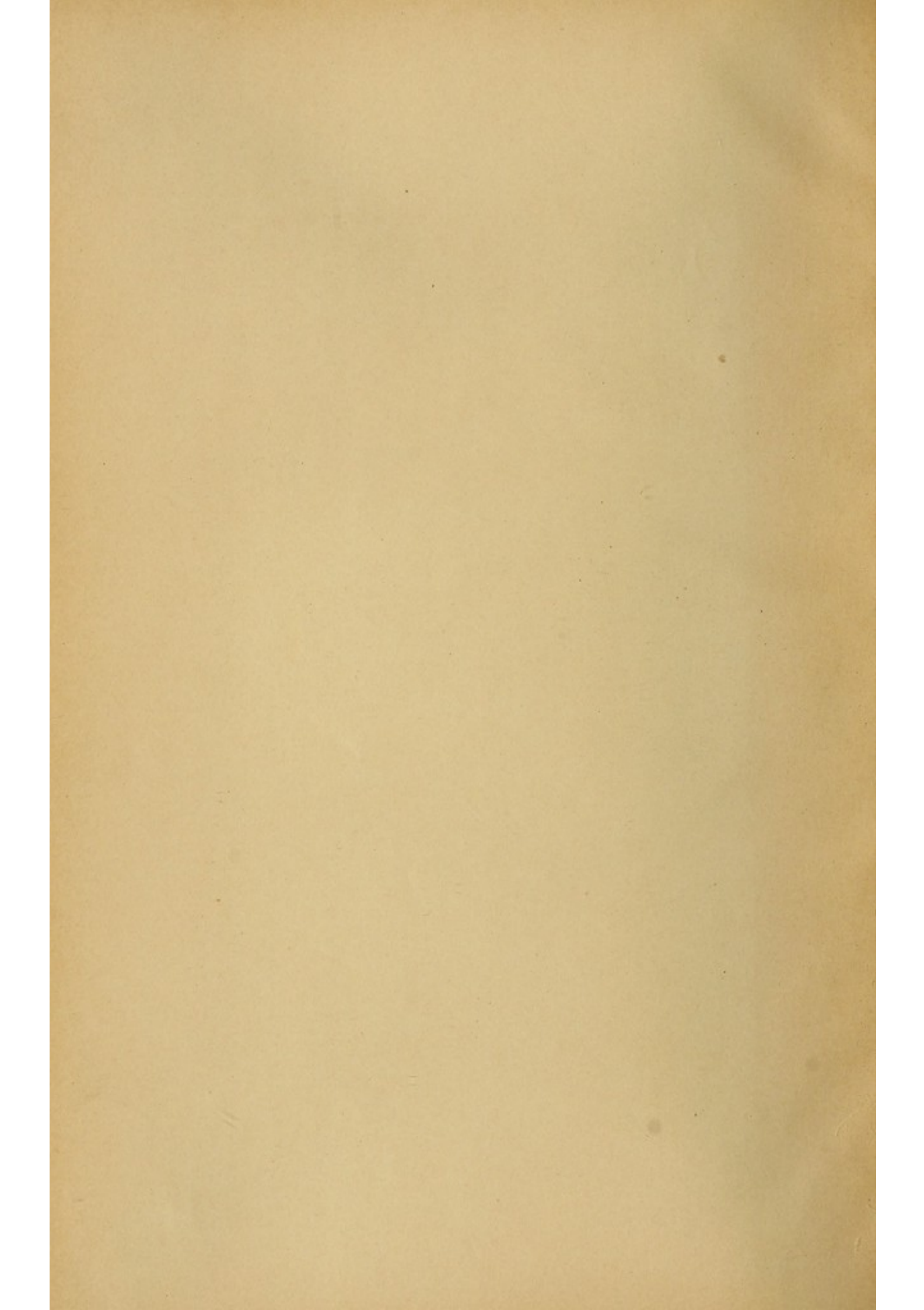
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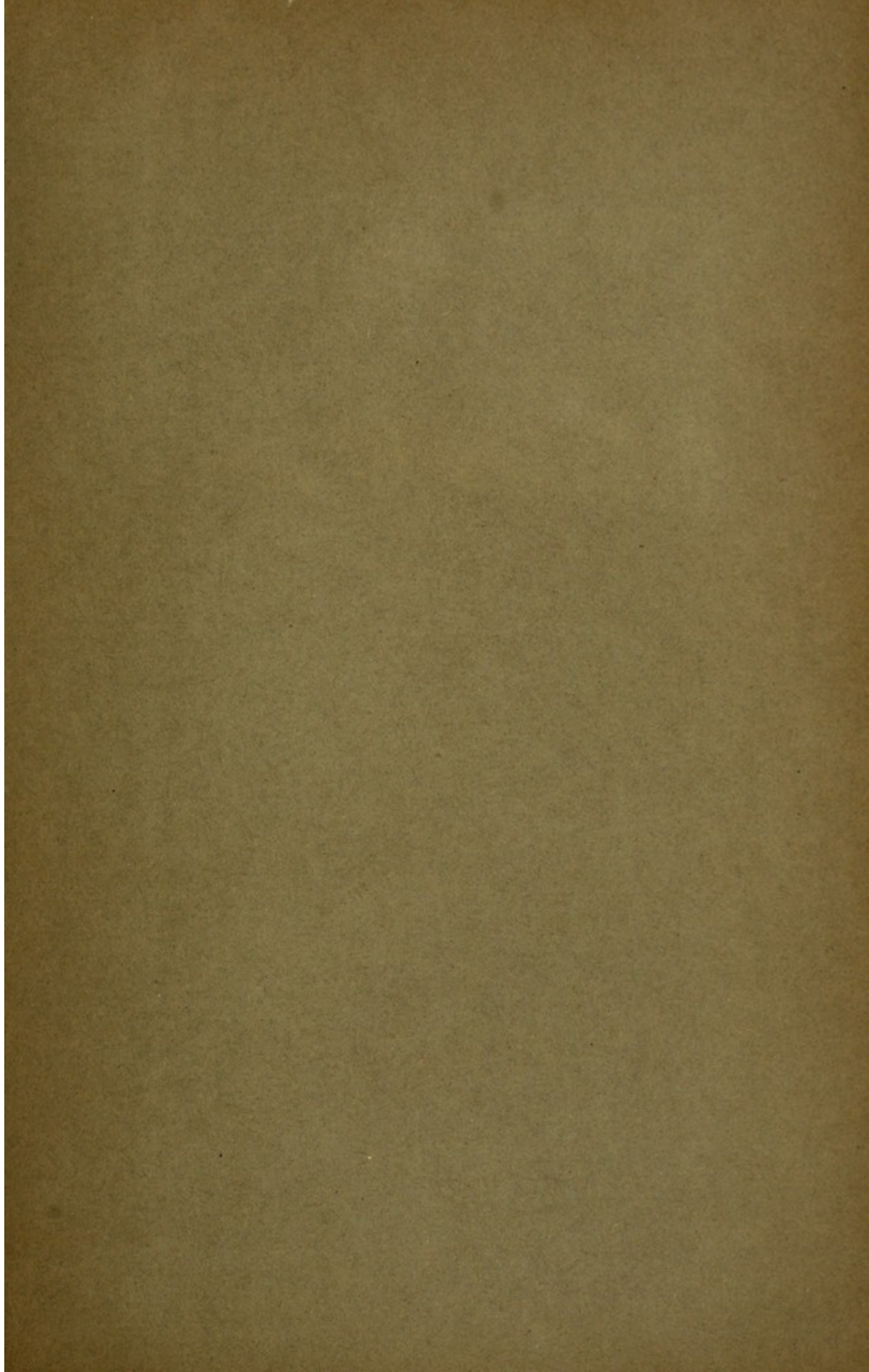
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