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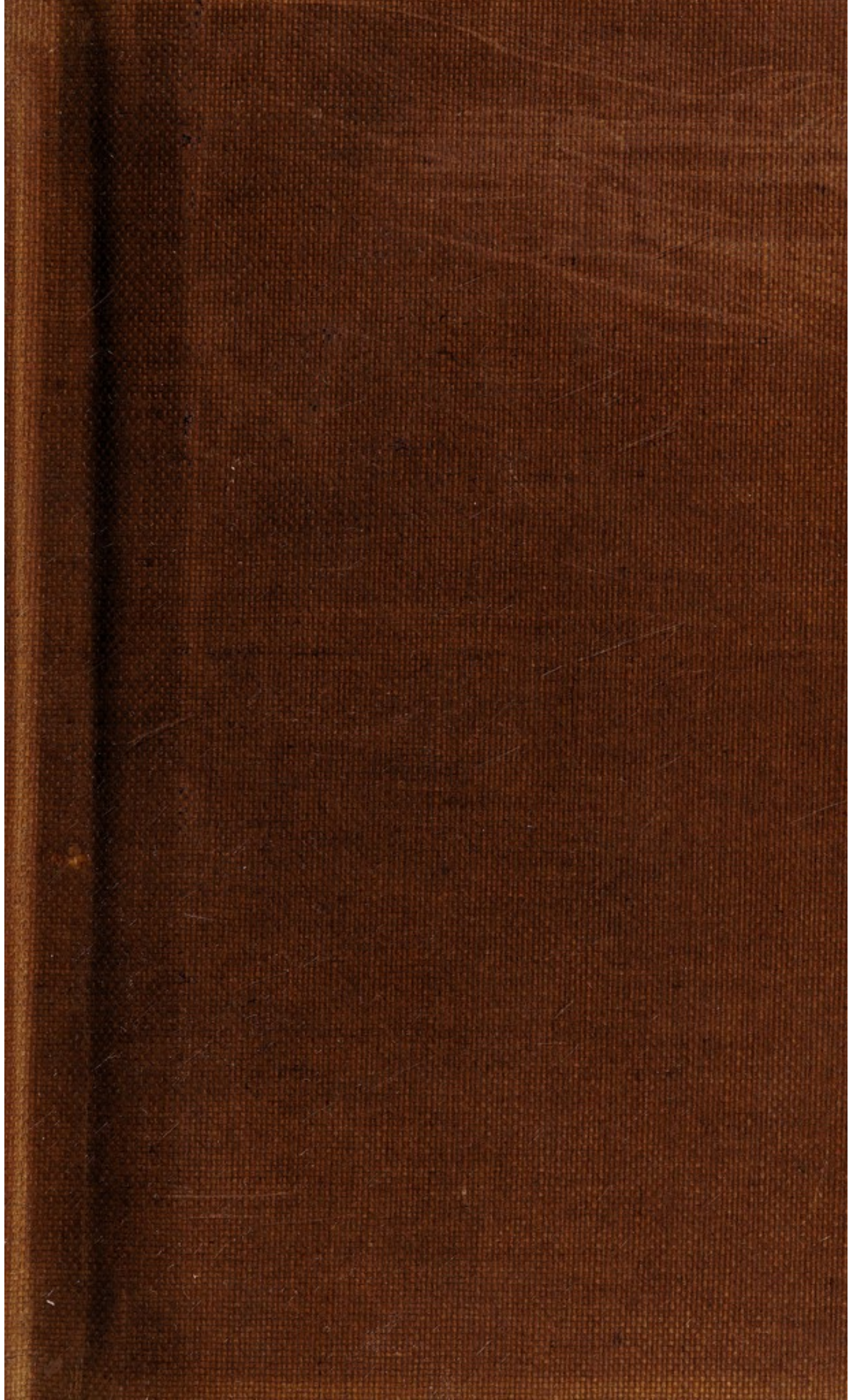
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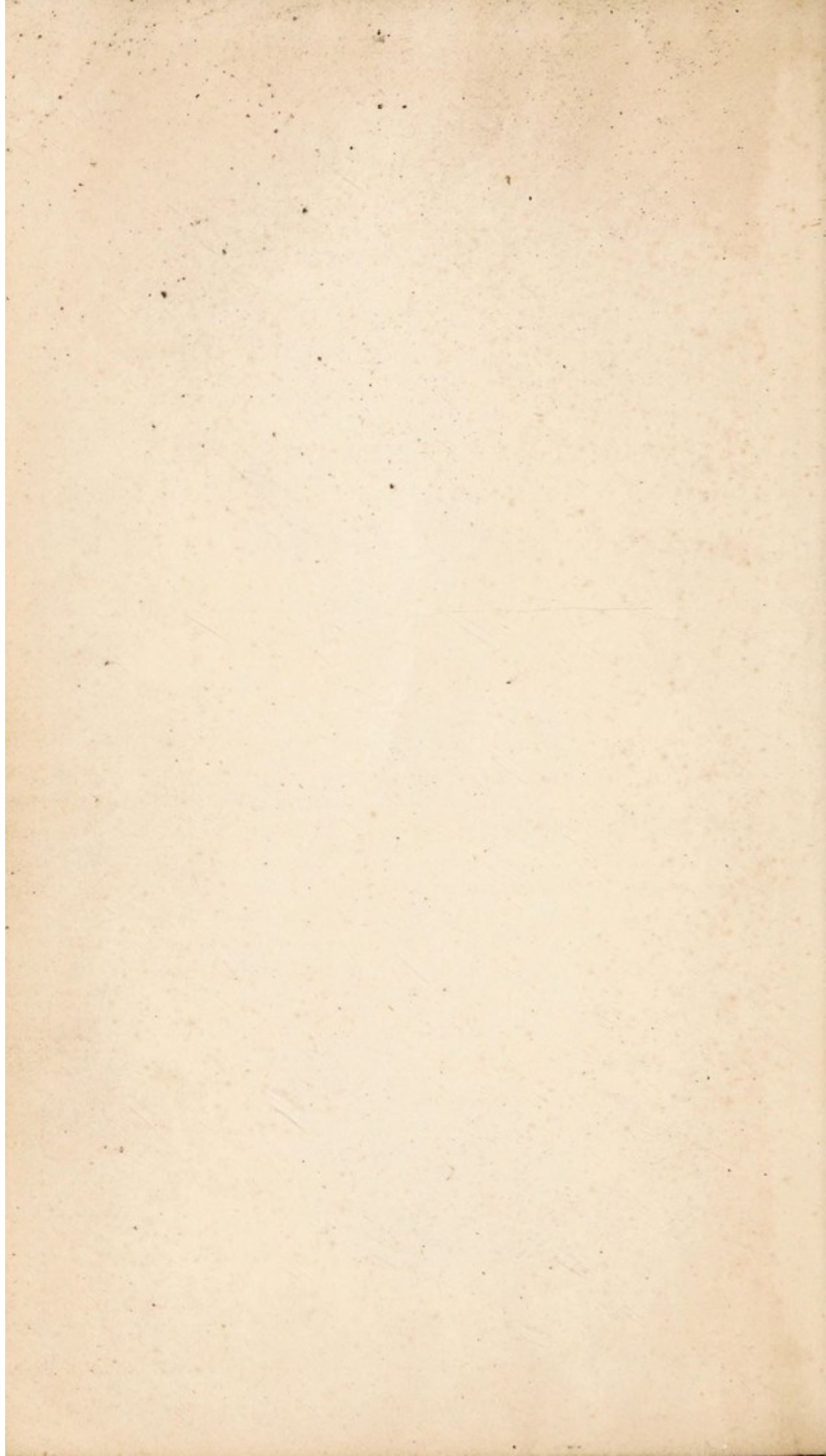
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D. I. Leech





A  
PRACTICAL HANDBOOK  
OF  
MEDICAL CHEMISTRY.



BY THE SAME AUTHOR.

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AN

INTRODUCTION TO PRACTICAL CHEMISTRY :

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A  
PRACTICAL HANDBOOK  
OF  
MEDICAL CHEMISTRY.

BY  
JOHN E. BOWMAN, F.C.S.,  
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EDITED BY  
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## PREFACE

TO THE

### FOURTH EDITION.

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DURING the seven years which have elapsed since the publication of the third edition of this work, considerable advances have been made in the practical part of Medical Chemistry.

The Editor has endeavoured to represent these as fully as is consistent with the concise and simple character which constitutes one of the great merits of Bowman's 'Handbook,' always remembering that the processes described should be such as can be carried out by the medical student, with the resources of the medical school and the hospital.

In the chapters on the Analysis of Urine, where



the greatest services are rendered by chemistry to clinical medicine, processes have been introduced for the quantitative determination of kreatinine and of ammonia, and the methods of determining them have been carefully revised at the laboratory table. The application of the volumetric principle to the analysis of urine has been extended as far as it appears to be safe, since the rapidity with which volumetric determinations may be executed, with great relative accuracy, and without demanding great skill on the part of the analyst, recommends this method strongly to the attention of the medical practitioner.

Short practical directions for the examination of the solid excrements, of bile, and of the liquids of muscular flesh, have been added; but chyle, lymph, &c., have been omitted, as not generally obtainable for analysis.

Much improvement remains to be made in the difficult examination of mixed fluids for the proximate constituents of animal bodies, though the recent researches of English and Continental chemists have enabled the editor to make some additions, which, it is hoped, will prove useful.

Since the examination for poisons in organic mixtures is comparatively seldom undertaken, except by the professional chemist, the additions which have been made to that part of the work presuppose considerable familiarity with chemical manipulations; though it has not been forgotten that, for the purposes of a judicial inquiry, the medical man often requires a knowledge of the best processes for the detection of poisons, though not desiring to carry them out himself.

Believing that the revival of the electrolytic method for the detection of metallic poisons will give greater confidence in chemico-legal investigations, the editor has fully described its application.

Short chapters have been added upon the detection of strychnia, nicotia, phosphorus, and alcohol, in organic mixtures.

The general systematic course for the detection of poisons in organic mixtures has been tested by mixing minute quantities of the poisonous substances to which it refers with articles of food, and proving that the directions given would certainly lead to their detection.



The concluding chapter contains some concise directions for the application of the elegant process of dialysis, introduced by Professor Graham, to the separation of poisons from organic mixtures.

KING'S COLLEGE, LONDON ;

*October, 1862.*



## PREFACE TO THE THIRD EDITION.

---

THE rapid sale of two large editions of this little work encourages me to hope that I was not altogether unsuccessful in supplying a deficiency in medical literature, which has been long felt by a large body of the Profession, as well as in furnishing a plain and trustworthy text-book for the Medical Student.

In the present edition I have endeavoured without materially adding to it, to embody all the recent discoveries in Medical Chemistry which have been announced up to the present time; and thus to keep pace with the rapid advance which is every year being made in this most important branch of medical science.

JOHN E. BOWMAN.

KING'S COLLEGE, LONDON;

*January, 1855.*

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# MEDICAL CHEMISTRY.

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## PART I.

---

### CHAPTER I.

#### HEALTHY URINE.

---

##### SECTION I.

1. HEALTHY human urine is an amber-coloured, watery fluid, holding in solution a great variety of substances, both organic and inorganic, and containing also in suspension a small quantity of mucus, derived from the bladder and urinary passages. The specific gravity (278) of the healthy secretion may be said to vary from 1003 to 1030, depending on the amount of solid and liquid food taken, the period of the day at which the urine is passed, and other circumstances which tend to increase or diminish the proportion of solid matter contained in it. Thus the urine which is passed shortly after drinking much water or other fluid, commonly called *urina potus*, is usually pale in colour, and of low specific gravity, varying from 1003 to 1009; while, on the other hand, that which is secreted soon after the digestion of a full meal, commonly called *urina chyli*, has most commonly a high specific gravity, frequently 1030; the urine which is passed immediately after a night's rest, called *urina sanguinis*, may generally be considered to furnish a fair specimen of the average density of the whole urine, and will in most cases be found to have a specific gravity varying from 1015 to 1025. The average density of the whole urine passed by an individual in the twenty-four hours is usually from 1015 to 1020; and the quantity passed during the same period varies from twenty to forty-eight or fifty ounces, holding in solution usually from 600 to 700 grains of solid matter (279).



2. While warm, urine has a slightly aromatic smell, which is not perceptible after cooling. It is usually slightly acid to test-paper, from the presence of acid phosphate of soda ( $\text{NaO}, \frac{1}{2}\text{HO}, \text{PO}_5$ ), but the experiments of Dr. Bence Jones show that when passed shortly after eating, the urine is often neutral, or even alkaline, becoming again gradually more and more acid, up to the time when the next meal is taken. When kept for some little time, it first becomes a little more acid (apparently from the formation of a little lactic and acetic acids), and deposits a few crystals of uric acid entangled in the cloudy deposit of mucus; but after a longer period it putrefies, becoming alkaline and ammoniacal, and deposits a sediment of earthy phosphates, previously held in solution by the free acid (43). If the urine be kept for a still longer time, it becomes more and more concentrated by spontaneous evaporation, deposits minute crystals of chloride of sodium, phosphates, and other salts, and eventually becomes covered with a grayish-coloured mould, containing minute fungi and animalcules.

3. Although chemists have not yet succeeded in insulating for examination all the ingredients of urine, nor even in ascertaining the general nature and character of several of the compounds which probably enter into its composition, still they have, by their researches, determined what appear to be the most important of its constituents; and it is to these only that the student need turn his attention, leaving the more problematical and obscure parts of the subject to be decided by the future labours of the physiological chemist.

4. The solid matters of the urine may be said to consist of the following—viz., *Urea*; *uric acid*; *hippuric acid*;  *kreatinine*; *grape-sugar*; *vesical mucus*, and *epithelial debris*; *animal extractive*; *ammoniacal salts*; *fixed alkaline salts*; and *earthy salts*.\*

5. The student will do well to test a little of the healthy secretion, which should, for this purpose, be that passed immediately after a night's rest (1), for these several substances, in the manner described under each, in the following sections; and if he has leisure and opportunity, he may prepare specimens of urea, uric and hippuric acids, and some of the other constituents.

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\* According to Campbell, urine also contains a minute quantity of formic acid ( $\text{C}_2\text{H}_2\text{O}_4$ ).



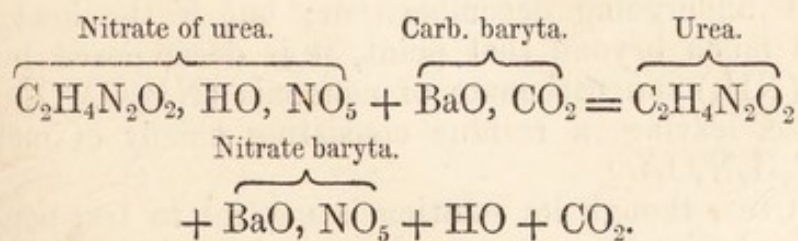
## SECTION II.

*Urea* ( $C_2H_4N_2O_2$ ).

6. This important ingredient of the urine, which appears to be the vehicle by which nearly the whole of the nitrogen of the exhausted tissues of the body is removed from the system, is a solid crystalline substance, colourless when in a state of purity, and easily separated from the other matters with which it is associated.

7. The presence of urea in the urine may be readily shown by concentrating a little of the secretion to about one half or one third its bulk, and mixing it with an equal quantity of pure nitric acid; when delicate crystalline rhomboidal plates of impure nitrate of urea ( $C_2H_4N_2O_2, HO, NO_5$ ) will be found to gradually to separate from the liquid (16).

8. Pure urea may be obtained from the nitrate thus separated. For this purpose, about a pint of urine, filtered from the mucus as soon as possible (11), is evaporated, at a heat below its boiling point, to two or three ounces; when cool, the concentrated urine is decanted from the deposited salts, and mixed with an equal bulk of colourless nitric acid (sp. gr. 1.25). After standing for some time, the pasty mass of nitrate of urea is pressed, to free it from the adhering liquid, dissolved in a little boiling water, and allowed to crystallize. The pure crystals are again dissolved in hot water, and finely powdered carbonate of baryta is added in small portions, as long as any effervescence is perceptible. The nitric acid has now combined with the baryta, whilst the carbonic acid, being incapable of combining with the urea, makes its escape.



The excess of carbonate of baryta is separated by filtration, and the clear liquid evaporated to dryness on the water-bath. The dry residue of urea and nitrate of baryta is boiled with a little alcohol, which dissolves only the urea, and, when decanted off and evaporated, deposits it in prismatic crystals resembling

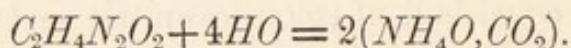


nitre, which may be purified, if necessary, by dissolving in water, decolorising with animal charcoal, and evaporating the filtered solution.

9. The crystals of urea, which, when obtained by slow evaporation, are four-sided prisms, and deliquesce slightly in air, are soluble in about their own weight of cold water, and in a much smaller quantity of hot; from which latter the urea separates on cooling, in the form of beautiful silky needles. It is soluble in about 4.5 parts of cold alcohol, and in less than half that quantity of hot; in cold ether it is nearly insoluble. Its taste is saline and cooling, somewhat resembling that of nitre.

10. The proportion of urea present in healthy urine appears to vary from twelve to upwards of thirty parts in 1000, about fourteen or fifteen being the average.

11. An aqueous solution of urea may be kept, provided it is pure and tolerably concentrated, for a considerable length of time, without undergoing chemical change; but if any albumen or mucus, or other fermentescible matter, is present, decomposition rapidly sets in, and in a short time the whole of the urea becomes transformed into carbonate of ammonia ( $NH_4O, CO_2$ ), the elements of water being at the same time assimilated.



In urine, this change speedily takes place, owing to the presence of mucus; the secretion thus acquiring, especially in warm weather, an alkaline reaction in the course of a few hours after being passed. Under the influence of the caustic alkalies, also, urea becomes gradually converted into carbonic acid and ammonia.

12. When heated on platinum foil to about  $250^\circ$ , urea fuses without undergoing decomposition; but if the heat be increased much beyond that point, it is decomposed into ammonia ( $NH_3$ ) and carbonate of ammonia ( $NH_4O, CO_2$ ), which volatilize, leaving a residue consisting chiefly of melanuric acid ( $C_6H_4N_4O_4$ ).

13. Urea, though its solution is neutral to test-paper, has decidedly basic characters, combining with acids to form salts, some of which are crystalline. Of these, the two which are of the most practical importance are the oxalate ( $C_2H_4N_2O_2, HO, C_2O_3$ ) and the nitrate ( $C_2H_4N_2O_2, HO, NO_5$ ), which, on account of their sparing solubility in water, supply a ready means of separating urea from the other matters co-existing in the urine.



14. *Oxalate of urea* ( $C_2H_4N_2O_2, HO, C_2O_3$ )\* may be prepared by concentrating urine on a water-bath to about one eighth its bulk, and filtering through muslin, in order to separate the insoluble sediment of phosphates and urates, which are gradually deposited during the evaporation. The liquid thus clarified is mixed with about an equal bulk of a strong solution of oxalic acid in hot water, or the solid acid in powder may be added as long as the liquid, heated to about  $190^\circ$  or  $200^\circ$ , continues to dissolve it. The mixture, on cooling, deposits an abundant crop of crystals of oxalate of urea, mixed with a little of the excess of oxalic acid, and coloured brown by the adhering impurities. The crystals are then gently expressed between folds of filtering paper, washed with a small quantity of ice-cold water, and purified by re-crystallization; the last traces of colouring matter being removed, if necessary, by boiling the solution with purified animal charcoal.†

15. The oxalate thus obtained is colourless, and in the form of tabular or prismatic crystals (fig. 1), which are readily soluble in hot water, but only sparingly so when cold, twenty-five parts of which dissolve not more than one part of the salt.

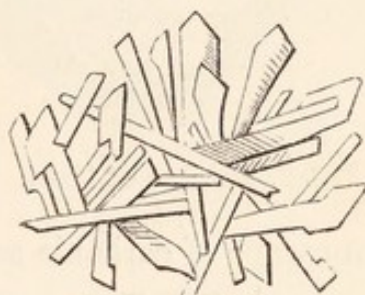
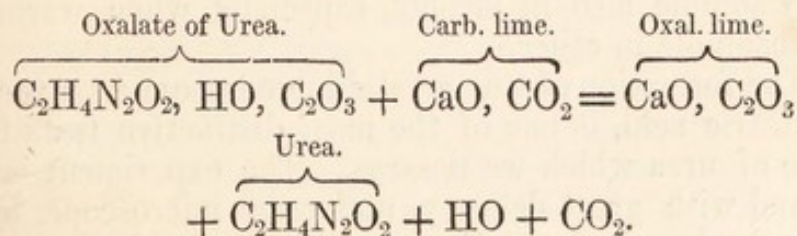


Fig. 1. Oxalate of Urea.

The oxalate of urea obtained from urine may be employed to furnish pure urea, by dissolving it in hot water, and adding powdered chalk as long as it causes effervescence. The insoluble oxalate of lime is then filtered off, and the solution of urea evaporated on a water-bath to crystallization.



\* The bibasic character of oxalic acid being now generally recognised, the formula of oxalate of urea should be written  $2(C_2H_4N_2O_2), 2HO, C_4O_6$ .

† Animal charcoal is purified from the phosphate and carbonate of lime by repeatedly boiling it with hydrochloric acid, till the acid liquid is not precipitated by ammonia; the charcoal must then be washed with water till the latter is no longer acid.



16. *Nitrate of urea* ( $C_2H_4N_2O_2, HO, NO_5$ ) may be obtained by adding strong, colourless nitric acid, free from nitrous acid, to urine previously concentrated by evaporation to about one third its bulk ; the nitrate gradually separates in irregular

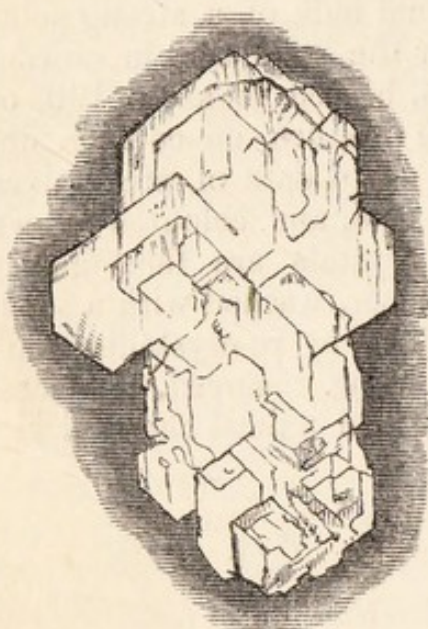
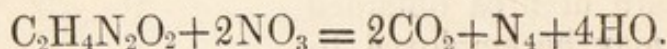


Fig. 2. Nitrate of Urea.

rhomboidal plates (fig. 2), more or less coloured and modified in form by the impurities present. The crystals are washed with a little ice-cold water, then pressed between folds of filtering paper, and redissolved in lukewarm water ; lastly, they are purified by re-crystallization, and if necessary, the last traces of colouring matter may be removed by boiling the solution with purified animal charcoal.

The absence of nitrous acid in the nitric acid employed for precipitating the urea is insisted on because this substance is immediately decomposed by nitrous acid, with violent effervescence, from escape of carbonic acid and nitrogen.



Even with colourless nitric acid, a slight effervescence always takes place, since a little nitrous acid is formed by the action of the urinary colouring matter upon the nitric acid.

17. Nitrate of urea is soluble in about eight times its weight of cold water, and in a much smaller quantity of hot. It is tolerably soluble also in alcohol, especially when warm ; but almost insoluble in ether.

18. The formation of this crystalline compound on the addition of nitric acid, is one of the most distinctive tests for the presence of urea which we possess. The experiment is made easily, and with great delicacy, under the microscope, by concentrating a drop or two of urine on a glass slide, and adding to it about an equal quantity of pure nitric acid ; the nitrate will gradually crystallize in delicate rhomboidal plates (fig. 2), the number and abundance of which will furnish some indication of the quantity of urea present in the secretion (181).



## SECTION III.

*Uric (or Lithic) Acid* ( $C_{10}H_4N_4O_6 = 2HO, C_{10}H_2N_4O_4$ ).

19. Uric acid, though usually present only in small quantity in human urine, appears to be one of the most important of its ingredients; and as the proportion varies considerably in many forms of disease, its determination, when in abnormal quantity, frequently affords much valuable assistance to the physician in diagnosis. The proportion present in the healthy secretion appears to vary from 0.3 to nearly 1.0 in 1000 parts, about 0.4 being the usual average. It probably exists, for the most part, in combination with alkalis, since, when uncombined, it requires nearly 15,000 times its weight of cold water to dissolve it, while the alkaline urates are considerably more soluble (22).

20. Uric acid may be obtained by adding to urine, previously concentrated to about half its bulk, a few drops of hydrochloric acid ( $HCl$ ), and allowing the mixture to stand for a few hours in a cool place.\* Minute reddish crystals of the acid gradually appear, having the forms shown in fig. 3, stained with the colouring matters co-existing in the urine. These crystals may then be dissolved in moderately dilute potash, and from the solution thus obtained the pure acid may be again precipitated in a crystalline and colourless state, by supersaturating it with hydrochloric acid.



Fig. 3. Uric Acid.

21. The crystalline forms in which uric acid is presented to us are very various (186), but they all appear to be modifications of the rhombic prism. Most of these crystals, when examined with the polarising microscope, develop very beautiful colours; and their forms are frequently characteristic, and indicative of the peculiar circumstances under which they may have been deposited.

22. Uric acid requires, according to Liebig, about 15,000 times its weight of cold, and nearly 2000 times its weight

\* Even without previous concentration, urine will generally deposit crystals of uric acid, if mixed with a little hydrochloric acid and set aside.



of hot water to dissolve it, forming, in the latter, a solution which is feebly acid to test-paper. It is insoluble in alcohol, and nearly so in dilute hydrochloric and sulphuric acids; it dissolves in the latter acid when concentrated, and is reprecipitated on the addition of water. It combines with bases, especially the alkalies and alkaline earths, forming salts (urates), which are for the most part insoluble, or very sparingly soluble in water. Of these the most soluble is the *urate of potash* ( $2\text{KO}, \text{C}_{10}\text{H}_2\text{N}_4\text{O}_4$ ), which dissolves in about 35 times its weight of hot water. On this account, uric acid dissolves with comparative facility in a dilute solution of potash. *Urate of soda* ( $2\text{NaO}, \text{C}_{10}\text{H}_2\text{N}_4\text{O}_4$ ) requires for its solution 124 times its weight of hot water; and *urate of ammonia* ( $\text{NH}_4\text{O}, \text{HO}, \text{C}_{10}\text{H}_2\text{N}_4\text{O}_4$ ) 243 times its weight of hot, and about 1720 of cold, water, to effect its solution. The presence of a small quantity of chloride of sodium, such as is contained in the urine, renders water capable of dissolving nearly twice as much urate of ammonia as is taken up by pure water.\*

23. The action of nitric acid ( $\text{HO}, \text{NO}_5$ ) upon uric acid is highly characteristic, and furnishes, perhaps, the most delicate test of its presence which we possess. If a little of the acid, in the state of powder, is placed in a drop or two of tolerably strong nitric acid, in a watch-glass or on a strip of glass, it will gradually dissolve; carbonic acid ( $\text{CO}_2$ ) and nitrogen being given off with effervescence, and leaving behind a mixture of alloxan† ( $\text{C}_8\text{H}_4\text{N}_2\text{O}_{10}$ ), alloxantine ( $\text{C}_8\text{H}_5\text{N}_2\text{O}_{10}$ ), urea, and some other compounds. This may then be evaporated nearly to dryness at a gentle heat, when a red residue will be left, which, *when cold*, should be moistened with a drop or two of ammonia, or exposed to ammoniacal fumes, which will develop a beautiful purplish-red colour, owing to the formation of murexide ( $\text{C}_{12}\text{H}_6\text{N}_5\text{O}_8$ ). If the mass be now moistened with solution of potash, a very beautiful purple colour will be produced. The potash may be applied at once to the residue left after evaporating the nitric acid, and is a far more delicate and characteristic test than the ammonia. The same effect is produced when urate of ammonia, or any other urate, is similarly treated.

\* Lithia forms one of the most soluble urates. Schilling has shown that the acid urate of lithia ( $\text{LiO}, \text{HO}, \text{C}_{10}\text{H}_2\text{N}_4\text{O}_4$ ) dissolves in 39 parts of boiling water, and 368 parts of cold water. The neutral urate would be still more soluble.

† Alloxan has been found by Liebig, on one occasion, in mucus from the intestines.



24. When heated before the blow-pipe, uric acid is decomposed, emitting a disagreeable smell, resembling that of burnt feathers, mixed with that of hydrocyanic acid, which, together with carbonate of ammonia and some other compounds, is formed during the decomposition.

## SECTION IV.

*Hippuric Acid* ( $\text{HO}, \text{C}_{18}\text{H}_8\text{NO}_5$ ).

25. A small quantity of hippuric acid appears to be generally present in healthy urine, and in certain forms of disease, especially in cases where a vegetable diet has been adopted, the quantity is found to increase considerably.\* In jaundice, according to Kühne, hippuric acid is entirely absent from the urine, even after the administration of benzoic acid, which is converted, in the normal state of the system, into hippuric acid.

26. Hippuric acid may be prepared from fresh human urine, or still more readily from the urine of the herbivora, in which usually contains it in much larger quantity than the human secretion. The urine is first evaporated at a gentle heat until it has the consistence of a syrup; it is then, after cooling, supersaturated with hydrochloric acid, which will dissolve the earthy salts, and cause after a time a crystalline precipitate of impure hippuric acid mixed with uric acid, colouring matters, and other substances, which give it a more or less dark brown or reddish colour. The precipitate is then dissolved in a small quantity of hot water, from which it again crystallizes on cooling. To obtain the pure acid, these crystals may be boiled with hydrate of lime and water, the solution of hippurate of lime ( $\text{CaO}, \text{C}_{18}\text{H}_8\text{NO}_5$ ), filtered and mixed with excess of hydrochloric acid, which precipitates the hippuric acid, in the form of minute tufts of needle-shaped crystals (fig. 4, *a* & *b*); these may be again dissolved in hot water, and allowed to cool gradually, when beautiful

\* According to Lücke, hippuric acid is often entirely absent from the urine of persons living upon a mixed diet, and can only be detected when the food is composed chiefly of vegetables. It is said to increase in fever, in chorea, and in diabetes. Dr. Bence Jones has recently determined the hippuric acid in the urine of two healthy men living upon a mixed diet. A large number of experiments led to the mean result that, in one case, the urine of 24 hours (1.25 pint) contained 4.96 grs. of hippuric acid, and in the other (2.37 pints) 6.5 grs., the quantities of uric acid passed in the same time being, respectively, 4.74 and 11.6 grs.



crystals (four-sided prisms), will be obtained, of considerable length, but so friable, as to fall into powder under the slightest

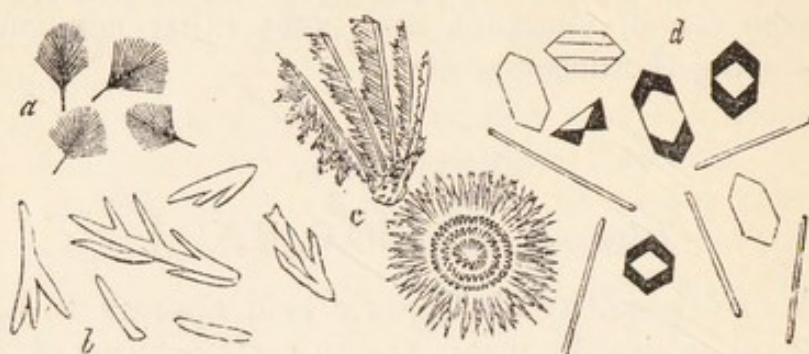


Fig. 4. Hippuric Acid.

pressure. A more minute examination for hippuric acid in human urine may be made by evaporating eight or ten ounces of urine to a syrup, acidulating with hydrochloric acid, and agitating with about an equal volume of ether, the separation of which is afterwards promoted by adding a little alcohol. If the solution in ether be evaporated, and the residue boiled with a little water, the hippuric acid will be dissolved, and deposited in crystals, when the solution is allowed to stand.

27. Hippuric acid is very sparingly soluble in cold water, requiring about 400 times its weight to dissolve it; in hot water, however, it is readily soluble, and, on cooling, crystallizes in beautiful silky tufts. It is very soluble in alcohol, but very slightly in ether.\*

28. When mixed with uric acid, it may be separated from that substance by treating the mixture either with hot water or alcohol, in both of which uric acid is insoluble or nearly so (22). It may be distinguished from uric acid also, by its giving no purple colour when tested with nitric acid and ammonia (23), and by its different crystalline form (26, 29, 186).

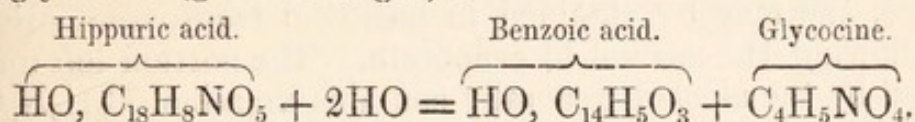
29. When an alcoholic solution of hippuric acid is allowed to evaporate slowly, the crystalline residue which is left has usually some such appearance as that shown in figure 4, *c*. When deposited from a hot aqueous solution, the crystals have more the appearance shown at *d* in the figure.

30. When heated in a tube, it is converted chiefly into

\* Thus distinguished from benzoic acid, which dissolves readily in ether.



benzoic acid ( $\text{HO}, \text{C}_{14}\text{H}_5\text{O}_3$ ), and benzoate of ammonia ( $\text{NH}_4\text{O}, \text{C}_{14}\text{H}_5\text{O}_3$ ), which sublime, together with a red oily matter (benzonitrile,  $\text{C}_{14}\text{H}_5\text{N}$ ), which has a peculiar and characteristic smell, resembling that of the Tonka bean. When boiled with acids or alkalies, hippuric acid is converted into benzoic acid and glycocine (gelatine sugar):



## SECTION V.

*Kreatinine* ( $\text{C}_8\text{H}_7\text{N}_3\text{O}_2$ ).

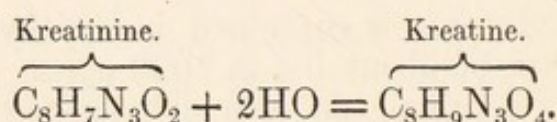
30 a. This substance is contained in healthy human urine, in the proportion of about 0.4 in 1000 parts, and in larger quantity in cow's urine, and, although its physiological and pathological relations have not yet been fully investigated, it must be regarded as a very important constituent of the excretion.

In order to extract the kreatinine, a pint of urine is neutralised with milk of lime, and chloride of calcium is added as long as it causes a fresh precipitate. The earthy phosphates are then separated by filtration, and the clear liquid evaporated on a water-bath to a small bulk, so that the salts begin to crystallize out on cooling. After it has been allowed to stand for some time, the liquid is poured off into another vessel (leaving the deposit), mixed with about  $\frac{1}{20}$ th of a saturated solution of chloride of zinc, well stirred with a glass rod, and set aside for three or four days. A crystalline precipitate will then be deposited, consisting of a compound of kreatinine with chloride of zinc ( $\text{C}_8\text{H}_7\text{N}_3\text{O}_2, \text{ZnCl}$ ). This is washed two or three times with small quantities of cold water, and dissolved in boiling water. The solution is boiled in a dish, and freshly prepared hydrated oxide of lead\* is added in small portions, till a yellow precipitate (oxychloride of lead), has separated, and the solution is decidedly alkaline. The kreatinine is thus set free and dissolved by the water, whilst the chloride of zinc is decomposed by the hydrated oxide of lead, with production of hydrated oxide

\* Prepared by precipitating nitrate of lead with a slight excess of potash, and rapidly washing, on a filter, so long as the washings are strongly alkaline.



of zinc and oxychloride of lead, which are both insoluble in water. The filtered liquid is boiled with a little animal charcoal, which removes the colouring matter, as well as any oxide of lead which may have dissolved, and after a second filtration is evaporated to dryness on the water-bath. On treating the residue with hot alcohol, the kreatinine is dissolved, and may be obtained in beautiful transparent prisms, by allowing the alcohol to evaporate. The portion left undissolved by the alcohol generally contains kreatine ( $C_8H_9N_3O_4$ ), a feeble organic base, which is found in the juice of flesh. It was formerly thought that kreatine was also a constituent of urine, but recent experiments have shown it to be formed from the kreatinine, during the evaporation of the urine, by the assimilation of the elements of water :



Since this change takes place, even in the cold, in alkaline solutions, the urine should be filtered as rapidly as possible after the addition of milk of lime, and a large excess of the latter should be avoided.

Kreatinine forms brilliant prismatic crystals, which dissolve in twelve parts of cold, and in a smaller proportion of hot water or of alcohol. The solution is alkaline to test-papers, and, even though moderately dilute, yields a characteristic crystalline precipitate with solution of chloride of zinc, especially on stirring. It combines with acids to form crystalline salts.

*Quantitative determination of kreatinine in urine.*—In order to determine the quantity of kreatinine in urine, the process given above requires some modification. The following is the method adopted by Neubauer for the estimation of kreatinine in the form of the double compound with chloride of zinc.

About 5000 grs. of urine are rendered slightly alkaline with milk of lime, and chloride of calcium is added as long as any fresh precipitate is formed. The filtered liquid is evaporated nearly to dryness on the water-bath, and mixed, while still warm, with about an ounce of strong alcohol (95 per cent.); the mixture is rinsed into a beaker, set aside for four or five hours and filtered, the residue upon the filter being washed with alcohol. The filtrate and washings are evaporated to about one and a half ounce, and mixed with a small quantity of



in a very strong alcoholic solution of chloride of zinc. After being briskly stirred, the mixture is set aside for three or four days, when the crystalline compound of kreatinine with chloride of zinc may be collected on a weighed filter, washed with alcohol, and dried at  $212^{\circ}$ . From the weight of the precipitate, that of the kreatinine may be found by the proportion :

Atc. wt. of kreati- nine and chloride of zinc.	Atc. wt. of kreatinine.			
181	113	::	Weight of precipitate	: $x$

## SECTION VI.

*Vesical Mucus and Epithelial Scales.*

31. The small traces of mucus and epithelial debris, which are always present in urine, and which do not generally amount to more than from 0.1 to 0.3, in 1000 parts of the healthy secretion, are derived from the internal surface of the bladder and urinary passages. The quantity is so small as to be scarcely visible in healthy urine, until, after standing a short time, it has subsided, in the form of a thin cloud, to the bottom of the liquid. It may be separated by passing the urine through a filter, on the sides of which it will be deposited in the form of a shining pellicle.

32. When examined under the microscope, mucus is found to consist of minute granular corpuscles (fig. 5, *a*) floating in the fluid, which are colourless, or nearly so, more or less round, and frequently oval in shape, and usually accompanied by epithelial scales. The mucus corpuscles dissolve when treated with strong nitric and acetic acids, forming a solution from which, after boiling, ferrocyanide of potassium throws down a white precipitate.



Fig. 5. Mucus Corpuscles and Scales of Epithelium. Magnified 200 diameters.



33. When treated with dilute acetic acid ( $HO, C_4H_3O_3$ ), these corpuscles become more transparent, lose their granular appearance, and show in the interior one or more distinct nuclei (662). The corpuscles are unaffected, or nearly so, by the dilute mineral acids, but readily dissolve in a solution of potash. For the further characters of mucus, see paragraphs 99, 153, 210, 247, 660, &c.

34. The epithelial scales found in the urine, associated with mucus, and derived from the epithelial covering of the organs through which the secretion has passed, are usually more or less torn and broken (fig. 5), but are occasionally met with uninjured, when they have the appearance shown at *b* in the figure.

## SECTION VII.

### *Extractive Matter.*

35. The term extractive matter is usually applied to those organic constituents of animal fluids, the nature of which cannot be exactly defined, and its use therefore becomes more restricted in proportion as analysis advances. Thus, within the last few years, the analyses of urine have disclosed, among the extractive matters, minute proportions of two substances, which are also found in the juice of muscular flesh, viz., kreatine ( $C_8H_9N_3O_4$ ), and kreatinine ( $C_8H_7N_3O_2$ ), as well as a little grape-sugar.\* The peculiar yellow colouring matter of the urine is also included under the head of extractive matters, together with minute quantities of fatty acids.

36. In stating the results of an analysis of urine, it is usual to distinguish between the *alcoholic extractive* matters, which are soluble in alcohol, and the *watery extractive*, which will dissolve only in water. The former averages about twelve parts in 1000 of normal urine, whilst the watery extract amounts to only two or three parts. The real nature of these matters is still very imperfectly understood; and until we shall have obtained further insight into them and their connexion with the animal functions, the student may consider them as so much undefined matter excreted from the body; without waiting to inquire whether lactic acid and other compounds, the presence of which may at present be considered as uncertain, are or are not contained in it.

\* Dr. Bence Jones ('Quar. Jour. Chem. Soc.,' April, 1861) has obtained as much as two grains of grape-sugar from forty ounces of healthy urine.



*Colouring matters obtained from urine.*—The yellow colouring matter of urine is characterised by the purple colour which it gives when heated with concentrated hydrochloric acid. A peculiar red colouring matter containing iron, and very similar in its character to the colouring matter of the blood, has been extracted from urine\* by the following process:—The urine was evaporated to a syrup, and treated with alcohol; the alcoholic solution boiled, and gradually mixed with milk of lime, until it was decolorised. The precipitate containing the colouring matter in combination with lime was filtered off and washed successively with water and ether; it was then dried and treated with a mixture of hydrochloric acid and alcohol, by which the colouring matter was dissolved. By shaking the alcoholic solution with an equal volume of ether during several days, the colouring matter was obtained in ethereal solution, and after washing the latter with water, and evaporating, was left as a dark red mass, consisting of the *urohæmatin* accompanied by a resinous substance.

Several observers have recorded the occasional presence of a blue colouring matter in urine, as well as the existence, in other cases, of some substance which gave rise to the separation of an insoluble blue matter, when the urine was mixed with sulphuric or hydrochloric acid and allowed to stand. Dr. Hassall showed that this substance corresponded, in its properties, to ordinary indigo-blue, and the more recent experiments of Schunck have demonstrated the existence in almost specimens of healthy urine of a substance possibly identical with the *indican* existing in woad, which is resolved, by the action of strong acids, into sugar and indigo-blue  $\text{K}(\text{C}_{16}\text{H}_5\text{NO}_2)$ .† Indigo was obtained from most of the samples of healthy urine by the following process:—sixteen ounces of urine are mixed with tribasic acetate of lead in excess, and filtered. The filtered liquid is mixed with an excess of ammonia, the precipitate collected and decomposed with cold dilute sulphuric acid. The solution is again filtered and set aside, when it deposits a blue precipitate. If this be filtered off, washed with a little caustic soda, and then with boiling

\* Harley, 'Journ. Pr. Chem.,' xliv, 264.

† Samples of urine in which no sugar could be detected by the copper-test, when heated with sulphuric or hydrochloric acid, deposited brown flakes, and the filtered liquid gave decided indications of sugar. The brown precipitate had the same composition as anthranilic acid ( $\text{C}_{14}\text{H}_7\text{NO}_4$ ), which is a product of the decomposition of indigo-blue; it dissolves in boiling alcohol, with a fine purple colour.



alcohol, the latter dissolves a purple-red colouring matter, which resembles the so-called purpurine, and leaves indigo-blue undissolved, which may be recognised by its evolving violet vapours, condensable to coppery scales, on heating, and by its dissolving, with a blue colour, in concentrated sulphuric acid.\*

The odour of urine is caused by minute quantities of certain volatile acids, among which *phenylic* or *carbolic acid* ( $C_{12}H_6O_2$ ) only is well known.

### SECTION VIII.

#### *Ammoniacal Salts.*

37. In perfectly fresh urine these are present in very small quantity. The urate of ammonia which has been already noticed (19), appears to be one form in which the uric acid present in the urine is held in solution, since the free acid requires for its solution a larger proportion of water than the secretion usually contains.

38. The presence of ammonia in urine is best shown by adding a little caustic baryta ( $BaO, HO$ )† to the residue left after evaporating the liquid nearly to dryness at a gentle heat, when the odour of ammonia will be perceptible, and a rod moistened with dilute hydrochloric acid, held over it, will give rise to the characteristic white fumes of hydrochlorate of ammonia. The proportion of ammonia contained in healthy urine appears to be very small; in some forms of disease, however, especially in certain kinds of fever, the quantity is found to increase considerably. Neubauer found, in the case of one person in health, about thirteen grains of ammoniac in the excretion of twenty-four hours. In another case, about eight grains.

### SECTION IX.

#### *Fixed Alkaline Salts.*

39. In order to obtain the fixed salts present in the urine, about eight ounces should be evaporated to dryness in an

\* Heller calls the yellow colouring matter of the urine *uroxanthin*, terming the red substance derived from it *urrodine*, and the blue *uroglauine*.

† Baryta is here to be used in preference to potash, since the latter would cause the evolution of ammonia by its action upon the urea, which, in presence of the alkalies, is converted into carbonate of ammonia (11).



in a porcelain dish, in which the residue is afterwards heated as long as any fumes escape; the resulting carbonaceous mass is powdered and introduced in small portions into a porcelain crucible heated to very low redness.\* In this way the carbon will be gradually burnt off, and a grey or white ash left, consisting of a mixture of the alkaline and earthy salts; the former may then be separated from the latter by dissolving in water, in which the earthy salts are insoluble (43). The composition of this ash, however, will not *exactly* represent that of the inorganic portions of the urine, on account of the chemical changes induced among its constituents during incineration.

40. The alkaline salts, which in the healthy secretion usually amount to from ten to twelve parts in 1000, consist of the sulphates of potash and soda ( $KO, SO_3$ ) and ( $NaO, SO_3$ ), chloride of sodium ( $NaCl$ ), chloride of potassium ( $KCl$ ), and phosphate of soda ( $2NaO, HO, PO_5 + 24Aq$ ). The crystalline residue left after slowly evaporating a few drops on a piece of glass, usually has the appearance represented in Fig. 6. The crosslets (*a*) consist of chloride of sodium, and the more plumose crystals (*b*) are probably phosphate of soda.



Fig. 6. Evaporated Residue of Healthy Urine.

41. The presence of these several salts may be shown by adding to the aqueous solution of the ash the following tests:

(a) *Nitrate of silver* ( $AgO, NO_5$ ) throws down a whitish precipitate, consisting of a mixture of chloride ( $AgCl$ ) and phosphate ( $3AgO, PO_5$ ) of silver. These may be separated from each other by warming the precipitate with a little nitric acid, when the phosphate will dissolve, leaving the insoluble chloride, which may then be tested with ammonia, in which it is readily soluble.

\* At a higher temperature, partial fusion might take place, rendering complete incineration impossible; some of the alkaline chlorides might also be lost.



(b) The acid solution separated from the chloride (a) must now be cautiously neutralised with ammonia, which will throw down a pale yellow precipitate of PHOSPHATE ( $3\text{AgO}, \text{PO}_5$ ), which may be again dissolved by adding a slight excess of nitric acid.

(c) *Chloride of barium* ( $\text{BaCl}$ ), or *nitrate of baryta* ( $\text{BaO}, \text{NO}_5$ ), throws down a white precipitate of sulphate of baryta ( $\text{BaO}, \text{SO}_3$ ), mixed with phosphate of baryta ( $2\text{BaO}, \text{HO}, \text{PO}_5$ ); which latter may be separated by dilute hydrochloric acid, which leaves the sulphate undissolved, proving the presence of SULPHURIC ACID. If the acid solution of the phosphate be neutralised with ammonia, the phosphate of baryta is again precipitated.

In order conclusively to prove the presence of phosphoric acid, another portion of the aqueous solution of the ash should be acidified slightly with acetic acid, and a drop of perchloride of iron ( $\text{Fe}_2\text{Cl}_3$ ) added, which will cause a yellowish-white precipitate of perphosphate of iron ( $\text{Fe}_2\text{O}_3, \text{PO}_5$ ).

(d) The absence of all bases except the alkalies, may be proved by testing the solution with hydrosulphate of ammonia, ( $\text{NH}_4\text{S}, \text{HS}$ ) and carbonate of soda ( $\text{NaO}, \text{CO}_2$ ), neither of which will be found to cause any precipitate.

(e) POTASH may be shown to be present by adding to a little of the strong solution about an equal quantity of bichloride of platinum ( $\text{PtCl}_2$ ), which will cause a yellow precipitate of the double chloride of platinum and potassium ( $\text{KCl}, \text{PtCl}_2$ ); and another portion may be stirred on a slip of glass with solution of tartaric acid, which will throw down a white crystalline precipitate of the bitartrate ( $\text{KO}, \text{HO}, \text{C}_8\text{H}_4\text{O}_{10}$ ).

(f) SODA may be identified by dipping a clean platinum wire into the solution, and heating it in the blow-pipe flame, which will be tinged golden yellow.

42. It is difficult to say in what exact state of combination these several bases and acids exist in the urine; but it is most probable that each base is divided among the several acids, and that a portion of each of the acids is combined with some of each of the fixed bases, and also of the ammonia (37, 40).



## SECTION X.

*Earthy Salts.*

43. The earthy salts, which form the insoluble portion of the ash, and which usually amount in healthy urine to about one part in 1000, consist of the phosphates of lime and magnesia, together with a small trace of alumina and silica. These earthy phosphates, which are insoluble in water, appear to be retained in solution in the urine by the small excess of acid (probably phosphoric) usually present in the healthy secretion, and may be immediately precipitated from it by supersaturating with ammonia. The precipitate thus formed consists of a mixture of PHOSPHATE OF LIME ( $3\text{CaO},\text{PO}_5$ ), and the DOUBLE PHOSPHATE OF AMMONIA and MAGNESIA ( $2\text{MgO},\text{NH}_4\text{O},\text{PO}_5 + 12\text{Aq}$ ), which is also called TRIPLE PHOSPHATE. If this precipitate be examined under the microscope, it will generally be found to consist of minute crystals of the triple phosphate, mixed with amorphous particles of phosphate of lime (Fig. 7). Collect the precipitate upon a filter, wash it several times with water, and drop a little nitrate of silver upon it. The formation of the bright yellow phosphate of silver ( $3\text{AgO},\text{PO}_5$ ), will indicate the presence of phosphoric acid.

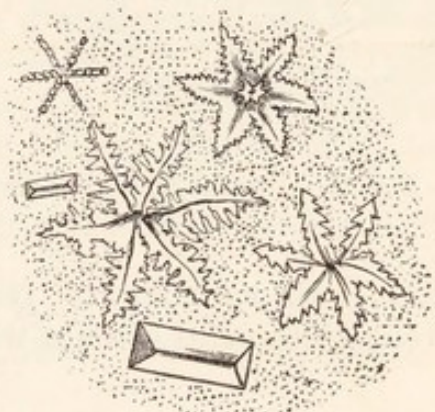


Fig. 7. Mixed Phosphates.

44. The crystalline form of the triple phosphate, as well as its chemical composition, depends upon the quantity of ammonia present in the liquid during its formation. When the urine is cautiously *neutralised* with the alkali, the crystals are prismatic (Fig. 8), and in a few rare cases, penniform\* (Fig. 9), and appear to consist of ( $\text{MgO},\text{NH}_4\text{O},\text{HO},\text{PO}_5$ ); while, if a decided excess of ammonia be added, the crystals are star-like and foliaceous, as shown in Fig. 10, and then consist of ( $2\text{MgO},\text{NH}_4\text{O},\text{PO}_5 + 12\text{Aq}$ ). When the urine gradually becomes alkaline, owing to the spontaneous formation

\* According to Dr. Hassall, these consist of phosphate of lime.



of ammonia from the urea (11), the triple phosphate is precipitated in the prismatic form, crystals of which are always to be detected in stale urine.

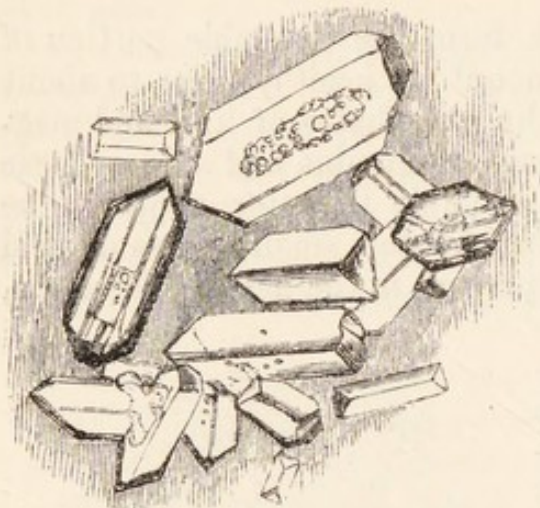


Fig. 8. Prismatic Crystals of Triple Phosphate.



Fig. 9. Penniform Crystals of Triple Phosphate.

45. Both varieties of triple phosphate will be found to develop beautiful colours when examined with polarised light.

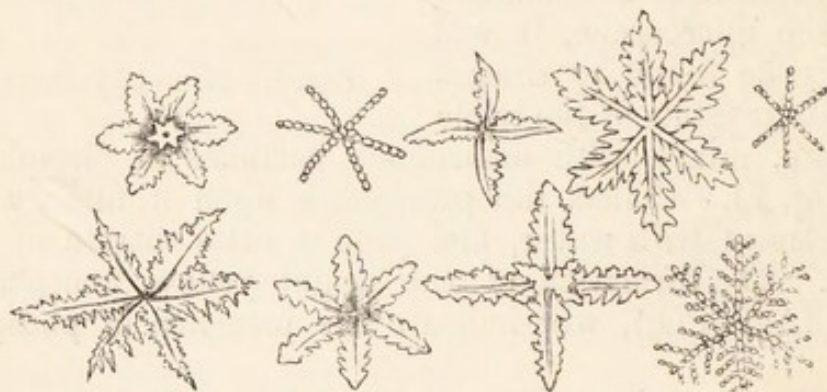


Fig. 10. Stellate Crystals of Triple Phosphate.

46. The presence of phosphoric acid, in combination with lime and magnesia, together with a trace of silica, in the insoluble portion of the ash, may be shown by digesting a considerable quantity of the latter in dilute hydrochloric acid, and filtering the solution from the insoluble residue. This insoluble portion,\* the amount of which is usually very small,

\* This residue generally contains carbon, which should be burnt off upon platinum foil.



may then be washed, and tested for SILICA, by fusion before the blow-pipe with carbonate of soda, with which it will form, when pure, a clear colourless bead (*Prac. Chem.* 138).

47. The acid solution of the phosphates, filtered from the silica, may then be divided into two portions, and tested as follows :

(a) Add ammonia in slight excess, redissolve the precipitate by adding acetic acid, and add a few drops of perchloride of iron; the yellowish-white precipitate ( $\text{Fe}_2\text{O}_3, \text{PO}_5$ ), indicates the PHOSPHORIC ACID.

(b) To the same portion add about twice its volume of water, and boil for a few seconds to precipitate the whole of the phosphate of iron; filter, and add oxalate of ammonia, which will precipitate the LIME as oxalate ( $\text{CaO}, \text{C}_2\text{O}_3$ ).

(c) The mixture (b) is boiled, and filtered from the oxalate of lime; after which the clear solution is well stirred with a decided excess of ammonia, which will in a short time cause a deposition of the crystalline double phosphate of ammonia and MAGNESIA, thus proving the presence of the latter base.

48. The same experiments (a, b, & c,) may also be made upon the phosphates which are thrown down by the addition of ammonia to fresh urine.

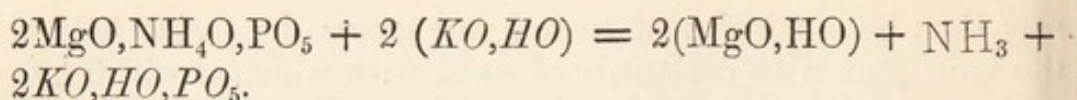
49. The earthy phosphates may also be distinguished by the following peculiarities, which may be readily seen either with or without the assistance of the microscope :

(a) When present in excess, they may frequently be precipitated from the urine in an amorphous form by boiling, thus behaving like albumen (139). The phosphatic deposit may be readily distinguished from the latter, by being soluble in a few drops of nitric acid, and in not being reprecipitated by any excess of that reagent (140).

(b) The earthy phosphates are readily soluble, without effervescence, in dilute acids, such as the hydrochloric, nitric, and acetic; and are reprecipitated by neutralising the acid solution with ammonia; that of lime being amorphous, and the triple phosphate, in a crystalline form, either prismatic or stellate (43).

(c) They are insoluble in a solution of potash. The triple phosphate, when warmed with an excess of the alkali, gives off ammoniacal fumes, which may be detected by the smell, and by the white cloud formed when a rod moistened with dilute hydrochloric acid is held at the mouth of the tube.





(*d*) When heated before the blow-pipe, phosphate of lime experiences little or no change, unless the heat be very intense, and continued for a long time, when it sometimes partially fuses. The triple phosphate, when heated, gives off ammonia and water; and the residual phosphate of magnesia ( $2\text{MgO}, \text{PO}_5$ ) fuses considerably more readily than the phosphate of lime. When the two phosphates are mixed in about equal proportion, they resemble in composition the fusible calculus, and fuse with extreme facility before the blow-pipe (392).

## CHAPTER II.

### QUANTITATIVE ANALYSIS OF HEALTHY URINE.

50. Counterpoise or weigh two Berlin porcelain evaporating basins, which, for the sake of distinction, may be marked A and B, each capable of holding about four ounces of water; and retain the counterpoises, marking them, in order to avoid confusion. Then weigh into each of the basins, 1000 grains of urine, and allow them to evaporate first on the water-bath, and afterwards in a hot-water oven, or chloride of calcium bath,\* until they cease to lose weight when weighed at intervals of an hour or two. (While the evaporation is going on, the experiments described in paragraphs 59, 66, &c., may be proceeded with. The specific gravity also may be determined (278), and the action of the urine on test-paper ascertained (277). Then accurately weigh them, and if the weights of both residues agree with each other, the loss experienced during evaporation will represent the quantity of WATER contained in the urine. If the weights do not agree, it is probable that the desiccation of at least one of the portions has been incomplete; in which case it is better to continue the heat a short time longer, until the results agree more closely.

51. The residue A may be first examined, retaining B for subsequent examination (62).

\* 'Pract. Chem.,' pp. 201, 212.



52. Warm the residue A with half an ounce or an ounce of alcohol of specific gravity about .833, stirring the mixture occasionally with a glass rod. Pour off the solution into another basin, and again warm the residue with a little more alcohol, fresh portions of which must be added until it ceases to dissolve anything more. Whether this is the case, may be known by evaporating a drop of the clear liquid on platinum foil or a slip of glass, when, if anything has been dissolved, it will be left behind as a residue. The alcoholic solution, which will contain the whole of the urea, contaminated with extractive matter and other impurities, is now to be evaporated to dryness on a water-bath, retaining the residue which proved insoluble in the alcohol for subsequent examination (57).

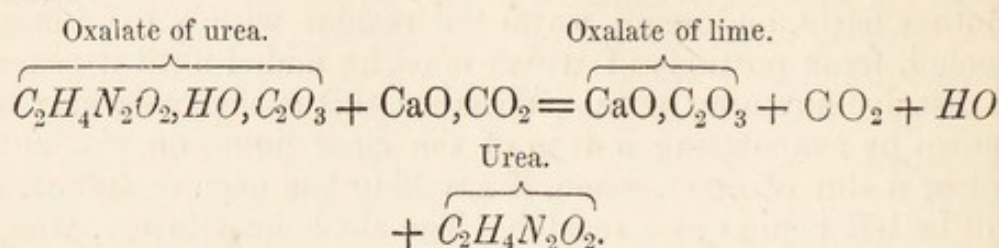
53. The residue, containing the urea, left after evaporating the alcoholic solution (52), is now to be dissolved in as small a quantity as possible of lukewarm water, and mixed with pounded oxalic acid ( $\text{HO}, \text{C}_2\text{O}_3 + 2\text{Aq}$ ), which may be added as long as the liquid, heated to about  $190^\circ$  or  $200^\circ$ , continues to dissolve it (14). The urea is thus converted into the oxalate  $\text{A}(\text{C}_2\text{H}_4\text{N}_2\text{O}_2, \text{HO}, \text{C}_2\text{O}_3)$ , which, as the solution cools, crystallizes out, mixed with some of the excess of oxalic acid employed, together with extractive matters and other impurities, which give the crystals a more or less intense brown colour. The crystals are to be washed in the basin with a very small quantity of cold distilled water, which may be poured off, and fresh water added to the crystals as long as it continues to become decidedly coloured; by which means most of the soluble salts and other foreign matters are removed.

54. The washings are now to be concentrated to a small bulk by evaporation on a water-bath, and left to cool, when a fresh crop of crystals will gradually separate. Care must be taken that an excess of oxalic acid is present in the liquid separated from the crystals, which may be known by its reddening litmus paper; if this is found on trial not to be the case, a little more of the pounded oxalic acid must be added to the solution, as otherwise, some of the urea, which, when uncombined, is very soluble in water, might escape separation.

55. When the whole of the oxalate of urea has been separated by successive crystallizations from the liquid, it must be gently pressed between folds of filtering paper, and dissolved in warm water; after which the solution is to be digested for a few hours, at the temperature of about  $100^\circ$ , with pounded carbonate of lime, stirring the mixture from



time to time with a glass rod, as long as any effervescence is produced. The oxalate is thus decomposed in the following manner :



56. The urea, which being soluble remains in solution, is to be separated by filtration from the insoluble oxalate and carbonate of lime, and carefully evaporated to dryness either on a water-bath or in vacuo over sulphuric acid. Its weight will then represent the proportion of UREA in 1000 grains of the specimen of urine under examination.\*

57. The portion of the residue which proved insoluble in the alcohol (52), containing the uric acid, vesical mucus, the extractive matter soluble in water, but insoluble in alcohol, the earthy salts, and most of the other saline matter, is now to be well stirred with successive small portions of warm water, which leaves undissolved the uric acid, mucus, and earthy salts. The insoluble matter is to be collected upon a filter, which has previously been dried and weighed, and then carefully dried on a water-bath, or in a hot-water oven, and weighed. The weight having been noted, the dry residue is to be ignited together with the filter in a crucible, until the incombustible ash becomes white, or very nearly so; when the crucible with its contents is to be again weighed. The difference between this weight and that of the dry residue previous to ignition, gives the amount of combustible matter, consisting of URIC ACID and VESICAL MUCUS; while that of the ash represents the EARTHY PHOSPHATES AND SILICA.

58. The portion of urine A will now have given us the weight of—1. The water; 2. Urea; 3. Uric acid and vesical mucus; and 4. Earthy phosphates and silica.

59. For the purpose of ascertaining the respective weights of the uric acid and vesical mucus, 2000 grains of the fresh urine may be concentrated by evaporation to about half its bulk, and mixed with twenty or thirty drops of hydrochloric

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\* The exact determination of the amount of urea in urine must be effected by an indirect method, which will be subsequently described.



acid.\* In the course of twenty-four hours, the whole of the uric acid will have been set free by the hydrochloric acid, and being insoluble (22), will be deposited in the form of minute crystals on the sides and bottom of the glass. These are to be collected on a weighed filter, and, after being washed with a little alcohol, dried in a hot-water oven or on a water-bath. The weight of this acid, divided by two (since it is derived from 2000 grains of urine), will represent the URIC ACID contained in 1000 grains of the secretion; and having already determined the quantity of uric acid and VESICAL MUCUS together (57), the weight of the latter is known by deducting from the combined weights that of the uric acid.

60. The proportion of uric acid and mucus may also be determined by evaporating to dryness 1000 grains of the urine, previously filtered from the mucus, and washing the residue first with dilute hydrochloric acid (containing one part of acid to eight or ten of water), and afterwards with a little alcohol. We thus dissolve out everything but the uric acid, which, after being washed with cold water, may be dried and weighed.

61. If it is required to determine the respective proportions of earthy phosphates and silica in the residue of earthy salts (57)—which, however, is seldom necessary, since the quantity of silica is always very small—it may be done in the following manner:—Moisten the residue with hydrochloric acid, and evaporate to dryness; then digest it with the aid of a gentle heat, in dilute hydrochloric acid, which will dissolve the phosphates, leaving the SILICA perfectly insoluble. The weight of the latter is then ascertained, and deducted from the gross weight of the earthy salts (57), when the difference will represent that of the EARTHY PHOSPHATES; or the phosphates may be precipitated from the hydrochloric acid solution by supersaturating it with ammonia, filtered, ignited, and weighed.

62. We have now to operate upon the residue left after the evaporation of the second portion of urine marked B (50), for the purpose of determining the weight of—1. The animal extractive and ammoniacal salts; and 2. The fixed alkaline salts.

63. The dry residue, after being accurately weighed, is to be

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\* In general, the uric acid may be determined in the urine without previous concentration, if it be acidulated and set aside for twenty-four hours.



incinerated (39) in a platinum or porcelain crucible, until the whole of the blackness (carbon) has disappeared, after which the weight of the ash is to be noted.\* The loss experienced during ignition being due to the combustion of the organic matters and the volatilization of the ammoniacal salts; and as we have already ascertained the weight of the urea, uric acid, and vesical mucus, we have only to deduct from the whole amount of loss the combined weights of those three substances, in order to determine the quantity of the ANIMAL EXTRACTIVE AND AMMONIACAL SALTS.

64. The ash obtained by ignition contains the whole of the inorganic matter, or, in other words, the fixed alkaline and earthy salts contained in the urine. By deducting from this the weight of the earthy salts already determined (57), we obtain the proportion of FIXED ALKALINE SALTS.

65. We shall thus have determined the proportion of the

Water,  
Urea,  
Uric acid,  
Vesical mucus,  
Animal extractive and ammoniacal salts,  
Fixed alkaline salts,  
Earthy phosphates,  
Silica,

which, when added together, ought to make up a fraction less than 1000 grains, some slight loss being unavoidable during the course of the analysis.

*Quantitative determination of Ammonia.*

65a. Place about an ounce of filtered urine in a shallow, flat evaporating dish, upon which is supported, by means of a triangle made of bent glass rod, a smaller flat dish containing half an ounce of dilute sulphuric acid, which is known to be exactly neutralised by a certain amount of a standard solution of soda. Add to the urine about half its volume of milk of lime, cover the whole with a bell glass, and allow it to stand for forty-eight hours. The ammonia which is set free by the lime will be absorbed by the sulphuric acid, and it only re-

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\* If an exact determination of the amount of ash be required, the dry residue must be carbonized by a low heat, until no more fumes are evolved, the soluble salts thoroughly extracted from it by boiling water, and their weight determined by evaporating the filtered liquid. The coal containing the insoluble salts is then dried and burnt, at a higher temperature, to a white ash.



remains to ascertain how much of the latter has been neutralised by the ammonia, which is easily done by means of the solution of soda above referred to. Every forty grains of sulphuric acid ( $\text{SO}_3$ ) neutralised, will represent seventeen grains of ammonia ( $\text{NH}_3$ ).<sup>\*</sup> Dark-coloured urine, which easily putrefies, must be precipitated with a mixture of acetate and tribasic acetate of lead, and filtered, before determining the ammonia.

*Quantitative determination of the Inorganic Salts.*

66. For the sake of practice in analysis, it will be well for the student to determine the proportions of the several bases and acids contained in the ash obtained in (39) and (63), but for purposes of diagnosis, it is more convenient to estimate the alkaline and earthy phosphates, the sulphuric acid, and the chlorine in the original urine, and this is the more necessary because the composition of the ash of any given sample of urine is liable to variation, according to the temperature at which the incineration was conducted.

67. For the quantitative analysis of the ash, about sixty grains will be required, which would be furnished by about 5000 grains (ten fluid ounces) of urine treated according to the directions given in (39); the ash should be thoroughly mixed together in a dry mortar whilst still warm, and transferred to a stoppered bottle.

68. *Determination of lime, magnesia, and phosphoric acid.*—Heat twenty grains of the ash with dilute hydrochloric acid for a few minutes, and filter the solution from the undissolved residue (carbon and silica), taking care to wash the latter as long as the washings are acid. Mix the filtered solution with ammonia in excess, stir it well, and allow it to stand for some time (if possible for twelve hours). Collect the precipitate of phosphate of lime ( $3\text{CaO}, \text{PO}_5$ ), and phosphate of magnesia and ammonia ( $2\text{MgO}, \text{NH}_4\text{O}, \text{PO}_5$ ) upon a filter, wash it with ammoniacal water as long as the washings leave any considerable residue, when evaporated upon a slip of glass, and save the filtrate and washings for further examination (72).

69. *Determination of lime.*—Dissolve the precipitate off the filter with a little warm acetic acid, taking care to wash the filter, and mix the solution with oxalate of ammonia. Allow it to stand for some time, that the oxalate of lime ( $\text{CaO}, \text{C}_2\text{O}_3$ )

<sup>\*</sup> This process was devised by Neubauer.



may separate, collect it upon a filter, wash it till the washings leave no residue on evaporation, dry, and ignite, together with the filter, in a weighed crucible, when the oxalate will be converted into carbonate of lime ( $\text{CaO}, \text{CO}_2$ ). Moisten the ignited precipitate with a little carbonate of ammonia, to convert any caustic lime into carbonate, dry at a moderate heat, and weigh. The amount of lime may then be calculated by the proportion—

$$\begin{array}{ccccccc} \text{Atc. wt. of carb.} & & \text{Atc. wt. of} & & \text{Wt. of carb. of lime} & & \text{Wt. of lime in} \\ \text{lime.} & & \text{lime.} & & \text{obtained.} & & \text{20 grs. of ash.} \\ \hline 50 & : & 28 & : : & a & : & x \end{array}$$

70. *Determination of magnesia.*—The filtrate and washings from the precipitate of oxalate of lime (69), are concentrated by evaporation, mixed with excess of ammonia, well stirred, and set aside for twelve hours; the precipitate of phosphate of magnesia and ammonia is collected upon a filter, washed with ammoniacal water, dried, ignited, and weighed. Since it is converted by ignition into pyrophosphate of magnesia ( $2\text{MgO}, \text{PO}_5$ ), the amount of magnesia will be calculated by the proportion—

$$\begin{array}{ccccccc} \text{Atc. wt. of} & & \text{Atc. wt. of} & & \text{Wt. } 2\text{MgO}, \text{PO}_5 & & \text{Wt. of magnesia in} \\ 2\text{MgO}, \text{PO}_5. & & 2\text{MgO.} & & \text{obtained.} & & \text{20 grs. of ash.} \\ \hline 111.4 & : & 40.4 & : : & a & : & x \end{array}$$

By deducting the weight of the magnesia from the total weight of the precipitate, we obtain the amount of phosphoric acid which was in combination with magnesia in the twenty grains of ash employed.

71. *Determination of phosphoric acid.*—In order to determine the phosphoric acid which was in combination with the lime, the filtrate and washings from the phosphate of magnesia and ammonia (70) may be concentrated by evaporation, and the phosphoric acid precipitated by adding a mixture of sulphate of magnesia, chloride of ammonium, and ammonia, and proceeding with the precipitate as in the last experiment, the amount of phosphoric acid being calculated by the proportion—

$$\begin{array}{ccccccc} \text{Atc. wt. of} & & \text{Atc. wt. of} & & \text{Wt. of } 2\text{MgO}, \text{PO}_5 & & \text{Wt. of phosphoric acid} \\ 2\text{MgO}, \text{PO}_5. & & \text{PO}_5. & & \text{obtained.} & & \text{in 20 grs. of ash.} \\ \hline 111.4 & : & 71 & : : & a & : & x \end{array}$$

72. To ascertain the amount of the phosphoric acid which



was in combination with the alkalies, the filtrate and washings from the first precipitate produced by ammonia (68) are evaporated to a small bulk, and the phosphoric acid determined precisely as in the last case.

73. *Determination of the chlorine.*—Dissolve twenty grains of the ash in a little dilute nitric acid with the aid of heat, filter, wash the filter till the washings are no longer acid, and mix the filtered solution with nitrate of silver as long as it causes a fresh precipitate; stir the solution briskly to favour the separation of the chloride of silver, collect the precipitate upon a small filter, wash it till the washings are no longer rendered turbid by hydrochloric acid, dry it, detach every particle from the filter, and fuse it carefully in a weighed porcelain crucible; burn the filter, allowing the ash to drop into the crucible, again ignite till all the carbon has burnt off, and weigh.\* From the weight of the chloride of silver that of the chlorine is calculated by the proportion—

Atc. wt. of chloride of silver.	Atc. wt. of chlorine.	Wt. of AgCl obtained.	Wt. of chlorine in 20 grs. of ash.
143·5	35·5	$a$	$x$

74. *Determination of sulphuric acid and the alkalies.*—Dissolve twenty grains of the ash in hydrochloric acid, filter the solution (68) and precipitate with excess of chloride of barium; collect the sulphate of baryta ( $\text{BaO}, \text{SO}_3$ ) upon a (Swedish) filter, wash it with hot water till the washings leave no considerable residue when evaporated, and save the filtrate and washings for further examination (75). Dry the sulphate of baryta, empty as much of it as possible into a weighed crucible, burn the filter so that the ash may fall into the crucible, ignite till all the carbon has burnt off, and weigh. The amount of sulphuric acid is calculated by the proportion—

Atc. wt. of $\text{BaO}, \text{SO}_3$ .	Atc. wt. of $\text{SO}_3$ .	Wt. of $\text{BaO}, \text{SO}_3$ obtained.	Wt. of sulphuric acid in 20 grs. of ash.
116·5	40	$a$	$x$

75. In the liquid filtered from the sulphate of baryta, the potash and soda have now to be determined. For this purpose,

\* If the precipitate be too small to be detached from the filter, the latter must be burnt with it, and the ash afterwards moistened with a few drops of nitric and hydrochloric acid, to convert the reduced silver into chloride; it is then again ignited and weighed.



ammonia in excess, and carbonate of ammonia must be added, and the solution gently heated. The excess of baryta, together with the lime, magnesia, and phosphoric acid, are thus precipitated, and must be filtered off, and washed till the washings leave no considerable residue on evaporation. The filtrate and washings are then evaporated to a small bulk, introduced into a weighed porcelain or platinum dish, carefully evaporated to dryness, and heated to dull redness, as long as any fumes (of chloride of ammonium) escape.

76. The residue after ignition, consisting merely of the chlorides of potassium and of sodium, is now to be weighed. It is then dissolved in a small quantity of water, mixed with a solution of bichloride of platinum, and the mixture is evaporated to dryness on a water-bath. The residue is treated with successive small portions of alcohol, which will dissolve out the excess of the bichloride of platinum, together with the chloride of sodium; leaving undissolved the double chloride of platinum and potassium ( $\text{KCl}, \text{PtCl}_2$ ). The latter is to be dried in a weighed filter, at a temperature of  $212^\circ$ , and weighed. From the weight of the double chloride thus obtained, we may then calculate that of the POTASH equivalent to it, as follows:—

Atc. wt. of the double chloride of platinum and potassium.	Atc. wt. of potash.	Wt. of the double chloride obtained.	Wt. of potash in 20 grs. of the ash.
244.1	47	$a$	$x$

77. From the weight of potash thus obtained, we are enabled to ascertain how much of the mixed chlorides (76) was chloride of potassium; and the difference between the latter and the gross weight will of course represent the quantity of chloride of sodium. The weight of chloride of potassium equivalent to the potash is for this purpose calculated as follows:

Atc. wt. of potash.	Atc. wt. of chlo- ride of potassium.	Wt. of pot- ash obtained.	Wt. of chloride of potassium con- tained in the mixed chlorides.
47	74.5	$a$	$x$

78. The weight of chloride of potassium thus calculated is then deducted from the weight of the mixed chlorides (76), and the difference will represent the weight of chloride of sodium; thus:



Weight of mixed chlorides . . . .  
 Deduct weight of chloride of potassium

Weight of chloride of sodium . . . .

79. The *whole* of the soda, however, does not exist in the urine as chloride of sodium, a portion of it being in combination with phosphoric, and, perhaps, also with some of the other acids present. We have, therefore, to calculate from the quantity of chlorine obtained in a former experiment (73) how much of the chloride of sodium obtained in paragraph 78 existed as such in the urine. This is done as follows :

Atc. wt. of chlorine.	Atc. wt. of chloride of sodium.	Wt. of chlorine in 20 grs. of ash.	Wt. of chloride of sodium in 20 grs. of ash.
35.5	58.5	$a$	$x$

80. The quantity of CHLORIDE OF SODIUM thus calculated is deducted from the whole weight of chloride of sodium previously obtained (78), and the difference will represent the amount of chloride of sodium equivalent to the SODA, which in the urine was combined with phosphoric or other acids; thus :

Atc. wt. of chloride of sodium.	Atc. wt. of soda.	Difference between the two amounts of chloride of sodium.	Soda existing as such in 20 grs. of the ash.
58.5	31	$a$	$x$

81. All the quantities obtained in the foregoing experiments (68 to 80), represent the amounts of the several saline ingredients contained in twenty grains of the ash; as, however, the organic ingredients were estimated as contained in 1000 grains of urine (65), the proportion of the inorganic constituents should also be reduced to the same scale. This may be done in the case of each constituent by the following calculation :

$$20 : \left\{ \begin{array}{l} \text{Quantity of in-} \\ \text{organic mat-} \\ \text{ter in 1000} \\ \text{grs. of urine.} \end{array} \right\} : : \left\{ \begin{array}{l} \text{Wt. of each consti-} \\ \text{tuent obtained} \\ \text{from 20 grs. of} \\ \text{the ash.} \end{array} \right\} : \left\{ \begin{array}{l} \text{Wt. of that consti-} \\ \text{tuent contained} \\ \text{in 1000 grs. of} \\ \text{urine.} \end{array} \right\}$$

82. *Determination of the alkaline and earthy phosphates, the sulphuric acid, and the chlorine in the original urine.*—Although



it would be very difficult to determine these constituents in the original urine with perfect exactness, it may be effected with sufficient accuracy for the purpose of diagnosis, since the results of the analysis of each specimen of urine are affected by the same sources of error, and may be compared with safety.

To ascertain the amount of earthy phosphates, 1000 grains of the urine may be mixed with excess of ammonia, and set aside for some hours. The precipitate is then filtered off and washed, as in (68), dried, ignited, and weighed.

The phosphoric acid contained in the solution filtered from this precipitate will, of course, be the measure of the alkaline phosphates present. In order to determine its amount, the filtrate and washings are precipitated with a mixture of sulphate of magnesia, chloride of ammonium, and ammonia, set aside for twelve hours, and further treated, as in (71).

*Volumetric determination of phosphoric acid in urine.*—The phosphoric acid in urine may be determined very easily, and, after a little practice, with considerable exactness, by ascertaining the amount of iron required to precipitate it as phosphate of sesquioxide of iron ( $\text{Fe}_2\text{O}_3, \text{PO}_5$ ) from the urine acidified by acetic acid.

A solution of sesquichloride of iron of known strength is prepared by dissolving twenty-eight grains of clean thin iron wire in half an ounce of hydrochloric acid, with the aid of heat, and gradually adding one drachm of nitric acid; the solution is evaporated very nearly to dryness at a gentle heat, and the residue dissolved in water. If the solution is not clear, it is rendered so by adding a very little hydrochloric acid, and it is then diluted to 2000 grains. 100 grains of this solution correspond to 1.775 grains of phosphoric acid.

In order to confirm this, 3.58 grains of pure crystallized phosphate of soda ( $2\text{NaO}, \text{HO}, \text{PO}_5 + 24\text{Aq}$ ), are dissolved in an ounce of water in a small beaker, the solution mixed with a little ammonia,\* then with an excess of acetic acid, and the solution of iron added from a burette until a trace of iron can be detected in the filtered liquid by ferrocyanide of potassium.

The most convenient mode of filtering a little of the fluid consists in employing a small tube about five inches long and

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\* This ammonia serves to neutralise the hydrochloric acid which is set free in the reaction, and which would otherwise dissolve the phosphate of iron;  $2\text{NaO}, \text{HO}, \text{PO}_5 + \text{Fe}_2\text{Cl}_3 = \text{Fe}_2\text{O}_3, \text{PO}_5 + 2\text{NaCl} + \text{HCl}$ .



half an inch wide, open at both ends, which should be turned over like the rim of a test-tube. Over one of these ends a circular piece of filtering paper is tightly bound by a platinum wire, so that its edges may overlap about two inches of the tube. The filter-paper having been moistened, it is dipped about an inch below the surface of the liquid containing the precipitate, when a few drops of the clear liquid will enter the tube, and may be poured into a test-tube containing a little dilute solution of ferrocyanide of potassium. As soon as it is found that, on making this experiment, a slight blue tinge is produced, it is known that a sufficient quantity of the iron solution has been added.

The 3.58 grains of phosphate of soda should have required forty grains of the iron-solution for complete precipitation.

In order to determine the phosphoric acid in urine, 500 or 1000 grains (according to the state of concentration) may be mixed with ammonia in excess, then with acetic acid in excess, and afterwards tested with the iron solution in the manner just described. From the number of grain measures of this solution required to complete the precipitation, the amount of phosphoric acid is calculated by the proportion—

Grs. of iron solution.	Phosphoric acid.				
100	1.775	::	Grs. of solution used	:	$x$

If it be desired to determine the amount of phosphoric acid existing in combination with lime and magnesia, 1000 grains of urine may be mixed with ammonia in excess, allowed to stand for an hour or two, filtered from the precipitated phosphates, the filtered liquid acidified with acetic acid, and tested, as before, with the iron solution. The difference between the amount of phosphoric acid thus obtained and the total quantity in 1000 grains, inferred from the former determination, will represent the phosphoric acid existing in the form of earthy phosphates.

83. For the determination of the sulphuric acid, 500 grains of the urine are acidified with hydrochloric acid, precipitated by chloride of barium and the precipitated sulphate of baryta treated as in (74).

The chlorine may be determined by acidifying 500 grains of urine with nitric acid, precipitating with nitrate of silver, and proceeding as in (73), avoiding as far as possible, the reducing



effect of light upon the silver-salt in the presence of organic matter.

84. If it be required to compare the acidity of different specimens of urine, it may be effected by adding to 1000 grains of the urine, from a graduated burette, a weak solution of carbonate of soda, of known strength, until it ceases to redden litmus paper. Of course the degrees of acidity will vary as the quantities of carbonate of soda employed.

### CHAPTER III.

#### AVERAGE COMPOSITION OF HEALTHY URINE.

85. The following analyses of healthy human urine will serve to give some idea of its average composition. Although in the amount of the several constituents they will be seen to differ considerably from each other, it will be found that these differences are not really quite so great as they at first sight appear, being in a great measure owing to variations in the relative proportions of water and solid ingredients (1).

#### *Analysis I. (Berzelius).*

Water	.	.	.	.	.	.	933.00	
Urea	.	.	.	.	.	.	30.10	
Uric acid	.	.	.	.	.	.	1.00	
Lactic acid, lactate of ammonia, and extractive matters	.	.	.	.	.	.	17.14	
Mucus	.	.	.	.	.	.	0.32	
Sulphate of potash	.	.	.	.	.	.	3.71	
Sulphate of soda	.	.	.	.	.	.	3.16	
Phosphate of soda	.	.	.	.	.	.	2.94	
Biphosphate of ammonia	.	.	.	.	.	.	1.65	
Chloride of sodium	.	.	.	.	.	.	4.45	
Muriate of ammonia	.	.	.	.	.	.	1.50	
Phosphates of lime and magnesia	.	.	.	.	.	.	1.00	
Silica	.	.	.	.	.	.	0.03	
							1000.00	

Fixed salts 15.29.

Solid matter 67.00.



*Analysis II. (Simon).**Specific gravity* 1012.

Water	.	.	.	.	.	.	956.000	
Urea	.	.	.	.	.	.	14.578	
Uric acid	.	.	.	.	.	.	0.710	
Extractive matters and ammoniacal salts	.	.	.	.	.	.	12.940	
Chloride of sodium	.	.	.	.	.	.	7.280	Fixed salts 13.77.
Sulphate of potash	.	.	.	.	.	.	3.508	
Phosphate of soda	.	.	.	.	.	.	2.330	
Phosphates of lime and magnesia	.	.	.	.	.	.	0.664	
Silica	.	.	.	.	.	.	a trace	
							998.000	Solid matter 44.00.

*Analysis III. (Dr. Miller.)**Specific gravity* 1020.

Water	.	.	.	956.8000		
Urea	.	.	.	14.2300		
Uric acid	.	.	.	0.3700	29.812	
Alcohol extractive	.	.	.	12.5270	Organic	
Water extractive	.	.	.	2.5204	matters.	
Vesical mucus	.	.	.	0.1650		
Chloride of sodium	.	.	.	7.2195		43.16
Phosphoric acid	.	.	.	2.1189		Solid matters.
Sulphuric acid	.	.	.	1.7020		
Lime	.	.	.	0.2101	13.349	
Magnesia	.	.	.	0.1198	Fixed salts.	
Potash	.	.	.	1.9260		
Soda	.	.	.	0.0536		
				999.9623		



*Analysis IV. (Marchand.)*

Water	.	.	.	.	.	933.199	
Urea	.	.	.	.	.	32.675	
Uric Acid	.	.	.	.	.	1.065	
Lactic acid	.	.	.	.	.	1.521	
Extractive matters	.	.	.	.	.	11.151	
Mucus	.	.	.	.	.	0.283	
Sulphate of potash	.	.	.	.	.	3.587	66.8 Solid matters.
Phosphate of soda	.	.	.	.	.	3.056	
Sulphate of soda	.	.	.	.	.	3.213	
Biphosphate of ammonia	.	.	.	.	.	1.552	
Chloride of sodium	.	.	.	.	.	4.218	
Muriate of ammonia	.	.	.	.	.	1.652	
Phosphates of lime and magnesia	.	.	.	.	.	1.210	
Lactates	.	.	.	.	.	1.618	
						<u>1000.000</u>	

*Analysis V. (Lehmann.)*

Water	.	.	.	.	.	937.682	
Urea	.	.	.	.	.	31.450	
Uric acid	.	.	.	.	.	1.021	
Lactic acid	.	.	.	.	.	1.496	
Water and alcohol extractives	.	.	.	.	.	10.680	
Lactates	.	.	.	.	.	1.897	62.318 Solid matters.
Chlorides of sodium and ammonium	.	.	.	.	.	3.646	
Alkaline sulphates	.	.	.	.	.	7.314	
Phosphate of soda	.	.	.	.	.	3.765	
Phosphates of lime and magnesia	.	.	.	.	.	1.132	
Mucus	.	.	.	.	.	0.112	
						<u>1000.195</u>	



*Analysis VI. (Becquerel.)**Showing the comparative composition of Male and Female Urine.*

	Mean composition of the urine of four healthy men.	Ditto of four healthy women.	General mean.
<i>Specific gravity</i> . . .	1018·9	1015·12	1017·01
Water . . .	968·815	975·052	971·935
Solid constituents . . .	31·185	24·948	28·066
Urea . . .	13·838	10·366	12·102
Uric acid . . .	0·391	0·406	0·398
Other organic matters	9·261	8·033	8·647
Fixed salts . . .	7·695	6·143	6·919
Consisting of—			
Chlorine . . . . .			0·502
Sulphuric acid . . . . .			0·855
Phosphoric acid . . . . .			0·317
Potash . . . . .			1·300
Soda, lime, and magnesia . . . . .			3·944

## CHAPTER IV.

## MORBID URINE.

86. The urine passed during a diseased state of the system, is almost invariably more or less altered in its composition, and frequently presents physical peculiarities, as of colour, opacity, &c., which are at once apparent on the most cursory examination. The variations which are found to occur in the chemical composition of morbid urine may be divided into two classes, viz. :

- 1st. Those in which no abnormal ingredient is present ; but in which one or more of the normal constituents is present either in greater or less proportion than is found in healthy urine, or is altogether absent.
- 2nd. Those in which one or more ingredients are present, which are not found in the healthy secretion.



I. *Urine containing no abnormal ingredient, but in which an excess or deficiency of one or more of its normal constituents is present.*

#### SECTION I.

##### *Urine containing Urea in abnormal quantity.*

87. Urine containing an excess of urea, is chiefly characterised by its high specific gravity, in which respect it resembles that secreted by diabetic patients (116). If the urea be present in large excess, it deposits irregular rhomboidal crystals of the nitrate ( $C_2H_4N_2O_2, HO, NO_5$ ), when the urine is either in its natural state, or especially when slightly concentrated, is mixed with an equal quantity of nitric acid (181). The proportion of urea present in the healthy urine passes during the twenty-four hours is usually about fourteen or fifteen parts in 1000 (10); while in disease it often amounts to thirty parts, or even more.

#### SECTION II.

##### *Urine containing Uric (or Lithic) Acid in abnormal quantity.*

88. When urine contains an excess of uric acid, it has usually rather a higher colour than the healthy secretion, either deep amber or reddish brown. Its specific gravity is seldom much higher than 1020 or 1025, unless an excess of urea is also present, which is not unfrequently the case. It generally has a decided acid reaction to test-paper; and if the uric acid is present in any considerable excess, it is partially deposited as the urine cools, in the form of a crystalline sediment, usually of a more or less decided red colour, and frequently mixed with urate of ammonia, mucus, and other matters.\* The crystalline forms in which uric acid is found in the urine, are represented in paragraph 186. This deposition of uric acid is greatly accelerated by the addition of a few drops of nitric or hydrochloric acid to the urine (20).

89. The urine of infants and young children not unfrequently deposits lozenge-shaped crystals of nearly pure uric acid, containing only a trace of yellow colouring matter. If

\* Hippuric acid has been known to deposit together with uric acid.



rarely happens that uric acid is deposited in the solid state previous to emission, being held in solution in the warm liquid, and gradually separating in the form of a sediment, as the secretion cools (186).

90. The quantity of uric acid, which, in the healthy secretion, is seldom more than from 0.3 to 1.0 in 1000 parts, varies in morbid urine from a scarcely perceptible trace to upwards of two parts in 1000.

### SECTION III.

#### *Urine containing an excess of Urate (or Lithate) of Ammonia.*

91. Urine containing an excess of urate of ammonia varies very much in colour and appearance, being sometimes pale and of low specific gravity, but more frequently high coloured, dense, and turbid. It is most commonly slightly acid, but is also met with neutral and even alkaline. The urate of ammonia is gradually deposited as the urine cools, in the form of an amorphous precipitate, which, with a high magnifying power, appears to consist of minute rounded particles, occasionally adhering together, and forming irregular linear masses (Fig. 11); frequently mixed with microscopic crystals of uric acid; and, occasionally, when the secretion is neutral or at all alkaline, with the earthy phosphates (106).



Fig. 11. Urate of Ammonia.

92. Urate of ammonia has been met with, in a few rare cases, in the form of globular masses of a larger size, and pierced with spicular crystals, probably of uric acid (Fig. 12). Like the other varieties of urate of ammonia deposit, it is usually found mixed with crystals of uric acid.

93. Urate of ammonia\* constitutes one of the most com-

\* The amorphous deposit formerly described and figured as urate of ammonia has been proved by Dr. Bence Jones to consist of different acid urates, among which, acid urate of potash occasionally predominates. On washing it with cold water, crystalline uric acid separates, although none could be perceived in the unwashed deposit.



mon of the urinary deposits. The colour of the sediment is found to vary considerably, being met with of all shades,

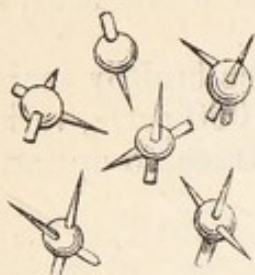


Fig. 12. Urate of Ammonia.\*

from pale fawn colour to reddish purple or pink, the latter colours being due to the admixture of purpurine, which is very frequently found associated with the urates (104, 217). Urate of soda, and traces of the urates of lime and magnesia, are not unfrequently found associated with urate of ammonia deposits.

94. A deposit of urate of ammonia readily dissolves when the urine containing it is gently warmed; and is again precipitated as the liquid cools. If, however, as is often the case, it contains also an admixture of free uric acid or earthy phosphates, the deposit will not wholly dissolve on the application of heat, those substances being nearly as insoluble in hot as in cold water. The presence of purpurine (104, 217) usually renders the urate less easily soluble when warmed.

95. When a deposit of urate of ammonia is treated with a little dilute hydrochloric or acetic acid, it is decomposed; and minute crystals of uric acid shortly appear, which may be readily distinguished under the microscope (194).

#### SECTION IV.

##### *Urine containing Urate (or Lithate) of Soda.*

96. The acid urate of soda ( $\text{NaO}, \text{HO}, \text{C}_{10}\text{H}_2\text{N}_4\text{O}_4$ ) is also a



Fig. 13. Urate of Soda.

frequent sediment in the urine, particularly in the urine of patients taking medicinally the carbonate or other salts of soda. It may generally be recognised without difficulty under the microscope, usually forming minute globular and sometimes granulated aggregations, with occasionally irregular and curved protuberances, as shown in Fig. 13.†

\* See note to (93).

† The acid urate of soda ( $\text{NaO}, \text{HO}, \text{C}_{10}\text{H}_2\text{N}_4\text{O}_4 + 2\text{Aq}$ ) has been obtained artificially in transparent granules, which might be mistaken, under the microscope, for fat globules, but when washed with water they assume the ordinary appearance of urate of soda.



97. It resembles the urate of ammonia in being soluble in hot water (22, 192), and also in most of its chemical characters; giving the same purple-coloured residue when tested with nitric acid and ammonia (23). It also yields crystals of uric acid, when treated with dilute hydrochloric acid (194). When warmed with potash, however, it does not of course give off ammoniacal fumes (377); and by this, and more especially by its behaviour before the blow-pipe (202), and by its microscopic appearance, it may be readily distinguished from the ammoniacal salt. The two salts are frequently found occurring together in the same deposit.

#### SECTION V.

##### *Urine containing an excess of Hippuric Acid.*

98. There is but little that can be said to be characteristic in the appearance of urine in which an excess of hippuric acid is present. It is most commonly either neutral or slightly acid to test paper, but occasionally alkaline; and is in most cases pale and whey-like, and of low specific gravity. The mode of its detection will be found described in paragraphs 206, &c.

#### SECTION VI.

##### *Urine containing an excess of Mucus.*

99. Mucous urine is most commonly very similar in colour to the healthy secretion. It deposits a viscid, tenacious sediment, usually of a dirty yellowish colour, consisting chiefly of mucus mixed with epithelium (328); which, when agitated, does not mix again uniformly with the fluid, but coheres together in tenacious, ropy masses, entangling and retaining numerous bubbles of air.

100. Urine containing an excess of mucus is generally neutral or slightly acid when passed, unless it has been retained some time in the bladder, when it is not unfrequently alkaline; and when this is not the case, it very speedily becomes so, owing to the rapid conversion of the urea into carbonate of ammonia under the influence of the mucus (11). This change takes place first in the portion of the fluid which is in contact with the mucous sediment: this may frequently be seen in specimens of slightly acid urine, the upper portions



of which redden litmus paper; but if the lower part, more exposed immediately in contact with the mucus, be tested, it will be found to restore the original blue colour.

101. Mucous urine differs from that containing pus, in theropy and tenacious character of the deposit; and also in not giving any sensible indication of albumen when tested with heat and nitric acid (254), unless the albumen be derived from some other independent source, which is sometimes the case (255). Minute traces of albumen, indeed, are present in the undiluted mucous fluid, but the quantity is so small, that, when mixed with urine, it is incapable of being detected (663).

102. The mucous deposit is frequently found mixed with a considerable quantity of earthy phosphates or urates; in which case its opacity renders it more liable to be mistaken for pus. The true nature of such a mixed deposit is, however, readily distinguished by microscopic examination, which should always be had recourse to in such cases (156, 211, 328).

## SECTION VII.

### *Urine containing an excess of Extractive and Colouring Matters.*

103. Urine containing extractive matters in excess is usually more highly coloured than the natural secretion, a large proportion of what is included under the title of extractive matters, consisting apparently, in most cases, of the peculiar colouring matters of the urine. When boiled, and subsequently mixed with a little hydrochloric acid, such urine becomes of a more or less decided red colour (215); and on cooling, usually deposits a quantity of brownish or bluish-black sediment, which is readily soluble in alcohol.

104. It is not unfrequently the case, that the peculiar red colouring matter called purpurine is present in considerable quantity in certain forms of morbid urine. This, when a deposit of urate of soda or ammonia is also present, is precipitated with the urate, giving the sediment a pink or red colour (217). When no deposit of urate exists, the purpurine remains in solution, giving the urine a more or less bloody appearance, which may sometimes lead to the suspicion that blood is present. For the methods of identifying purpurine see paragraphs 216 to 221.



## SECTION VIII.

*Urine containing an abnormal proportion of Fixed Alkaline Salts.*

105. When these salts are present in excess, they tend to raise the specific gravity of the secretion. The quantity of insoluble saline matter may be readily estimated in the mass, by incinerating the dry residue left after evaporating a known weight of the urine, and treating the ash with water, which will dissolve out the alkaline salts, leaving the earthy phosphates and silica undissolved.\* The aqueous solution is then evaporated to dryness, ignited, and weighed. The individual proportion of the several salts, which is sometimes a point of considerable interest, may be determined in the manner described in paragraphs 66 to 84.

## SECTION IX.

*Urine containing the Earthy Phosphates in abnormal quantity.*

106. The physical characters of urine containing an excess of earthy phosphates vary considerably. The colour is most uncommonly pale, and the specific gravity rather low, but it is also occasionally dark, and of high specific gravity, especially when urea is present in large quantity (87, 301). It is generally slightly acid when passed, but shortly becomes neutral or alkaline (43), when the phosphates are precipitated, often in large quantity, in the form of a crystalline sediment, the colour of which varies from white and grey to a yellow or reddish brown. When white or grey, the sediment will probably be found to consist chiefly of phosphates mixed with mucus; when yellowish or red, it will probably be found to contain, in addition, a certain amount of uric acid, or urate of soda, or ammonia, most commonly one of the latter.

107. It must be borne in mind that the spontaneous occurrence of a precipitate of earthy phosphates, is not of itself a proof that they are present in excess; nor, on the other hand, is the non-occurrence of a deposit a proof that a small quantity only is present. When the urine is acid, as in health, they

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\* See note to (63).



may be retained in solution in considerable quantity, without forming any solid sediment; while if the secretion is neutral or alkaline, a comparatively small amount of earthy phosphates may be precipitated in the form of a deposit.

108. When examined with the microscope, deposits of the earthy phosphates will frequently be found to contain both the crystalline triple phosphate ( $\text{MgO}, \text{NH}_4\text{O}, \text{HO}, \text{PO}_5$ ), and also phosphate of lime, in the form of an amorphous powder, or in minute, irregular, rounded particles (43, 44). Minute dumbbells, like those of oxalate of lime (Fig. 25), have also been met with.

Crystallized phosphate of lime (Fig. 14.) ( $2\text{CaO}, \text{HO}, \text{PO}_5$ )

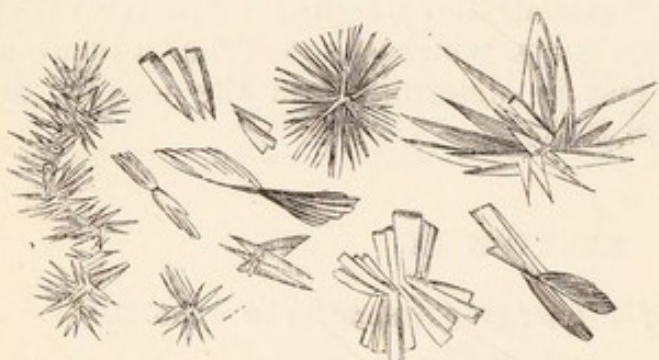


Fig. 14. Crystallized Phosphate of Lime.

is by no means uncommon in urine. It may always be obtained by *partial* neutralising the fresh urine with ammonia, and setting it aside.

109. The quantity of earthy phosphates which in healthy

urine, is usually about one part in 1000, varies, in diseases, from a scarcely perceptible trace to 5.5 in 1000 parts, and is occasionally even higher. When present in excess, they may generally be partially precipitated by warming the urine (49).

110. It sometimes happens, in certain forms of disease, that the earthy phosphates are secreted in much smaller quantities than is found in healthy urine, and in some rare cases they appear to be altogether absent. Whether this is the case in any specimen of the secretion, may be ascertained by adding to it a slight excess of ammonia, when, if present only in very small proportion, or not at all, no precipitation will take place, or the ash of the urine may be digested in dilute hydrochloric or nitric acid, and the clear acid solution supersaturated with ammonia, when, if no precipitate is produced, it may be concluded that no perceptible trace of earthy phosphate is present.



II. *Urine containing one or more abnormal ingredients.*

111. The abnormal matters usually found in morbid urine are:—1, sugar;\* 2, albumen; 3, blood; 4, biliary matter; 5, pus; 6, fat and chylous matter; 7, semen; 8, oxalate of lime; 9, cystine and other foreign matters. Besides the substances just enumerated, various others may be occasionally detected in urine, such as arsenic, antimony, and many other mineral and organic matters, which having been taken into the system medicinally or otherwise, and being incapable of assimilation, have passed through either unchanged, or more or less modified in composition.

## SECTION X.

*Urine containing Sugar ( $C_{12}H_{14}O_{14}$ ).*

112. The variety of sugar always present in the urine of diabetic patients, and hence called diabetic sugar, has the same chemical composition as that contained in most kinds of fruit, commonly known as grape sugar, or glucose. It appears to contain two equivalents of water of crystallization, which may be expelled at a temperature of  $212^{\circ}$ ; so that its composition may be more correctly expressed by the formula ( $C_{12}H_{12}O_{12} + 2Aq$ ).

113. Diabetic sugar may be obtained by concentrating the urine containing it, by evaporation on a water-bath, until it begins to deposit a crystalline sediment; the mass is then allowed to cool, on which the greater part of the sugar crystallizes out. It is then filtered; and when most of the liquid has passed through, the crystals are to be pressed between folds of filtering paper, and washed with a small quantity of cold strong alcohol, which serves to remove the greater part of the impurities, without dissolving much of the sugar. The crystals are then dissolved in hot water, and purified by successive crystallizations, or, if necessary, by boiling with animal charcoal.

114. Diabetic sugar differs from cane sugar ( $C_{12}H_{22}O_{11}$ ), in being considerably less sweet to the taste, harder, and less soluble in water; one part requiring about one and a half of

\* Recent researches (35) have removed sugar from the list of strictly abnormal ingredients of urine, but this does not affect the practical value of the classification here given, or of tests described in the following section.



cold water to dissolve it. In dilute alcohol, on the other hand, it is somewhat more soluble than the cane variety; but is insoluble in absolute alcohol and ether. It is usually in the form of granular crystals; but when crystallized out of a considerable mass of syrup, it is often obtained in needle-like tufts. When crystallized from its solution in dilute alcohol, it usually separates in the form of hard transparent cubes, and occasionally in square plates.\*

115. Strong sulphuric acid dissolves grape sugar, forming a pale yellowish solution; cane sugar, on the contrary, is almost instantly charred and blackened by the strong acid.

116. Urine containing sugar is usually characterised by its high specific gravity, which is frequently from 1030 to 1045, and occasionally as high as 1050 and 1055. If, however, the sugar is present only in small quantity, the specific gravity may not be higher than usual; so that a moderately low specific gravity is of itself no proof of the absence of sugar.

117. Diabetic urine has usually, after standing a short time in a warm atmosphere, a white scum, somewhat resembling flour, on the surface, consisting of minute, oval-shaped convolvoid vesicles (132), which is highly characteristic of the presence of sugar, and occasionally leads to its detection before it has been secreted in sufficient abundance to raise the specific gravity of the urine to a suspicious extent.

118. This variety of urine is usually paler than the natural secretion, and frequently possesses a faint greenish tint. It is most commonly slightly turbid. When fresh, it has a faint and rather agreeable odour, somewhat resembling that of hay.

119. The proportion of urea in diabetic urine is usually much smaller than that found in the healthy secretion; but whether the absolute amount secreted differs materially from the normal average, or whether the apparent deficiency is merely owing to the large quantity of water passed by diabetic patients thus largely diluting the urea, has not yet been satisfactorily decided, owing to the difficulty of correctly estimating the quantity of urea when mixed with any considerable amount of sugar (334).

120. The proportion of sugar in diabetic urine varies from a mere trace to from 50 to 80 parts in 1000; and has been known to amount to as much as 134 parts in 1000.

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\* Vohl has met with a case in which a part of the diabetic sugar in the urine was replaced by inosite. It is also stated that acetone has been found in diabetic urine.



§ 121. Several tests have been proposed for the detection of sugar in urine. Of these, the following only need here be noticed, viz., *Trommer's test*, *Maumené's test*, *Moore's test*, *the fermentation test*, and the test afforded by the growth of a microscopic conservoid vegetation, called the *torula*.

§ 122. *Trommer's Test*. \*—This excellent test is founded on the circumstance, that when a solution containing diabetic or grape sugar (112), is boiled with a mixture of potash ( $KO$ ), and sulphate of copper ( $CuO, SO_3$ ), the oxide of copper ( $CuO$ ) contained in the latter becomes reduced to the state of suboxide ( $Cu_2O$ ), which is precipitated in the form of a reddish ochre-coloured granular powder.

§ 123. A little of the urine suspected to contain sugar is placed in a tolerably large test-tube, and mixed with a drop or two of a solution of sulphate of copper, which should be added only in sufficient quantity to give the mixture a very pale blue tint. This will probably cause a slight precipitation of pale blue phosphate of copper, owing to the presence of soluble phosphates in the urine (40); this, however, need not be regarded, as it will not afterwards interfere with the indications of the test. A solution of potash is now added in large excess,† or in quantity equal to about half the volume of urine employed; this will first throw down a pale blue precipitate of hydrated oxide of copper ( $CuO, HO$ ), which, if sugar be present, will immediately redissolve, forming a purplish-blue solution, something similar to that caused in a very dilute solution of copper by ammonia.

§ 124. The mixture is now to be carefully heated over a lamp, and gently boiled; when, if sugar is present, a reddish or yellowish brown precipitate of suboxide of copper ( $Cu_2O$ ) will be deposited in the liquid, generally before the boiling point is reached. If no sugar is present, a black precipitate of the common oxide of copper ( $CuO$ ) will be thrown down, totally distinct in appearance from the suboxide. It is important, in this experiment, not to add too much of the sulphate of copper, because, in that case, the suboxide might be mixed with some of the black oxide (the sugar being capable

\* If diabetic urine be not procurable, a few grains of grape sugar may be dissolved in normal urine for the demonstration of these tests. Grape sugar may be obtained by boiling white sugar with water and a few drops of sulphuric acid for a few minutes, neutralising with chalk, filtering, and evaporating.

† Or the potash may be added, and the solution filtered from any deposit of earthy phosphates that may be thrown down, before the addition of the sulphate of copper.



of reducing only a certain definite quantity), which would more or less mask the characteristic colour and appearance of the suboxide. This test is extremely delicate, and is capable of detecting very small traces of sugar in the urine. If very much sugar be present, the action of the potash upon it will produce a very dark brown colour, which may disguise the suboxide of copper. If this be the case, another portion of the urine may be diluted before being tested. Since other substances are occasionally found in urine\* which reduce the oxide of copper, this test cannot be considered as conclusive, except as to the absence of sugar. Since the presence of ammonia in any considerable quantity interferes with this test (as Dr. Beale originally pointed out), recourse must be had to the yeast test, if much ammonia be smelt in this experiment.

125. *Maumené's test*.—This test is founded on the circumstance, that when sugar is moderately heated in contact with the bichloride of tin ( $\text{SnCl}_2$ ), it is decomposed, and a brownish-black compound, somewhat resembling caramel, is formed. The most convenient method of applying this test is to saturate strips of merino, flannel, or some other *woollen* tissue,† with a solution of bichloride of tin (prepared by dissolving the salt in about twice its weight of water and filtering), after which they may be dried at a gentle heat on a water-bath, and kept ready for use. On moistening one of these strips with urine, or any other liquid containing sugar even in a highly diluted state, and holding it near a fire, or over a lamp, so as to heat it about  $270^\circ$  or  $300^\circ$  Fahr., it immediately assumes a brownish-black colour. The delicacy of this test is stated to be so great, that though ordinary healthy urine causes no change of colour, if ten drops of diabetic urine be diffused through half a pint of water, the mixture will immediately give decided indications of sugar.

126. *Moore's test*.—Mix a little of the suspected urine in a test-tube, with about half its volume of liquor potassæ, and

\* Even uric acid produces the same effect, but in general its quantity is too small in normal urine to allow of its being mistaken for sugar.

† It is necessary in this test to avoid the use of cotton or linen, since those substances, being analogous to sugar in composition, undergo also a similar decomposition when warmed with the bichloride of tin; and would consequently become blackened, even though no sugar were present. It will be seen that the utility of this test depends upon the circumstance that the prepared strips can be carried about so as to render the test available in a clinical examination. In the laboratory the other tests are more convenient.



boil the mixture gently for a minute or two. If sugar is present, the liquid will assume a brownish or bistre tint; while little or no heightening of colour takes place when the urine is free from saccharine matter.

127. *Böttger's test*.—Add to the urine suspected to contain sugar a few drops of a somewhat dilute solution of nitrate of bismuth ( $\text{BiO}_3, 3\text{NO}_5$ ) in nitric acid, then add carbonate of soda till the solution is alkaline, and boil for three or four minutes, when, if sugar be present, the mixture assumes a dark colour from the reduction of bismuth, and when set aside, deposits a grey or black precipitate. With healthy urine a white precipitate of phosphate and carbonate of bismuth is obtained.

128. *Fermentation test*.—Fill a test-tube with the suspected urine, having previously mixed with it a few drops of fresh yeast, or still better, a little of the dried German yeast; close the open end with a small saucer or evaporating dish, and while gently pressing the latter upon the tube, invert the tube, when they will be in the position shown in the figure (Fig. 15). A little more of the urine is then poured into the saucer, in order to prevent the escape of any of the liquid from the tube; and if any bubbles of air have accidentally been allowed to enter, the exact height of the upper surface of the liquid in the tube must be marked with ink, or with a strip of gummed paper. The tube, with its contents, is then set aside in a warm place, having a temperature of about  $70^\circ$  or  $80^\circ$ , for twenty-four hours. As bubbles of gas are sometimes given off by the yeast itself, it is a good precaution to put the same quantity of yeast into a second tube of equal size, and fill it up with pure water. The amount of gas, if any, derived from the yeast, will thus be rendered apparent, and may afterwards be deducted from the volume of gas in the tube containing the urine.

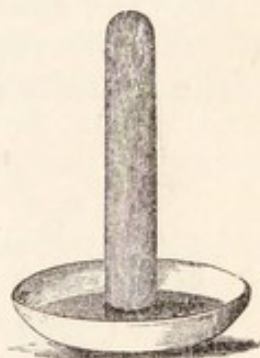
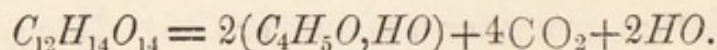


Fig. 15. Fermentation test.

129. If sugar is present, it begins almost immediately to undergo the vinous fermentation, by which it becomes converted into alcohol ( $\text{C}_4\text{H}_5\text{O}, \text{HO}$ ) and carbonic acid ( $\text{CO}_2$ ), each equivalent of sugar giving rise to the formation of two equivalents of alcohol, four of carbonic acid, and two of water; thus:





The carbonic acid thus formed rises in minute bubbles, causing gradual and gentle effervescence, and collects in the upper part of the tube; at the same time displacing the liquid, which escapes through the open end of the tube into the saucer.\*

130. That the gas thus formed is really carbonic acid, may be proved by decanting a little of it under water into a clean tube, and testing it with lime-water, which will instantly become milky, owing to the formation of the insoluble carbonate of lime ( $CaO,CO_2$ ). When the quantity of sugar present is at all considerable, the urine, after fermentation, will be found to possess a faint vinous smell, due to the alcohol formed during the process.

131. If, on the contrary, the urine is free from sugar, of course no fermentation will take place, and no gas will be formed in the tube.

132. *Test afforded by the growth of the torula.*—During the process of the vinous fermentation of a liquid containing sugar, a delicate white scum gradually collects on the surface, which,

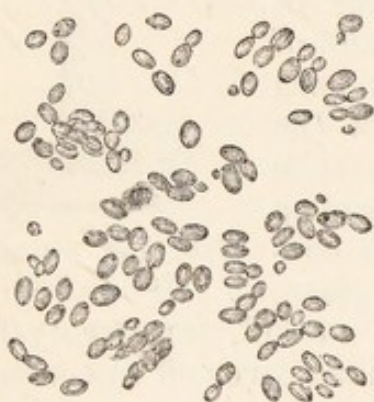


Fig. 16. Torula Vesicles, magnified 400 diameters.

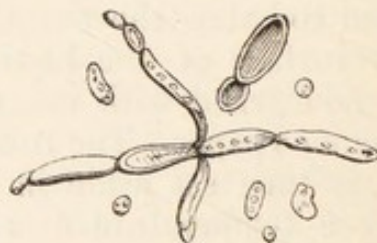


Fig. 17. Torula Stem.

when seen merely with the naked eye, is so highly characteristic an indication of the presence of sugar, as frequently to lead to its detection, when present only in very small quantity. If a little of this scum be examined under the microscope, with a magnifying power of four or five hundred diameters, it will be found to consist of minute oval vesicles (Fig. 16), which in the course of a few hours rapidly change

\* This has been lately proved to be only one of the changes which are involved in the fermentation of sugar, other substances, such as succinic acid and glycerine, being formed at the same time.



in their form, becoming longer and more tubular, and giving rise to new vesicles, which shoot out from the parent body, forming an irregularly jointed confervoid stem (Fig. 17). These again gradually break up into a great number of oval vesicles, which eventually separate, and fall to the bottom, where they may be detected by microscopic examination.

*Precipitation by tribasic acetate of lead and ammonia.*—When the proportion of sugar present in urine is very minute, the best process for extracting it consists in precipitating the urine, first with acetate of lead, then with tribasic acetate of lead added in excess, and lastly, after filtering, with ammonia. This last precipitate is collected upon a filter, washed, suspended in water, and decomposed by sulphuretted hydrogen. After filtering from the sulphide of lead, and evaporating, the sugar may be detected by any of the tests above described.\*

*Precipitation by potash and alcohol.*—If urine containing sugar be mixed with four volumes of absolute alcohol, allowed to stand for some time, filtered, mixed with a little alcoholic solution of potash, and set aside for a day or two, a combination of grape-sugar with potash separates as a deposit which adheres to the side of the vessel. The alcoholic liquid having been drained off, the deposit may be dissolved in water, and tested by any of the above tests.†

132*a*. A very remarkable substance has occasionally been observed in urine, which answers to the test with the alkaline copper solution in precisely the same manner as sugar, but does not possess the property of reducing the oxide of bismuth, or of fermenting in contact with yeast. Its most striking feature is that of absorbing oxygen from the air, and producing a brown colour, when an alkali is added to the solution containing it, so that urine in which this substance is present becomes brown at once when shaken with potash, whilst saccharine urine does not become brown until it is boiled. Boedeker,‡ who first directed attention to this substance, which he named *alkapton*, has isolated it in the following manner:—The urine was precipitated by acetate of lead and filtered. The filtrate was mixed with tribasic acetate of lead,

\* This process has been employed for the demonstration of the presence of sugar in normal urine, but the circumstance that the indigo-producing body described by Schunck (36) is precipitated in the same way, and yields sugar as a product of its decomposition, much diminishes the value of the evidence which it affords.

† According to Brücke, sugar may be detected in healthy urine by this process.

‡ 'Ann. Ch. Pharm.,' Jan., 1861.



avoiding an excess, the precipitate washed, suspended in water, and decomposed by sulphuretted hydrogen. The solution filtered from the sulphide of lead was evaporated to dryness on the water-bath, and the residue extracted with ether. On evaporating the ethereal solution, the alkaptone was obtained as a golden-yellow, resinous, deliquescent substance, very easily soluble in water and alcohol. Its solution reddens litmus slightly, and is unchanged by exposure to air, unless an alkali is added. The precipitate produced by tribasic acetate of lead, also becomes brown when exposed to the air. Alkaptone contains nitrogen, but its exact composition has not yet been ascertained.\*

#### SECTION XI.

##### *Urine containing Albumen.†*

133. This substance, which is contained, as is well known, in large quantity in many of the tissues of the body, and especially in the serum of the blood (466), is not unfrequently present in morbid urine. Albuminous urine varies very considerably in appearance and general characters, being found alkaline, acid, and neutral; high coloured, and pale; of high specific gravity and the contrary; so that no general rule can be laid down as to its usual physical peculiarities, likely to lead to its detection; though, when its presence is once suspected, its detection is easy and simple (139).

134. The quantity of albumen found in urine varies very much; a mere trace only being sometimes present, and at others as much as ten or twelve parts in 1000.

135. The most remarkable property of albumen is, that when a solution containing it is heated to a temperature of about  $170^{\circ}$ , or higher, it coagulates, and separates completely from the liquid; and when this change has once taken place, it becomes quite insoluble in water. The coagulated albumen is readily soluble in potash and other alkaline solutions; and when an excess of alkali is present, no coagulation takes place on boiling.

136. Albumen is precipitated from its solution by nitric

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\* Dr. Johnson has observed the occurrence of alkaptone in the urine of an infant, his attention being called to it by the brown stains produced on the linen.

† For the purpose of demonstrating the tests, a very little well-beaten white of egg may be mixed with some normal urine.



land hydrochloric acids, but not by phosphoric, acetic, or, tartaric acids, which, indeed, appear to exercise a decided solvent action upon it, and, when present, prevent its coagulating on the application of heat.

137. It is also readily precipitated, even from an acetic acid solution, by ferrocyanide ( $K_2FeCy_3 + 3Aq$ ) and ferridecyanide ( $K_3Fe_2Cy_6$ ) of potassium; and the precipitates thus formed are easily soluble in alkaline solutions.

138. Chloride of mercury ( $HgCl$ ), alum ( $Al_2O_3, 3SO_3 + KO, SO_3 + 24Aq$ ), and many other of the metallic salts, also cause precipitates in albuminous solutions, which are compounds of the salt with albumen. It is precipitated, too, by alcohol, creosote, tannin, and many other substances.

139. The detection of albumen in urine containing it is very easy. The suspected urine may be gently boiled in a test-tube, when, if albumen is present, it will coagulate, and form a more or less copious white precipitate. If the albumen is present only in minute quantity, it may cause merely a delicate opalescence; or, when in larger quantity, it may separate in curdy flakes; and if very abundant, may cause the liquid to gelatinize, and become nearly solid (142).

140. The appearance of a white precipitate on boiling is not, however, of itself, a sure proof of the presence of albumen in urine, since a white precipitate is also produced by boiling, when the secretion, free from albumen, contains an excess of earthy phosphates (49a). It is therefore necessary to add a few drops of nitric acid, which, in case the precipitate consists of phosphates, immediately redissolves it, but if albuminous, leaves it still insoluble.

141. To prevent the possibility of error, it is always advisable to test a separate portion of the urine also with dilute nitric acid, by which the albumen, if present, will instantly be thrown down. If the quantity of albumen is very small, it is possible that the milkiness first caused by the acid may disappear, but if a few drops more of the acid be added, the precipitate will again separate, and remain insoluble. If both heat and nitric acid cause a white precipitate, there can be no doubt of the presence of albumen.

141a. The coagulum may be further tested by *Millon's test*, which consists in gently heating it with solution of nitrate of mercury ( $HgO, NO_3$ ), prepared by dissolving 200 grains of mercury in 5 drachms of ordinary concentrated nitric acid, with the aid of heat. Albumen acquires an intense red colour when



heated with this solution, whilst normal urine is only rendered faintly and transiently pink. Fibrin, casein, and other members of the same group of bodies (protein compounds) exhibit the same behaviour.

142. In testing for albumen, it must be borne in mind that, if the liquid is alkaline to test-paper, the albumen, though present, will probably not be coagulated on the application of heat, since coagulated albumen is readily soluble in alkaline solutions (135). On this account the urine should first be examined with turmeric or reddened litmus paper (277), and, if found to be alkaline, neutralized with nitric acid before boiling.

143. It should also be remembered that, when the albumen is present only in small quantity, the addition of a very slight excess of nitric acid may redissolve it, and thus lead to the supposition that the precipitate is phosphatic. A few drops more of the acid, however, will instantly cause it to reappear, if albuminous; while, if really phosphatic, no excess of the acid would cause it to do so.\*



Fig. 18. Fibrinous Cast.  
(Dr. G. Johnson.)

epithelium, and fatty globules, have usually the appearance shown in figure 18.†

144. The peculiar casts of urinary tubes, found in the urine of patients suffering from Bright's disease, consisting of fibrinous or albuminous matter, and entangling blood corpuscles,

## SECTION XII.

### *Urine containing Blood.*

145. Urine frequently contains, in addition to albumen, one or more of the other constituents of the blood (450), and is often more or less highly coloured red or brown, by the pre-

\* The result of these tests for albumen may be confirmed by acidulating another portion of urine with acetic acid, and adding ferrocyanide of potassium (137). Chloride of mercury may be added to another portion (138).

In a case of mollities ossium, an albuminoid substance has been observed in the urine, which was precipitated by nitric acid like albumen, but the coagulum dissolved on heating, and reappeared as the solution cooled. Ferrocyanide of potassium also precipitated this substance. Tannic acid gave a precipitate in this urine.

† The presence of inosite in the urine has been noticed in a case of Bright's disease.



presence of the corpuscles and red colouring matter. When the fibrin, in its soluble form, is present, it usually coagulates spontaneously on cooling, and causes the urine to become more or less gelatinous soon after it is passed. This spontaneous coagulation on cooling may be considered of itself an insufficient proof of the presence of the fibrin of the blood.

146. The blood corpuscles may generally be detected both in the coagulum and also in the superincumbent fluid, when examined under the microscope (451); occasionally, however, they are almost entirely disintegrated, so that little or no trace of their characteristic form remains. They are sometimes found adhering together, forming little thread-like aggregations; but more frequently floating detached from each other, looking like little transparent rings (Fig. 19).

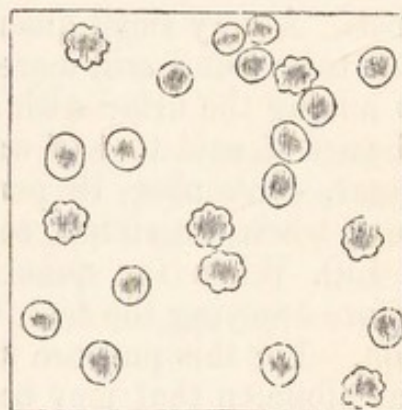


Fig. 19. Blood in Urine.

147. In urine containing blood, the albumen may in all cases be readily detected by the tests already mentioned (139)—viz., heat and nitric acid; but when any of the colouring matter of the blood is also present, it will coagulate with the albumen, giving the coagulum a more or less decided red or brown colour.

### SECTION XIII.

#### *Urine containing Biliary matter.\**

148. When biliary matter is present in urine,† it generally gives a more or less decided yellowish brown colour, both to the liquid and also to any sediment that may be deposited from it. The taste also of such urine is remarkably bitter—a peculiarity which furnishes a ready indication of its presence when other tests are not at hand; though it must not be implicitly relied on, since small traces may exist in the secretion, without communicating to it any very decided taste.

149. *Pettenkofer's test.* Perhaps the best test for the pre-

\* A little ox-gall may be added to the urine for the purpose of applying the tests.

† According to Scherer, biliary colouring matter is occasionally present in healthy urine.



sence of bile, is that known as Pettenkofer's. If the urine contains albumen, it should first be freed from that substance by coagulation and filtration (135, 151); because albumen, when present in considerable quantity, would give, with sulphuric acid and sugar, a colour resembling that caused by bile. Dissolve a grain or two of white sugar in the urine to be tested, and add, drop by drop, about two thirds of its volume of strong sulphuric acid. If biliary matter be present, a very distinct and characteristic violet-red colour will be produced, which becomes redder and more intense on the application of heat. A very small quantity of the biliary matter responds to this test; but a still more delicate mode of applying it consists in mixing the urine with a single drop of dilute sulphuric acid (1 part of acid to 4 of water), adding a trace of a solution of sugar, containing 10 per cent., and evaporating at a gentle heat, when the violent colour becomes evident.

150. When the quantity of bile is small, it is advisable, before applying the test, to concentrate the urine by evaporation. For this purpose it is first boiled, in order to coagulate any albumen that may be present (151), and afterwards evaporated nearly to dryness on a water-bath. The residue is then treated with a small quantity of boiling water or alcohol; and the solution thus formed, containing any biliary matter that may be present, is allowed to cool and tested as above.

151. The experiment known as *Heller's test* is made as follows:—Mix with a little of the suspected urine a few drops of the serum of blood or white of egg, or of any liquid containing albumen in solution; and having shaken them well together, add a slight excess of nitric acid, which will cause the precipitation of the albumen (136). If bile is present, the coagulum thrown down by the acid will have a more or less distinct dull green or bluish colour, quite different from the white or pale fawn colour which it would otherwise have. When only a small quantity of biliary matter is present, the urine may be concentrated, as in Pettenkofer's test (150), the serum or white of egg being subsequently added to the cold concentrated aqueous solution of the evaporated residue.

152. *Gmelin's test*.—Pour a few drops of the suspected urine upon a clean white plate or dish, so as to form a thin layer of the liquid, and then carefully drop into the centre, with a pipette, five or six drops of nitric acid. When bile is present in any considerable quantity, the liquid becomes successively pale green, violet, pink, and yellow, the colour rapidly changing



When the acid mixes with the urine. When the bile is present only in small quantity, these colours are not distinctly visible; but unless the proportion is very minute, a greenish tint is generally perceptible. On concentrating the urine by evaporation, the appearance may be seen to greater advantage when only small traces of bile are present (150). The action of this test appears to depend on the presence of the peculiar brown colouring matter of bile, called biliphæin, or cholepyrrhin.\*

## SECTION XIV.

*Urine containing Pus.*

153. Pus is a substance which in many respects closely resembles mucus, both in its behaviour with reagents, and still more in its appearance under the microscope; so that it is not always easy to distinguish between them; and when mixed together in the urine, it is frequently quite impossible to say with certainty whether or not both are present. Like mucus, it consists of minute round or oval granular corpuscles (Fig. 20), floating in the fluid, from which they separate on standing, and gradually sink to the bottom. These form, in urine containing pus, a pale greenish yellow or cream-coloured layer at the bottom of the fluid; and, if shaken, the sediment readily breaks up, and diffuses itself through the liquid, again gradually subsiding to the bottom when allowed to stand. If the urine, however, be decidedly alkaline, the character of the purulent deposit is changed, and it assumes nearly the same appearance as mucus (251, 680).



Fig. 20. Pus in Urine, magnified 400 diam.

154. Urine containing pus is met with sometimes neutral, acid, and alkaline. It always contains albumen in solution, which may be recognised in the filtered urine by the usual tests, heat and nitric acid (139). This albumen is derived from the *liquor puris*, in which it is always present (254, 677). The absence of albumen, therefore, in the urine, may be con-

\* Brücke recommends a useful modification of this test, which consists in mixing the urine in a tube with dilute nitric acid, and carefully pouring in sulphuric acid, so as to form a layer at the bottom of the urine. The rings of colour commence from the junction of the two layers.



sidered as a strong indication of the absence of pus; though the presence of albumen is of itself no kind of proof of the existence of pus, since it may be derived from other independent sources. Traces of blood are by no means unfrequent in purulent urine, giving the sediment a brown or reddish colour (145).

155. The chemical and microscopic characters of pus, and the modes of distinguishing it from mucus, will be more fully described further on (247 to 258, 674).

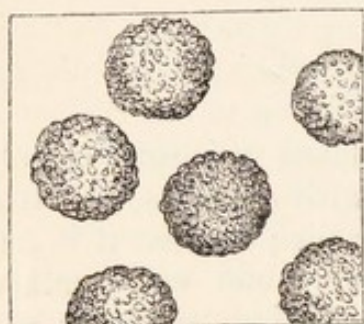


Fig. 21. Large Organic Globules, magnified 400 diameters.

viscid fluids, which are characteristic respectively of pus and mucus (676, 661). Their general appearance is shown in Fig. 21.

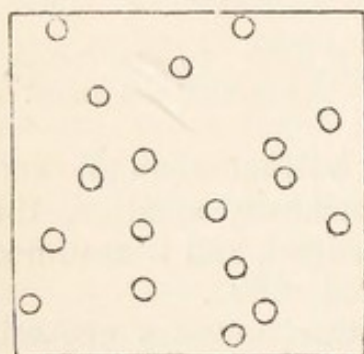


Fig. 22.  
Small Organic Globules.

156. The peculiar granular corpuscles, which have been called *large organic globules*, and which are not unfrequently met with in certain conditions of the urine, especially in that of pregnant women, closely resemble the corpuscles of mucus and pus, being granular on the exterior, and, on the addition of acetic acid, develop internal nuclei. They are, however, larger, and are un-

accompanied by the albuminous and

157. The small circular bodies, which have been occasionally, though much more rarely, found in certain morbid conditions of the secretion, and called *small organic globules*, are represented in Fig. 22. They are spherical and smooth on the surface, no appearance of granular structure being apparent, and considerably smaller than the large organic globules (156). They are unaffected by acetic acid.

## SECTION XV.

### *Urine containing Fat and Chylous matter.*

158. Urine containing fatty or chylous matter is usually more or less turbid, and frequently has an almost milky ap-



appearance. Little is known as to the precise nature of the fatty matter which is thus occasionally met with in urine, although it is probable that its composition varies with the circumstances under which it is formed. It sometimes exists dissociated with albumen and chylous matter, sometimes alone. Numerous minute oily globules may in many cases be seen under the microscope (325), but it is often so intimately mixed with the albuminous matter also present, forming a kind of emulsion, that no trace of oily globules can be detected even with a high magnifying power. In such cases, the urine may be agitated with a little ether, which will dissolve the fat; and the ethereal solution thus formed will separate from the watery liquid, forming a distinct stratum floating on the surface. If the ethereal solution be evaporated at a gentle heat, the fat will be left, and may be readily recognised by the physical peculiarities of fatty substances; such as immiscibility with water; breaking up into minute globules when agitated with hot water, &c. Fibrin is said to be discoverable in urine of this description.

Chylous urine frequently contains minute round corpuscles, resembling the white globules of the blood or lymph, which at first sight have a good deal the appearance of oil globules, or which they have probably been in some cases mistaken. Their insolubility in ether, however, shows that they are not always composed of fatty matter.

159. The peculiar form of mucilaginous or caseous matter, usually present in the urine of pregnancy, and which has received the name of Kiestein, gives the urine containing it a cloudy appearance; and after the lapse of a few days, gradually forms on the surface a more or less shining pellicle, which in three or four days, as the urine becomes ammoniacal, breaks up into minute particles, which subside to the bottom. When examined under the microscope, the pellicle is found to consist of minute granular particles, usually mixed with great numbers of prismatic crystals of triple phosphate (44), to which latter the peculiar shining appearance, somewhat resembling spermaceti, seems to be due. A few globules of oily matter, resembling butter, are also occasionally present.\*

Cholesterin has been found in urine in Bright's disease by Dr. Beale.

\* Since Kiestein is not a definite substance, but merely a deposit of a fungoid growth together with triple phosphate, consequent upon the rapid alkalescence of the urine, it is not regarded as a conclusive evidence of pregnancy.



## SECTION XVI.

*Urine containing Semen.*

160. When semen is present in urine, it may easily be detected under the microscope, by the appearance of minute animalcules, always found in the spermatic fluid,

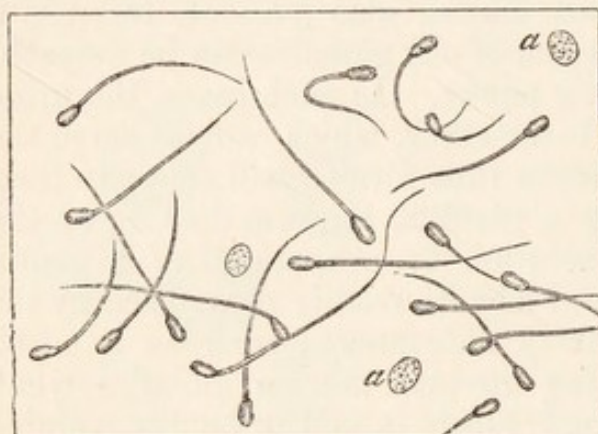


Fig. 23. Spermatozoa, and Spermatic Granules, magnified 400 diameters.

and hence called *spermatozoa*. They are more or less oval in form, and are furnished with long and delicate tails, as shown in Fig. 23. These spermatozoa, while in their native fluid, enjoy an active existence, and move about at will. In urine, however, unless a considerable quantity of

pus is also present, they are never found alive, the secretion proving apparently fatal to them.

161. In addition to the spermatozoa, there may generally be recognised in seminal urine a few minute granular corpuscles, of a round or oval form (*a*, Fig. 23), and rather larger than the bodies of the animalcules. Traces of albumen also may generally be detected in urine containing semen (264).

## SECTION XVII.

*Urine containing Oxalate of Lime ( $\text{CaO}, \text{C}_2\text{O}_3 + 2\text{Aq.}$ )*

162. Urine containing much oxalate of lime is usually, though by no means always, of a dark amber, and often of a pale greenish, or citron colour. It is in most cases decidedly acid to test-paper, and is frequently found to contain an unusually large quantity of epithelial debris. It often contains an excess of uric acid and urates, and almost invariably also an abnormally large quantity of urea. Its specific gravity is not often materially different from that of the healthy secretion, viz., about 1020.

163. Oxalate of lime appears to exist very frequently in



urine, generally in the form of minute and well-defined octohedral crystals (Fig. 24); but unless carefully looked for, it may readily escape detection, owing to the crystals, which were very transparent, having almost exactly the same refractive power as the urine itself, so that it is not always easy to distinguish them as they float in the liquid. The crystals have also nearly the same specific gravity as urine, in consequence of which they generally remain suspended in the fluid some considerable time, before they form a sedimentary deposit at the bottom of the containing vessel.

164. The best way of detecting them is to allow the urine suspected to contain them to stand a few hours, that the oxalate may, in some measure, subside; though frequently it remains several days without doing so completely, in which case the urine may be passed through a filter, when most of the crystals will be retained by the paper, and may be warmed with a little distilled water, in the manner described below (165). The greater part of the liquid is then carefully poured off, and the lower stratum is placed in a watch-glass or small porcelain dish and gently heated over a lamp. In this way the liquid will become specifically lighter, and in consequence, the crystals, if present, will gradually subside to the bottom, especially if a slight rotatory motion be given to the liquid. It is now allowed to stand a few minutes, and the clear liquid is carefully poured off, or removed by means of a pipette.

165. A little distilled water may now be added, when the sediment will become much more distinctly visible, owing to the refractive power of the water differing more decidedly from that of the crystals. The mixture is again heated, when anyurate of ammonia, which is often also present, will be dissolved; and by pouring off the liquid, after standing a few minutes, the crystals will be left at the bottom, and may be removed for the purpose of microscopic examination, or for testing with reagents.

166. Oxalate of lime, as found in the urine, is usually in the form of beautifully defined octohedral crystals (Fig. 24), of sizes vary-

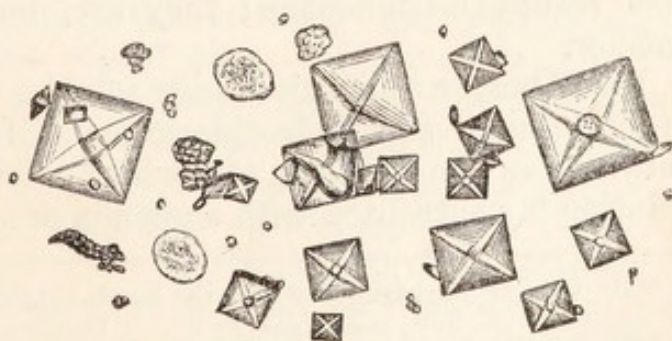


Fig. 24. Octohedral Crystals of Oxalate of Lime.



ing from  $\frac{1}{750}$ th to  $\frac{1}{3600}$ th of an inch in diameter. When examined with polarised light, these octohedra will be found to have little or no action upon it, and remain invisible, or nearly so, when the field is dark.



Fig. 25. Octohedra of Oxalate of Lime; seen when dry.

167. When allowed to dry upon thin glass, each crystal appears under the microscope, especially if the magnifying power is not very high, like a black cube having in the centre a small white square opening, as shown in Fig. 25. This curious appearance is owing to the rays of light, from the greater part of the crystal being refracted beyond the field of vision. On again moistening them the crystals reappear as before in their true octohedral form.

168. Oxalate of lime is not unfrequently met with in the urine having the forms shown in Fig. 26, more or less resembling dumb-bells, with finely striated surfaces. This form of oxalate



Fig. 26. Dumb-bells of Oxalate of Lime.

of-lime sediment, unlike the octohedral variety (166), appears beautifully coloured and striated when examined with polarised light.\* If these "dumb-bells" be kept in any liquid medium for a length of time, they gradually pass into octohedra, which is their more natural form, so that when it is wished to preserve the dumb-bells, they should be put in balsam in which they will continue to retain their peculiar form. There are

occasionally to be seen also, mixed with the octohedra and dumb-bells, a few minute, flat, disk-shaped particles, having a good deal the appearance of blood-corpuscles (451), for which they may readily be mistaken; they are, however, usually much smaller.

169. Oxalate of lime is readily soluble, without effervescence in dilute nitric and hydrochloric acids, from which it is again thrown down in the form of a white precipitate, when the acid solution is neutralized with ammonia or potash.

\* Dr. Golding Bird imagined that the dumb-bells consisted, not of *oxalate*, but of *oxalurate* of lime ( $\text{CaO}, \text{C}_6\text{H}_3\text{N}_2\text{O}_7$ ). ('Urinary Deposits,' fourth edition, p. 219), but the observation that oxalate-of-lime calculi consist often of aggregations of similar dumb-bells renders such an assumption unnecessary.



170. It is insoluble in both cold and hot water; also in acetic and oxalic acids; and in solution of potash.

171. When gently ignited before the blow-pipe, it undergoes little or no blackening, and becomes converted into carbonate of lime ( $\text{CaO}, \text{CO}_2$ ), which, when treated with dilute hydrochloric or nitric acid, dissolves with effervescence (399). The solution thus obtained by dissolving the carbonate in acid, gives, when neutralized, a white precipitate with oxalate of ammonia, but none with ammonia. If the oxalate be kept intensely heated for some little time before the blow-pipe, the carbonate itself is decomposed, and caustic lime is formed (402).

### SECTION XVIII.

#### *Urine containing Cystine ( $\text{C}_6\text{H}_6\text{NO}_4\text{S}_2$ ).*

172. Cystine has occasionally, though but rarely, been found both as a crystalline deposit in urine, and also in the form of small calculi; in one of which latter it was first discovered by Dr. Wollaston.

A deposit of cystine, when examined under the microscope, usually appears as a mass of minute irregularly formed crystals, having the appearance shown in Figure 27. To the naked eye, the deposit has a good deal the appearance of pale fawn-coloured urate of ammonia (93), from which it may be readily distinguished by being insoluble, or nearly

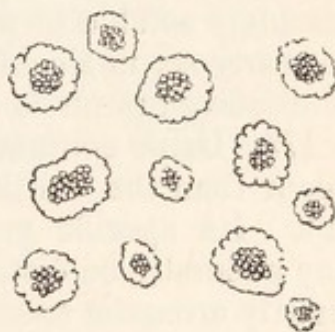


Fig. 27. Cystine.

insoluble, in warm water, and consequently not disappearing when the urine containing it is gently warmed (94).

173. One of the most characteristic properties of cystine is the readiness with which it dissolves in ammonia. If a little of the ammoniacal solution, thus formed, be allowed to evaporate spontaneously on a slip of glass, the cystine is deposited in minute hexagonal crystals, having the form and appearance as shown in Figure 28. It must be remembered that occasionally chloride of sodium crystallizes in octohedral masses (Fig. 29), which in some positions may have at first sight very much the appearance of cystine. The ready solubility of the chloride in water is, however, sufficient to prevent such a mistake. The crystals of cystine, too, when examined with polarised light, appear beautifully coloured, unless very thick, which is not the



case with chloride of sodium. The triangular crystals of triphosphosphate (44), which in some positions somewhat resemble

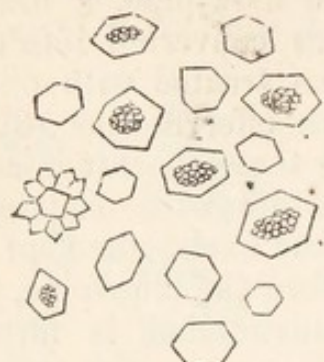


Fig. 28. Cystine Crystallized from an Ammoniacal solution.

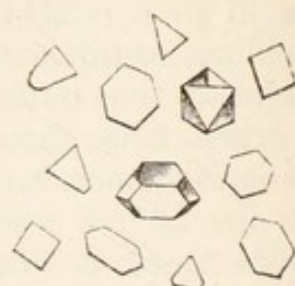


Fig. 29. Crystals of Chloride of Sodium, resembling Cystine.

cystine, may be at once distinguished by their ready solubility in dilute acids (49, 174).

174. Cystine is insoluble in a solution of carbonate of ammonia, but soluble in the fixed alkaline carbonates. It is very sparingly soluble in water, even when warmed, and insoluble or nearly so, in alcohol. In acetic acid it is insoluble, but may be dissolved in nitric and hydrochloric acids.

175. Urine containing cystine has usually a somewhat paler colour than the healthy secretion, with occasionally a greenish tint. Its specific gravity is most commonly rather low. It may generally be distinguished, when fresh, by a peculiar and slightly aromatic smell, a good deal resembling that of sweet briar: this gradually gives place to a fetid, disagreeable odour owing to the occurrence of putrefactive decomposition.

176. Cystic urine is, in most cases, slightly turbid when passed, and becomes considerably more so as it cools, the cystine being less soluble in the cold liquid. A small quantity of the cystine, however, is still held in solution, and may be precipitated by adding a little acetic acid to the filtered urine.

## SECTION XIX.

### *Urine containing Iodine and other foreign matters.*

177. When the compounds of iodine, as the iodide of potassium, are taken internally, it is generally found that nearly the whole of the iodine is carried off by the kidneys, and may be detected, in some form of combination, in the urine. 1



may readily be identified by adding to the secretion a drop or two of yellow nitric acid or very weak chlorine water, and then testing with a solution of starch; when, if iodine is present, the liquid will assume a more or less intense purple colour (807, 810).

178. Many other substances, taken into the system either as food or medicinally, pass into the urine unchanged, and may frequently be distinguished by their peculiar properties. This is especially the case with many of the vegetable colouring matters, as those of indigo,\* madder, beetroot, gamboge, logwood, &c. Some of these may occasionally give rise to the suspicion of the presence of blood, but their real nature may generally be ascertained by examination under the microscope.

179. Besides these colouring matters, various other substances, both organic and inorganic, are occasionally found in urine. Thus, when any metallic preparation has been taken internally, traces of the metal, in some state of combination, may usually be found. The inorganic, and some of the organic acids also, are frequently to be detected; though, when neutral salts of the latter have been taken, carbonates of the bases are more usually found. In addition to these, the odorous principles of many vegetables appear to pass off unchanged in the urine, where they may often be recognised by their peculiar smell.

## CHAPTER V.

EXAMINATION OF URINE SUSPECTED TO CONTAIN EITHER AN UNNATURAL PROPORTION OF SOME ONE OR MORE OF THE USUAL INGREDIENTS, OR ELSE SOME ABNORMAL MATTER.

180. It often happens that, owing to some peculiarity of colour and appearance, either of the liquid or sedimentary portion of morbid urine, or from some other circumstance, such as its high specific gravity, we are led to form some conjecture as to its real nature. When such is the case, one or two well-

\* A deposit of indigo has been found in the urine in cases in which that substance had not been taken into the system.



selected experiments, such as those about to be described, will generally be found sufficient to decide whether or not the suspected peculiarity really exists. When, however, the observer is unable to form a tolerably strong opinion as to the nature of the urine he is about to examine, he had better proceed to test it according to the directions given in Chapter VI.

### SECTION I.

#### *Examination of Urine suspected to contain Urea in abnormal quantity.*

181. When the presence of an excess of urea is suspected, either on account of the high specific gravity of the urine (301), or from any other cause, a drop or two of the liquid should be placed on a slip of glass, and mixed with about an

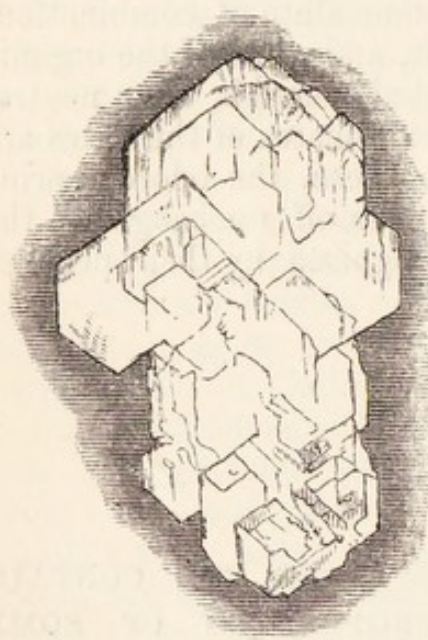


Fig. 30. Nitrate of Urea.

equal quantity of pure colourless nitric acid. If the urea is present in large excess, there will probably be a deposition of minute rhomboidal crystals of the nitrate in the course of a few minutes (fig. 30), and if no trace of crystallization is visible to the naked eye, the mixture should be examined under the microscope. If no crystals appear in the course of half an hour or an hour, a few drops of the urine may be slightly concentrated by evaporation on a slip of glass, at a gentle heat; and when cool, mixed as before, with an equal quantity of nitric acid. Crystals of the nitrate will

now separate if any considerable quantity of urea is contained in the urine; and from the rapidity with which the crystals form, together with their abundance, the student will be able, after a little practice, to form a tolerably accurate opinion as to the relative amount of urea present in the urine. If a microscope is not at hand, the experiment may be made, though less delicately, without it. It must be remembered that variations in the atmospheric temperature affect the crystallization of this salt very materially; in cold weather, a speci-



When of urine will consequently often be found to afford an abundant crop of crystals, which, in warm weather, would furnish little or none. For this reason it is often advisable to cool the mixture artificially, by immersing the glass containing it, either in cold water or a freezing mixture; which latter may be readily made by mixing a little pounded nitrate of ammonia with an equal weight of water. The nitric acid may very conveniently be added to the urine in a thin watch-glass in which it has been previously cooled by floating the glass upon water. Very brilliant leaflets of the nitrate will be deposited if excess of urea be present.

### *Quantitative Estimation of the Urea.*

182. *Liebig's method.*—This is founded on the circumstance that urea is capable of combining with nitric acid and peroxide of mercury, to form a nearly insoluble compound ( $\text{HC}_2\text{H}_4\text{N}_2\text{O}_2, \text{NO}_5, 4\text{HgO}$ ), which is immediately precipitated when a solution of urea is mixed with a solution of nitrate of mercury containing no free acid. But since this reaction does not take place with the bichloride of mercury which is formed, by double decomposition, when the nitrate of mercury is added to urine containing chloride of sodium, it is necessary to remove the chlorine previously to determining the urea; or a larger quantity of the mercury-solution would be employed than was necessary to precipitate the urea. The removal of the chlorine is effected by means of nitrate of silver, its quantity having been previously determined by an ingenious application of the principle above-stated, that nitrate of mercury will not precipitate urea, in the presence of common salt, until a sufficient quantity of the mercury-salt has been added to convert all the chloride of sodium into nitrate of soda.

The test solutions required for this purpose are:—

The solution of nitrate of mercury, No. 1, for determining the chlorine;

The solution of nitrate of silver, for removing the chlorine;

The solution of nitrate of mercury, No. 2, for determining the urea.

*Preparation of the solution of Nitrate of Mercury, No. 1, employed for determining the Chlorine.*—Pure crystals of proto-nitrate of mercury are dissolved in moderately strong nitric acid, and the solution heated until a sample is no longer rendered turbid by chloride of sodium; the solution is eva-



porated, on a water-bath, to a syrupy consistence, and diluted with about 10 times its bulk of water; it is then set aside for twenty-four hours, and, if necessary, filtered.\* In order to graduate the solution, it is requisite to prepare a saturated solution of common salt: pure chloride of sodium (colourless rock-salt, is powdered, and digested with water (at the ordinary temperature) for twenty-four hours, with occasional shaking; so much salt must be employed that a considerable quantity may remain undissolved.† One hundred and fifty grain-measures of this solution (= 47.76 grs. of chloride of sodium) are poured into a small beaker, and mixed with 45 grs. of a solution of urea (containing about 4 per cent. of urea), and with 75 grs. of a cold saturated solution of pure sulphate of soda; to this mixture the solution of nitrate of mercury is added, from a burette, with constant stirring, until a distinct precipitate is permanently formed.‡ The strength of the mercury-solution having been thus ascertained, such a proportion of water must be added to it that 100 grain-measures may correspond to 1 gr. of chloride of sodium.

*Preparation of the solution of nitrate of silver employed for removing the chlorine.*—174.36 grs. of fused nitrate of silver are dissolved in water, and diluted till the solution amounts to 6000 grain-measures; 100 grain-measures of this solution are equal to one grain of the chloride.

*Preparation of the solution of nitrate of mercury, No. 2, employed for determining the urea.*—A solution of nitrate of mercury is prepared, according to the directions given above, so as to contain about 25 grs. of nitrate of mercury in 1800 grain-measures. In order to graduate this solution, 20 grs. of pure urea are dissolved in water, and diluted till the volume of the solution amounts to exactly 1000 grs.; 1500 grain-measures of this solution are poured into a beaker, and the mercury-solution is added from a burette till a few drops on

\* An easier process for the preparation of this solution consists in adding finely powdered red oxide of mercury to moderately strong nitric acid, as long as it is dissolved. An ounce of ordinary nitric acid (sp. gr. 1.42) will dissolve 540 grs. of oxide of mercury, and may then be diluted with 52 ounces of water.

† One hundred grain-measures of this solution contain 31.84 grs. of chloride of sodium.

‡ If any crystalline precipitate should be formed, it may be redissolved by adding a little water. In this process the sulphate of soda is added for two reasons; firstly, because the compound of nitrate of mercury and urea is less soluble in saline solutions (as urine for example) than in pure water; and, secondly, in order that the free nitric acid in the nitrate of mercury may be neutralised by a portion of the soda from the sulphate, which is thus converted into bisulphate.



a watch-glass produce a distinct yellow colour with carbonate of soda. This should be the case after the addition of 300 grain-measures of the mercury solution, but if the latter be prepared of the above strength, less than that quantity will be required, and so much water must be added to the solution as will bring it to the proper standard; thus, suppose only 296 grain-measures had been used, then to every 296 grs. of the solution, 4 grs. of water must be added; 100 grs. of this solution correspond to 1 gr. of urea.

For the expeditious determination of urea in urine, the analyst should be provided with the following measures, accurately graduated, for the solutions employed: \*—

1. A pipette (Fig. 31), with a mark upon the tube indicating the level at which 225 grs. of distilled water would stand. This is employed for measuring the urine after precipitation with baryta.

2. A burette (Fig. 32), capable of containing 100 grs. of distilled water, for the mercurial solution, No. 1. This should be graduated as accurately as possible.

3. A tall narrow glass measure, capable of containing 1000 grs. of distilled water.

4. A graduated burette, containing 1000 grs., for the mercurial solution, No. 2.

Having the test solutions ready prepared, it is necessary, before determining the urea in urine, to remove the phosphoric acid, which is effected by means of a mixture of 2 vols. of cold saturated baryta-water, and 1 vol. of a cold saturated solution of nitrate of baryta.† A glass cylinder, of about 1 oz. capacity, is filled to overflowing with urine, the excess being made to flow off by covering the cylinder with a glass



Fig. 31.  
A Pipette.

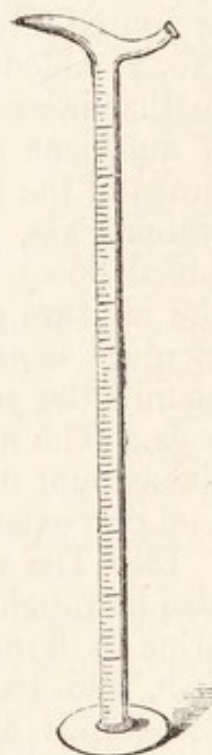


Fig. 32.  
A Burette.

\* These may be obtained from Negretti & Zambra, 11, Hatton Garden.

† The former to neutralise the free acid of the urine, the latter to decompose the alkaline phosphates. If much ammonia be present in the urine, it must be expelled by evaporation, with an excess of baryta, on the water-bath, as it would interfere with the determination of the urea.



plate; two such cylinderfuls are poured into a beaker, and mixed with one cylinderful of the baryta-solution; the precipitate is filtered off, and the amount of chloride of sodium contained in 225 grain-measures of the filtrate (= 150 grs. of urine) is then determined by slightly acidulating with nitric acid, and adding the standard solution of mercury, No. 1, till the appearance of a cloudiness; 450 grs. more of the filtrate (= 300 grs. of urine), are then measured off, acidulated with nitric acid, and mixed with a quantity of the standard solution of silver equal to twice that of the mercury solution employed in the preceding experiment; the liquid is filtered, and half the sum of the mixed liquors is taken for the determination of the urea. This quantity (= 150 grs. of urine), is poured into a beaker, and the graduated mercurial solution, No. 2, added from a burette, with frequent stirring, until no further increase of the precipitate is perceptible; to ascertain if sufficient of the mercury solution has been added, a few drops of the turbid liquid are removed with a pipette into a watch-glass, and a few drops of carbonate of soda carefully added down the edge of the glass;\* if, after some minutes, the mixture retain its white colour, a further quantity of the mercury solution is to be added, until a fresh sample exhibits plainly the yellow colour after the addition of carbonate of soda. The number of grains employed is then read off, and the amount of urea calculated, 100 grs. of the mercurial solution corresponding to one grain of urea.†

183. The absolute quantity of urea present in urine, may also be roughly ascertained by evaporating 1000 grains of the urine to dryness on a water-bath, in a counterpoised porcelain dish, and treating the residue in the manner described in paragraphs 52 to 56, or by precipitating the concentrated urine (500 grs.) with nitric acid (16), and weighing the nitrate after washing with very cold water, and drying at 212°.

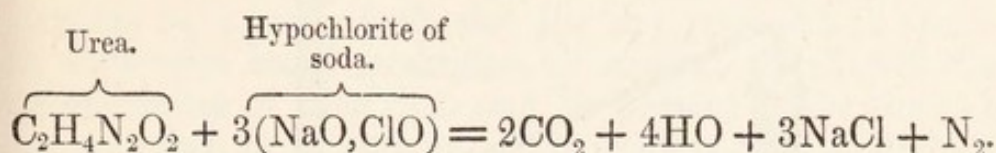
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\* It is a good plan to place a row of drops of carbonate of soda upon a glass plate, and to add, from time to time, a drop of the liquid under examination, until it begins to give a yellow tinge.

† It has been found that, in analyses of urine, when the amount of urea is increasing, an error is committed, tending to diminish the apparent amount of urea; in order to remove this error, an addition has to be made—for 225 grain-measures of urine, and before the test is applied—of 7.5 grs. of water for every 15 grs. of solution of mercury which have been used over and above 450 grain-measures, in a preliminary determination. To obviate an error in the opposite direction, in the more dilute urines, a deduction has to be made of 1.5 grain-measures for every 75 grs. of mercury solution used less than 450 grs.



183a. A very simple method of determining the proportion of urea in urine consists in decomposing it by oxidation with solution of chloride of soda (hypochlorite of soda), and measuring the nitrogen evolved.



To prepare the solution of hypochlorite of soda, 500 grs. of good chloride of lime (bleaching powder) are stirred with boiling water, filtered, and the residue washed once or twice with the boiling water. 1000 grs. of crystallized carbonate of soda are dissolved in a little water and added to the solution, which is then filtered, and made up to 20 oz. with water. Before determining the urea in urine, the uric acid and some of the nitrogenised extractive matters must be precipitated by tribasic acetate of lead. 300 grain-measures of urine are mixed with tribasic acetate of lead till no fresh precipitate is obtained; the solution is boiled and filtered, the precipitate being washed once or twice with water;\* the filtrate and washings are mixed with a solution of 50 grs. of carbonate of soda in a little water, boiled, and again filtered, the precipitate being washed as before. One half of the cold filtrate and washings is introduced into a flask (capable of holding from five to six ounces) which is then rapidly filled up to the brim with the solution of chloride of soda, so that when a cork is inserted, with a narrow bent tube for collecting the gas, the tube may be filled with the liquid, to the exclusion of air. The flask is then placed in a water-bath, with the tube dipping beneath the water in a pneumatic trough, so that the gas may be collected in a tube graduated to fractions of a cubic inch (Fig 33). Heat is then applied to the water-bath, and when the evolution of gas begins to slacken, the flask itself may be heated with a spirit lamp until the volume of the gas in the graduated tube exhibits no increase after a minute or two. The flask is then removed, the graduated tube sunk, as far as possible, in the trough, and when the temperature has fallen to 60° Fahr., the volume of the nitro-

\* If a very rapid determination be required, the mixture containing the precipitate may be measured before filtration, and half that measure taken for determining the urea, so that the washing may be dispensed with.



gen\* is carefully read off, being corrected, by calculation, from

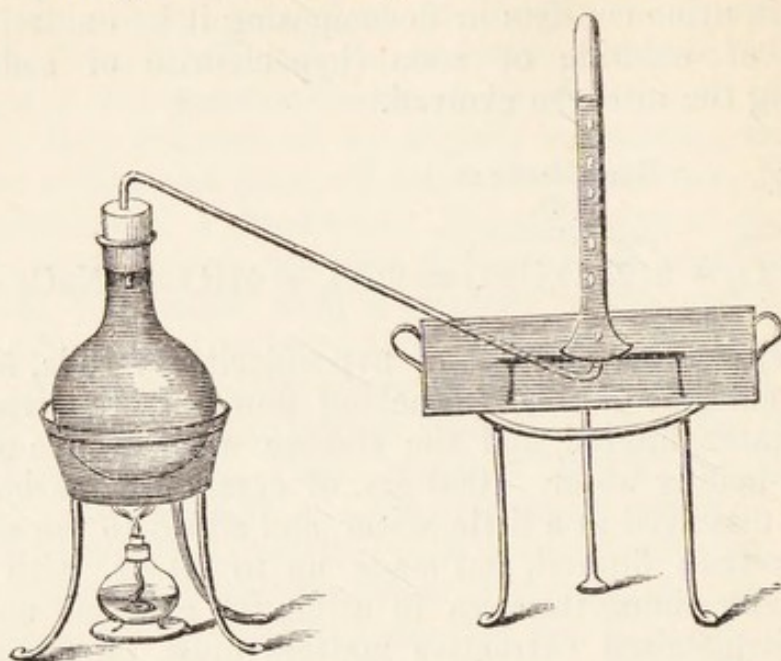


Fig. 33.

any deviation of the barometric pressure from thirty inches. 1.549 cubic inches of nitrogen represent 1 gr. of urea.\*

If ammonia be present in the urine, its nitrogen being evolved, will increase the apparent amount of the urea. After the experiment, the liquid in the flask should be tested with a little sulphuric acid, to ascertain (from the evolution of chlorine) that an excess of chloride of soda was present.†

The same principle may be more easily, though less accurately, applied in the following manner (E. Davy). A measuring tube twelve or fourteen inches long is provided, easily closed by the thumb, and graduated to tenths and hundredths of a cubic inch. This tube is filled rather more than one-third full of mercury, and a measured quantity (50 or 60 grs.) of urine poured into it. The tube is then quickly filled to the brim with solution of chloride of soda, closed by the thumb, and inverted under a saturated solution of common salt (which, being heavier than the solution in the tube, prevents its escape) contained in a small mortar. The tube is allowed to stand for three or four hours, or until the volume of the nitrogen ceases to increase, and the amount of urea is then

\* The carbonic acid having been retained by the large excess of carbonate of soda employed.

† This method of estimating urea was devised by Leconte.



uncalculated as above. In this process, the carbonic acid is retained by the excess of chloride of soda employed.

31 184. When it is suspected that the urea is present in a smaller quantity than in the healthy secretion, or is even altogether absent, 2000 grs. of the urine are to be evaporated to dryness on a water-bath, and the dry residue well stirred with successive small quantities of alcohol, which will dissolve any traces of urea that may be present. The alcoholic solution is then to be evaporated to dryness on a water-bath, and the residue which it leaves is afterwards treated in the manner described in paragraphs 341 and 342, in order to separate the whole of the urea, which may, if necessary, be weighed.

## SECTION II.

*Examination of Urine suspected to contain Uric (or Lithic) Acid in abnormal quantity.*

31 185. When urine is suspected to contain an excess of uric acid, it may be examined in the following manner. Pour off the clear liquid from any solid deposit that may have subsided to the bottom, and retain both the solid and liquid portions for examination.

31 186. A little of the sediment is placed on a slip of glass, and examined under the microscope; when, if uric acid is present in it, either alone, or mixed with the amorphous or rounded particles of urate of ammonia (193), or other matters, it may be distinguished by its peculiar crystalline forms, most of the modifications of which are shown in the annexed figure (Fig. 34).

31 187. If the sediment consists of uric acid, it will prove insoluble when the liquid is warmed. If urate of ammonia is also present, however, the latter will readily dissolve on the application of heat (192), leaving the crystalline uric acid unaffected.

31 188. Uric-acid sediment is insoluble in dilute hydrochloric and acetic acids, but dissolves readily in a solution of potash, owing to the formation of the soluble urate of potash (22).

31 189. When uric acid is moistened with a little tolerably strong nitric acid, and the residue, after evaporation at a gentle heat, is treated, when cold, with a drop or two of ammonia, or exposed to ammoniacal fumes, a beautiful purple



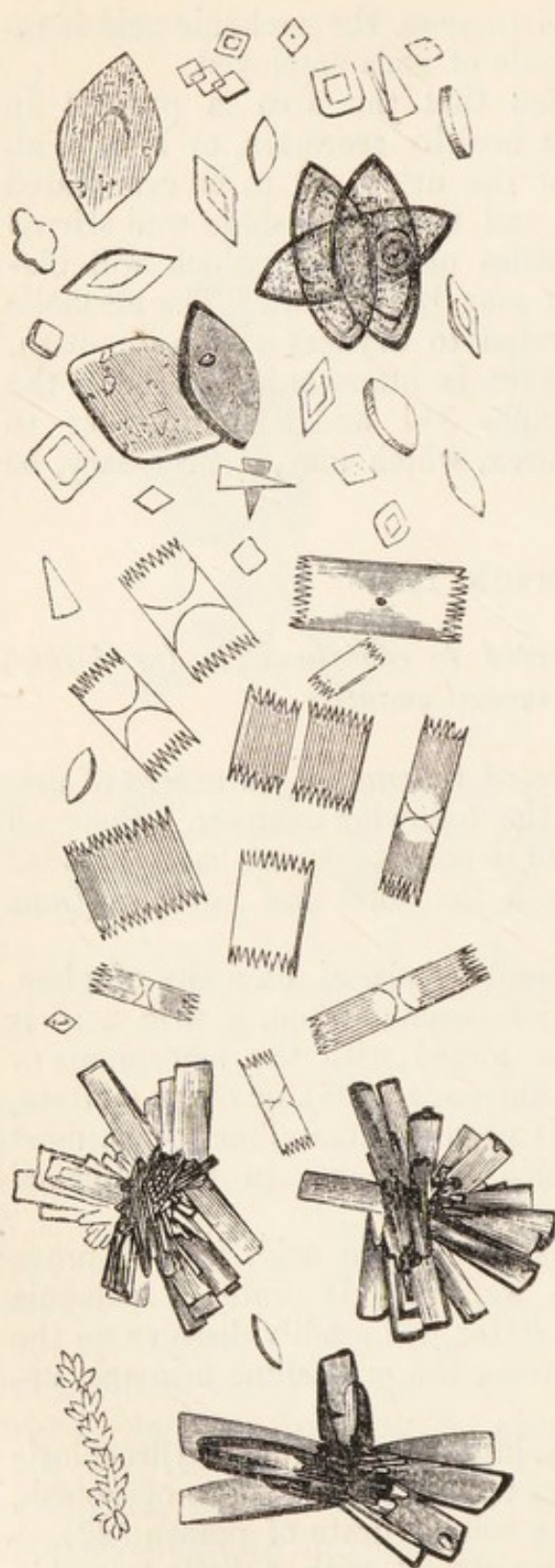


Fig. 34. Crystalline Forms of Uric Acid.

colour is developed, owing to the formation of murexide (23).

190. The clear urine, separated from the uric acid sediment (185), being still saturated with the acid, the latter may be gradually precipitated by adding a few drops of nitric or hydrochloric acid. The uric acid thus precipitated usually has the crystalline forms shown in the upper and middle parts of the figure.

191. When a deficiency of uric acid is suspected, the best way of ascertaining whether or not such is the case, is to filter one or two thousand grains of the urine, in order to separate the mucus and any other solid matter which it may contain, and which may be separately examined for uric acid under the microscope (186), or with nitric acid and ammonia (189). The filtered urine is then evaporated nearly to dryness, on a water-bath, and the residue digested with dilute hydrochloric acid, containing one part of strong acid to eight or ten of water. Any uric acid that may be present will thus be left undissolved and may be examined under the microscope, or otherwise; and, if necessary, weighed, after being first

dried at a temperature of  $212^{\circ}$  on a water-bath.



## SECTION III.

*Examination of Urine suspected to contain an excess of Urate (or Lithate) of Ammonia.*

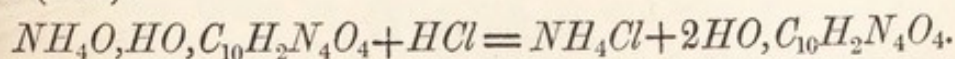
192. When a sediment is suspected to consist, either wholly or partially, of urate of ammonia, a little of the urine containing it is to be warmed over a spirit lamp. If it consists of urate of ammonia unmixed with other matters, it will readily dissolve as the liquid becomes warm, and, on cooling, will be again precipitated. When purpurine is present (104), the urate will probably not dissolve quite so readily on the application of heat as when it is unmixed with the colouring matter.

193. Under the microscope, urate of ammonia appears as an amorphous powder, frequently interspersed with minute round particles larger than the rest, some of which are occasionally found adhering closely together. (See Fig. 11, paragraph 91.) More rarely, it is found in the form of large masses, containing spiculæ (Fig. 12, paragraph 92).

194. It must be remembered that phosphate-of-lime sediment usually has a very similar appearance under the microscope (108), and may consequently be mistaken for urate of ammonia, if the microscopic appearance alone be relied upon. All that is necessary, in order to distinguish between them, is to add a drop of dilute hydrochloric acid to a little of the deposit on a slip of glass. If it consists of phosphate of lime, it will instantly dissolve on the addition of the acid (49,322); while, if urate of ammonia, it will be acted on much more slowly, and in a short time minute crystals of uric acid (Fig. 35) will gradually appear, having been displaced from the urate by the action of the hydrochloric acid (196).



Fig. 35. Uric Acid.



195. When uric acid coexists in a sediment with urate of ammonia, which is of very common occurrence, it may be distinguished under the microscope, by its crystalline forms (186). The uric acid would also be left undissolved when the liquid



is warmed, and may then, if necessary, be separated by filtration, and further examined.

196. Urate of ammonia deposits are not unfrequently found mixed with the earthy phosphates, especially when the urine has at all an alkaline reaction. These will be left undissolved when the liquid is warmed, and may be examined under the microscope, and tested with dilute hydrochloric acid (317, 322).

197. When albumen is present in urine containing a sediment which is supposed to consist of urate of ammonia, it may, by coagulating when heated, disguise the solubility of the urate, and thus lead to an erroneous opinion as to the nature of the deposit. If, however, the heat be applied very gradually, the urate of ammonia will be found to dissolve some time before any of the albumen coagulates; so that, with care, this source of error may be avoided. Or if the urine has been inadvertently allowed to boil, and a precipitation of albumen has taken place, the liquid may be filtered, *while hot*, and the clear filtered solution will, on cooling, again deposit the urate of ammonia; which may then, if necessary, be further examined (94, 192).

198. If pus or mucus be contained in the sediment, together with urate of ammonia, the urine will not become perfectly clear on the application of heat; nor will those substances dissolve on the addition of dilute hydrochloric acid. They may, however, be distinguished with the aid of the microscope (328, 329).

199. When it is required to estimate the quantity of urate of ammonia in a urinary sediment, a portion of the latter, derived from a known quantity of the secretion, is to be boiled with water, and filtered while hot; when the soluble urate will be separated from any uric acid, earthy phosphates, &c., that may be also present with it. The solution is then concentrated by evaporation at a gentle heat, and allowed to cool; when the urate of ammonia will again separate in the solid form, and after drying on a water-bath, may be weighed.

#### SECTION IV.

##### *Examination of Urine suspected to contain Urate (or Lithate) of Soda.*

200. When gently warmed, the deposit dissolves, like urate of ammonia, and reprecipitates on cooling.



201. Under the microscope, it usually appears in the form of small circular, and sometimes semi-crystalline grains, encumbered occasionally with irregularly formed spiculæ, or granular protuberances, as shown in Fig. 13, paragraph 96.

202. When ignited before the blow-pipe on platinum foil, it leaves an abundant white fusible residue of carbonate of soda, which is readily soluble in water, forming a solution in which is strongly alkaline to test-paper.

203. If the ignited residue be treated, on a slip of glass, with a drop of dilute hydrochloric acid, it dissolves with effervescence, forming chloride of sodium; which, if the liquid be expelled by gentle evaporation, is gradually deposited in minute cubical crystals, on the glass, and may be easily recognised with a lens or a microscope (Fig. 36).

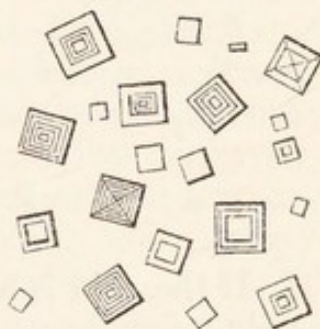


Fig. 36. Chloride of Sodium.

204. When a little of the deposit, previous to ignition, is placed in a drop of nitric acid on a slip of glass, and the residue, after evaporation, treated with a little ammonia, in the manner described in paragraph 23, a purple colour is developed, similar to that caused under the same circumstances, with uric acid and urate of ammonia.

205. Urate of soda may be distinguished from urate of ammonia, which in chemical properties it much resembles, by its microscopic appearance (91, 96); by not being entirely dissipated by ignition (202, 375); by giving no ammoniacal odours when warmed with a solution of potash (377); and by the ignited residue yielding, with hydrochloric acid, cubical crystals of chloride of sodium (203).

## SECTION V.

### *Examination of Urine suspected to contain an excess of Hippuric Acid.*

206. When urine is suspected to contain an excess of hippuric acid, an ounce or so of the liquid is evaporated on a water-bath to the consistence of a syrup; which is then mixed with about half its bulk of strong hydrochloric acid.

The mixture is set aside, and examined after the lapse of a few hours. If any considerable excess of hippuric acid is



present, it will gradually crystallize at the bottom of the dish, in fine tufts of needle-like crystals, often coloured pink by the admixture of purpurine, and having the form shown at *a*, Fig. 37.

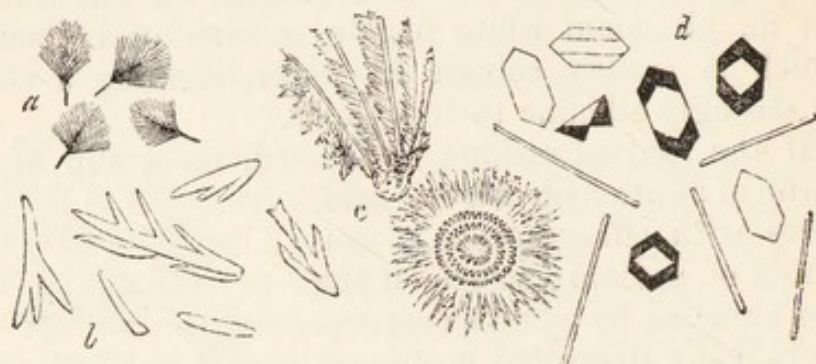


Fig. 37. Hippuric Acid.

207. If the acid is present in smaller quantity, there may be merely a few detached microscopic needle-like or branched crystals, deposited here and there upon the glass, as shown at *b* in the figure.

208. Hippuric acid is readily soluble in alcohol; the alcoholic solution leaving, after evaporation, a crystalline residue, which has usually the appearance shown at *c*, Fig. 37.

209. It is nearly insoluble in cold water, but readily soluble in hot. On cooling, the aqueous solution deposits the acid in well-defined prismatic crystals, which are either detached, as in *d* (Fig. 37), or in tufts, as shown at *a*. These crystals form very beautiful objects under the microscope; and when examined with polarized light, develop colours of great variety and brilliancy.

## SECTION VI.

### *Examination of Urine suspected to contain an excess of Mucus.*

210. Mucous urine always deposits a viscid, tenacious mass, having an alkaline reaction (100), and consisting chiefly of mucus, often mixed with the earthy phosphates, oxalate of lime, and other matters. If the urine be shaken, the deposit does not again mix uniformly with the liquid, but remains cohering in ropy masses, which are very characteristic.

211. When, owing to the admixture of a large quantity of earthy phosphates, the deposit has no longer the property of



adcohering together, the microscope must be resorted to, in order to determine whether or not much mucus is present; the appearance and abundance of the peculiar granular corpuscles (315, 328) furnishing a rough index of the quantity present.

212. It is possible that pus may also be present, in which case, unless in very small quantity, it may generally be detected in the manner described further on (247, 258), where will be found the means of distinguishing between pus and mucus.

213. If it is wished to determine the amount of mucus contained in a deposit, in which it is mixed with earthy phosphates, urates, &c., the sediment must be filtered, and washed with a little boiling water, in order to dissolve out the urates; it may then be treated with a little very dilute hydrochloric acid, which will dissolve out the earthy phosphates, when the residue of mucus may, after careful washing and drying on a water-bath or in a hot-water oven, be weighed.

## SECTION VII.

### *Examination of Urine suspected to contain an abnormal proportion of Extractive Matter.*

214. It is often of some importance to be able to identify the presence of an excess of the peculiar yellow colouring matter, of which the bulk of the extractive matter of urine appears to consist; and also that of purpurine, which is probably a morbid modification of the yellow substance.

#### *Yellow Colouring Matter.*

215. An excess of the yellow colouring matter may be recognised by boiling a little of the suspected urine, and then adding to it a few drops of hydrochloric acid. A more or less intense red colour is in this way produced; the intensity of the colour indicating the comparative amount of the yellow colouring matter present. In healthy urine, a faint lilac or pinkish tint only is caused by the hydrochloric acid; while, if the colouring matter is in large excess, an exceedingly intense crimson is produced.

#### *Purpurine.*

216. The presence of purpurine, or the red colouring matter so often met with in cases even of very slight derange-



ment of the system, is easily ascertained. Owing to its solubility in water or urine, it is never met with as a deposit *per se*.

217. Purpurine, however, has a remarkable tendency to unite with urate of ammonia (104), and whenever a deposit of that substance is formed in urine containing purpurine, the latter is invariably precipitated with it, giving the sediment, which would otherwise be white, or nearly so, a more or less decided pink or red colour. When purpurine is present in a deposit of urate of ammonia, the latter is not so easily soluble in hot water, so that the red deposit does not disappear so readily on the application of heat, as when no purpurine is present (94).

218. If a deposit of urates, coloured with purpurine, be digested in warm dilute alcohol, the purpurine will dissolve, leaving the deposit nearly colourless, and forming a solution of a yellowish-pink colour.

219. Urine containing purpurine, when no excess of urates is present, has a more or less decided pink or red colour, which may appear at first sight very similar to blood.

220. Purpurine may be distinguished from blood, when present in a sediment, by microscopic examination, when the true nature of the uric deposit will be at once apparent (318, 323), together with the absence of blood-discs (330). When treated with warm alcohol also, the colouring matter will be dissolved out (218).

221. Purpurine, when contained in solution in urine, may be precipitated by adding a little warm aqueous solution of urate of ammonia, which will, on cooling, separate from the liquid, carrying with it nearly the whole of the colouring matter, forming a pink deposit, and leaving the urine nearly colourless (217).

## SECTION VIII.

### *Examination of Urine suspected to contain an abnormal proportion of Fixed Alkaline Salts.*

222. When an excess or deficiency of any of the fixed alkaline salts is suspected to be present, a known weight of the urine may be taken, from which the proportion of the substance in question is estimated in the manner described in Chapter II, paragraphs 66 to 84.



## SECTION IX.

*Examination of Urine suspected to contain an abnormal proportion of Earthy Phosphates.*

§ 223. If the suspected urine is neutral or alkaline to test-paper, a sediment of earthy phosphates may be precipitated even in cases where they do not exist in larger proportion than in the healthy secretion; so that the mere occurrence of a small phosphatic deposit is not *necessarily* a proof of their excess (107).

§ 224. On warming the urine, the sediment, if phosphatic, remains undissolved (94, 229).\*

§ 225. The earthy phosphates are readily soluble in most of the dilute acids, especially hydrochloric, nitric, and acetic.

§ 226. If the acid solution thus formed be neutralized or supersaturated with ammonia, the earthy phosphates are immediately reprecipitated (49*b*).

§ 227. They are quite insoluble in potash, ammonia, and the alkaline carbonates (49*c*).

§ 227*a*. When collected on a filter, washed with water, and moistened with nitrate of silver, the earthy phosphates assume a bright yellow colour.

§ 228. A deposit of earthy phosphates may generally be immediately recognised under the microscope. The crystalline forms of the triple magnesian phosphate have been already noticed (44), and these are often mixed with the amorphous phosphate of lime (fig. 338). If a drop of dilute hydrochloric or acetic acid be bladded, while the sediment is in the field of the microscope, the crystals will be seen rapidly to dissolve, leaving the liquid clear, unless uric acid or some other matter insoluble in the acid be also present in the deposit.

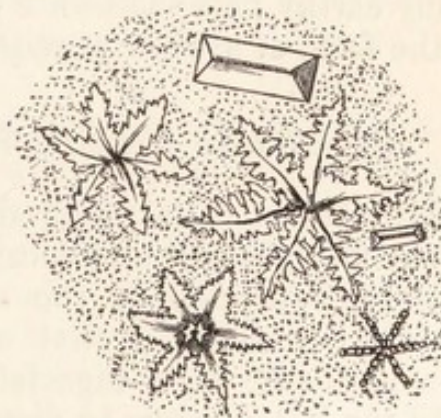


Fig. 38. Mixed Phosphates.

\* If the urine contains albumen, the deposit must be filtered off and washed before being tested.



229. When urine, containing in solution an excess of earthy phosphates, is boiled, a portion of them is usually precipitated, giving the liquid a turbid appearance, resembling the coagulation of a small trace of albumen under similar circumstances (49, 139). It may readily be distinguished from albumen, by adding a drop or two of dilute nitric or hydrochloric acid, which will immediately redissolve the precipitate, if it consists of phosphates, but if albuminous, will not affect it. When the precipitate is found to dissolve on the addition of the first drops of acid, it is advisable, before concluding that albumen is not present, to acidify the mixture more strongly, since the coagulum of albumen, when very small in quantity, occasionally dissolves on the first application of acid, but is wholly reprecipitated on the addition of a few drops more of the acid (140—143).

230. If the absence or a deficiency of the earthy phosphates is suspected, the urine may be treated with a slight excess of ammonia, when, if no precipitate occurs, it may be inferred that they are either altogether absent or else present in very small quantity.

231. In order to ascertain, in such a case, whether or not any traces of them are present, a pint or two of the urine may be evaporated to dryness, and the residue, after incineration, digested with dilute hydrochloric acid, which will dissolve out the earthy salts, if any are present. The acid solution thus obtained is then filtered, and supersaturated with ammonia, when, if any earthy phosphates are present, they will be thrown down in the form of a white precipitate (496).

#### *Quantitative determination of the Earthy Phosphates.*

232. When it is required to estimate the proportion of earthy phosphates in a deposit containing uric acid and other matters, a portion of the sediment, derived from a known quantity of urine, is first washed with a dilute solution of ammonia, and then digested with dilute hydrochloric acid, until the latter ceases to dissolve anything further. The acid solution of the earthy salts, thus obtained, is separated from the insoluble matter by filtration, and then supersaturated with ammonia, which will throw down the whole of the earthy phosphates. The mixture, after standing a short time, to allow the magnesian phosphate wholly to separate, is to be filtered; and the precipitate, after drying at a gentle heat, is to be



weighed, when its weight will represent the amount of deposited earthy phosphates in the quantity of urine from which it was derived.

## SECTION X.

*Examination of Urine suspected to contain Sugar.*

233. When urine is suspected to contain sugar, it may be examined by paragraphs 122 to 130. If any white scum or sediment is present, it should also be examined for the torula vesicles, under the microscope (132).

234. The method of estimating the quantity of sugar contained in diabetic urine will be fully described in Chapter VII.

## SECTION XI.

*Examination of Urine suspected to contain Albumen.*

235. A little of the suspected urine is to be gently boiled in a test-tube. If any albumen is present, it will be coagulated, forming a more or less copious white deposit in the liquid. The precautions necessary for the success of this experiment have been already noticed in paragraphs 139 to 143.

236. To another portion of the urine add a few drops of nitric acid, observing the precautions mentioned in paragraph 143. If a precipitate or milkiness be produced by the acid, and also by boiling (235), the presence of albumen in the urine may be considered certain (141).

237. The proportion of albumen in urine may be estimated with tolerable accuracy by boiling a known quantity of the secretion, and separating the coagulum by filtration; the insoluble matter is then washed with a little dilute nitric or hydrochloric acid, in order to dissolve out any earthy phosphates that may have been precipitated (140), dried on a chloride of calcium bath, at a temperature of  $240^{\circ}$  or  $250^{\circ}$ , and weighed.

238. If the quantity of albumen is so small as not to form a tolerably decided coagulum when boiled, but only to render the liquid opalescent, it will be hardly necessary to proceed with the quantitative determination.

239. The method of making a complete quantitative analysis of albuminous urine will be fully described in Chapter VIII.



## SECTION XII.

*Examination of Urine suspected to contain Blood.*

240. When, from its peculiar red or brown colour, or from other circumstances, the presence of blood is suspected in urine, it may first be examined under the microscope for any blood-corpuscles that may be contained in it (146). If no coagula have separated (145), the liquid should be allowed to repose for a short time, in order to let the corpuscles subside to the bottom; and a drop then taken from the bottom of the vessel will generally be found to contain an abundance of the corpuscles, more or less modified in form and appearance (456).

241. When so much blood is present as to give the urine a decidedly red colour, it will probably be unnecessary to wait for the subsidence of the corpuscles; and a drop of the liquid taken indiscriminately will usually be found to contain sufficient for microscopic examination.

242. If the blood has coagulated, either in the bladder or subsequent to emission, it is most probable that the greater portion of the blood-corpuscles will have been entangled in the coagula, and may be forced out by gentle pressure under a strip of thin glass, so as to be made visible with the help of the microscope.

243. The urine should also be tested for albumen by heat and nitric acid, in the manner already described (139—143). The coagulated albumen will probably, in this case, be more or less highly coloured, owing to the presence of the colouring matter of the blood (147, 455). If the urine already contains coagula, or other solid matter, it should be separated from them by filtration, before being tested for albumen; as their presence would tend to mask the appearance of coagulation.

244. If the urine contains much blood, it may probably become spontaneously gelatinous, owing to the coagulation of the dissolved fibrine (145, 448). This coagulum should be examined under the microscope, since a somewhat similar gelatinous character might be occasioned by the presence of a considerable quantity of mucus (101); or, if the urine be alkaline, of pus (251, 680). The coagulum of fibrine, when pressed between glasses, is usually found to be composed of minute amorphous particles, with a few red blood-corpuscles;



quite different in character from the granular mucus-corpuscles (146, 328).

§ 245. Urine containing bile or purpurine (148, 104), has sometimes nearly the same colour and appearance as when blood is present, and may, without care, be inadvertently mistaken for it. If no trace of blood-corpuscles can be detected under the microscope, we should, before deciding that blood is present, prove that the colour of the secretion is not due to purpurine or biliary matter, by applying the tests described for the detection of those substances, in paragraphs 219—221, 246, &c.

### SECTION XIII.

#### *Examination of Urine suspected to contain Biliary Matter.*

§ 246. When urine is suspected to contain biliary matter, it may be examined by Pettenkofer's and Heller's tests, described in paragraphs 149 and 151. If these fail to afford indications of it in the urine, the latter should be concentrated by evaporation on a water-bath, and the strong aqueous or alcoholic solution of the evaporated residue again tested (150).

### SECTION XIV.

#### *Examination of Urine suspected to contain Pus.*

§ 247. When pus is contained in urine, unmixed with any considerable quantity of mucus, it may readily be distinguished under the microscope by its containing the peculiar nucleated pus-granules (153, 678). These particles, when the urine is allowed to stand a short time, gradually subside to the bottom of the liquid; and when shaken, again mix readily with the urine, in which respect a deposit of pus differs essentially from one of mucus; the latter forming, on agitation, tenaciousropy masses, which do not again mix uniformly with the liquid (99).

§ 248. As purulent deposits frequently appear to the naked eye very similar to those of the earthy phosphates (106), and as it is often difficult to distinguish between pus and mucus when they coexist in a specimen of urine, I will mention the more characteristic tests by which purulent deposits may be most readily identified.



249. It must be remembered that the form and general appearance of the pus- and mucus-corpuscles vary considerably under different pathological conditions of the patient; so that it is not unfrequently impossible to distinguish between them. The granules of pus appear, indeed, to be identical with those of mucus; the difference between the two substances being in the composition of the fluid in which the particles float (661, 676).

250. Under the microscope, with a power of about 4000 diameters, the pus-granules have the appearance represented at *a*, figure 39; and on the addition of a little dilute acetic acid, they become much more transparent, and in each corpuscle one or more internal nuclei are rendered visible, having

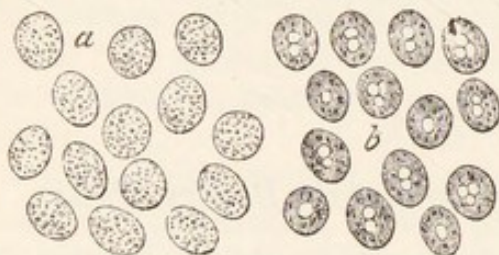


Fig. 39. Pus-Granules.

the appearance shown at *b* in the figure. The granules of pus will be found to float about freely in the liquid (678, 156).

251. When the urine is alkaline, the character of the pus contained in it is different; being then thick and gelatinous, closely resembling mucus (680).

252. The granules of mucus present almost precisely the same appearance under the microscope as those of pus, but are usually, perhaps, rather smaller, and less distinctly granular on the surface. The addition of dilute acetic acid renders visible the interior nuclei, as in the case of pus (250). The acid, however, coagulates the fluid portion of the mucus, owing, probably, to the precipitation of the mucin, before held in solution by a small quantity of alkali (663). In the case of urine containing only a small quantity of mucus, it is uncertain whether this phenomenon of coagulation will be seen, on account of the dilution of the mucous fluid, and also because the coagulation may have been already occasioned by the presence of the large quantity of water (663). When, however, the quantity of mucus is tolerably abundant, the coagulation by acetic acid furnishes a very characteristic reaction.

253. The earthy phosphates, which to the naked eye sometimes closely resemble pus, may be at once distinguished under the microscope by their crystalline form (43), and also by being readily soluble on the addition of dilute acetic acid (228).

254. The *liquor puris*, in which the pus-granules float,



we always contains albumen in solution (676). This may be readily detected by the tests of heat and nitric acid, already described (139); unless, indeed, the quantity of urine is so large, compared with that of the pus contained in it, as to have rendered it too dilute.

255. The fluid portion of mucus, on the contrary, contains no albumen, or merely a minute trace (663), and consequently when diluted with urine undergoes no coagulation when heated, or tested with nitric acid. It is, however, very possible that urine containing an excess of mucus, and no pus, may also contain albumen; so that the mere presence of albumen in the secretion is not necessarily a proof of the presence of pus (101).

256. A certain quantity of fatty matter, readily soluble in ether, is always present in pus (676, 678), but seldom, and in much smaller proportion, in mucus (663). If, therefore, the deposit, or the residue after evaporation, be boiled with a little ether, and the ethereal solution thus obtained is found to yield, on evaporation, small globules of yellowish fat, it is probable that pus is present.

257. A deposit of pus, when treated with a solution of ammonia or potash, becomes converted into a thick gelatinous mass, often sufficiently tenacious to allow the tube containing it to be inverted without any of the mixture flowing out. This reaction is very characteristic.

258. Urine containing pus is most commonly either neutral or slightly acid, and becomes alkaline very slowly. Mucous urine, on the contrary, even if acid, when it is passed, quickly becomes ammoniacal, and alkaline to test-paper (100). Tubercular matter deposited in the urine might at first be mistaken for pus; a minute examination, however, will show débris of cells, and sometimes crystals of cholesterin.

#### SECTION XV.

##### *Examination of Urine suspected to contain Fat or Chylous Matter.*

259. Urine suspected to contain fat, may be examined with a tolerably high power under the microscope, when it is occasionally found to contain minute oil-globules (158, 325). This, however, is not always the case; so that the best way of proving the presence of fatty matter, is to agitate a little



of the suspected urine with about half its bulk of ether; which will separate the fat from the watery fluid, forming, usually, a yellowish solution, which gradually rises to the surface. The ethereal solution thus obtained may then be cautiously evaporated on a water-bath, when the fat or oily matter will, if present, be left behind; and may, if necessary, be tested as to its oily nature, by shaking up with hot water; when, if oil or fat, it will break up into minute globules, immiscible with the water (158).

260. Chylous urine is usually so peculiar in appearance that it can hardly be mistaken for any other morbid condition of the secretion. Under the microscope, it appears to be chiefly composed of amorphous albuminous matter in a minute state of division, mixed occasionally with globules resembling those found in the lymph and chyle. On agitation with ether, it will yield abundant traces of fatty matter, and distinct oily globules may occasionally be distinguished.

261. This form of urine always contains albumen in solution. A portion of this, or more probably a little soluble fibrin (145), not unfrequently coagulates spontaneously after emission, giving the urine a gelatinous or semi-solid consistence. The presence of albumen may be shown by applying to the urine, rendered clear by filtration, the tests of heat and nitric acid (235).

262. If it is required to ascertain the quantity of fatty matter in any specimen of urine, a known weight of the secretion may be agitated with successive small quantities of ether; and the ethereal solution thus obtained will leave, after evaporation, the fatty matter which it had dissolved. This is to be dried on a water-bath until it ceases to lose weight.

#### SECTION XVI.

##### *Examination of Urine suspected to contain Semen.*

263. Microscopic examination is the only trustworthy means of determining whether or not any traces of semen are contained in urine. The urine should be well shaken, and then left to stand a short time, in order to allow the flocculi of mucus and spermatozoa to subside. The greater part of the fluid is then poured off, and a drop containing the sediment, taken from the bottom, and examined under the microscope, with a magnifying power of at least four or five hun-



hundred diameters. If semen is present, the spermatozoa always contained in that secretion will then be visible (160), together, probably, with the peculiar seminal granules also found in the spermatic fluid (161).

264. Traces of albumen, also, may generally be detected in seminal urine, by the application of heat and nitric acid (235).

## SECTION XVII.

### *Examination of Urine suspected to contain Oxalate of Lime.*

265. When the presence of oxalate of lime is suspected, the urine should be allowed to stand some little time, in order that the sediment may partially subside. A little of the liquid taken from the bottom of the vessel is then treated in the manner described in paragraph 164, and examined under the microscope; when, if present, the oxalate will be seen in the form of octohedral crystals (166, 168), or of the dumb-bell shape.

266. Oxalate of lime is insoluble in acetic acid, but dissolves without effervescence in dilute hydrochloric acid, and is again precipitated unchanged, when the acid solution is neutralized or supersaturated with ammonia or potash.

267. If the oxalate of lime deposit be gently ignited, and the residue after ignition treated with dilute hydrochloric acid, it will be found to dissolve with effervescence, having been converted, during ignition, into the carbonate of lime (399).

268. When it is required to estimate the amount of oxalate of lime sediment, it may, if unmixed with other deposits, be separated by filtration from a known quantity of urine, and weighed. When mixed with earthy phosphates or urates, the deposit, after filtration, may be washed with a little dilute acetic acid to dissolve out the phosphates (496); the mixture is then filtered, and the insoluble portion digested in dilute hydrochloric acid, which will dissolve the oxalate of lime, leaving undissolved any uric acid that may be present. The acid solution is then filtered, if necessary, and supersaturated with ammonia; by which the oxalate will be again precipitated. It may then be collected on a weighed filter, dried on a water-bath, and weighed.



## SECTION XVIII.

*Examination of Urine suspected to contain Cystine.\**

269. The presence of cystine may generally be identified by means of the microscope (172), especially after the deposit has been dissolved in ammonia, and allowed to crystallize, either spontaneously or with the aid of a very gentle heat, from the ammoniacal solution (270).

270. Treat a portion of the suspected deposit with a little solution of ammonia; if it is cystine, it will be found readily to dissolve. Place a drop of the ammoniacal liquid on a strip of glass, and allow it to evaporate spontaneously. The peculiar hexagonal tabular crystals of cystine thus obtained, are very characteristic (173).

271. Neutralize the rest of the ammoniacal solution formed in 270, with acetic acid; the cystine, if present, will be precipitated (174).

272. Cystine may be distinguished from urate of ammonia, which it often closely resembles in external appearance, by being insoluble, or nearly so, when the urine containing it is warmed; while urate of ammonia readily dissolves (172, 94).

273. It may be distinguished from the earthy phosphates by its insolubility in acetic acid (174); by its appearance under the microscope (317, 320); and also by its ready solubility in ammonia (173). From chloride of sodium, cystine may be distinguished by its sparing solubility in water (173).

274. If cystine be boiled with a little caustic potash, and the solution tested with acetate of lead, a black precipitate of sulphide of lead will be produced; in consequence of the large amount of sulphur contained in the cystine ( $C_6H_6NO_4S_2$ ).†

## SECTION XIX.

*Examination of Urine suspected to contain other Foreign Matters not included in the foregoing sections.*

275. When the presence of any other kind of foreign

\* In cases where cystine has been excreted, it has generally been found to be most abundant in the morning urine.

† According to Städeler, tyrosine ( $C_{18}H_{11}NO_6$ ) has been found in the form of a sediment in urine, in extreme derangement of the liver. It dissolves in boiling water, and is deposited in fibrous crystals on cooling. Its solution gives a red flocculent precipitate with pernitrate of mercury.



matter is suspected in the urine (180), such as metallic salts, iodine, inorganic or organic acids, &c., a few tests, such as hydrosulphuric acid, hydrosulphate of ammonia, &c., will generally lead to their detection without much difficulty. (See Parts IV and V; also my 'Introduction to Practical Chemistry,' Parts II and III.) If the suspected substance is organic, either the urine itself or the evaporated residue may be tested; but when a non-volatile inorganic substance is to be looked for, it is generally advisable to incinerate the evaporated residue, and test the ash for the substance in question.

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## CHAPTER VI.

### EXAMINATION OF MORBID URINE, THE NATURE OF WHICH IS ALTOGETHER UNKNOWN.

276. When a specimen of urine is suspected to differ in some respect from the healthy secretion, it will generally be found easy, by means of a very few simple experiments, such as those which I am about to describe, not only to ascertain whether or not such is the case, but also to discover the nature of the particular morbid condition in question; whether it be that one or more of the normal constituents of healthy urine is present in an abnormal proportion, or whether it be due to the presence of some substance which is never found in the healthy secretion. In such an examination, the microscope will be found to afford most valuable and ready assistance, the simple microscopic inspection of a deposit often rendering its true nature at once apparent. Whenever, therefore, the student has access to one, he will do well to avail himself of it as much as possible; and he will soon find that, with a little experience, he will be able readily to discriminate between the more common forms of urinary deposits.

For the method of distinguishing the several forms of deposit under the microscope, see paragraphs 315 to 332.



## SECTION I.

*Examination of Urine containing some Solid Deposit.*

277. The urine may be first tested with blue litmus paper, which should be allowed to remain for some time in the urine; if acid, the colour will change to red, or reddish purple. Should the blue colour remain unchanged, test it with yellow turmeric or reddened litmus paper; if the urine is alkaline—owing, probably, to the conversion of urea into carbonate of ammonia (11)—the turmeric will become brown, and the reddened litmus blue; while, if the colour in both cases remain unaltered, the urine may be considered neutral.

278. The specific gravity of the urine may then be taken.

279. This is most readily done by means of the urinometer, which is a little instrument constructed on the principle of the hydrometer, the usual form of which is shown in the annexed figure. The tube, when used, is simply immersed in the urine at the temperature of 60° Fahr.; and when it has come to rest, the number on the graduated scale, which stands at the level of the liquid, when added to 1000, will represent the specific gravity of the fluid. For example, if the level of the liquid stands at 5 on the scale, the specific gravity of the urine will be 1005; if at 30, it will be 1030, and so on (301).

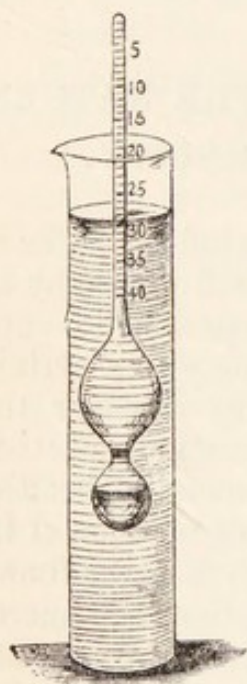


Fig. 40.

280. If a urinometer is not at hand, the specific gravity of the urine may be taken by means of a bottle, or even with a small piece of glass.\*

281. The deposit may now be for the most part separated from the urine, by allowing it to subside for a short time in a tall glass, and then pouring off the clear liquid, or drawing it off with a syphon or pipette. The portion of urine containing the sediment in suspension may be first examined. For the mode of examining the clear liquid separated from it, see paragraphs 300 to 314.

\* See 'Introduction to Practical Chemistry,' fourth edition, p. 56.



*Examination of the Solid Deposit.*

282. If, owing to some characteristic peculiarity in the appearance of the deposit, or of the urine containing it, or from other circumstances, the observer has reason to suspect the nature of the sediment, he may at once proceed to apply the tests for the suspected substance, according to the directions given in Chapter V. At first, however, and until he has had some little experience on the subject, he will do well to adopt some such method of examination as the following.

283. In the great majority of cases, the deposits contained in urine will be found to consist of one or other of the following substances—viz., earthy phosphates, uric acid, urate of soda or ammonia, or oxalate of lime; sometimes alone, sometimes two or more mixed with each other, or with mucus or other matters. The first experiments, therefore, should be directed to the detection of these four substances.

284. Put a little of the urine containing the deposit into a test tube, and warm it gently over a lamp. IF IT READILY DISSOLVES, it is probably URATE OF SODA OR AMMONIA (192, 200); in which case one or two of the more characteristic tests for those substances may be applied, and the deposit may be examined under the microscope, in order to confirm or correct the first result. If purpurine is present with the urate, which may be known by its pink or reddish colour, the deposit will probably not dissolve so immediately on warming, as when the colouring matter is absent (192). If the deposit does not dissolve when gently warmed, nor yet when heated nearly to boiling, it must be further tested as follows.

285. IF THE DEPOSIT DOES NOT DISSOLVE WHEN WARMED, add to a few drops of the sedimentary urine in a test tube, a little acetic acid.

286. If the DEPOSIT DISSOLVES IN ACETIC ACID, it probably consists of EARTHY PHOSPHATES; the nature of which, whether consisting of phosphate of lime, or triple phosphate, or a mixture of both, may be distinguished by submitting a little of the deposit to microscopic examination (228, 317, 322). (Confirm 47, 225—227).

287. IF THE DEPOSIT PROVES INSOLUBLE IN ACETIC ACID, test another portion with a little dilute hydrochloric acid. If it DISSOLVES IN THE ACID, and the acid solution thus obtained gives, when neutralized with ammonia, a white precipitate, it is probably OXALATE OF LIME (266). (Confirm 319, 267.)



288. IF THE HYDROCHLORIC ACID FAILS TO DISSOLVE THE DEPOSIT, it may be tested for URIC ACID by means of nitric acid and ammonia, in the manner described in paragraph 23. Uric acid may also be readily distinguished under the microscope (318). (Confirm 187, 188.)

289. If the deposit proves to consist neither of earthy phosphates, uric acid, urate of ammonia, nor oxalate of lime, it must be examined for the other matters which are occasionally, though less frequently, met with in morbid urine, and which have been already noticed in Chapters IV and V. It must be remembered that, in perhaps the majority of cases, urinary deposits do not consist *exclusively* of any one substance, but contain two or more mixed together; as when the earthy phosphates occur associated with an excess of mucus. The action of the several tests may frequently in this way be more or less masked, and when taken alone, may lead to erroneous conclusions. In such cases, the microscope will be found of infinite value, and should always, when available, be employed (315).

290. If the deposit sinks readily to the bottom of the vessel, forming a PALE GREENISH YELLOW SEDIMENT, which, on agitation, is again diffused readily and uniformly in the liquid, it probably consists of PUS (247). (Confirm 250, 254, 256, 257, 156.)

291. If, on the other hand, the deposit is TENACIOUS AND STICKY, not mixing uniformly with the liquid when shaken, it probably contains an excess of MUCUS (210). (Confirm 211, 100, 156.)

292. If the deposit is DARK COLOURED, brown, or red, and has been found not to consist of urate of ammonia coloured with purpurine (284), it probably contains BLOOD; in which case the clear portion of the urine (281) will give indications of albumen when heated, or when tested with nitric acid (243). (Confirm 240, 242, 245.)

293. When the deposit is WHITE OR NEARLY SO, having proved insoluble when warmed (284), and also when treated with dilute hydrochloric and acetic acids (285, 286); and is found to be readily SOLUBLE IN A SOLUTION OF AMMONIA, the ammoniacal solution yielding on evaporation HEXAGONAL CRYSTALLINE PLATES, it is probably CYSTINE (272, 270, 273).

294. If the deposit is PALE YELLOW, tolerably soluble when warmed (200), but does not appear to consist of urate of ammonia, owing to its yielding no ammonia when warmed



with a solution of potash (205), and appearing under the microscope, not as an amorphous sediment, but in small irregularly shaped roundish or oval particles, with or without projecting protuberances (324), it is probably URATE OF SODA. (Confirm 202, 203, 204.)

295. If, when a little of the urine is agitated with a little ether in a test-tube, and the ethereal solution, after separating from the watery portion on which it floats, is found to leave, after evaporation at a gentle heat, a residue of fat or oily matter, the presence of FAT may be inferred (259). (Confirm 325.)

296. If the urine is OPAQUE AND ALMOST MILKY in appearance, yielding traces of fat when treated with ether; and is found, when examined under the microscope, to contain an abundant white amorphous or granular deposit of albumen or fibrin, together probably with small round colourless corpuscles, it probably contains CHYLOUS MATTER (260). (Confirm 261, 326.)

297. If, on examination under a microscope of high magnifying power, minute ANIMALCULES are visible, having the appearance shown in figure 23, page 60, it is probable that SEMEN is present (160). (Confirm 161, 264.)\*

298. The following table may serve to facilitate the examination of deposits with reagents. It must, however, be borne in mind, that until the observer has had some little experience in the action of the several tests, he must not depend too much on the result of any one experiment; but must in all cases confirm his suspicions by one or more corroborative tests.

### TABLE

*For facilitating the Examination of Urinary Deposits,  
by means of Chemical Tests.*

299. Test first for the earthy phosphates, uric acid, urates of soda and ammonia, and oxalate of lime (283.)

#### 1. THE SEDIMENT DISSOLVES WHEN WARMED ; *Urate of soda*

\* Specimens of urine are occasionally met with holding in suspension a deposit (either amorphous or crystalline) which is insoluble in acids and alkalies as well as in alcohol and ether; but since it is at least diminished by shaking with hydrochloric acid and ether, it appears to consist of an earthy salt of one of the fatty acids.



or ammonia (200, 284). NOT SOLUBLE WHEN WARMED ;  
See 2.

2. SOLUBLE IN ACETIC ACID ; *Earthy phosphates* (286).  
INSOLUBLE IN ACETIC ACID ; See 3.
3. SOLUBLE IN DILUTE HYDROCHLORIC ACID ; *Oxalate of lime* (287). INSOLUBLE IN DILUTE HYDROCHLORIC ACID ; See 4.
4. PURPLE WITH NITRIC ACID AND AMMONIA ; *Uric acid* (288).

If the deposit proves to be neither of the above, it is probably one of the following :

5. GREENISH YELLOW DEPOSIT, EASILY DIFFUSED ON AGITATION ; *Pus ?* (290).
6. ROPY AND TENACIOUS ; *Mucus ?* (291).
7. RED OR BROWN ; NOT SOLUBLE WHEN WARMED ; THE FLUID PORTION COAGULABLE BY HEAT AND NITRIC ACID ; *Blood ?* (292).
8. SOLUBLE IN AMMONIA ; THE SOLUTION LEAVING, ON EVAPORATION, HEXAGONAL CRYSTALS ; *Cystine ?* (293).
9. ETHER YIELDS, AFTER AGITATION, AN OILY OR FATTY RESIDUE ; *Fatty matter* (295).
10. MILKY APPEARANCE ; *Chylous matter* (296).

## SECTION II.

*Examination of Urine containing no Solid Deposit ; or from which a Deposit has been separated* (281).

300. Test the urine with litmus and turmeric paper (277).  
If ALKALINE, it must be tested for ALBUMEN with nitric acid (305, 306).

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\* If these experiments had been already made before the separation of the sedimentary and non-sedimentary portions of the urine (281), they need not be repeated. When the alkalinity is due to ammonia or carbonate of ammonia, red litmus paper, which has been rendered blue by it, will regain its colour when dried by a gentle heat.



301. Take the specific gravity (279).\* If the SPECIFIC GRAVITY IS HIGHER THAN 1025, the urine may perhaps be found to contain either SUGAR or an EXCESS OF UREA (302, 304). If the specific gravity is not higher than 1025, pass on to 305. See also 304.

302. Whether UREA be present in excess, may be ascertained by mixing a little of the urine in a watch-glass, with an equal bulk of pure nitric acid, keeping the glass cool by allowing it to float in cold water. If any excess of urea is present, a more or less abundant crop of crystals of nitrate of urea will, in a short time, appear in the mixture (181). (Confirm 183.)

303. When a microscope is at hand, we can in this manner detect even a very slight excess of urea. A drop of the suspected urine is placed on a slip of glass, and mixed with a drop of pure nitric acid. If even a small excess of urea is present, minute crystals of the nitrate may generally be seen, after a short time, with a very moderate magnifying power.

304. To prove the presence of SUGAR, a little of the urine may be examined by paragraphs 122 and 126. (Confirm 113). It must here be borne in mind, that very decided traces of sugar may exist in urine without raising the density to a suspicious extent, so that the mere circumstance of the specific gravity of the urine being below 1025 is no proof whatever of the absence of sugar; and in any doubtful case it should be carefully looked for by means of the tests above referred to.

305. Boil a little of the urine in a test-tube. If the liquid remains clear, pass on to 307; but if a PRECIPITATE IS PRODUCED, it may be owing to the presence either of albumen (235), or of an excess of earthy phosphates (109). To distinguish between them, add to the boiled portion a few drops of nitric acid. If the PRECIPITATE DISSOLVES, and is not reprecipitated by the addition of a few more drops of the acid, it probably consists of EARTHY PHOSPHATES (229) (confirm 228, 226); while, if it either does not dissolve, or after being dissolved by the first drop or two of the acid, again precipitates when the liquid is more strongly acidified, ALBUMEN is indicated (143). (Confirm 137, 138.)

306. It must be remembered that when the urine is alkaline ALBUMEN may be present in it without being coagulated by boiling (142). Such urine should therefore be tested for albumen, by means of nitric acid (141).

\* See note, p. 96.



307. Add to a little of the suspected urine a few drops of nitric acid. If a PRECIPITATE IS PRODUCED, either immediately or after a short time, none having been occasioned by boiling (305), an EXCESS OF URIC ACID is probably present (190). (Confirm 23, 288.) If the urine is alkaline, the precipitate thus occasioned may consist of ALBUMEN, since that substance would not then be precipitated by boiling (306).

308. Evaporate a little of the urine on a water-bath, to the consistence of a syrup, and add about half its bulk of strong hydrochloric acid. If, after the lapse of a few hours, tufts or branches of NEEDLE-SHAPED CRYSTALS are visible, either to the naked eye or when examined under the microscope, an excess of HIPPURIC ACID is probably present (206). (Confirm 208, 209.)

309. If THE URINE IS HIGHLY COLOURED, it is probable, either that it contains an excess of yellow colouring matter, or, that blood, biliary matter, or purpurine is present.\* To determine which of these it is—

310. Boil a little of the urine; if it contains BLOOD, the albumen will COAGULATE, mixed with some of the colouring matter (243). (Confirm 240, 245.)

311. If an excess of YELLOW COLOURING MATTER is present, the boiled urine, when mixed with a little hydrochloric acid, will assume a more or less decided RED COLOUR (215).

312. The presence of biliary matter may be proved by Pettenkofer's and Heller's tests (149, 151). (Confirm 152.)

313. If PURPURINE is present in solution, the urine usually has a more or less decided pink colour; and when a little warm aqueous solution of urate of ammonia is mixed with it, that salt precipitates as the liquid cools, and carries with it nearly the whole of the purpurine, which gives the precipitate a PINK COLOUR (221). (Confirm 218, 220.)

314. The following table may be found useful for reference (298).

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\* It will be remembered that many vegetable colouring matters taken into the stomach make their appearance in the urine, and might, by a careless examination, be mistaken for blood, &c.



## TABLE

*For facilitating the Examination of the clear liquid portion of Urine, by means of Tests.*

1. SPECIFIC GRAVITY HIGHER THAN 1025; See 2 & 3.
2. CRYSTALS WITH NITRIC ACID; *Excess of urea* (302).
3. HEAT WITH SULPHATE OF COPPER AND POTASH; *Sugar* (304).
4. IF NEUTRAL OR FEEBLY ACID TO TEST-PAPER, see 5, &c.  
IF ALKALINE, see 7.
5. PRECIPITATE FORMED ON BOILING; SOLUBLE IN NITRIC ACID; *Excess of earthy phosphates* (305).
6. PRECIPITATE FORMED ON BOILING; INSOLUBLE IN NITRIC ACID; *Albumen* (305).
7. PRECIPITATE FORMED BY NITRIC ACID; *Excess of uric acid or albumen* (307).
8. CONCENTRATED URINE YIELDS NEEDLE-SHAPED CRYSTALS WITH HYDROCHLORIC ACID; *Hippuric acid* (308).
9. IF THE URINE IS HIGHLY COLOURED, see 10, 11, 12, & 13.
10. DARK COAGULUM FORMED ON BOILING; *Blood?* (310).
11. RED COLOUR WITH HYDROCHLORIC ACID; *Excess of colouring matter* (311).
12. PINK PRECIPITATE WITH WARM SOLUTION OF URATE OF AMMONIA; *Purpurine* (313).
13. CHANGE OF COLOUR WITH NITRIC ACID, &c.; *Biliary matter* (152, 312).



## SECTION II.

*Microscopic Examination of Urinary Deposits (276, 289).*

315. Place a drop of the urine containing the deposit (after being allowed to stand a short time, that the sediment may subside) on a strip of glass; cover it with a small square of thin glass,\* and examine it with a magnifying power of about two hundred diameters. Observe whether the particles are CRYSTALLINE, AMORPHOUS, or ORGANIZED. If CRYSTALLINE, refer to paragraph 316; if AMORPHOUS, to paragraph 321; and if ORGANIZED, pass on to paragraph 327. When, as is frequently the case, the deposit appears to consist of a mixture of two or more different forms of matter, each of these should in succession be examined, until the nature of the whole of the deposit is clearly understood.

316. IF THE DEPOSIT IS CRYSTALLINE, it is probably either URIC ACID, TRIPLE PHOSPHATE, or OXALATE OF LIME; or possibly CYSTINE.

317. If the crystals are STELLATE (Fig. 41), or TRIANGULAR PRISMS (Fig. 42), instantly disappearing on the addition of acetic acid, they consist of the TRIPLE PHOSPHATE.

318. IF THE CRYSTALS ARE LOZENGE-SHAPED, OR POSSESS ANY OF THE FORMS SHOWN IN FIGURE 43, being insoluble in dilute acids, but tolerably soluble in a solution of potash, they are probably uric acid. (Confirm 288.)

319. If the crystals are OCTOHEDRA (Fig. 44), or some modification of the DUMB-BELL form (Fig. 45), insoluble in acetic acid, but really soluble in dilute hydrochloric acid, they are probably OXALATE OF LIME. (Confirm 287.)

320. If the crystals are MULTANGULAR PLATES, having the rosette-like form shown in Fig. 46, insoluble, or nearly so, in water and dilute acids, but readily soluble in ammonia, the ammoniacal solution leaving, on evaporation, HEXAGONAL CRYSTALLINE PLATES (Fig. 47), they are probably CYSTINE (172).

321. IF THE DEPOSIT IS AMORPHOUS, OR IN MINUTELY ROUNDED PARTICLES, it probably consists of PHOSPHATE OF LIME, or URATE OF AMMONIA; or possibly URATE OF SODA, FAT, or CHYLOUS MATTER. See also 327, &c.

322. If it is INSOLUBLE WHEN WARMED, BUT DISSOLVED

\* Except for high powers, the thin glass may generally be dispensed with.



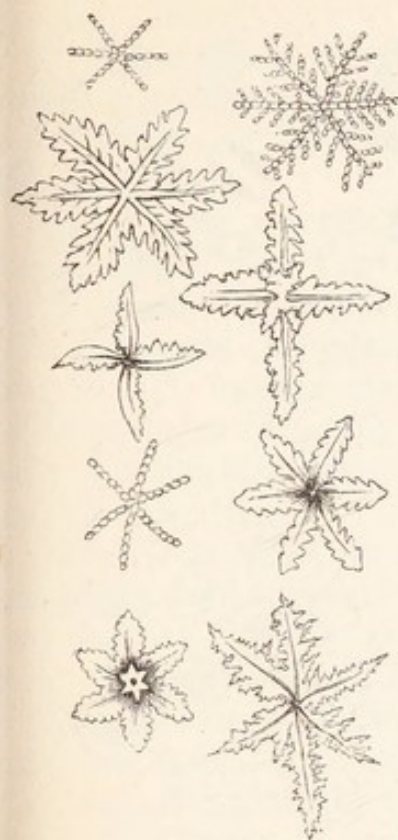


Fig. 41.

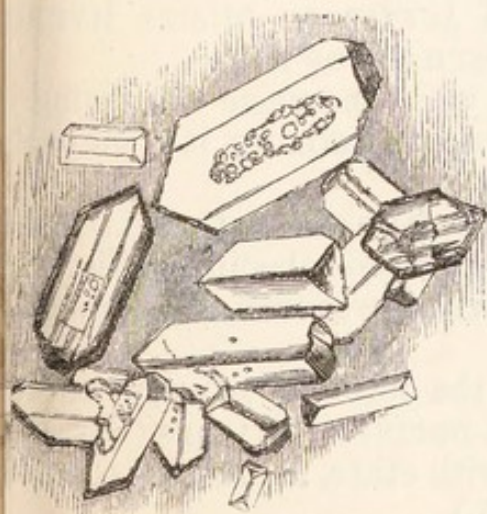


Fig. 42.

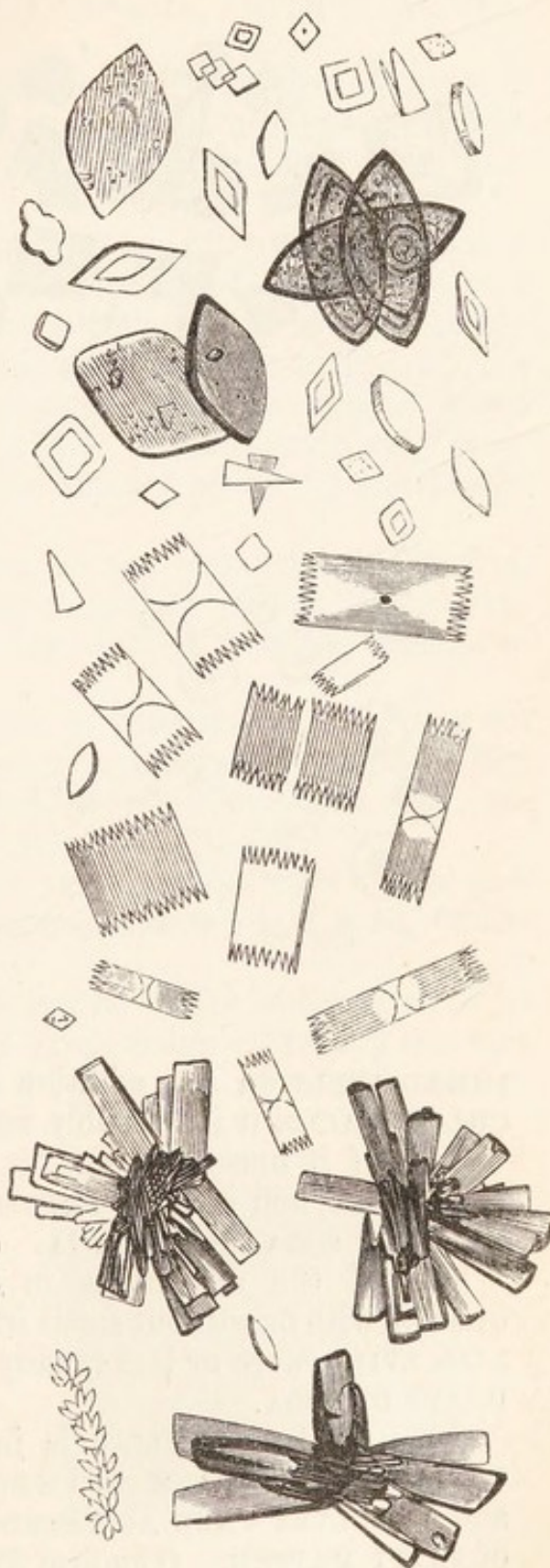


Fig. 43.



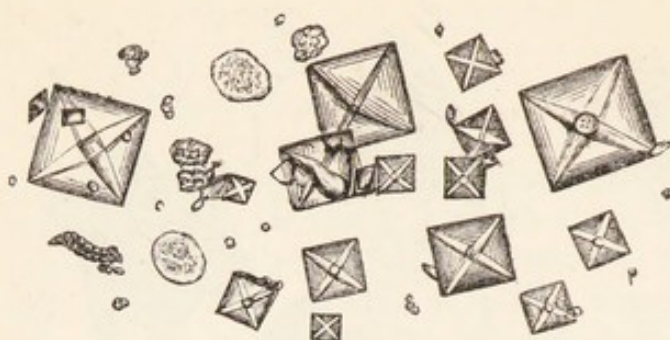


Fig. 44.



Fig. 45.



Fig. 46.

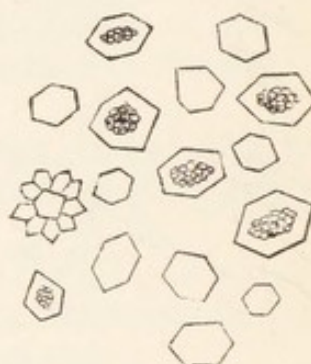


Fig. 47.

IMMEDIATELY on the addition of ACETIC OR DILUTE HYDRO-  
CHLORIC ACID, it is probably PHOSPHATE OF LIME.

323. If it DISSOLVES READILY when the urine containing it  
IS WARMED, and is again DEPOSITED ON COOLING, it is probably  
URATE OF SODA OR AMMONIA.

324. If the deposit is in the form of PALE-YELLOWISH  
GRAINS, with or without small irregular protuberances (Fig. 48),  
DISSOLVING more or less readily WHEN WARMED, it is probably  
URATE OF SODA.

325. If the substance is in the form of MINUTE ROUND  
GLOBULES, WITH DARK AND WELL-DEFINED OUTLINES (Fig. 49),  
and DISSOLVES WHEN AGITATED with ether, it probably consists  
of FATTY MATTER. (Confirm 295.)

326. If the urine is OPAQUE AND MILKY in appearance,  
yielding fatty matter when agitated with ether, and containing  
minute amorphous, albuminous particles, and perhaps also



colourless globules, it probably contains CHYLOUS MATTER. (Confirm 296.)

327. IF THE DEPOSIT CONSISTS OF ORGANIZED PARTICLES, it probably consists either of MUCUS (which is usually mixed with more or less EPITHELIUM), PUS, BLOOD, or SEMEN. See also paragraph 132.

328. If the PARTICLES ARE ROUND, OR NEARLY SO, AND GRANULATED on the surface, ENTANGLED in TENACIOUS, STRINGY MASSES, which do not break up and mix uniformly with the liquid on agitation, it is probably MUCUS (Fig. 50, *a*). EPITHELIAL DÉBRIS may be recognised by the peculiar forms of its particles (Fig. 50, *b*). (156.) Mucous urine very generally contains also a considerable amount of earthy phosphates and other matters.

329. If the particles are ROUND AND GRANULAR (Fig. 51), not being held together by any tenacious matter, but FLOATING FREELY IN THE LIQUID, the deposit probably consists of PUS. (Confirm 290, 156.)

330. If the particles appear as CIRCULAR AND SLIGHTLY CONCAVE DISCS, the outlines being occasionally irregular (Fig. 52), and of a more or less decided yellowish colour, it is probable that BLOOD is present. (Confirm 292.)

331. If the particles, or any among them, have the form of seminal animalcules, or SPERMATOOA, shown in Fig. 53, SEMEN is probably present.

332. The table on page 104 may be useful to the student for reference, in the microscopical examination of urinary deposits.



Fig. 48.

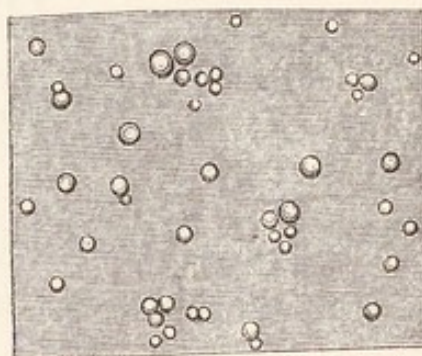


Fig. 49.





Fig. 50.



Fig. 51.

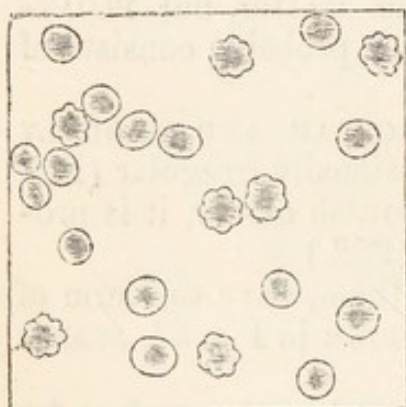


Fig. 52.

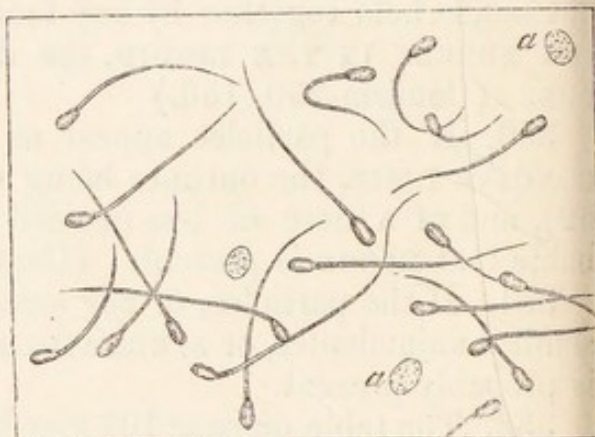


Fig. 53.

## TABLE

*For facilitating the Microscopical Examination of Urinary Deposits.*

1. IF THE DEPOSIT IS CRYSTALLINE, see 4 to 7.
2. IF AMORPHOUS, OR ROUNDED PARTICLES, see 8 to 11.
3. IF ORGANIZED PARTICLES, see 12 to 16.

*Crystalline.*

4. LOZENGE-SHAPED CRYSTALS, AND OTHER FORMS SHOWN in figure 43; *Uric acid* (318).



5. STELLÆ, or THREE-SIDED PRISMS (Figs. 41 and 42); *Triple phosphate* (317).
6. OCTOHEDRA, or DUMB-BELLS (Figs. 44 & 45); *Oxalate of lime*.
7. ROSETTE-LIKE TABLES (fig. 46); *Cystine* (320).

*Amorphous or Rounded Particles.*

8. SOLUBLE WHEN WARMED; *Urate of soda or ammonia* (91, 92, 96).
9. SOLUBLE IN ACETIC ACID; *Phosphate of lime*.
10. ROUND GLOBULES WITH DARK EDGES (Fig. 49); *Fatty matter* (325).
11. WHITE AND MILKY; *Chylous matter?* (326).

*Organized Particles.*

12. GRANULATED CORPUSCLES, IN STRINGY AGGREGATIONS (Fig. 50); *Mucus* (328).
13. IRREGULARLY-SHAPED SCALES (Fig. 50, *b*); *Epithelium*.
14. DETACHED GRANULATED CORPUSCLES (Fig. 51); *Pus* (329).
15. BLOOD-CORPUSCLES (Fig. 52); *Blood* (330).
16. SPERMATOOZOA (Fig. 53); *Semen*.

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## CHAPTER VII.

### QUANTITATIVE ANALYSIS OF DIABETIC URINE.

333. In the quantitative examination of diabetic urine, it is generally sufficient to estimate merely the quantity of sugar, since the determination of the other constituents is of comparatively small practical importance in diagnosis. When this is the case, all that is necessary is, to ferment 250 grs. of the urine in the manner described below (336); and from



the amount of carbonic acid evolved, to estimate the quantity of sugar which yielded it.

334. It is, however, frequently of importance to be able to determine the proportion of some of the other matters co-existing in the urine, especially the urea (119), which has been supposed by some to diminish, and by others to increase, materially in quantity, simultaneously with the appearance of sugar. The exact estimation of small quantities of urea, when mixed, as in diabetic urine, with a large amount of sugar, is attended with considerable practical difficulty; and, indeed, the results hitherto obtained must be regarded merely as approximations to the truth. By the method of analysis which I am about to describe, the proportions of the following substances may, without much difficulty, be determined; or the inquiry may be limited to the estimation of the sugar and the urea (335, 341):—1, water; 2, sugar; 3, urea; 4, uric acid and vesical mucus; 5, animal extractive and ammoniacal salts; 6, fixed alkaline salts; and 7, earthy salts.

335. Two portions of the urine, A weighing 1000 grs., and B weighing 500 grs. are to be evaporated to dryness (50), in weighed or counterpoised dishes, on a water or chloride of calcium bath; or, still better, in vacuo, over sulphuric acid. While the evaporation of A and B is going on, a third portion, C, consisting of 250 grs. of the urine, may be weighed out, for the purpose of estimating the sugar, which is done in the following manner (336).

336. *Treatment of the portion C.*\*—Put 250 grs. of the urine into a small wide-mouthed bottle, capable of holding an ounce and a half or two ounces of water; to the mouth of which is adapted a cork, fitted with tubes of the form shown in the figure (Fig. 54). The bottle should be graduated in cubic inches and tenths, in order to enable the experimenter to estimate the amount of carbonic acid which is retained in solution by the liquid at the close of the operation (338). The

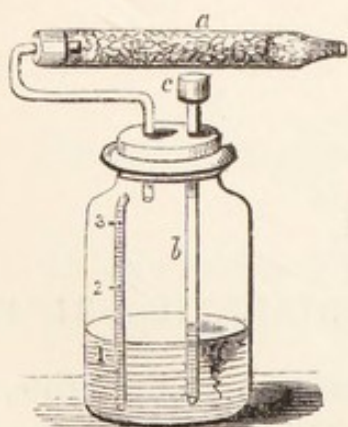


Fig. 54.

tube *a* is nearly filled with small fragments of dry chlorid.

\* Although this method will furnish only a rough estimate of the sugar present, the experiment is an instructive one for the student.



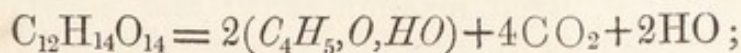
of calcium, which are prevented from falling out by a loose plug of cotton wool placed at each end. The tube, *b*, which reaches nearly to the bottom, is made open at both ends; the top, however, being accurately closed by means of a small bit of cork or wax, *c*, during the process of fermentation.

337. Mix a few drops of fresh yeast, or, still better, about fifty grains of dry German yeast (128), with the urine in the bottle; and having placed the cork, with its tubes, firmly in the neck, weigh the whole apparatus, with its contents, as accurately as possible. Allow the apparatus to stand a day or two in a warm place, having a temperature of about  $70^{\circ}$  or  $80^{\circ}$ ; and when the fermentation appears to have entirely ceased, remove the small plug of cork or wax from the tube *b*, and suck air gently from *a*, for the purpose of expelling the carbonic acid contained in the bottle, and replacing it with common air. The small plug is then attached to the tube *b*, as before, and the whole apparatus is again weighed.

338. The amount of loss will indicate the quantity of carbonic acid which has escaped through the tube *a*; but as carbonic acid is soluble, at ordinary temperatures, in about its own bulk of water, the portion of acid held in solution by the liquid must be added to that which has escaped. This amount is readily known, since each cubic inch of liquid, which may be supposed to be saturated with the acid, must contain about a cubic inch of the gas, weighing rather less than half a grain.\*

339. The whole amount of carbonic acid formed during fermentation, therefore, is determined by adding to the loss of weight half a grain for every cubic inch of liquid contained in the bottle, the quantity of which is known by the graduations on the surface (336). Thus, supposing the loss of weight during fermentation to have been 4.1 grs., and the volume of liquid in the bottle 1.2 cubic inch, the weight of the carbonic acid formed must be  $4.1 + 1.2 \times \frac{1}{2}$ , or 4.7 grs.

340. Now, since every equivalent of diabetic sugar ( $C_{12}H_{14}O_{14}$ ) is converted, during fermentation, into two equivalents of alcohol ( $C_4H_5O, HO$ ), four equivalents of carbonic acid ( $CO_2$ ), and two equivalents of water ( $HO$ );†—



\* One hundred cubic inches of carbonic acid weigh 47.30 grains; one cubic inch, consequently, weighs 0.47 of a grain.

† See note to 129.



it follows that every 198 parts by weight of sugar (one equivalent) give rise to the formation of 88 parts of carbonic acid (four equivalents); so that every 88 grs. of carbonic acid would indicate 198 grs. of sugar, or, in other words, one gr. of carbonic acid will represent 2.25 grs. of sugar. Therefore, by multiplying the weight of carbonic acid by 2.25, we obtain the weight of SUGAR present in the quantity of urine operated on. Thus, in the above example, 4.7 grs. multiplied by 2.25 ( $=10.57$ ) gives the weight of sugar in 250 grs. of urine; which, when multiplied by four ( $250 \times 4 = 1000$ ), represents the proportion in 1000 grs. of the secretion.

341. *Treatment of the portion A.*—The dry residue left after the evaporation of the 1000 grs. marked A (335), is to be used for estimating the urea, which is usually present only in minute proportion in diabetic urine. For this purpose, the residue is treated with successive small quantities of alcohol, stirring the mixture with a glass rod, until it ceases to dissolve anything more. The alcoholic solution is now to be evaporated to dryness on a water-bath, and the residue treated with strong alcohol (absolute alcohol, if possible, 114), which will dissolve out the urea, leaving undissolved most of the sugar and other matters. The alcoholic solution thus obtained is to be again evaporated to dryness on a water-bath, and the residue treated, as long as anything dissolves, with warm distilled water, which will separate the urea from most of the other matters which are less soluble in water.

342. The impure aqueous solution of urea thus obtained is evaporated to a small bulk, and while at a temperature of about  $190^{\circ}$  or  $200^{\circ}$ , mixed with as much pounded oxalic acid ( $\text{HO},\text{C}_2\text{O}_3 + 2\text{Aq}$ ) as will dissolve in the liquid (14). The mixture, after cooling, is immersed in a freezing mixture,\* when the whole of the oxalate of urea, together with the excess of oxalic acid, will crystallize out. The liquid is now to be poured off, and the crystals further treated as in 55.

343. *Treatment of the portion B.*—The residue left after the evaporation of the 500 grs. of urine marked B, may now be examined, for the purpose of estimating, 1, the water; 2, uric acid and vesical mucus; 3, animal extractive and ammoniacal salts; 4, fixed alkaline salts; and 5, earthy salts. For this purpose it is to be carefully evaporated until it ceases

\* A little pounded ice or snow, mixed with about half its weight of common salt; or, in the absence of ice, a mixture of equal weights of nitrate of ammonia and water, will be found the most convenient freezing mixture.



to lose weight, either on a water or chloride of calcium bath, or, still better, in vacuo over sulphuric acid; since by long exposure to a high temperature a portion of the sugar loses five equivalents of water, and becomes converted into a kind of uncrystallizable caramel, thus causing the residue to weigh less than it ought to do. It is generally a matter of considerable difficulty to expel the last traces of water from the residue of diabetic urine: for ordinary purposes, however, this is not of much importance, since the small error which it here occasions affects only the proportion of the water and animal extractive, and not that of the two substances of most importance—viz., the sugar and the urea.

344. The dry residue B is to be weighed; and by deducting its weight from that of the urine before evaporation (500 grs.), the proportion of water is determined; which, when multiplied by two ( $500 \times 2 = 1000$ ), gives the proportion of WATER in 1000 grs. of the secretion.

345. The weight of the dry residue having been carefully noted, it is to be treated with water as long as anything appears to dissolve. In this way the sugar, urea, animal extractive, and alkaline salts are dissolved out, leaving a small insoluble residue, consisting of vesical mucus, uric acid, earthy phosphates, and traces of silica.

346. The aqueous solution thus formed is to be evaporated to dryness on a water-bath, and retained for subsequent experiments (349).

347. The weight of the matter insoluble in water (345), having been noted after careful drying, it is to be incinerated until the residue becomes white or pale grey. The ash thus obtained is to be weighed; and its weight, multiplied by two, furnishes the proportion of EARTHY SALTS in 1000 grains of the urine.

348. The difference between the weight of the ash and that of the dry insoluble residue previous to ignition (347), represents the quantity of insoluble organic matter, consisting of URIC ACID and MUCUS, in 500 grains of the urine; which must be multiplied by two, as in the former cases, in order to give the proportion in 1000 grains of the secretion.

349. The dry residue obtained by evaporating the aqueous solution (346), consisting of the soluble matters of the urine, is now to be weighed. It consists of two portions, the organic or combustible, and the inorganic or incombustible. The relative amounts of these two portions are determined by incineration;



the weight of the ash representing the FIXED ALKALINE SALTS in 500 grains ; which, as before, is to be multiplied by two.

350. The loss of weight experienced during incineration (349), which is that of the soluble combustible matters—viz., sugar, urea, animal extractive, and ammoniacal salts, is also to be multiplied by two. Now, since we know from our experiments with the other portions of urine A and C, the weight of the sugar and urea (340, 342), we can, by deducting their combined weights from the amount of loss during ignition, obtain the proportion of ANIMAL EXTRACTIVE and AMMONIACAL SALTS contained in 1000 grains of the urine.

351. Thus we shall have determined the proportions of the several ingredients of the urine, which together should amount to a fraction less than 1000, viz.—

Water	.	.	.	.	.	.	.
Sugar	.	.	.	.	.	.	.
Urea	.	.	.	.	.	.	.
Uric acid and mucus	.	.	.	.	.	.	.
Animal extract and ammoniacal salts	.	.	.	.	.	.	.
Fixed alkaline salts	.	.	.	.	.	.	.
Earthy salts	.	.	.	.	.	.	.
Loss	.	.	.	.	.	.	.
							<hr/> 1000·00 <hr/>

352. One of the best methods of estimating the sugar is founded upon its property of reducing the oxide of copper ( $\text{CuO}$ ), in an alkaline solution, to the state of suboxide ( $\text{Cu}_2\text{O}$ ), one equivalent of grape-sugar ( $\text{C}_{12}\text{H}_{24}\text{O}_{14}$ ) effecting the reduction of ten equivalents of the oxide.

To prepare the alkaline solution of oxide of copper, advantage is taken of the circumstance that the presence of tartaric acid or a tartrate enables the fixed alkalies to retain the oxide in solution ; and if there be a sufficiently large excess of caustic alkali present, the solution may be boiled without alteration.

315·16 grs. of crystallized sulphate of copper are dissolved in about four ounces of water, and 1420 grs. of the tartrate of potash and soda\* ( $\text{KO}, \text{NaO}, \text{C}_8\text{H}_4\text{O}_{10} + 8\text{Aq}$ ) are powdered, and added gradually to the solution. 480 grs.

\* 950 grs. of pure cream of tartar (bitartrate of potash,  $\text{KO}, \text{HO}, \text{C}_8\text{H}_4\text{O}_{10}$ ) might be substituted for the Rochelle salt, but the latter is preferable, as being more easily obtained in a pure state.



of carbonate of potash are then added, and  $8\frac{1}{2}$  fluid ounces of a solution of hydrate of soda (caustic soda) of sp. gr. 1.12. The volume of the mixture is made up to 10,000 grs. by adding water, the whole boiled for a few minutes, and, if necessary, filtered. 1000 grain-measures of this solution should correspond to 5 grs. of grape-sugar ( $C_{12}H_{22}O_{11}$ ).

In order to ascertain its exact strength, 4.32 grs. of pure cane-sugar\* (white sugar-candy,  $C_{12}H_{22}O_{11}$ ) are dissolved in a little water, in a flask, a few drops of dilute sulphuric acid added, and the mixture boiled for an hour (replacing the water as it evaporates), in order to convert the cane-sugar into grape-sugar. The solution is then rendered slightly alkaline with carbonate of soda, and its volume is made up to 1000 grs. with water. 500 grain-measures of the alkaline copper solution are heated to boiling in a beaker or dish, and the solution of sugar gradually added from a burette or graduated glass (Fig. 32), until the disappearance of the blue colour, and the non-occurrence of any fresh precipitate prove that the whole of the oxide of copper has been reduced. If the copper solution was correctly prepared, 500 grs. of the sugar solution should have been required; but if more or less than this have been found necessary, it is easy to calculate the exact strength of the copper solution, which should then be recorded upon the label of the bottle, together with the date of the experiment. For example, suppose 455 grs. of the sugar solution to have completed the reduction, then

$$\begin{array}{ccccccc} \text{Grs. of sugar-solution.} & & \text{Grape-sugar.} & & \text{Sugar-solution used.} & & \\ \hline \underbrace{\hspace{2cm}} & : & \underbrace{\hspace{2cm}} & :: & \underbrace{\hspace{2cm}} & : & x \\ 1000 & & 5 & & 455 & & \end{array}$$

The value of  $x$  will represent the weight of grape-sugar to which 500 grain-measures of the copper solution correspond. The solution must be kept in a well-stopped bottle, which should be nearly filled by it, and set aside in a dark place.

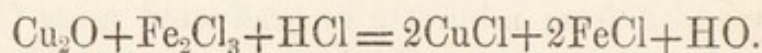
To determine the amount of sugar in diabetic urine, 750 grain-measures of the urine are precipitated by a solution of tribasic acetate of lead, added in very small portions, with occasional stirring, as long as any fresh precipitate is observed. The precipitate (phosphate, sulphate, and urate of lead, with extractive matters) is filtered off, and washed with a little water, so as to make the total volume of the filtrate and washings up

\* Which will yield 5 grs. of grape-sugar. It would be better to employ 5 grs. of pure grape-sugar, but this is not so easily obtained.



to 1000 grs. 500 grain-measures of the alkaline copper solution are then heated to boiling, and the purified urine added from a burette, until the blue colour of the solution has disappeared. A simple calculation will then give the amount of sugar contained in the 750 grs. of urine originally employed.

A very excellent method of controlling the result of this experiment consists in adding to the solution from which the blue colour has disappeared enough of the alkaline copper solution to restore the blue colour, boiling for a few seconds, and collecting the precipitated suboxide of copper upon a filter. It is then rapidly washed, as long as the washings are alkaline, and dissolved by pouring over the filter a hot solution of perchloride of iron, acidulated with hydrochloric acid, when the suboxide of copper ( $\text{Cu}_2\text{O}$ ) is converted into the chloride ( $\text{CuCl}$ ):



The filter is washed with much water, and the amount of iron in the state of protochloride ( $\text{FeCl}$ ) is determined by adding a solution of permanganate of potash of known strength\* from a burette, until a permanent faint rose colour is produced.

Now, each atom (198 parts) of grape-sugar ( $\text{C}_{12}\text{H}_{14}\text{O}_{14}$ ) precipitates five atoms (357 parts) of suboxide of copper ( $\text{Cu}_2\text{O}$ ), and these, when dissolved in perchloride of iron, produce ten atoms of protochloride of iron ( $\text{FeCl}$ ), containing 280 parts of iron. Hence every grain of iron indicated by the permanganate of potash represents 0.707 gr. of grape-sugar.

A slight error occurs in the determination of sugar in urine according to this process, from the precipitation of a little sugar by the tribasic acetate of lead; for although a pure solution of sugar is not precipitated by that reagent, the precipitate which it causes in saccharine urine is always accompanied by a little sugar. In most cases this error is too slight to be of any consequence, but its extent may be easily ascertained by boiling the lead precipitate with oxalic acid, filtering, mixing the solution with an excess of potash, and determining the sugar by the alkaline copper solution. This will give a slight error in the opposite direction, on account of the reduction of the oxide of copper by the uric acid.

\* The strength of this solution is readily determined by dissolving 5 grs. of pure iron wire in hydrochloric acid, diluting largely with water, and adding the permanganate solution, from a burette, till the permanent rose colour is seen. About 1000 grs. of the solution should be required for 5 grs. of iron.



The following analyses of diabetic urine will serve to illustrate its usual composition in 1000 parts :

*Analyses I & II. (Simon.)*

	I.	II.
Specific gravity . . . . .	1018	1016
Water . . . . .	957.00	960.00
Solid constituents . . . . .	43.00	40.00
Urea . . . . .	traces	7.99
Uric acid . . . . .	traces	traces
Sugar . . . . .	39.80	25.00
Extractive matter and soluble salts .	2.10	6.50
Earthy phosphates . . . . .	0.52	0.80
Albumen . . . . .	traces	traces

*Analyses III, IV, & V. (Dr. Percy.)*

	III.	IV.	V.
Specific gravity . . . . .	1042	1035	1039
Water . . . . .	894.50	918.30	898.90
Solid constituents . . . . .	105.50	81.70	101.10
Urea . . . . .	12.16	30.32	2.39
Uric acid . . . . .	0.16	0.26	not isolated
Sugar . . . . .	40.12	17.15	79.10
Extractive matters } and soluble salts }	53.06	32.59	19.52
Earthy phosphates . . . . .		1.30	0.09

*Analysis VI. (Bouchardat.)*

Water . . . . .	837.58
Solid constituents . . . . .	162.42
Urea . . . . .	8.27
Uric acid . . . . .	not isolated
Sugar . . . . .	134.42
Extractive matters and soluble salts .	20.34
Earthy phosphates . . . . .	0.38



## CHAPTER VIII.

## QUANTITATIVE ANALYSIS OF ALBUMINOUS URINE.

353. In the quantitative analysis of albuminous urine, it is usual to estimate the following ingredients; though for many purposes it is sufficient merely to determine the proportion of albumen, either with or without that of the urea:—1, water; 2, urea; 3, albumen, with traces of uric acid;\* 4, vesical mucus; 5, animal extractive and ammoniacal salts; 6, fixed alkaline salts; and 7, earthy salts.

354. *Treatment of the portion A.*—Two portions of the urine, marked respectively A and B, each weighing 5000 grains, are to be evaporated to dryness on a water-bath,† The portion A will serve for the estimation of the urea; and the portion B for that of the other substances above enumerated.

355. The residue left after the evaporation of A is treated with hot alcohol, to dissolve out the urea. The alcoholic solution is evaporated to dryness on a water-bath, and redissolved, as far as it is capable, in hot distilled water; the aqueous solution thus obtained is evaporated to a small bulk, and mixed with pounded oxalic acid in the manner described in the analysis of diabetic urine (342). The oxalate of urea is afterwards decomposed by means of carbonate of lime in the manner already detailed; the weight of the urea obtained being multiplied by two, in order to represent the proportion of UREA in 1000 grains of the urine.‡

356. *Treatment of the portion B.*—The residue left after the evaporation of B is now to be examined. When it has ceased to lose weight by exposure on the water-bath, the weight of the residue is to be noted; and the loss which it has sustained during evaporation, multiplied by two, will represent the amount of WATER in 1000 grains of urine.

\* Or the uric acid may be estimated separately. See paragraph 363.

† If it is intended to estimate the uric acid separately, a third portion of urine, weighing 1000 grs., will also be required (363).

‡ A far more accurate result would, of course, be obtained by heating the urine (acidified, if necessary, with acetic acid) to coagulate the albumen, and determining the urea in the filtered liquid according to (182).



357. The dry residue, when cold, is to be carefully reduced to powder in a clean dry mortar, which should be placed on a large sheet of white paper, in order to catch any particles that may be projected out of the mortar during the pounding. The powder is to be warmed with distilled water, which will dissolve out the urea, animal extractive, and soluble salts; leaving an insoluble residue of coagulated albumen, uric acid, mucus, and earthy salts. The mixture is then filtered. The solution thus obtained we call M, and the insoluble matter N.

358. The solution M is to be evaporated to dryness on a water-bath, and subsequently examined in the manner described below (361). While the evaporation is going on, the insoluble matter N may be operated on (359).

359. The insoluble matter N, consisting of albumen, uric acid, mucus, and earthy salts, is to be carefully detached from the filter whilst still moist. It is then warmed for a few seconds with a little dilute nitric acid (consisting of one part of strong acid, and about ten parts of water,) and well stirred with a glass rod, in order to dissolve out the earthy phosphates. The insoluble portion is to be washed with a little warm water (360), and the acid solution, together with the washings, then evaporated to dryness on a water-bath. The dry residue is weighed, incinerated, and weighed again; when the weight of the incombustible matter, multiplied by two, will represent the proportion of EARTHY PHOSPHATES in 1000 parts of the urine; while the loss which the mixture sustained during the incineration, also multiplied by two, will represent the amount of VESICAL MUCUS.

360. The portion of N which proved insoluble in the dilute nitric acid (359), consisting of albumen, with probably a little uric acid, is to be dried on a water-bath, and weighed. The weight, multiplied by two, will represent the proportion of ALBUMEN and URIC ACID in 1000 grains of the urine.

361. The evaporated residue left by the solution M (358), containing the urea, animal extractive, and soluble salts, must now be examined. After its weight has been ascertained, the dry residue is to be gently ignited until the incombustible matter becomes white or pale grey. The ash thus obtained is then weighed; and its weight, multiplied by two, will represent the proportion of FIXED ALKALINE SALTS in 1000 grains of the urine.\*

\* During this ignition, traces of the alkaline chlorides are always volatilized, causing a slight loss.



362. The loss of weight which the residue sustained during incineration (361) being due to the combustion of the urea and animal extractive, and the volatilization of the ammoniacal salts, derived from 500 grains of urine; we obtain, by doubling it, the amount of those substances contained in 1000 grains. From this we deduct the proportion of urea, which we have already ascertained (355), and the difference will represent the amount of ANIMAL EXTRACTIVE and AMMONIACAL SALTS contained in 1000 grains of the secretion.

363. If it is required to estimate the proportion of uric acid in albuminous urine, which, however, is seldom necessary, since there is not often more than a small trace of it present, a separate portion of urine must be used for the experiment. For this purpose, 1000 grains are to be boiled for about a quarter of an hour, and filtered from the coagulated albumen. The filtered liquid is then concentrated to about one fourth of its bulk, by evaporation on a water-bath, and, after the addition of a few drops of hydrochloric acid, set aside in a cool place for forty-eight hours. The URIC ACID, if present in any notable quantity, will gradually crystallize out, mixed possibly with traces of hippuric acid (25), which may be washed off with a little alcohol (28). The weight of the residue will then, after drying on a water-bath, represent the proportion of the acid in 1000 grains of urine.

364. Thus we shall have completed the analysis, having determined the proportion of the several ingredients proposed, which, when added together, should amount to a fraction less than 1000 grains, viz.—

Water	.	.	.	.	.	.	.
Urea	.	.	.	.	.	.	.
Albumen	.	.	.	.	.	.	.
Uric acid	.	.	.	.	.	.	.
Vesical mucus	.	.	.	.	.	.	.
Animal extractive and ammoniacal salts	.	.	.	.	.	.	.
Fixed alkaline salts	.	.	.	.	.	.	.
Earthy salts	.	.	.	.	.	.	.
Loss	.	.	.	.	.	.	.
							1000·000

365. The following analyses of albuminous urine, in case



of Bright's disease, will serve to show its usual composition in 1000 parts :

*Analyses I & II. (Simon.)*

	I.	II.
<i>Specific gravity</i> . . . . .	1014	1022
Water . . . . .	966.10	933.50
Solid constituents . . . . .	33.90	66.50
Urea . . . . .	4.77	10.10
Uric acid . . . . .	0.40	0.60
Fixed salts . . . . .	8.04	10.00
Extractive matters . . . . .	2.40	—
Albumen . . . . .	18.00	33.60

*Analysis III. (Dr. Percy.)*

<i>Specific gravity</i> . . . . .	1020
Water . . . . .	946.82
Solid constituents . . . . .	53.18
Urea . . . . .	7.68
Uric acid and indeterminate animal matter . . . . .	17.52
Fixed soluble salts . . . . .	5.20
Earthy phosphates . . . . .	0.14
Albumen . . . . .	22.64



## PART II.

### CALCULI AND CONCRETIONS.

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#### CHAPTER I.

#### URINARY CALCULI.

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##### SECTION I.

366. URINARY calculi are composed, in the great majority of cases, of substances which are contained in healthy urine, such as uric acid, urate of ammonia, and the phosphates of lime and magnesia; they are, however, occasionally composed of substances which are met with only in morbid urine, such as oxalate of lime, cystine, &c. Other substances also, which may strictly be called accidental, are occasionally contained in calculi; such as fragments of sand, or other hard bodies, which have accidentally found their way into the kidneys or bladder, and there formed nuclei, round which the earthy phosphates, or other matters, have gradually been deposited. Calculi always contain, in addition to the ingredients of which they mainly consist, more or less animal matter, such as dried blood and urine, vesical mucus, &c.

367. Calculi are found to consist occasionally almost entirely of one ingredient only, but more frequently of two or more different constituents arranged together in irregular concentric layers. On this account it is impossible to determine, with any degree of certainty, the nature of the mass of a calculus, by merely examining the external coating, since the more central portion may be of a nature wholly different. The best



way is to divide the calculus into two equal parts, which is easily done by carefully cutting it through the centre with a fine saw. Fig. 55 represents a mixed calculus divided in this manner; the darker layers consisted, in the specimen from which the drawing was made, of oxalate of lime, and the lighter rings of uric acid. When a calculus is thus found on examination to consist apparently of two or more kinds of matter, fragments of each kind should be carefully detached and separately examined (411).\*



Fig. 55. Alternating Calculus.

## SECTION II.

*Uric (or Lithic) Acid* ( $C_{10}H_4N_4O_6$ ).

368. Uric acid calculi are usually smooth or slightly tuberculated on the surface (Fig. 56), and of colours varying from pale yellowish fawn to reddish or brown. When sawn through, the layers will generally be found to be tolerably regular, although of different thicknesses, and nearly parallel to the outline of the section. This is the most common of all the urinary calculi.



Fig. 56. Uric Acid Calculus.

369. Heat a small fragment of the calculus on platinum foil; it immediately blackens, owing to the charring of the animal matter, emitting, at the same time, a disagreeable smell, resembling that of burnt feathers, mixed with that of hydrocyanic acid ( $H, C_2N$ ), which, together with carbonate of ammonia and some other compounds, is formed during the decomposition. If the heat be continued, the charred residue is gradually consumed, leaving only a slight trace of ash, which is usually alkaline to test-paper, consisting of phosphate or carbonate of soda.

\* \* A small fragment of the calculus, about the size of a pin's head, is generally sufficient for each experiment, and will be found more convenient in practice than a larger quantity.



Traces of the earthy phosphates, also, are almost always to be found in this and most other varieties of calculi.

370. Uric acid is sparingly soluble in water, and in cold dilute acids (22).

371. A little of the calculus in powder is placed in a drop or two of tolerably strong nitric acid, in a watch-glass, or on a strip of glass or platinum; it dissolves with effervescence, carbonic acid and nitrogen being given off, and a mixture of alloxan ( $C_8H_4N_2O_{10}$ ), alloxantine ( $C_8H_5N_2O_{10}$ ), and some other compounds, remains. This is evaporated to dryness at a gentle heat, when a red residue is left, which, *when cold*, and treated with a drop of ammonia, or exposed to ammoniacal fumes, becomes purple, owing to the formation of murexide ( $C_{12}H_6N_5O_8$ ).

372. Uric acid calculus dissolves in a dilute solution of potash, leaving only a few shreds of animal matter (366); and when the mixture is warmed, no smell of ammonia is perceptible, thus differing from the urate of ammonia (377). On neutralizing the alkaline solution with any acid, as hydrochloric, a white precipitate of pure uric acid is thrown down, which, when separated by filtration, may be tested with nitric acid and ammonia, as described in 371.

373. If the precipitated uric acid be examined under the microscope, it will be found to consist of minute crystals, having the form shown in Fig. 3, page 7.\*

### SECTION III.

#### *Urate (or Lithate) of Ammonia* ( $NH_4O, HO, C_{10}H_2N_4O_4$ ).

374. It is not often that we meet with calculi composed wholly of urate of ammonia, that substance being more commonly found alternating with uric acid, earthy phosphates, or other matters. These calculi are generally small in size, smooth,

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\* *Xanthic* or *Uric Oxide* or *Xanthine* ( $C_{10}H_4N_4O_4$ ), which composes a very rare form of calculus, also dissolves in potash, and is reprecipitated by hydrochloric acid. When dissolved in nitric acid, however, it leaves a yellow residue on evaporation, which is not reddened by ammonia. Xanthic oxide has also been found in normal urine, and in the spleen, pancreas, brain, and liver of oxen and other animals. *Hypoxanthine* ( $C_{10}H_4N_4O_2$ ) is very similar in its properties, and has also been found in the spleen. Dr. Bence Jones has observed, in one case, in the urine, a deposit which had the chemical characters of xanthic oxide, and appeared, under the microscope, in lozenge-shaped crystals, resembling some of the forms of uric acid.



or slightly tuberculated (Fig. 57), and pale slate or clay colour, sometimes inclining to brown. The concentric layers are usually thinner, and less distinctly marked, than those of uric acid.



Fig. 57.  
Urate of Ammonia Calculus.

375. When heated, urate of ammonia usually decrepitates, gradually disappears, and in other respects behaves like uric acid (369). It dissolves tolerably well in hot water; but being insoluble, or nearly so, in cold, is deposited again when the solution cools, as an amorphous precipitate. If a dilute acid, as hydrochloric, be added to a hot solution of urate of ammonia, the latter is decomposed, and the uric acid set free, which, being sparingly soluble even in hot water, is precipitated in the form of minute crystals (Fig. 3, page 7).

376. With nitric acid and ammonia, urate of ammonia produces the same results as uric acid (371).

377. Urate of ammonia dissolves readily in a warm dilute solution of potash, giving off at the same time ammoniacal fumes by which it may be distinguished from uric acid and urate of soda. The addition of a dilute acid to the hot solution causes a crystalline precipitate of uric acid (373).

#### SECTION IV.

##### *Phosphate of Lime* ( $3\text{CaO}, \text{PO}_5$ ).

378. Calculi of phosphate of lime are most commonly smooth and even polished on the surface. The concentric laminæ are generally arranged with considerable regularity (Fig. 58); and when the calculus is broken, these separate from each other with great facility, forming detached crusts. The colour is usually pale fawn or stone colour.



Fig. 58.  
Phosphate of Lime Calculus.

379. Before the blow-pipe it chars, owing to the presence of a little animal matter, and gradually becomes white as the carbonaceous matter burns away. It is almost infusible, requiring



for its fusion so intense and prolonged a heat, that few can succeed in fusing it.

380. The residue, after ignition, is neutral to test-paper.

381. It is soluble, without effervescence, in dilute nitric or hydrochloric acid (49).

382. To the solution in nitric acid formed in the last experiment add ammonia in slight excess; the phosphate of lime will be precipitated as a gelatinous precipitate. Re-dissolve this in a little acetic acid, and divide the solution into two parts.

383. To one part of the acetic solution add a drop of perchloride of iron, which will cause a yellowish white precipitate of perphosphate of iron ( $\text{Fe}_2\text{O}_3, \text{PO}_5$ ).

384. To the second part of the solution add oxalate of ammonia, when the white precipitate of oxalate of lime will be formed.

385. If a little of the powdered phosphate of lime be mixed with about twice its bulk of the double phosphate of ammonia and magnesia, or triple phosphate ( $\text{MgO}, \text{NH}_4\text{O}, \text{HO}, \text{PO}_5$ ), and heated before the blow-pipe on platinum wire, it readily fuses. The *fusible calculus* is composed of a similar mixture of the two salts (391).

## SECTION V.

### *Phosphate of Ammonia and Magnesia, or Triple Phosphate* ( $\text{MgO}, \text{NH}_4\text{O}, \text{HO}, \text{PO}_5$ ).

386. Calculi composed entirely of triple phosphate are of somewhat rare occurrence; but mixed, or alternating with other matters, and indeed constituting the great bulk of the concretion, this substance is very common. Such calculi are sometimes found to have been deposited in concentric layers, and sometimes consist of an aggregated mass of prismatic crystals. They are usually nearly colourless, or slightly tinged with drab or stone colour. The surface is most commonly rough and uneven, and often covered with small, shining crystals.

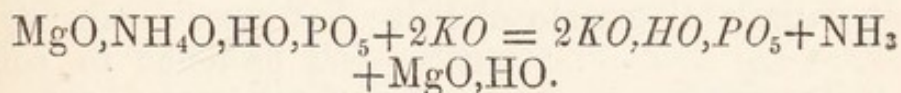
387. The triple phosphate calculus, when heated before the blow-pipe, chars, and gives off the smell of ammonia; swells up, gradually becomes grey as the carbonaceous matter is consumed, and ultimately fuses.

388. It is almost insoluble in water, but if boiled, a small quantity will be found to dissolve.



389. It dissolves readily in dilute hydrochloric and most other acids, and is again thrown down in the form of a crystalline precipitate, when the solution is neutralized with ammonia. If the precipitate thus obtained be examined under the microscope, it will be found to consist of well-defined crystals, which, if the solution has been supersaturated with the ammonia, are stellate (Fig. 10, page 20); but if merely neutralized, they are prismatic (Fig. 8, page 20) (44).

390. When heated with a solution of potash, it is decomposed, the potash combining with the phosphoric acid, and setting free the ammonia and the magnesia. The former volatilizes, and may be detected by the smell, while the magnesia is precipitated (49).



#### SECTION VI.

*Fusible Calculus, which is a mixture of Phosphate of Lime ( $3\text{CaO}, \text{PO}_5$ ), and the Triple Phosphate ( $\text{MgO}, \text{NH}_4\text{O}, \text{HO}, \text{PO}_5$ ).*

391. The fusible matter of which this form of calculus is composed, is, next to uric acid, the most common of the ingredients of calculi. It sometimes constitutes the entire mass of the calculus; is also frequently found alternating with other ingredients; and very commonly forms the outer crust of calculi composed of uric acid and other matters. Fusible calculi are generally oval or irregular in form (Fig. 59); white, soft, and friable, resembling chalk; though occasionally they are compact and hard.



Fig. 59. Fusible Calculus.

392. This calculus is chiefly characterised by the readiness with which it fuses before the blow-pipe, without being consumed; in which respect it differs from all other kinds of calculus. During the ignition, the ammonia and water are expelled, leaving a mixture of the phosphates of lime and magnesia.

393. If a portion of the calculus be dissolved in dilute hydrochloric acid, and ammonia added in slight excess, the



mixed phosphates are precipitated and may be recognised under the microscope (43).

394. If the precipitate be redissolved in acetic acid, and the solution mixed with oxalate of ammonia, the lime will be separated as oxalate, and if this be filtered off (after boiling), the phosphate of magnesia and ammonia may be obtained as a crystalline precipitate by adding an excess of ammonia.

#### SECTION VII.

##### *Oxalate of lime Calculus* ( $\text{CaO}, \text{C}_2\text{O}_3$ ).

395. Calculi are not unfrequently met with, composed almost entirely of oxalate of lime; but more commonly the nucleus will be found to consist of uric acid or urate of lime. Oxalate of lime calculi are usually very dark in colour, either brown or dark olive, or a kind of dirty purple. Their surface is much more irregular and rugged than that of other descriptions of calculi; and when sawn asunder, they exhibit an irregular and angular structure, as shown in Fig. 60.

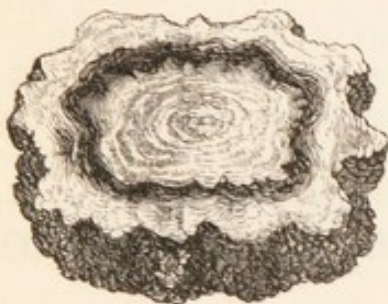


Fig. 60.  
Oxalate of Lime Calculus.

From their resemblance to the fruit of the mulberry, this variety is commonly known as the *mulberry calculus*.

396. There is also another form in which oxalate of lime calculi are occasionally met with, commonly called *hemp-seed* calculi. These are small, round, or oval, and very smooth and polished on the exterior; they generally contain also a little urate of ammonia.

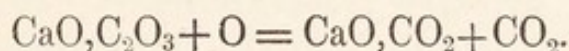
The general form and appearance of these oxalate of lime calculi are usually so peculiar and characteristic, that they may be, in most cases, easily recognised by simple inspection.

397. Powdered oxalate of lime dissolves without effervescence in dilute nitric and hydrochloric acids, and is again thrown down unchanged, in the form of a white precipitate, when the acid solution is neutralized with ammonia; the precipitate is insoluble in acetic acid. Occasionally a little carbonate of lime is found mixed with the oxalate, in which case slight effervescence will, of course, take place on the addition of the acid.



398. Oxalate of lime is insoluble in acetic and oxalic acids.

399. When heated, it blackens, and gives off a disagreeable smell, resembling that of burnt feathers. If the heat be continued a short time, the residue becomes white, and then consists of carbonate of lime, into which the oxalate is converted; carbonic acid being also, with other gaseous matters, at the same time given off.



400. Treat the residue formed in the last experiment, with dilute hydrochloric acid: it readily dissolves, with effervescence, showing that it has been changed into the carbonate.

401. The solution of chloride of calcium (*CaCl*) thus formed, may be neutralized with ammonia, and tested for lime with oxalate of ammonia, which will throw down the oxalate of lime ( $\text{CaO}, \text{C}_2\text{O}_3 + 2\text{Aq}$ ), in the form of a white precipitate (171).

402. If the oxalate of lime be kept intensely heated for some little time, the carbonate which is at first formed is reduced to the state of caustic lime ( $\text{CaO}$ ); which may be proved by placing the residue, when cold, on a piece of moistened turmeric paper, the yellow colour of which will be turned to brown.

#### SECTION VIII.

##### *Urate (or Lithate) of Lime* ( $\text{CaO}, \text{HO}, \text{C}_{10}\text{H}_2\text{N}_4\text{O}_4$ ).

403. This substance, though never found composing entire calculi, is not unfrequently present in small quantities in concretions which consist chiefly of uric acid, oxalate of lime, or other matters.

404. Urate of lime is nearly insoluble in cold water, but dissolves in hot, though somewhat less readily than urate of ammonia (375). The hot aqueous solution deposits it again on cooling, generally in the form of minute needle-shaped crystals.

405. Like the other urates, it is decomposed by hydrochloric acid. If the acid be added to a hot aqueous solution of the salt, a crystalline precipitate of uric acid is thrown down (377, 373), and chloride of calcium remains in solution.

406. When tested with nitric acid and ammonia, in the manner described in paragraph 371, urate of lime behaves



like uric acid and the other urates, yielding the rich purple colour of murexide.

407. As this is the only salt of lime found in calculi which is soluble in hot water, it may be supposed to be present when, after boiling a little of the powdered calculus in water, the *hot* aqueous solution gives a white precipitate of oxalate of lime ( $\text{CaO}, \text{C}_2\text{O}_3 + 2\text{Aq}$ ), when tested with oxalate of ammonia.

#### SECTION IX.

##### *Cystine* ( $\text{C}_6\text{H}_6\text{NO}_4\text{S}_2$ ).

408. Calculi of cystine are of rather rare occurrence. They are usually more or less crystalline in structure, not deposited in laminæ, soft, and of a pale brownish-yellow or greenish tint. Small calculi composed almost exclusively of this substance have been occasionally found in the dog.

409. The chemical characters of cystine, and the methods of distinguishing it by tests, will be found described in the chapters on urine (172, 269, &c.).

410. The following directions will facilitate the identification of the several varieties of urinary calculi.

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## CHAPTER II.

### QUALITATIVE EXAMINATION OF URINARY CALCULI, THE COMPOSITION OF WHICH IS UNKNOWN.

411. When a calculus has to be examined with a view to ascertaining the nature of its ingredients, a very few simple experiments, conducted on some such plan as the following, will generally furnish the required information. The calculus should first be sawn through, and the loose dust gently brushed away. If the several laminæ of which the mass is composed appear to be homogeneous, and to consist of the same kind of matter, a small fragment may be taken from any part of it for examination (412); but if, as is more frequently the case, there appear to be two or more different kinds of matter contained in the several layers (367), fragments of



each of them should be carefully detached from the mass, and examined separately in the following manner.

412. Place a small fragment on platinum foil, and heat it to redness before the blow-pipe, until the blackness of the charred animal matter disappears. Observe whether—

(a) IT BURNS AWAY, LEAVING ONLY A MINUTE TRACE OF ASH (413); or

(b) IT PROVES INCOMBUSTIBLE, WITHOUT MATERIALLY LESSENING IN BULK (414); or

(c) IT IS PARTIALLY CONSUMED, leaving, however, a considerable residue of incombustible matter (415).

413. IF IT BURNS AWAY, leaving only a minute trace of incombustible ash, it is probably either uric acid, urate of ammonia, or cystine; or possibly a mixture of two or more of them. See 416—419.

414. IF IT IS INCOMBUSTIBLE, not materially lessening in bulk during the ignition, it is probably either phosphate of lime, triple phosphate, fusible matter (391), oxalate of lime (converted into carbonate by the heat), urate of lime (also converted into carbonate); or, perhaps, two or more of those substances mixed together. See 420—425.

415. IF THE FRAGMENT IS PARTIALLY CONSUMED, it will probably be found to consist of a mixture of one or more of the combustible substances mentioned in paragraph 413, with some of those enumerated in paragraph 414. See 426—428.

#### *Examination of Combustible Calculi (413).*

416. If the calculus (in powder) is found to be sparingly SOLUBLE IN WARM WATER; SOLUBLE IN DILUTE SOLUTION OF POTASH, without the evolution of ammonia; and to form, when tested with nitric acid and ammonia, a PURPLE RESIDUE; it is probably URIC ACID (370, 372, 371). (Confirm 373.)

417. If it is found to be SOLUBLE IN HOT WATER; SOLUBLE IN DILUTE SOLUTION OF POTASH, with the evolution of ammoniacal vapours; and to yield, with nitric acid and ammonia, a PURPLE RESIDUE; it is probably URATE OF AMMONIA (375, 377, 376). (Confirm 373.)

418. If it is found to be INSOLUBLE IN WARM WATER; readily SOLUBLE IN AMMONIA; the ammoniacal solution yielding, on slow evaporation, HEXAGONAL CRYSTALLINE PLATES, it is probably CYSTINE (174, 173). Confirm 174, 271, 273.)

419. If it is suspected that more than one of the above



substances are present, a little of the powder may be boiled with water, and if any portion remains undissolved, the mixture filtered *while hot*.

(a) If the clear filtered liquid DEPOSITS, ON COOLING, AN AMORPHOUS PRECIPITATE, URATE OF AMMONIA is probably present (375). (Confirm 417.)

(b) If the insoluble portion gives a PURPLE COLOUR when tested with nitric acid and ammonia, URIC ACID is probably present (371). (Confirm 416.)

(c) If the insoluble portion is wholly or partially SOLUBLE IN AMMONIA: the ammoniacal solution, yielding, on evaporation, HEXAGONAL PLATES, CYSTINE is probably present (173).\*

*Examination of Incombustible Calculi (414).*

420. If the matter of the calculus is INFUSIBLE BEFORE THE BLOW-PIPE; SOLUBLE IN DILUTE HYDROCHLORIC ACID; the acid solution of the substance after ignition, yielding, when neutralized with ammonia, an AMORPHOUS PRECIPITATE, it is probably PHOSPHATE OF LIME (379, 381, 382). (Confirm 383, 384.)

421. If it is TOLERABLY FUSIBLE before the blow-pipe; SOLUBLE IN DILUTE HYDROCHLORIC ACID; the acid solution giving, when neutralized with ammonia, a CRYSTALLINE PRECIPITATE, it is probably TRIPLE PHOSPHATE (387, 389). (Confirm 390.)

422. If it is readily FUSIBLE before the blow-pipe; SOLUBLE IN DILUTE HYDROCHLORIC ACID; the acid solution yielding, when supersaturated WITH AMMONIA, A PRECIPITATE, which, when examined under the microscope, is found to contain both AMORPHOUS PARTICLES and also CRYSTALLINE STELLÆ, it is probably composed of the MIXED OR FUSIBLE PHOSPHATES (392, 394).

423. If the substance, before ignition, is SOLUBLE WITHOUT EFFERVESCENCE in dilute hydrochloric acid; the acid solution yielding a WHITE PRECIPITATE WHEN NEUTRALIZED WITH AMMONIA; and after gentle ignition, is SOLUBLE WITH EFFERVESCENCE in the dilute acid; the acid solution, moderately diluted, now yielding NO PRECIPITATE when neutralized with

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\* A very rare combustible calculus, discovered by Heller, is called urostealith. It is soft and elastic when fresh, but becomes hard after drying. When heated it evolves a resinous odour, somewhat similar to that of benzoin. It is readily soluble in ether, and sparingly in alcohol.



ammonia, it is probably OXALATE OF LIME (397, 400, 401). (Confirm 398, 402.)

424. If the hot aqueous solution, formed by boiling a little of the powdered calculus with water, gives a WHITE PRECIPITATE WITH OXALATE OF AMMONIA, the presence of URATE OF LIME is indicated (407). (Confirm 404, 405, 406.)

425. If it is suspected that more than one of the above substances are present in the portion of calculus under examination, it may be gently ignited, and then treated with dilute hydrochloric acid.

(a) IF EFFERVESCENCE ENSUES (the calculus before ignition, not causing effervescence with the acid), oxalate (or possibly urate (c),) of lime is present (397, 400).

(b) Supersaturate the acid solution with ammonia; and if any PRECIPITATE IS PRODUCED, examine it under the microscope for PHOSPHATE OF LIME and TRIPLE PHOSPHATE (382, 389).

(c) Boil a little of the powdered calculus with water; and test the *hot* aqueous solution thus obtained, with oxalate of ammonia. If a WHITE PRECIPITATE is produced, URATE OF LIME is probably present (407). (Confirm 405.)

#### *Examination of Partially Combustible Calculi (415).*

426. When the calculus, or any portion of it, is found to be partially consumed when ignited, it is probably a mixture of one or more of the combustible matters enumerated in paragraph 413, associated with one or more of the incombustible ingredients mentioned in paragraph 414.

427. A portion of the calculus, before ignition, may first be examined for the organic or combustible ingredients, in the manner described in paragraph 419, *a*, *b*, & *c*.

428. Another portion of the calculus may then be gently ignited on platinum foil, and the residue examined for the inorganic matters, according to the directions given in paragraph 425, *a*, *b*, & *c*.

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## CHAPTER III.

### BILIARY CALCULI OR GALL-STONES.

429. Biliary calculi are usually of a pale yellow or brownish colour; soft, soapy to the touch, and easily crushed into small



fragments by pressure; and the texture of the mass is in most cases decidedly crystalline. The size most commonly met with



Fig. 61.  
Biliary Calculi.

is about that of a pea; but they are frequently found much smaller, and occasionally almost as large as a pigeon's egg. The form is generally irregular and somewhat angular, as shown in Fig. 61.

430. They usually contain from fifty to eighty per cent. of cholesterin ( $C_{52}H_{104}O_2$ ), the rest of the concretion being made up of biliary resin and colouring matters, mucus, and traces of other animal matters,\* with a small quantity of inorganic salts. The per-centage composition of three specimens analysed by Brande was as follows:

	I.	II.	III.
Cholesterin . . .	81.25	69.76	81.77
Biliary resin . . .	3.12	5.66	3.83
Bile-pigment . . .	9.38	11.38	7.57
Albumen and salts extractable by water . . .	—	—	3.83
Biliary mucus . . .	6.25	13.20	—

431. Heat a small fragment of gall-stone on platinum foil; it will fuse and burn with a bright but smoky flame, leaving a small fixed residue, consisting of inorganic salts.

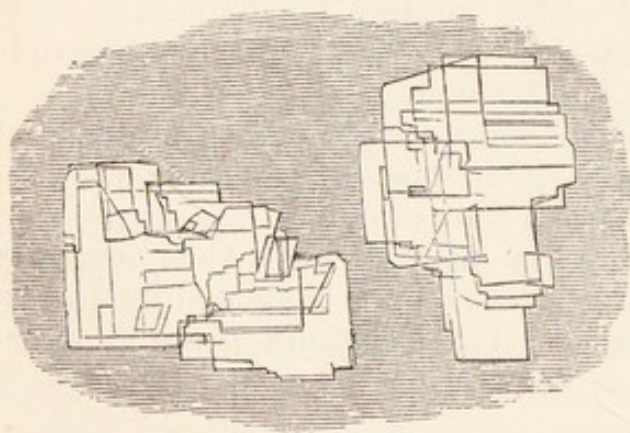


Fig. 62. Cholesterin.

432. When coarsely powdered, it dissolves readily in boiling alcohol; and on cooling, the cholesterin crystallizes out in the form of fine scaly crystals. (Fig. 62) while the biliary resinous and colouring matters remain in solution, giving the liquid a yellowish tinge.

433. It is insoluble in dilute nitric and hydrochloric acids. It is insoluble also in a solution of potash; thus differing from other fatty and oily substances, which cholesterin resembles in many respects.

\* Among which stearate and palmitate of lime have been noticed.



Cholesterin is found in large quantity in the fluid in ovarian dropsy, and in hydrocele.

434. For a more complete analysis of biliary calculi, Dr. Thudichum\* recommends the following process: The powdered calculus is digested with hot benzole, which dissolves the cholesterin. The residue, having been washed with alcohol, and dried, is treated with ether and a little nitric acid, by which the fatty acids are removed. On washing the residue on the filter with water, phosphates and nitrates of lime and magnesia are extracted (with occasionally a little copper). The final residue consists of biliary colouring matter, and a small quantity of earthy salts.

## CHAPTER IV.

### GOUTY CONCRETIONS.

435. These earthy concretions, which form in the joints of gouty persons, are usually white, or nearly so, soft and friable, closely resembling chalk in appearance, and hence commonly known as *chalk stones*. They seem to vary a good deal in composition; but in the great majority of those which have been analysed, acid urate of soda ( $\text{NaO}, \text{HO}, \text{C}_{10}\text{H}_2\text{N}_4\text{O}_4$ ) appears to form the principal and most characteristic ingredient. They contain also a considerable quantity of chloride of sodium and dried cellular tissue; with occasionally urate of lime ( $\text{CaO}, \text{HO}, \text{C}_{10}\text{H}_2\text{N}_4\text{O}_4$ ), phosphate of lime ( $3\text{CaO}, \text{PO}_5$ ), and chloride of potassium. The presence of a large quantity of uric acid may be shown by the formation of the purple-coloured murexide, when a little of the concretion, in powder, is treated with nitric acid and ammonia, in the manner described in paragraph 371.

#### *Qualitative Examination of Gouty Concretions.*

436. Reduce the concretion intended for analysis to tolerably fine powder, and digest it in cold water to dissolve out the chlorides of sodium and potassium. Filter the solution

\* 'Chem. Soc. Quar. Journ.,' July, 1861.



from the insoluble portion, which must be reserved for subsequent examination (440).

437. Test a few drops of the aqueous solution thus formed with nitrate of silver. A white curdy precipitate, which is readily soluble in ammonia, but insoluble in nitric acid, will show the presence of CHLORINE (chloride of potassium or sodium) (41, *a*).

438. Mix the rest of the aqueous solution with bichloride of platinum; evaporate the mixture to dryness, or nearly so, on a water-bath; and observe the yellow, needle-shaped crystals of the double chloride of sodium and platinum ( $\text{NaCl}, \text{PtCl}_2$ ), showing the presence of SODIUM (chloride of sodium).

439. Add a little alcohol to the evaporated residue, and observe whether any small, yellow, sandy-looking crystals remain undissolved, indicating the presence of POTASSIUM (41, *e*).

440. The portion which proved insoluble in cold water (436), may now be treated with hot water, and gently boiled with successive small quantities of the liquid as long as anything appears to dissolve. The urate of soda is thus slowly dissolved, together with any urate of lime that may be present (97, 404). The matter which proves insoluble in the hot water is to be retained for subsequent examination (444).

441. Hydrochloric acid is now added in slight excess to the hot aqueous solution, and the mixture set aside until it cools, in order to allow the uric acid, which will have been displaced from the soda and lime by the hydrochloric acid (405), to separate completely from the solution. The uric acid is thus precipitated; leaving in solution chloride of sodium, and also, in case any urate of lime was present in the concretion, a little chloride of calcium.

442. The mixture thus obtained is filtered. The URIC ACID may be examined with the microscope and with other tests; (373, 371); and a little of the aqueous solution may be neutralized with ammonia, and tested for LIME with oxalate of ammonia (171).

443. The rest of the aqueous solution may be evaporated at a gentle heat with bichloride of platinum; when the yellow needles of the double chloride of sodium and platinum will prove the presence of a large quantity of SODA derived from the urate (435).

444. The remaining portion of the concretion, which resisted the action of the hot water (440) may now be examined. It will probably be found to consist chiefly of dried cellular



matter, with perhaps a little phosphate of lime (435). The animal matter may be burnt away, by keeping it at a red heat until the blackness disappears; after which the incombustible residue may be examined in the manner described in paragraph 425, and will probably be found to consist of phosphate of lime.

445. The following is an analysis by T. J. Herapath of some concretions taken from the joints of the fingers of a man suffering from gout :

Fat . . . . .	1.123
Chloride of sodium . . . . .	} traces
Phosphate of soda . . . . .	
Extractive matter . . . . .	
Albumen . . . . .	
Urate of soda, with some urate of potash	43.973
Urate of lime . . . . .	14.769
Phosphate of lime . . . . .	34.141
Perphosphate of iron . . . . .	traces
Water and loss . . . . .	5.994
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## CHAPTER V.

### SOLID EXCREMENTS.

445a. The separation of the proximate principles contained in the solid excrements is effected by Dr. Marcet, by the following process :

The fæces are exhausted by boiling alcohol, and rapidly strained through a cloth. The alcoholic solution, on standing for a short time, yields a deposit which is partly dissolved by boiling alcohol; the insoluble portion of this deposit is boiled with potash, which dissolves it almost entirely, and the alkaline solution, neutralized with hydrochloric acid, gives a deposit of *margaric acid*, whilst the acid filtrate, neutralized by ammonia, yields a precipitate of *phosphate of lime*.

The alcoholic solution, after longer standing, deposits some *margarate of magnesia*, and if exposed to cold for some hours, it gives crystals of *excretine*.



If the solution obtained by boiling the first deposit with alcohol be evaporated to dryness, the residue extracted with ether, and the ethereal solution heated with alcohol and lime-water, a precipitate is formed which, when treated with hydrochloric acid and ether, yields, on evaporating the ethereal solution, an olive-coloured substance named *excretolic acid*.

If the alcoholic liquid containing the excretine (or from which the excretine has been separated by cooling) be mixed with lime, a yellowish-brown deposit is formed, which yields excretine to boiling ether. If the portion left undissolved by ether be treated with alcohol and hydrochloric acid, it gives a port-wine-coloured solution, which deposits margaric acid on standing. If water be then added, and the solution concentrated by evaporation, a brown substance separates, which may be purified by solution in ether and washing with water. It then much resembles the colouring matter of blood and that extracted by Dr. Harley from urine (36).

*Excretine* ( $C_{78}H_{78}O_2S$ ). This new proximate principle crystallises in four-sided prisms, which are insoluble in water, and sparingly soluble in cold alcohol, but dissolve readily in ether. Its solution has a feeble alkaline reaction. It fuses at a little below  $212^{\circ}$  and is not dissolved by boiling with solution of potash.

*Excretolic acid*, the composition of which has not yet been determined, is a very fusible olive-coloured body, which is insoluble in water and in boiling potash, dissolves sparingly in cold alcohol, but readily on heating, and is very soluble in ether. Its solutions have a marked acid reaction.

As far as they have yet been examined, healthy human excrements contain—

Excretine.\*

Excretolic acid.

Peculiar red colouring matter.

Margarates of lime and magnesia.

Butyric acid.

Taurine.

Phosphate of lime.

Phosphate of magnesia and ammonia.

Phosphate of potash.

Insoluble and undigested matters derived from the food.

\* Dr. Marcet estimates the average amount of excretine in each evacuation at about 2.8 grs. In the fæces of an infant cholesterine was found, but no excretine. The fæces of a man with a diseased pancreas contained a large proportion of bisulphate of soda.



## PART III.

### BLOOD.

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#### CHAPTER I.

#### HEALTHY BLOOD.

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##### SECTION I.

##### *General Characters of Blood.*

446. THE general appearance of blood, as it flows from the vessels through which it circulates in the living body, is familiar to every one, as an opaque, slightly viscous fluid, of a more or less brilliant red colour; that from the arteries being brighter and more scarlet than that from the veins. It has, while warm, a faint though characteristic odour, differing in the blood of different animals, and a saline and disagreeable taste. The specific gravity of healthy blood appears to vary from 1050 to 1058, the average being about 1055. It is always alkaline\* to test-paper, from the presence of an alkaline carbonate.

447. While circulating in the vessels, blood consists of a nearly colourless and transparent liquid, in which float myriads of minute vesicular bodies or corpuscles, of which by far the greater number are of a bright red colour; and these, being so small as to be individually quite invisible without the aid of a tolerably good microscope, give the blood, when seen with the naked eye, the appearance of being a homogeneous red fluid (451). A few of the corpuscles are colourless, and differ also in other respects from the red ones (464). The fluid portion of the blood, in which the corpuscles float, is usually called the *liquor sanguinis*.

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\* In certain morbid conditions an acid reaction has been observed in the blood, due, it is said, to the presence of free lactic acid.



448. The most remarkable peculiarity presented by the blood is the spontaneous coagulation which it begins to undergo almost immediately after being drawn, gradually separating into a more or less firm and solid red coagulum or *clot*, consisting of coagulated fibrin mixed with the corpuscles, and a pale yellowish, transparent, watery liquid, called the *serum*, holding in solution all the other solid matters of the blood. The nature and cause of this phenomenon will be more fully explained further on (473). The specific gravity of the serum is lower than that of the entire blood, being about 1029.

449. The chemical composition of the blood is highly complex; and though the nature of the principal ingredients is now tolerably well understood, our knowledge of the more obscure parts of its history is still very imperfect. The following substances appear to enter into its composition (Simon), and probably further researches will reveal the presence of other compounds, and, perhaps, also prove the non-existence of some of those now included in the list.

	Water,
Protein compounds	{ Albumen,
	{ Fibrin,
	{ Globulin,
Colouring matters	{ Hæmatin,
	{ Hæmaphæin,
Extractive matters	{ Alcohol extractive (containing traces of urea),
	{ Water extractive,
	{ Cholesterin,
	{ Serolin,
Fatty matters .....	{ Margaric acid,
	{ Oleic acid,
	{ Red and white solid fats, containing phosphorus,
	{ Oxide of iron,
	{ Albuminate of soda,
	{ Phosphates of lime, magnesia, and soda,
	{ Sulphates of potash and soda,
Saline matters .....	{ Carbonates of lime, magnesia, and soda,
	{ Chlorides of sodium and potassium,
	{ Lactate, urate, and probably hippurate, of soda,
	{ Oleate and margarate of soda,



Gases\* ..... { Oxygen,  
Nitrogen,  
Carbonic acid,  
Sulphur,  
Phosphorus.

450. It will, however, be more convenient for our present purpose, to consider the constituents of the blood as arranged in the following manner, the more important substances only being placed separately, and the others being, for the sake of simplicity, grouped together :

Water,  
Red and white corpuscles,  
Albumen,  
Fibrin,  
Alcohol extractive,  
Water extractive,  
Oily fats,  
Crystalline or solid fats,  
Fixed saline matters.

A short description of each of these substances and groups, will assist in rendering the subsequent analytical operations, both qualitative and quantitative, more simple and intelligible to the student.

## SECTION II.

### *Blood-Corpuscles.*

451. If freshly-drawn blood, previous to coagulation, be examined under the microscope, it will be found to consist of a transparent and nearly colourless fluid, in which float innumerable minute, circular, disk-shaped bodies or corpuscles, of which by far the greater number appear of a pale yellowish colour, though they are in reality red; the paleness of the colour being caused by the red rays from each of the corpuscles being spread over so large a surface. It is to these corpuscles that the red colour and opacity of the blood are due; the *liquor sanguinis* or fluid portion of the blood, in which they float, being nearly colourless and perfectly transparent.

452. These minute bodies, which, when the blood is first

\* These gases appear to be contained, at least chiefly, in the blood-corpuscles; the serum has been shown to have very little power of absorbing gases.



drawn, float freely in the *liquor sanguinis*, occasionally adhere together, forming little aggregations resembling strings of

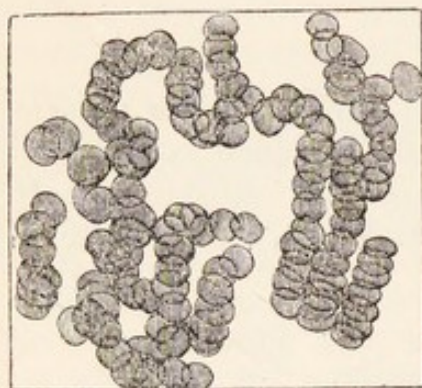


Fig. 63. Blood-corpuscles, magnified 400 diameters.

beads or rolls of coin (Fig. 63); this arrangement, however, is not always permanent, and the corpuscles gradually become again disunited and scattered. The tendency to aggregate together is usually greater during the inflammatory state, frequently causing the red corpuscles to collect in irregularly shaped masses, which sink more rapidly than when they are detached from each other.

This is one of the causes which tend to produce what is known as the *buffy coat*, which was formerly supposed to be always indicative of inflammation, but which has since been found to be formed almost whenever the fibrin, from whatever cause, coagulates more slowly, or the corpuscles subside more rapidly, than in healthy blood (454, 473).

453. The red corpuscles of human blood have an average diameter of about  $\frac{1}{3000}$ th of



Fig. 64. Blood-corpuscles, magnified 400 diameters.

an inch. They are nearly circular, flattened disks, each being slightly depressed and concave in the centre; their thickness is usually about one fourth or one fifth of their diameter (Fig. 64).\*

454. When, owing to the solidification of the fibrin, the blood coagulates (473), the corpuscles gradually be-

come entangled in the network of the solidifying clot, which is, in consequence, of a bright red colour; while the serum, or defibrinated *liquor sanguinis*, is left nearly colourless as the clot subsides. In consequence of the corpuscles being slightly heavier than the liquid in which they float, they begin very slowly to subside almost immediately after the blood is drawn; so that the lower portion of the clot usually contains a larger propor-

\* For further particulars relative to the structure of the blood-corpuscles, see Todd and Bowman's 'Physiological Anatomy and Physiology of Man.'



tion of them, and has consequently a deeper colour than the upper. This is the case to a remarkable extent in certain morbid conditions of the blood, which will be noticed further on (589).

455. The red corpuscles appear to consist of delicate membranous vesicles, filled with the red fluid to which they owe their peculiar colour, which fluid is supposed to consist of a colouring matter containing a considerable quantity of iron, to which the name of hæmatin has been given, associated with a protein compound, in many respects analogous to albumen, and called globulin. The enclosing membrane, which is highly elastic, appears to be composed either of coagulated fibrin or albumen, or of some other modification of protein closely allied to them.

456. When placed in solutions of different densities, the phenomena of endosmosis and exosmosis presented by the corpuscles are very curious and interesting, and may be seen with great facility with the help of a tolerable microscope. As long as the fluid in which they float is of the same density as that which they contain—such, for instance, as the *liquor sanguinis*—the corpuscles experience little or no change of form. But if the external liquid is less dense than that contained in the corpuscles, the latter will become more or less distended and globular, owing to the lighter fluid, in obedience to the well-known laws of endosmosis, passing through the membranous vesicles into the interior more rapidly than the heavier fluid within can pass outwards. If, on the other hand, the external liquid be more dense than that contained within the corpuscles, the contrary effect will be produced, and the corpuscles will immediately begin to collapse, and assume a wrinkled appearance (Fig. 65). This change of form not unfrequently takes place spontaneously, while a drop of blood, placed between two surfaces of glass, is being examined under the microscope, especially near the edges, where, owing to evaporation, the liquid with which the corpuscles are in contact, gradually becomes more concentrated, and consequently more dense.

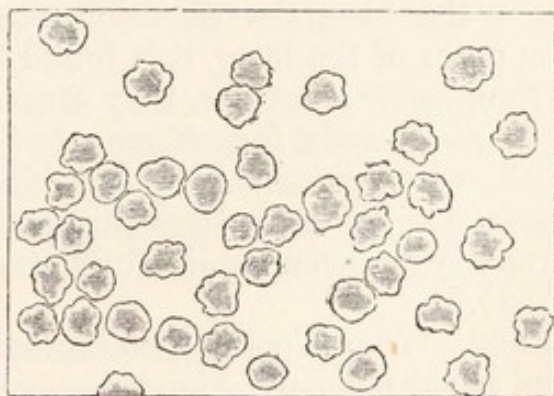


Fig. 65. Blood-corpuscles collapsed, magnified 400 diameters.



457. The liquor sanguinis, or fluid portion of the blood, as it exists in the living body, and before it undergoes coagulation, appears to possess the same density as the red fluid contained in the vesicles; so that, as long as it continues so, no change takes place in the form of the corpuscles. When, however, the fibrin, which was before dissolved in the liquor sanguinis, has coagulated, the resulting serum becomes less dense, in consequence of its holding in solution a smaller amount of solid matter (448). The effect of this upon the blood-corpuscles is to cause them, when in contact with the serum of coagulated blood, gradually to enlarge in size, in consequence of the increased rapidity with which the less dense serum enters through the membranous integument.

458. If the red corpuscles be brought in contact with water, the change is extremely rapid; they instantly swell to a much larger size, the vesicles becoming less and less distinct, until at length, unless the quantity of water is very small, they almost entirely disappear.

459. When, owing to the action of water, or some other liquid of comparatively low specific gravity, the corpuscles have become distended, they may, if the distension has not been allowed to go too far, be again brought back almost to their original size, and even be made to assume a wrinkled appearance, by bringing them in contact with a tolerably strong solution of sugar, or of certain salts, as chloride of sodium or chloride of ammonium.

460. The corpuscles readily dissolve in a solution of potash, ammonia, acetic acid, and some other fluids.

461. Although we are unable to separate the corpuscles from the blood by filtration, since they pass readily through the pores of the filter, it is found that when mixed with certain strong saline solutions, they are retained by it. A solution of sulphate of soda, for example, having a specific gravity of about 1.13, when mixed with the blood, effectually prevents the passage of the corpuscles through the filter. This remarkable property has been applied by Figuier to the purposes of analysis (582).\*

462. When blood is allowed to dry at common temperatures, and is subsequently moistened, even after the lapse of a considerable time, with some liquid having a specific gravity similar

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\* According to Dumas, oxygen should be passed through the liquid during filtration, and solution of sulphate of soda allowed to drop into it, in order to prevent the obstruction of the pores of the filter.



to that of the serum (448), the corpuscles are found to have retained their characteristic form and appearance, and may be readily distinguished under the microscope. This circumstance has been ingeniously applied for the purpose of solving a question which in some medico-legal inquiries is one of grave importance, viz., whether the stains found on clothing or elsewhere are or are not stains of blood.

463. For this purpose the stain is to be moistened, and gently rubbed with a little fresh white of egg, or some other fluid having a specific gravity of about 1030 to 1050. It is then scraped off, and a little of the mixture examined under the microscope with a tolerably high power, when, if the stain consisted of blood, the characteristic corpuscles will, in most cases, be distinctly visible.

It is, of course, desirable to obtain chemical as well as microscopical evidence of the presence of blood in the stain under examination. With this view, the stained material should be soaked in a little water for an hour or two, when, unless the stain be of long standing, a portion of the blood will be extracted, imparting a dingy reddish colour to the liquid, to which the following tests should then be applied :

(a) Boil a portion in a test-tube; a dirty red coagulum should be formed. On dissolving this in boiling potash, the solution should be green by transmitted and red by reflected, light.

(b) Chlorine-water should decolorise the liquid, and produce a white flocculent precipitate.

(c) Nitric acid should produce a brown coagulum of albumen and colouring matter. The albumen may also be tested for with chloride of mercury, and with acetic acid and ferrocyanide of potassium (137, 138).

(d) Evaporate the remainder of the solution to dryness, and heat the residue, observing whether the offensive odour of burnt blood is evolved. Incinerate the residue completely, and boil the ash with a few drops of hydrochloric acid; dilute the solution with water, and test for iron with ferrocyanide of potassium.

If the blood has not been extracted by soaking in water, the piece of stuff with the stain should be placed in a stout glass tube, closed at one end, about half an inch in diameter, and five or six inches long. About a drachm of distilled water should be poured upon it, and the tube drawn out and sealed before the blow-pipe, at about two inches from the open end.



The sealed tube is then heated to about 300° to 310° Fahr. for about an hour.\* At this temperature the fibrin as well as the albumen of the stain will dissolve in the water. When the tube has cooled, it may be opened with a file, and the liquid examined. It should have a yellow or reddish colour, and a feeble alkaline reaction to reddened litmus-paper. Of course, it would not be coagulated by heat, but nitric acid, chloride of mercury, and ferrocyanide of potassium (after acidifying with acetic acid) should yield precipitates.

The fabric from which the soluble matter has thus been extracted will generally retain some of the iron of the blood, and may be examined, for further confirmation, by the following tests:

(a) If it be boiled in water containing a little tannic acid or tincture of galls, it acquires a black or grey colour.

(b) The same piece may be boiled with a little dilute hydrochloric acid, the solution diluted, and tested with ferrocyanide of potassium for iron.

(c) Another piece may be boiled with hydrochloric acid, and the solution tested for iron with ammonia and hydrosulphate of ammonia.

It is always advisable to repeat the above experiments with an unstained portion of the same fabric, especially if the latter be made of a coloured material.

*Identification of blood-spots upon iron.*—Spots of blood are generally much more easily detached than those of ordinary rust, and may be recognised by the following tests:

(a) Digest in water at about 100° Fahr. A recent stain will be dissolved, and the solution may be tested by boiling, by nitric acid, and by chlorine (see above).

(b) If water does not dissolve the stain,† boil it with a dilute solution of potash, and test the solution with nitric acid and chlorine.

(c) Heat a portion of the suspected rust in a tube, and observe the odour.

(d) Heat another portion in a tube with a fragment of solid hydrate of potash or soda; notice if any ammonia is evolved during the fusion. All rust would evolve a little ammonia; but an abundant evolution of this substance would afford some

\* This may be effected either by suspending it in oil which is afterwards heated to that temperature, or by placing it in a hot-air bath.

† Rose ('Journ. Pharm.,' xxviii, 436) has proved by direct experiment that hydrated peroxide of iron (rust) forms an insoluble compound with the colouring matter of the blood.



evidence of the presence of blood. A part of the nitrogen contained in the blood would be converted into cyanide of potassium, which may be detected by dissolving the fused mass in water, adding a little solution of protosulphate and perchloride of iron, followed by an excess of acetic acid, which should leave a blue precipitate (Prussian blue).

464. *White corpuscles of the blood.*—In addition to the red corpuscles, there are always present in the blood a few colourless particles,\* somewhat larger than the coloured ones, and otherwise differing from them in general appearance and structure (Fig. 66). They are of irregular forms, sometimes spherical, slightly granular on the surface, and appear to be identical, or nearly so, with the peculiar corpuscles always present in the lymph and the chyle. When treated with acetic acid, the granular exterior becomes transparent, as in the corpuscles of pus, and one or more internal nuclei are rendered visible.

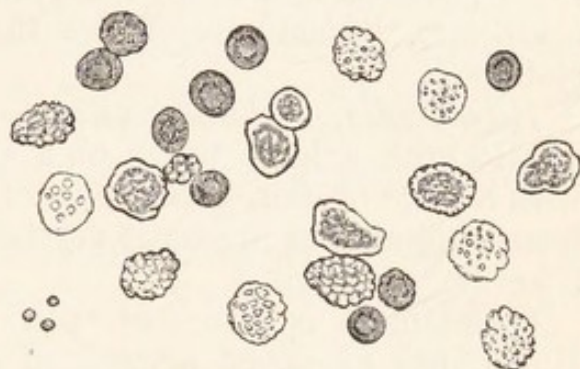


Fig. 66. White Corpuscles of the Blood, magnified 400 diameters.

465. The proportion of corpuscles present in healthy blood is usually about 130 parts in 1000 (573).†

465a. *Hæmatin.*—In order to separate the peculiar colouring matter of the blood-globules, freshly drawn blood, which has been defibrinated by stirring, is mixed with eight volumes of a saturated solution of sulphate of soda, and set aside, in order that the globules may subside. The deposit is collected upon a filter, washed with solution of sulphate of soda, boiled with alcohol containing a little sulphuric acid, and filtered while hot. The globulin is then removed from the solution by precipitation with carbonate of ammonia, the filtered solution evaporated to dryness on a water-bath, and all soluble matters removed from the residue by boiling water, alcohol, and ether. By treating the residue with ammoniacal alcohol, the hæmatin is dissolved, and may be further purified by evaporating the clear solution to dryness, and washing the residue with water.

\* These have a lower specific gravity than the red corpuscles, and, therefore, accumulate on the upper surface of the clot.

† According to Denis, they contain one part of solid matter and 1.8 of water.



The dark brown colouring matter thus obtained is remarkable for its containing a large proportion (6.6 per cent.) of iron, its composition being represented (according to Mulder) by the formula  $C_{44}H_{22}N_3O_6Fe$ . If it be digested with cold strong sulphuric acid, it forms a brown liquid, and if this be diluted, hydrogen is evolved, and a dark brown substance left, whilst sulphate of iron ( $FeO,SO_3$ ) is found in solution. Chlorine also removes the iron, but at the same time destroys the colour, producing a white coagulum. In its general chemical characters, hæmatin resembles the albuminous class of substances.

*Hæmatoidin, or blood-crystals.*—When a drop of blood is diluted with a little water on a slip of glass, lightly covered with a piece of thin glass, and left in the sunlight for some hours, minute red prismatic crystals of hæmatoidin will sometimes be perceived.

Hæmatoidin is also met with in old extravasations, and Robin\* has examined a mass of crystals of this substance, weighing about fifty grains, obtained from a hydatid cyst in the liver. An analysis of these crystals led to the formula  $C_{14}H_8NO_2 + HO$ , and Robin believes them to represent hæmatin which has lost all its iron, and acquired an atom of water. The substance obtained by treating hæmatin with sulphuric acid has almost exactly the same composition. Hæmatoidin is very sparingly soluble in water, and insoluble in alcohol; but it dissolves in ammonia, yielding a red solution.

Lehmann obtained a red crystalline substance from the blood of guinea-pigs, rats, or mice, by washing the clot with water, filtering the red liquid, and passing first a current of oxygen, and afterwards carbonic acid. After a time the crystals separate, and may be washed with a little water. This substance, which has been named hæmato-crystallin, dissolves sparingly in water, the solution being coagulated by heat, but not by chloride of mercury. In composition it somewhat resembles albumen, containing more nitrogen and less sulphur. Much obscurity still remains to be cleared up with respect to these crystalline bodies obtained from blood.

Dr. Carter ('Ed. Med. Journal,' August, 1859) obtained from serum a substance yielding indigo by decomposition, similar to that extracted from urine (page 15).

\* 'Compt. Rend.,' xli, 506.



## SECTION III.

*Albumen.*

466. This is one of the most important of the constituents of the blood, and, with the exception of the red corpuscles, is present in larger quantity than any of the other solid matters contained in it. It is held in solution in the serum, where it may readily be shown to exist by gently boiling in a tube a little of the clear, colourless fluid from which the coagulated clot of fibrin and corpuscles has subsided. As soon as the temperature reaches about  $170^{\circ}$ , the albumen begins to coagulate, and on being boiled for a short time, separates entirely in the insoluble form.

467. It may also be precipitated from its solution in the serum, by adding to the clear fluid a few drops of dilute nitric or hydrochloric acid (136, 141). Acetic acid fails to precipitate it; but if ferrocyanide of potassium be added to the acidified solution, a dense white precipitate is produced, even when the albuminous liquid is very dilute.

468. When gently warmed with strong hydrochloric acid, albumen dissolves, forming a purple-coloured solution, in which respect it resembles fibrin and casein.

469. When moistened with strong nitric acid, albumen becomes yellow, owing to the formation of xanthoproteic acid ( $2\text{HO}, \text{C}_{34}\text{H}_{24}\text{N}_4\text{O}_{12}$ ), which, together with oxalic acid ( $\text{HO}, \text{C}_2\text{O}_3$ ), ammonia ( $\text{NH}_3$ ), nitric oxide ( $\text{NO}_2$ ), and nitrogen, is always formed by the action of strong nitric acid on the so-called compounds of protein.

469a. Heated with a solution of nitrate of mercury ( $\text{HgO}, \text{NO}_5$ ), albumen becomes intensely red (141a).

470. It appears from the results of numerous analyses that the average amount of dry albumen present in healthy blood is rather more than 70 parts in 1000 (573).

471. The composition of albumen is usually expressed by Mulder's formula ( $\text{C}_{400}\text{H}_{310}\text{N}_{50}\text{O}_{120}\text{S}_2\text{P}$ ); but considerable uncertainty still hangs over the real nature of this class of bodies.\* It is even doubtful whether any phosphorus exists in albumen, except in the state of phosphoric acid, as a constituent of the ash. By some chemists albumen is regarded as containing

\* The per-centage composition of the three so-called *protein compounds*, albumen, fibrin, and casein, is as follows:



$C_{144}H_{110}N_{18}S_2O_{42}$ ; and since the serum of blood always contains nearly 2 per cent. of soda, they consider it to consist of an albuminate of soda, in which one atom of the above albumen is combined with each atom of soda. As the more important peculiarities of albumen have been already noticed in the chapter on morbid urine (133, 235, &c.), they need not be again described.

## SECTION IV.

*Fibrin.*

472. This substance, of which muscular fibre is chiefly composed, is closely allied in chemical composition and general properties to albumen; and it is, indeed, not improbable that both are, in their chemical relations, merely modifications of the same compound, which, from the circumstance of its being apparently the basis, not only of albumen and fibrin, but also of casein (625) and some other analogous substances, has been called *protein*, from  $\pi\rho\omega\tau\epsilon\acute{\upsilon}\omega$ , *I am first*.\*

473. While circulating in the vessels, the fibrin of the blood is held in a state of solution in the *liquor sanguinis*; but no sooner is the blood removed from the system, than it begins to separate in a solid state, after which it becomes quite insoluble in water. This solidification of the fibrin is the cause of the well-known phenomenon of coagulation,

	Albumen.	Fibrin.	Casein.
Carbon.....	55.46	54.45	54.66
Hydrogen ...	7.20	7.07	7.15
Nitrogen.....	16.48	17.21	15.72
Oxygen .....	18.27	19.35	21.55
Sulphur .....	2.16	1.59	.92
Phosphorus...	.43	.33	
	<hr/> 100.00 <hr/>	<hr/> 100.00 <hr/>	<hr/> 100.00 <hr/>

The mineral constituents, such as phosphate of lime, from which these substances can never be completely separated, have been excluded from this calculation.

A peculiar modification of albumen (*albuminose* or *peptone*) has been observed in the liver and in blood, differing from normal albumen in not coagulating when heated, and from casein in not being precipitated by acids.

\* The existence of protein as an independent proximate principle appears very doubtful. Mulder represented fibrine as a compound of ten atoms of protein ( $C_{40}H_{31}N_5O_{12}$ ) with one atom of sulphur and one of phosphorus, but the presence of the latter, except in the ash of fibrin, is disputed. The fibrin of muscle does not appear to be identical with that of blood, the latter containing a smaller proportion of sulphur.



which blood experiences almost immediately after it is drawn; and although the coagulum or clot contains the blood-corpuscles in addition to the fibrin, these have merely been entangled in the network of coagulating fibrin, and do not themselves play any active part in the process of coagulation.\*

474. The coagulation of blood may be retarded, and even altogether prevented, by the presence of certain salts and other substances. The alkalies, for example, and their carbonates and acetates, entirely prevent it; and tolerably strong solutions of sulphate of soda, nitrate of potash, nitrate of lime, chloride of ammonium, and some other salts, retard it for a considerable time. The latter salt, indeed, gradually dissolves fibrin, after it has been allowed to coagulate. Most of the dilute acids, also, cause blood to retain its fluidity, though it becomes, under their influence, more viscous and syrupy in its consistence.

475. Contact with certain animal membranes also appears to exercise a retarding influence on the coagulation of the blood. When infused into the cellular tissue, it has been known to continue uncoagulated for some weeks; and even in a tied artery, it remains some hours without coagulating.

476. It appears from the experiments of M. Denis, that if moist fibrin be digested in a solution of nitrate of potash containing a little soda, at a temperature of about 100° Fahr., it becomes gradually converted into a substance in almost every respect identical with albumen†; being soluble in water, and coagulable by heat. This change is said to be most readily produced when the fibrin employed in the experiment has been obtained from venous blood, by allowing it to coagulate spontaneously; while, if it is separated by agitation, or if the blood be arterial, it scarcely experiences any alteration in the saline solution. By drawing blood into a solution of sulphate of soda, so as to prevent the coagulation of the fibrin, filtering off the globules, and saturating the filtrate with chloride of sodium, Denis obtained a precipitate of fibrin, which dissolved in water, forming a solution which gave, after a short time, a transparent coagulum of fibrin.

477. Pure fibrin may be obtained without difficulty, by receiving the blood, as it flows from the body, in a clean

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\* According to Dr. Richardson, the coagulation of the fibrin is due to the escape of ammonia, by which it was previously held in solution.

† Fibrin suffers a similar change when heated with water to about 300° Fahr., in a sealed tube.



porcelain dish, and stirring it well for some little time with a glass rod; or the blood may be shaken with a few small fragments of lead, in a closed glass flask. The fibrin, as it coagulates, collects in loose fibrous masses round the rod or fragments of lead, coloured slightly red, owing to the imprisonment of a few corpuscles within the network of fibrin. These may be removed by tying the coagulum in a piece of fine muslin, and washing it under a stream of cold water until the mass becomes colourless. In this state, it still contains traces of fatty matter and inorganic salts, together with a considerable amount of water. To obtain the fibrin, therefore, in a state of perfect purity, the washed coagulum must be dried on a chloride of calcium bath at a temperature of about  $250^{\circ}$ , and the dry mass then reduced to fine powder in a mortar. The pounded fibrin may afterwards be washed successively with alcohol, ether, and dilute hydrochloric acid; and lastly, macerated with cold or lukewarm water, until all the soluble matter is removed; after which it may be dried as before at a temperature of about  $250^{\circ}$ .

478. If the blood from which we wish to extract the fibrin has already coagulated, the clot is first gently pressed between folds of bibulous paper, in order to squeeze out the greater part of the adhering serum, and then cut into thin shreds with a sharp knife. The finely-divided clot is then washed in a muslin bag under a gentle stream of cold water, until it becomes colourless, by which means the imprisoned corpuscles are washed out of the fibrous mass. The latter is then dried, and reduced to powder, and subsequently purified by washing and drying in the manner above described (477).

479. Fibrin thus prepared is a pale yellowish, horny-looking substance, hard, brittle, and, if all traces of fat have been removed, transparent. It is perfectly tasteless, and insoluble in water, alcohol, and ether; if kept for a short time in water, however, it gradually softens, swells up, and reassumes the appearance it had previous to desiccation. When digested with acetic and most of the other acids, fibrin becomes gelatinous, and is in that state soluble in water. The acid solution, when treated with ferrocyanide of potassium, gives a copious white precipitate, similar to that caused in albuminous solutions. Fibrin is also dissolved by digestion in a dilute solution of nitre, the liquid coagulating when heated. Like albumen, and the other modifications of protein, it forms, when gently warmed with strong hydrochloric acid, a purple-



coloured solution. With nitric acid, also, fibrin behaves like the other protein compounds, forming the yellow xanthoproteic acid (469). It also becomes red when heated with solution of nitrate of mercury (141a).

479a. The fibrin of muscle (sometimes called *syntonin*) differs somewhat in properties from that of blood. It does not dissolve in a dilute solution of nitre, but may be dissolved by digestion in very dilute hydrochloric acid (1 of acid to 1000 of water), whilst blood-fibrin swells up and becomes transparent, but does not dissolve.

480. When examined under the microscope, coagulated fibrin appears to consist of a rude network of amorphous threads, together with detached aggregations of irregular form, similar to albumen.

481. The average proportion of dry fibrin present in healthy blood appears to be rather more than two parts in a thousand (573).

## SECTION V.

### *Extractive Matters.*

482. Of the real chemical nature of the substances included under the name of extractive matters, little is yet definitely known, though they have frequently engaged the attention of chemists. It is probable, however, that further researches will, ere long, throw new light upon this at present obscure class of substances. They include all the undefined, uncrySTALLIZABLE organic matters which are soluble in water; or, in other words, the extractive matters of the blood may be said to include all the organic substances contained in it, with the exception of the corpuscles, albumen, fibrin, and fatty matters.

483. Extractive matters are usually divided into *alcohol extractive* and *water extractive*; the first including that portion which is soluble both in water and alcohol; and the latter, that which is soluble in water and insoluble in alcohol. They are of a brown or yellowish colour, and are characterised by their solutions giving brown precipitates with acetate of lead, but none with bichloride of mercury. A solution of the alcohol extractive is precipitated by an infusion of galls, which reagent causes little or no change in the water extractive.

484. Traces of urea are probably always present in the



blood, and would be contained in the alcohol extractive. The method of detecting it will be described further on (598). The minute traces of uric acid which appear to be usually present even in healthy blood, would be contained in the water extractive; the mode of detecting them is described in paragraph 604.\*

485. The amount of extractive matters present in healthy blood seems to vary from one to three parts in a thousand.

## SECTION VI.

### *Fatty Matters.*

486. Our knowledge of the fatty matters contained in the blood is at present far from being complete. They are usually divided into *oily fats* and *crystalline fats*; the first being soluble in cold alcohol, and the latter insoluble. The oily fats appear to consist chiefly of oleic ( $HO, C_{36}H_{33}O_3$ ) and margaric ( $HO, C_{34}H_{33}O_3$ ) acids; the crystalline fatty matter is probably a mixture of serolin with traces of cholesterin ( $C_{52}H_{44}O_2$ ), together with one or more solid fats containing phosphorus.†

487. To obtain these fatty matters, a quantity of blood is evaporated to dryness on a water-bath, and the dry residue, after being reduced to powder, is digested in hot ether, successive portions of which must be added as long as anything appears to be dissolved by it. The ethereal solution is then evaporated to dryness on a water-bath, and the residue, consisting of the mixed fats, treated with cold alcohol, which will dissolve out the oily fats, and leave the crystalline matters undissolved. The first may be obtained by evaporating the alcoholic solution on a water-bath; and the undissolved crystalline fats may be dissolved in boiling alcohol, from which they will almost entirely separate, as the liquid cools, in the form of small crystalline scales.

488. The quantity of fatty matters present in healthy blood appears to vary from 1·5 to 2·5 in 1000 parts (573).

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\* Sugar has been discovered in minute proportion in normal blood. Campbell has also indicated the presence of a little formic acid in the serum.

† The serum also contains minute proportions of one or more of the volatile fatty acids (*e.g.* butyric or caproic,) as shown by the odour which is developed on mixing the blood with sulphuric acid, and varies in different animals.



## SECTION VII.

*Fixed Saline Matters.*

489. The ash left after the incineration of the dry residue of evaporated blood appears to contain the following substances:—viz., the chlorides of sodium and potassium; the phosphates of lime, magnesia, and soda; the sulphates of potash and soda; and oxide of iron derived from the hæmatin (455). If the ash has been obtained by the incineration of the serum, traces of alkaline and earthy carbonates will probably be rendered apparent by the effervescence caused by the addition of an acid; but if the ash has been obtained by the incineration of the entire blood, no trace of carbonates will be observable on the addition of the acid. The cause of this appears to be, that some of the fatty matters present in the clot contain traces of phosphorus (486), which, during combustion, is converted into phosphoric acid ( $\text{PO}_5$ ); and the phosphoric acid thus formed decomposes the small quantity of carbonates derived from the serum, converting them into phosphates.

490. The saline matters of the blood may be conveniently divided into the *alkaline salts*, which readily dissolve in water, and the *earthy salts*, which require an acid for their solution. The alkaline portion of the ash consists of the chlorides of sodium and potassium; the sulphates of potash and soda; and phosphate, with possibly traces of carbonate (489) of soda. The earthy or insoluble portion contains the phosphates of lime and magnesia; oxide of iron derived from the red colouring matter; and possibly a little earthy carbonate (489). The presence of the bases and acids contained in these several salts may be shown by the following experiments.

491. Digest from twenty to thirty grains of the ash in warm water, in order to dissolve out the alkaline salts, and filter the solution from the insoluble portion. The aqueous solution thus obtained may be first tested, retaining the earthy residue for subsequent examination (499).

492. If the aqueous solution is at all dilute, it should first be concentrated by evaporation. To a little of the concentrated solution, add a slight excess of tartaric acid ( $2\text{HO}, \text{C}_8\text{H}_4\text{O}_{10}$ ), and agitate the mixture with a glass rod. A colourless crystalline precipitate of the bitartrate shows the presence of POTASH.



493. To another portion of the solution add a solution of bichloride of platinum ( $PtCl_2$ ), and allow the mixture to evaporate to dryness, either spontaneously or at a very gentle heat. Minute, yellow, granular crystals of the double chloride of platinum and potassium ( $KCl, PtCl_2$ ) will be found deposited, also showing the presence of POTASH. In addition to these will be seen long, yellow, needle-shaped crystals of the double chloride of platinum and sodium, proving the presence of SODA. If the bichloride of platinum has not been added in sufficient quantity to combine with the whole of the soda, a few detached cubical crystals of chloride of sodium will also be deposited, which may be proved to be such by their well-known taste.

494. The presence of soda may also be shown by adding to a little of the strong aqueous solution a few drops of antimoniate of potash ( $KO, SbO_5$ ), which will gradually cause a colourless crystalline precipitate of antimoniate of soda ( $NaO, SbO_5$ ).

495. To another portion of the aqueous solution of the ash, add a solution of chloride of barium, or nitrate of baryta, as long as it causes any precipitate. The sulphuric, phosphoric, and (if any (489)), carbonic acids are thus thrown down in combination with baryta. The mixture containing the precipitate thus produced is now strongly acidified with hydrochloric acid, and warmed. If effervescence occurs on the acid of the acid, CARBONIC ACID is probably present. The presence of SULPHURIC ACID is shown by a portion of the precipitate (sulphate of baryta) proving insoluble in the acid.

496. Filter the acid mixture formed in (495), and neutralize the filtered liquid with ammonia. The phosphate of baryta ( $2BaO, HO, PO_5$ ), which had been dissolved by the acid, is reprecipitated, indicating the presence of PHOSPHORIC ACID (498).

497. Acidify another portion of the aqueous solution of the ash with nitric acid; add a slight excess of nitrate of silver, and filter the liquid from the white precipitate occasioned by the silver salt. This precipitate may be proved to consist of chloride of silver (HYDROCHLORIC ACID), by being readily soluble in ammonia, and insoluble in nitric acid.

498. Accurately neutralize the acid solution formed in (497), with dilute ammonia; the pale yellow phosphate of silver ( $3AgO, PO_5$ ), which had been held in solution by the ex-



cess of acid, will now be precipitated, showing the presence of PHOSPHORIC ACID (496).\*

499. The earthy portion of the ash, which proved insoluble in water (491), may now be examined. It is to be dissolved in as small a quantity as possible of dilute hydrochloric acid, a gentle heat being applied if necessary. If effervescence occurs on the addition of the acid, CARBONIC ACID is present (489).

500. A little of the acid solution may now be nearly neutralized with dilute ammonia, which should not be added in sufficient quantity to cause any precipitate. The liquid is then tested with a drop or two of a solution of ferrocyanide of potassium, which will cause, either at once, or in the course of a few minutes, a blue colour, owing to the formation of the ferrocyanide of iron ( $\text{Fe}_4\text{FeCy}_3$ ), showing the presence of IRON.

501. The rest of the acid solution of the earthy portion of the ash may now be supersaturated with ammonia, which will throw down a white gelatinous precipitate of earthy phosphates. A little of this precipitate may be examined under the microscope, when it will be found to consist chiefly of amorphous particles of phosphate of lime ( $3\text{CaO},\text{PO}_5$ ), with a few crystals of the double phosphate of ammonia and magnesia ( $2\text{MgO},\text{NH}_4\text{O},\text{PO}_5+12\text{Aq}$ ). The precipitate thrown down by the ammonia may also be examined for LIME, MAGNESIA, and PHOSPHORIC ACID, by redissolving it in acetic acid, and digesting the solution in the manner described in paragraph 47.

502. The quantity of alkaline salts usually present in healthy blood, varies from about six to ten parts in 1000; and that of earthy salts from 0.5 to 1.5 in 1000 parts.

## CHAPTER II.

### QUANTITATIVE ANALYSIS OF BLOOD.

503. A complete quantitative analysis of the blood, including the separation from each other and estimation of *all* the ingredients, would be, even if our knowledge and resources were much less limited than they are, in the highest degree

\* The phosphoric acid may be detected with greater certainty by perchloride of iron, or the mixture of sulphate of magnesia, chloride of ammonium and ammonia (41).



complicated and difficult; while at present it may be said to be altogether impracticable. For most purposes, however, a comparatively incomplete analysis, embracing the determination of the more important ingredients, is all that is required; and in the majority of cases, a knowledge merely of the proportion of fibrin, the corpuscles, and the solids contained in the serum, is what the medical practitioner chiefly requires.

504. I will first describe the mode of conducting such an analysis, by which the amount of water, corpuscles, fibrin, and solids contained in the serum, may, with very little difficulty, be ascertained; and subsequently go through a somewhat more complete scheme, by which, in addition to the above substances, the more important constituents of the serum may also be individually estimated. (See sections 3 and 4.)

505. When the blood intended for analysis can be collected in the proper vessels as it flows from the body, the process is somewhat simpler than when it has been allowed to coagulate and the results are generally more accurate. As, however, this is frequently impracticable, I will also describe the method by which the analysis of coagulated blood may be effected.

#### SECTION I.

*Quantitative Analysis of Uncoagulated Blood, including the estimation of the water, corpuscles, fibrin, and the solid matter contained in the serum.*

506. Before proceeding to collect the blood as it flows from the body, for the purpose of analysis, the experimenter should provide himself with three vessels, the exact weight of each of which is to be carefully ascertained and noted. These vessels are:

1. A six- or eight-ounce bottle, provided with a stopper; this bottle should be perfectly clean and dry, and of known weight. Eight or ten small strips of thin sheet lead, about half an inch square, the weight of which should also be known, are put into the bottle, which will then be ready to receive the blood (507). This bottle is used for effecting the separation of the fibrin.
2. A small platinum or Berlin porcelain capsule, capable of holding from half an ounce to an ounce of water. This is used for estimating the proportion of water in the blood (508).



3. A rather tall, upright beaker, or cylindrical glass, capable of holding about six ounces of water.

507. The blood may now be collected. About five or six ounces of the fluid are first poured into the bottle containing the fragments of lead, which should then be tightly closed with the stopper, and kept gently agitated for about a quarter of an hour, in order to allow the whole of the fibrin to coagulate, and attach itself to the pieces of lead (477). This portion of blood we will call A (510).

508. Two or three drachms of blood are collected in the capsule, which is then again accurately weighed, and the weight of the empty capsule, previously ascertained (506), deducted from the gross weight, in order to determine the exact quantity of blood contained in it. It may then be placed on a water-bath, and evaporated to dryness. This portion we will call B (514).

509. The beaker, or cylindrical glass, is to be nearly filled with the freshly-drawn blood, covered with a glass plate, and set aside in a tolerably cool place for twenty-four hours; at the end of which time it will be found to be thoroughly coagulated, and separated into a firm clot and clear serum. This portion we will call C (516).

510. *Treatment of the portion A.*—When the blood has been recently shaken for about a quarter of an hour, immediately on being placed in the bottle (507), the fibrin will be found to have separated, and collected round the fragments of lead which have been previously introduced. The outside of the bottle is then cleaned with a wet cloth, and wiped dry.

511. The weight of the bottle, with its contents, is now taken, in order to ascertain the exact quantity of blood employed in the experiment, which is known by deducting from the gross weight that of the empty bottle and the lead, the difference being the weight of blood contained in it.

512. The stopper is now removed, and the contents of the bottle poured out upon a piece of fine muslin placed in a small basin or saucer. The liquid portion is carefully drained off, and may be thrown away; after which the fibrin adhering to the lead is to be washed with a gentle stream of cold water, until it becomes colourless, in order to separate from it the whole of the corpuscles and serum. During the washing, the spongy aggregations of fibrin may be gently pressed occasionally between the fingers, care being taken that none of the fragments are lost. When clean, the fibrin is to be placed in



a small evaporating dish, and dried on a chloride of calcium bath, at a temperature of  $220^{\circ}$  or  $230^{\circ}$ , until it ceases to lose weight. It is unimportant whether it is dried and weighed with the pieces of lead, or first separated from them, since the weight of the lead, being known (506), may be deducted from the gross weight of the lead and fibrin, the difference being that of the fibrin.

513. The weight thus obtained represents the proportion of FIBRIN in the quantity of blood used in the experiment; the proportion in 1000 parts of blood may afterwards be ascertained by the following calculation :

$$\left\{ \begin{array}{l} \text{Wt. of blood} \\ \text{employed.} \end{array} \right\} : \left\{ \begin{array}{l} \text{Wt. of fibrin} \\ \text{obtained.} \end{array} \right\} :: 1000 : \left\{ \begin{array}{l} \text{Quantity of fibrin in} \\ \text{1000 parts of blood.} \end{array} \right.$$

514. *Treatment of the portion B.*—The capsule containing the portion B, after being accurately weighed (508), is allowed to remain on the water-bath (or still better, on a chloride of calcium bath, heated to about  $220^{\circ}$  or  $230^{\circ}$ ), until it ceases to lose weight on being weighed at intervals of half an hour or an hour, care being taken to wipe the outside clean and dried each time. When the weight becomes constant, it may be concluded that the whole of the water has been expelled.

515. From the weight thus obtained, that of the empty capsule is now to be deducted; the difference being the weight of the ENTIRE SOLID MATTER contained in the quantity of blood operated on. The difference between the weight of this dry residue and that of the blood before evaporation, or, in other words, the loss which it has experienced during the evaporation, will then represent the amount of WATER contained in the quantity of blood employed in the experiment. The proportion of solid matter present in 1000 parts of the blood, may therefore be calculated in the following manner :

$$\left\{ \begin{array}{l} \text{Wt. of} \\ \text{blood} \\ \text{evaporated.} \end{array} \right\} : \left\{ \begin{array}{l} \text{Wt. of} \\ \text{dry} \\ \text{residue.} \end{array} \right\} :: 1000 : \left\{ \begin{array}{l} \text{Proportion of solid} \\ \text{matter in 1000} \\ \text{parts of the blood.} \end{array} \right.$$

516. *Treatment of the portion C.*—The third portion of blood which was collected in the beaker (509), is allowed to stand for about twenty-four hours, or until it separates into a firm clot and clear serum. Two or three drachms of the clear serum are carefully poured off from the clot into a small platinum or porcelain capsule, similar to that before used (506), the weight of which has been previously accurately noted. The capsule with the serum is now weighed, to ascertain the



quantity of the latter employed in the experiment, and then evaporated to perfect dryness on a chloride of calcium bath at a temperature of about 230°, until it ceases to lose weight, the loss of weight which it experiences during evaporation, represents the amount of water in the quantity of serum used; while the weight of the dry residue shows the amount of solid matter contained in the same quantity of serum.

¶ 517. From the numbers now obtained, we are enabled to calculate the proportion of the SOLID MATTERS OF THE SERUM in 1000 parts of blood, in the following manner. Knowing, as we do, the quantity of water in 1000 parts of the blood (515); and assuming that the water of the blood exists wholly in the form of serum;\* knowing also the proportion of water and of solid matter contained in the serum (516); we may, from the quantity of water in the blood, estimate the quantity of solids held in solution in the serum, thus:

$$\left\{ \begin{array}{l} \text{Wt. of water} \\ \text{in the quan-} \\ \text{tity of serum} \\ \text{emp.} \end{array} \right\} : \left\{ \begin{array}{l} \text{Wt. of solid mat-} \\ \text{ter in the quan-} \\ \text{tity of serum} \\ \text{emp.} \end{array} \right\} :: \left\{ \begin{array}{l} \text{Water in} \\ \text{1000 pts.} \\ \text{of the} \\ \text{blood.} \end{array} \right\} : \left\{ \begin{array}{l} \text{Solids of} \\ \text{serum in} \\ \text{1000 pts. of} \\ \text{the blood.} \end{array} \right\}$$

¶ 518. We have now determined the proportion of water, fibrin, and solid matters of the serum, contained in the blood, and have only to ascertain the weight of the CORPUSCLES in order to complete the analysis. This is done by adding together the weights of the fibrin and the solids of the serum contained in 1000 parts of blood, and deducting the sum of them from the weight of the entire solid matter, which consists of fibrin, solids of the serum, and corpuscles; the difference, therefore, will represent the proportion of the latter in 1000 parts of the blood.

¶ 519. The several results now obtained may be recorded thus; and the numbers, when added together, should amount to within a fraction of 1000.

Water	.	.	.	.	.
Corpuscles	.	.	.	.	.
Fibrin	.	.	.	.	.
Solid matters of serum	.	.	.	.	.
<hr/>					
					1000·00
					<hr/>

\* Of course this assumption introduces an error into the analysis, since the water belonging to the contents of the globules is imputed to the serum.



## SECTION II.

*Quantitative Analysis of Coagulated Blood, including the estimation of the water, corpuscles, fibrin, and the solid matters contained in the serum.*

520. The portion of blood intended for analysis, which may consist of about ten fluid ounces, should be collected in a weighed or counterpoised glass beaker, or other cylindrical vessel, and accurately weighed; or if it has been accidentally collected in any vessel of which the weight has not previously been determined, it may be weighed as before, and the weight of the containing vessel, ascertained after the blood has been removed, deducted from the gross weight; the difference being, of course, the weight of the blood employed. The blood, after being collected, is to be set aside in a tolerably cool place for about twenty-four hours, to allow it to coagulate; the top of the glass being covered with a glass plate or small dish, to preserve it from dust and prevent evaporation.

521. About two or three fluid drachms of the clear serum are to be drawn off with a pipette, or carefully poured off, into a small weighed platinum or porcelain capsule; after being accurately weighed, it is to be evaporated, until it ceases to lose weight, on a chloride of calcium bath, kept at a temperature of about  $220^{\circ}$ . When dry, the weight is noted; the loss during evaporation representing the amount of water in the quantity of serum operated on, and the weight of the dry residue being that of the solid matter contained in the same. The relative proportions of solid matter and water which form the serum, are thus ascertained.

522. While the evaporation of the serum (521) is going on, the examination of the rest of the coagulated blood may be proceeded with. The serum is first poured off from the clot with great care, avoiding the escape of any portion of the coagulum; the last portions of the liquid being removed by means of a fine-pointed pipette, or by introducing one end of a folded piece of bibulous paper, which will suck up the liquid until it is saturated, and may then be replaced by another. This serum, although it will probably not be wanted for any subsequent experiments, had better be for the present retained, in case of any accident happening to the portion already taken for evaporation (521).

523. The clot, thus separated from the greater part of the



serum, is now to be divided, by means of a sharp knife, into two portions of equal weight;\* the weight of both being accurately made to correspond by weighing, and adding or taking off small slices, as necessity may require. When this is done, each portion will contain one half the fibrin and corpuscles of the quantity of blood operated on, together with a certain amount of serum. One of these equal portions of the clot we will call A, and the other B.

524. *Treatment of the portion of clot A.*—This is to be cut into thin shreds with a clean, sharp knife, carefully avoiding any loss of the fragments of the coagulum. The finely sliced clot is then tied up in a piece of fine muslin, or calico, and washed under a gentle stream of cold water, with the assistance of occasional pressure between the fingers and thumb, until the whole of the serum and corpuscles are removed from the interstices of the coagulum, and the fibrin is left quite clean and colourless. It is then taken out of the muslin, and dried on a chloride of calcium bath, until it ceases to lose weight. The weight thus obtained represents the fibrin contained in half the clot, and when multiplied by two, gives the proportion of FIBRIN in the quantity of blood employed. †

525. *Treatment of the portion of clot B.*—The weight of the portion B having been noted, it is to be evaporated to dryness on a chloride of calcium bath in a counterpoised or weighed capsule. The loss of weight which it experiences during evaporation, shows the quantity of water contained in half the clot, which, when multiplied by two, gives the amount of water present in the entire clot; while the weight of the solid residue, also multiplied by two, shows the quantity of solid matter which the entire clot contains.

526. From the data thus obtained, we are enabled to calculate the proportion of the several constituents, in the following manner. Having ascertained the weight of the whole solid matter of the clot (525), which consists of fibrin, corpuscles, and solids contained in the portion of serum with which the clot is saturated, we first calculate how much of the weight is due to the solids of the serum. To do this, we assume that the whole of the water present in the clot is due to serum; then,

\* The division must be made *vertically*, since the horizontal layers of the clot contain different proportions of globules, being richer towards the bottom. The exact equality of the two parts of the clot is only necessary to save subsequent calculation.

† To obtain an exact result, this fibrin should be treated with ether to remove oil, then with alcohol, and again dried and weighed.



knowing, from a previous experiment (521), the relative proportions of water and solid matter in the serum, and knowing also the quantity of water contained in the clot (525), we calculate the amount of solid matters in the clot, which belong to the serum, as follows :

$$\left\{ \begin{array}{c} \text{Wt. of water} \\ \text{in quan-} \\ \text{tity of} \\ \text{serum} \\ \text{evaporated.} \end{array} \right\} : \left\{ \begin{array}{c} \text{Wt. of solid} \\ \text{matter in} \\ \text{quantity of} \\ \text{serum} \\ \text{evaporated.} \end{array} \right\} :: \left\{ \begin{array}{c} \text{Wt. of} \\ \text{water} \\ \text{in the} \\ \text{entire} \\ \text{clot.} \end{array} \right\} : \left\{ \begin{array}{c} \text{Wt. of solid} \\ \text{matters of} \\ \text{serum con-} \\ \text{tained in the} \\ \text{entire clot.} \end{array} \right\}$$

527. The weight, thus calculated, of solid matters of serum present in the clot, is deducted from the weight of the entire solid matter contained in the clot (525), and the difference will represent the weight of the fibrin and corpuscles. Having, therefore, previously determined, by a separate experiment (524), the amount of fibrin, we have only to deduct that number, in order to obtain the proportion of CORPUSCLES in the quantity of blood operated on.

528. Knowing now the amount of the fibrin and corpuscles, we can, by deducting their combined weights from that of the entire blood, learn the quantity of serum which it contained, since the blood is wholly composed of fibrin, corpuscles and serum.

529. From the weight of serum thus obtained, assuming that the whole of the water in the blood is due to the serum, we can calculate that of the WATER and SOLID MATTERS OF THE SERUM contained in the entire blood, in the following manner, since we have before determined, by experiment (521), their relative proportions :

*For the Water.*

$$\left\{ \begin{array}{c} \text{Wt. of serum} \\ \text{which was} \\ \text{evaporated} \\ \text{to dryness.} \end{array} \right\} : \left\{ \begin{array}{c} \text{Loss of wt.} \\ \text{during} \\ \text{evapora-} \\ \text{tion.} \end{array} \right\} :: \left\{ \begin{array}{c} \text{Wt. of serum} \\ \text{in quantity} \\ \text{of blood} \\ \text{used.} \end{array} \right\} : \left\{ \begin{array}{c} \text{Proportion} \\ \text{of water in} \\ \text{quantity of} \\ \text{blood used.} \end{array} \right\}$$

*For the Solid Matters of the Serum.*

$$\left\{ \begin{array}{c} \text{Weight of} \\ \text{serum which} \\ \text{was evapo-} \\ \text{rated to dry-} \\ \text{ness.} \end{array} \right\} : \left\{ \begin{array}{c} \text{Weight of} \\ \text{solid} \\ \text{residue of} \\ \text{serum after} \\ \text{evaporation.} \end{array} \right\} :: \left\{ \begin{array}{c} \text{Weight of} \\ \text{serum in} \\ \text{quantity} \\ \text{of blood} \\ \text{used.} \end{array} \right\} : \left\{ \begin{array}{c} \text{Wt. of solid} \\ \text{matters of} \\ \text{serum in} \\ \text{quantity of} \\ \text{blood used.} \end{array} \right\}$$

530. We shall now, therefore, have ascertained the proportions of the four several constituents required, in the quantity of blood employed in the analysis, viz. :



Water . . . . .	
Corpuscles . . . . .	
Fibrin . . . . .	
Solid matters contained in the serum	

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which, when added together, should amount very nearly to the weight of the blood used.

531. In order to determine the proportion of the several constituents present in 1000 parts of the blood, the following calculation will in each case be necessary :

$$\left\{ \begin{array}{l} \text{Wt. of blood} \\ \text{employed} \\ \text{in the} \\ \text{analysis.} \end{array} \right\} : 1000 :: \left\{ \begin{array}{l} \text{Weight of} \\ \text{each con-} \\ \text{stituent} \\ \text{obtained.} \end{array} \right\} : \left\{ \begin{array}{l} \text{Proportion of that} \\ \text{constituent in} \\ \text{1000 parts of the} \\ \text{blood.} \end{array} \right\}$$

### SECTION III.

*Quantitative Analysis of Uncoagulated Blood, including the determination of the water, corpuscles, albumen, fibrin, alcohol extractive, water extractive, oily fats, crystalline or solid fats, and fixed saline matters.*

532. The vessels required for this analysis are nearly the same as those already described in the shorter scheme of analysis (506), viz.—

1. A six- or eight-ounce stoppered bottle, the weight of which is accurately known ; and in which are placed a few small strips of thin sheet lead, the weight of which also is known.
2. A weighed platinum capsule or crucible, capable of holding rather more than an ounce of liquid ; or, in default of this, a thin Dresden porcelain crucible, of about the same capacity. And
3. A tall upright beaker or cylindrical glass, capable of holding about eight ounces of liquid. The weight of this need not be taken.

533. The three vessels being in readiness, the blood is first to be collected. About six ounces of the fluid are allowed to flow into the bottle, which should immediately be closed with the stopper, and gently shaken for a quarter of an hour or twenty minutes, at the end of which time the fibrin will be



found to have separated from the liquid, and attached itself round the fragments of lead. This portion of blood we will call A (536).

534. About an ounce of blood is collected in the weighed capsule or crucible, and, after being weighed for the purpose of ascertaining the exact quantity of blood employed, it is placed on a water-bath or chloride of calcium bath, and allowed to evaporate. This portion we will call B (539).

535. From six to eight ounces of blood are allowed to flow into the beaker, and set aside to coagulate in a tolerably cool place for about twenty-four hours. This portion we call C (541).

536. *Treatment of the portion A.*—As soon as the fibrin is supposed to have separated completely from the blood, and become attached to the pieces of lead, the outside of the bottle is to be wiped clean and dry, and the whole is weighed; whereupon the difference between the combined weights of the empty bottle and the lead, and that of the whole when filled, will represent the quantity of blood employed in the experiment.

537. The contents of the bottle are now to be emptied out upon fine muslin, in a small evaporating basin, and the fibrin is to be carefully separated from the fragments of lead, to which it adheres loosely. It is then washed, under a gentle stream of cold water, from the serum and corpuscles with which it is saturated, carefully avoiding the loss of any particles of the fibrin.

538. When quite clean and colourless, the fibrin is placed in a platinum or thin porcelain crucible of known weight, and dried on a chloride of calcium bath, at a temperature of about  $220^{\circ}$  or  $230^{\circ}$ , until it ceases to lose weight.\* When dry, the weight is noted. As the fibrin, in its present state, contains traces of earthy phosphates, which add slightly to its apparent weight, it may now be incinerated in the crucible, until the ash becomes white or grey. The loss of weight which the dry fibrin experiences during incineration, represents the amount of pure FIBRIN in the quantity of blood that was contained in the bottle. The proportion present in 1000 parts of the blood may then be calculated as follows:

$$\left\{ \begin{array}{c} \text{Weight of} \\ \text{blood} \\ \text{employed.} \end{array} \right\} : \left\{ \begin{array}{c} \text{Weight of} \\ \text{fibrin} \\ \text{obtained.} \end{array} \right\} :: 1000 : \left\{ \begin{array}{c} \text{Proportion of} \\ \text{fibrin in 1000} \\ \text{parts of blood.} \end{array} \right\}$$

\* For greater accuracy, it is well to weigh the piece of muslin, dried at  $220^{\circ}$  before collecting the fibrin upon it, so as to avoid the necessity of removing the fibrin before drying.



539. *Treatment of the Portion B.*—This portion of the blood, after being weighed, is allowed to remain on a chloride of calcium bath, heated to about  $220^{\circ}$ , until it ceases to lose weight; when it may be concluded that the whole of the water has been expelled. When this is the case, the weight is noted; and the proportion of SOLID MATTERS OF THE BLOOD, contained in 1000 parts of the fluid, may be calculated as follows :—

$$\left\{ \begin{array}{l} \text{Wt. of blood} \\ \text{evaporated} \\ \text{to dryness.} \end{array} \right\} : \left\{ \begin{array}{l} \text{Weight of} \\ \text{dry} \\ \text{residue.} \end{array} \right\} :: 1000 : \left\{ \begin{array}{l} \text{Proportion of solid} \\ \text{matter in 1000} \\ \text{parts of blood.} \end{array} \right\}$$

540. The dry residue (539), after being weighed, is to be incinerated in the capsule or crucible, until the whole of the charcoal of the organic matter is burnt away, and the ash becomes of a pale red colour.\* The weight of the ash thus obtained, shows the amount of FIXED SALINE MATTER in the quantity of blood evaporated; and from this, the proportion contained in 1000 parts of the blood may be thus estimated:

$$\left\{ \begin{array}{l} \text{Weight of} \\ \text{blood} \\ \text{evaporated.} \end{array} \right\} : \left\{ \begin{array}{l} \text{Wt. of ash} \\ \text{after in-} \\ \text{cineration.} \end{array} \right\} :: 1000 : \left\{ \begin{array}{l} \text{Proportion of fixed} \\ \text{saline matter in} \\ \text{1000 pts. of blood.} \end{array} \right\}$$

541. *Treatment of the portion C.*—This portion of blood is allowed to stand for about twenty-four hours, in order that it may coagulate spontaneously, and divide itself into a firm clot and perfectly clear serum.

542. Two or three fluid drachms of the serum are first removed from the surface, and placed in a small platinum or porcelain capsule; the exact quantity of serum taken, being ascertained by again weighing the capsule and its contents. It is then placed on a chloride of calcium bath, and, when perfectly dry, again weighed, in order to determine the relative proportions of solid matter and water in the serum; the weight of the dry residue, and the amount of loss during evaporation, representing respectively the proportion of solids and of water in the quantity of serum employed.

543. From the numbers thus obtained, we are able (assuming that the whole of the water in the blood exists in the form of serum) to estimate the quantity of SERUM contained in 1000 parts of the blood, since we have before ascertained the amount of water in 1000 parts of blood (539), and also the

\* A muffle heated to very low redness will be found very convenient for this incineration, which takes place very slowly over a lamp. A small charcoal fire is better than the latter.



relative proportion which the serum bears to the water contained in it (542), thus :

$$\left\{ \begin{array}{l} \text{Wt. of water} \\ \text{in the quan-} \\ \text{tity of serum} \\ \text{that was} \\ \text{evaporated} \\ \text{to dryness.} \end{array} \right\} : \left\{ \begin{array}{l} \text{Weight of} \\ \text{serum} \\ \text{which was} \\ \text{evapo-} \\ \text{rated to} \\ \text{dryness.} \end{array} \right\} :: \left\{ \begin{array}{l} \text{Weight of} \\ \text{water} \\ \text{in 1000} \\ \text{parts} \\ \text{of} \\ \text{blood.} \end{array} \right\} : \left\{ \begin{array}{l} \text{Weight of} \\ \text{serum} \\ \text{in 1000} \\ \text{parts} \\ \text{of} \\ \text{blood.} \end{array} \right\}$$

544. Another portion of the clear serum, weighing exactly 500 grains, is now to be weighed out in a platinum or porcelain capsule, and evaporated to dryness on a water-bath. This will serve for the estimation of the albumen, oily and crystalline fats, and alcohol and water extractives.

545. The dry residue is to be detached, with the aid of a knife, from the capsule, and reduced to fine powder in a mortar. As it is impossible to effect this without loss, the weight of the powder must be ascertained. A small lighting flask, (perfectly dry) is weighed, and the powder introduced into it, the increase of weight being noted. Half an ounce of ether is then poured into the flask and a gentle heat applied by a water-bath\* for about ten minutes, when it may be carefully poured off and replaced by a fresh portion.

546. The ethereal solution thus obtained, containing the fatty matters, both oily and crystalline, is to be evaporated in a capsule of known weight, on a water-bath, until the whole of the ether is expelled. The residue is now weighed, by which the whole amount of fatty matters is ascertained. It is then treated with cold alcohol, which will dissolve out the oily fat. The weight of the residue left on evaporating the alcoholic solution, therefore, will represent the amount of OILY FAT in the serum; and the difference between this and the weight of the whole fatty matter shows the quantity of SOLID or CRYSTALLINE FATTY MATTER in the same serum. The proportion of each of these, which is contained in 1000 parts of blood, may then be calculated as follows :

$$\left\{ \begin{array}{l} \text{Wt. of solid} \\ \text{residue of} \\ \text{serum} \\ \text{employed.} \end{array} \right\} : \left\{ \begin{array}{l} \text{Weight of} \\ \text{fatty} \\ \text{matter} \\ \text{obtained.} \end{array} \right\} :: \left\{ \begin{array}{l} \text{Total solid} \\ \text{from 500} \\ \text{grs. of} \\ \text{serum.} \end{array} \right\} : \left\{ \begin{array}{l} \text{Fatty mat-} \\ \text{ter in 500} \\ \text{grs. of} \\ \text{serum.} \end{array} \right\}$$

\* The flask should be fitted with a cork carrying a glass tube about three feet long and a quarter of an inch in diameter, to diminish the loss of ether by evaporation.



*For the Oily Fat.*

$$500 : \left\{ \begin{array}{c} \text{Wt. of oily} \\ \text{fat in 500} \\ \text{grs. of serum.} \end{array} \right\} :: \left\{ \begin{array}{c} \text{Wt. of serum} \\ \text{in 1000 parts} \\ \text{of blood.} \end{array} \right\} : \left\{ \begin{array}{c} \text{Proportion of oily} \\ \text{fat in 1000 parts of} \\ \text{blood.} \end{array} \right\}$$

*For the Crystalline Fatty Matter.*

$$500 : \left\{ \begin{array}{c} \text{Wt. of crystal-} \\ \text{line fat in 500} \\ \text{grs. of serum.} \end{array} \right\} :: \left\{ \begin{array}{c} \text{Wt. of serum} \\ \text{in 1000 parts} \\ \text{of blood.} \end{array} \right\} : \left\{ \begin{array}{c} \text{Proportion of crys-} \\ \text{talline fat in 1000} \\ \text{parts of blood.} \end{array} \right\}$$

547. The residue which proved insoluble in the ether (545), is now to be warmed, in order to expel any traces of ether that may still be present, and then treated with water which is gradually heated to boiling, this will coagulate the albumen, thus rendering it insoluble; while the extractive matters are dissolved out (549). The mixture is then filtered through a dried and weighed filter, and the insoluble residue of albumen washed on the filter with hot water, until a drop of the filtered liquid causes no precipitate, or merely a very slight opalescence, when tested with a solution of nitrate of silver.

548. The albumen, thus freed from extractive and soluble saline matters, is to be dried and weighed; but as some traces of inorganic matter are always associated with the albumen, the dry mass is to be incinerated, and the weight of the ash deducted from it; when the difference will represent the amount of pure ALBUMEN in the serum. The proportion in 1000 parts of blood may then be calculated thus:—

$$\left\{ \begin{array}{c} \text{Wt. of solid} \\ \text{residue of} \\ \text{serum} \\ \text{employed.} \end{array} \right\} : \left\{ \begin{array}{c} \text{Weight of} \\ \text{albumen} \\ \text{obtained.} \end{array} \right\} :: \left\{ \begin{array}{c} \text{Total solid} \\ \text{from 500} \\ \text{grs. of} \\ \text{serum.} \end{array} \right\} : \left\{ \begin{array}{c} \text{Albumen} \\ \text{in 500} \\ \text{grs. of} \\ \text{serum.} \end{array} \right\}$$

$$500 : \left\{ \begin{array}{c} \text{Wt. of albu-} \\ \text{men in 500 grs.} \\ \text{of serum.} \end{array} \right\} :: \left\{ \begin{array}{c} \text{Wt. of serum} \\ \text{in 1000 parts} \\ \text{of blood.} \end{array} \right\} : \left\{ \begin{array}{c} \text{Proportion of albu-} \\ \text{men in 1000 parts} \\ \text{of blood.} \end{array} \right\}$$

549. The aqueous solution filtered from the albumen (547), containing the extractive matters and soluble salts, is now to be evaporated to dryness in a capsule of known weight, on a water-bath, and weighed. The dry residue is then treated with alcohol, which should be poured off and renewed as long as anything continues to be dissolved by it. The alcoholic solution is evaporated to dryness on a water-bath, and weighed; it is then incinerated, and the weight of the ash is deducted from that of the dry mass previous to incineration.



The number thus obtained represents the amount of ALCOHOL EXTRACTIVE in 500 grs. of serum, which may be reduced to the proportion in 1000 parts of blood by a calculation similar to the above.

550. The portion of the dry residue which proved insoluble in alcohol (549), is now to be dried, weighed, and ignited; the weight of the ash being deducted from that of the dry mass previous to ignition. This will give the weight of the WATER EXTRACTIVE in 500 grs. of serum; from which the quantity in 1000 parts of blood may be estimated as in the former cases :—

$$500 : \left\{ \begin{array}{l} \text{Wt. of water} \\ \text{extract. in 500} \\ \text{grs. of serum.} \end{array} \right\} :: \left\{ \begin{array}{l} \text{Wt. of serum} \\ \text{in 1000 parts} \\ \text{of blood.} \end{array} \right\} : \left\{ \begin{array}{l} \text{Proportion of water} \\ \text{extract. in 1000} \\ \text{parts of blood.} \end{array} \right\}$$

551. We shall now have estimated the proportion of water and of all the solid constituents, with the exception of the CORPUSCLES. The proportion of these is known by deducting the sum of the several solid matters, the weights of which are already determined (including everything but the corpuscles), from the weight of the whole solid matter contained in 1000 parts of blood (539), the difference representing the proportion of corpuscles present in 1000 parts of the fluid.

552. The results of the analysis may then be recorded as follows, and should, when added together, amount to a fraction less than 1000.

Water	.	.	.	.	.	.
Corpuscles	.	.	.	.	.	.
Albumen	.	.	.	.	.	.
Fibrin	.	.	.	.	.	.
Alcohol extractive	.	.	.	.	.	.
Water extractive	.	.	.	.	.	.
Oily fats	.	.	.	.	.	.
Crystalline or solid fats	.	.	.	.	.	.
Fixed saline matter	.	.	.	.	.	.

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## SECTION IV.

*Quantitative Analysis of Coagulated Blood, including the estimation of the water, corpuscles, albumen, fibrin, alcohol extractive, water extractive, oily fats, crystalline or solid fats, and fixed saline matters.*

553. About ten or twelve ounces of blood having been collected in a beaker, or other rather tall vessel of known weight, it is to be covered over to prevent evaporation, and set aside in a cool place for about twenty-four hours, when it will be found to have separated into a firm clot and clear serum. The weight of the whole blood is to be accurately determined either before or after coagulation. Three or four fluid drachms of the clear serum are first drawn off with a pipette, weighed in a platinum or porcelain crucible of known weight, evaporated to dryness on a chloride of calcium bath, and the weight of the dry residue ascertained. The loss of weight during evaporation representing the water, we thus determine the relative proportions of SOLID MATTER AND WATER IN THE SERUM.

554. The dry residue of the serum (553), is now to be incinerated, until the ash becomes white or grey; and the latter is then weighed. The proportion of FIXED SALINE MATTER OF THE SERUM is thus ascertained.

555. The greater part of the remaining clear serum is now to be carefully poured off from the coagulum, and retained for further examination (565). The last portions of the liquid are to be removed by means of a fine pipette, or by sucking it up with little rolls of bibulous paper (522), carefully avoiding the removal of any portions of the clot.

556. The coagulum, thus separated as completely as possible from the serum, is now to be divided vertically into two portions of exactly equal weight (523), each of which will then contain one half of the fibrin and corpuscles present in the quantity of blood operated on, together with a certain amount of serum. These two equal portions of clot we will distinguish as A and B.

557. *Treatment of the portion of clot A.*—This portion of the clot is to be cut with a sharp knife into fine slices, carefully avoiding any loss. These are then tied up in a piece of fine muslin, and washed, until they become quite colourless, when



it may be concluded that the whole of the corpuscles and serum has been washed out. The fibrin is now dried on a chloride of calcium bath at a temperature of about  $230^{\circ}$ , and weighed. It still, however, contains traces of earthy salts, the quantity of which is known by incinerating the dry fibrin, and deducting from it the weight of the ash. The loss of weight during incineration represents the quantity of fibrin contained in one half the clot, and this, when multiplied by two, gives the proportion of FIBRIN in the quantity of blood employed.

558. *Treatment of the portion of clot B.*—This half of the clot is to be weighed in a capsule of known weight, and evaporated to dryness on a chloride of calcium bath. The residue is now weighed, and the loss of weight during evaporation will show the amount of water present in half the clot; which, when multiplied by two, gives the quantity of WATER CONTAINED IN THE ENTIRE CLOT; while the weight of the dry residue, also multiplied by two, represents the amount of SOLID MATTER PRESENT IN THE ENTIRE CLOT.

The dry residue of B is to be retained for subsequent incineration (563).

559. Having thus determined the weight of the whole solid matter of the clot, which consists of fibrin and corpuscles, together with the solids contained in the portion of serum with which the clot is saturated; we now have to calculate how much of the weight is due to the solids of the serum. Assuming that the whole of the water present in the clot is due to the serum, and knowing the relative proportions of water and solid matter in the serum (553); knowing also the quantity of water present in the entire clot (558); the amount of solid matters in the clot which belong to the serum may be calculated in the following manner:

$$\left\{ \begin{array}{l} \text{Wt. of water} \\ \text{in quantity} \\ \text{of serum} \\ \text{evaporated.} \end{array} \right\} : \left\{ \begin{array}{l} \text{Wt. of solid} \\ \text{matter in} \\ \text{quantity of} \\ \text{serum evap.} \end{array} \right\} :: \left\{ \begin{array}{l} \text{Wt. of} \\ \text{water} \\ \text{in entire} \\ \text{clot.} \end{array} \right\} : \left\{ \begin{array}{l} \text{Wt. of solid mat-} \\ \text{ters of serum} \\ \text{contained in the} \\ \text{entire clot.} \end{array} \right\}$$

560. The weight of solid matters of the serum thus found to be present in the clot, is to be deducted from the weight of the entire solid matter of the clot (558), when the difference will represent the weight of the fibrin and corpuscles; the weight of the fibrin, however, having been already ascertained



by a separate experiment (557), we have merely to deduct that amount, in order to determine the proportion of CORPUSCLES in the quantity of blood employed in the analysis.

561. Now, since the blood may be said to consist wholly of fibrin, corpuscles, and serum; and knowing, as we do (557, 560), the weight of the fibrin and corpuscles; we can, by deducting the combined weights of those two substances from the weight of the entire blood, learn the proportion of SERUM in the quantity of blood operated upon.

562. But we have before determined the relative proportions of solid matter and water in the serum (553); so that, assuming that the whole water of the blood is due to the serum, we can, from the quantity of serum obtained in paragraph 561, estimate the proportion of WATER in the blood, thus :

$$\left\{ \begin{array}{l} \text{Wt. of serum} \\ \text{which was} \\ \text{evaporated} \\ \text{to dryness.} \end{array} \right\} : \left\{ \begin{array}{l} \text{Loss of wt.} \\ \text{during eva-} \\ \text{poration} \\ \text{(water).} \end{array} \right\} :: \left\{ \begin{array}{l} \text{Wt. of serum} \\ \text{in quantity} \\ \text{of blood} \\ \text{used.} \end{array} \right\} : \left\{ \begin{array}{l} \text{Proportion} \\ \text{of water in} \\ \text{quantity of} \\ \text{blood used.} \end{array} \right\}$$

563. The dry residue of the portion of the clot B (558), is now to be incinerated. The weight of the ash thus obtained, multiplied by two, will give the amount of the inorganic salts contained in the clot. A certain portion of this weight, however, is due to the salts of the serum which was contained in the clot, the amount of which may be learnt by the following calculation, since we have before determined the relative proportions of solid matter and inorganic ash in the serum (553, 554).

$$\left\{ \begin{array}{l} \text{Wt. of solid mat-} \\ \text{ter in quantity} \\ \text{of serum evapo-} \\ \text{rated to dry-} \\ \text{ness.} \end{array} \right\} : \left\{ \begin{array}{l} \text{Wt. of ash} \\ \text{derived} \\ \text{from same} \\ \text{quantity} \\ \text{of serum.} \end{array} \right\} :: \left\{ \begin{array}{l} \text{Wt. of solid} \\ \text{matters} \\ \text{of serum} \\ \text{in the} \\ \text{clot.} \end{array} \right\} : \left\{ \begin{array}{l} \text{Wt. of ash} \\ \text{derived} \\ \text{from the} \\ \text{serum in} \\ \text{the clot.} \end{array} \right\}$$

By deducting this number from the weight of the ash of the whole clot, we ascertain the amount of inorganic saline matter derived from the fibrin and corpuscles.

564. In order to determine the whole amount of fixed salts in the blood, we must now reckon how much the whole of the serum contains. This is done as follows :

$$\left\{ \begin{array}{l} \text{Weight of} \\ \text{serum evapo-} \\ \text{rated to} \\ \text{dryness.} \end{array} \right\} : \left\{ \begin{array}{l} \text{Wt. of ash} \\ \text{from same} \\ \text{quantity} \\ \text{of serum.} \end{array} \right\} :: \left\{ \begin{array}{l} \text{Wt. of se-} \\ \text{rum in} \\ \text{the entire} \\ \text{blood.} \end{array} \right\} : \left\{ \begin{array}{l} \text{Wt. of fixed} \\ \text{salts in} \\ \text{the whole} \\ \text{serum.} \end{array} \right\}$$



By adding together the ash of the serum thus obtained, and that derived from the fibrin and corpuscles (563), we ascertain the proportion of FIXED SALINE MATTER in the quantity of blood employed in the analysis.

565. *Estimation of the albumen, extractives, and fatty matters.*—Five hundred grains of the clear serum (555), are to be weighed out in a platinum or porcelain evaporating basin, and evaporated to dryness on a water-bath. The residue is then treated as described in paragraphs 545—550.

566. The results of the analysis may then be summed up as follows; and if the experiments have been conducted with care, the numbers will, when added together, coincide very nearly with the whole quantity of blood employed in the analysis.

Water	.	.	.	.	.	.
Corpuscles	.	.	.	.	.	.
Albumen	.	.	.	.	.	.
Fibrin	.	.	.	.	.	.
Alcohol extractive	.	.	.	.	.	.
Water extractive	.	.	.	.	.	.
Oily fats	.	.	.	.	.	.
Crystalline or solid fats	.	.	.	.	.	.
Fixed saline matters	.	.	.	.	.	.
<hr/>						
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567. In order to reduce these several amounts to the proportion contained in 1000 parts of the blood, the following calculation must be made in each case:

$$\left\{ \begin{array}{l} \text{Wt. of} \\ \text{blood} \\ \text{used.} \end{array} \right\} : 1000 :: \left\{ \begin{array}{l} \text{Wt. of each} \\ \text{constituent} \\ \text{obtained.} \end{array} \right\} : \left\{ \begin{array}{l} \text{Proportion of that con-} \\ \text{stituent in 1000 parts} \\ \text{of blood.} \end{array} \right\}$$

The several quantities thus obtained should, when added together, amount to a fraction less than one thousand.



## SECTION V.

*Average Composition of Healthy Blood.*

568. The following analyses will serve to show the usual composition of healthy blood in 1000 parts.

569. *Analysis I. Healthy Venous Blood. (Dumas).*

130 Clot	{	Fibrin . . . . .	3
		Globules { Hæmatin . . . . .	2
		Albuminous matter . . . . .	125
		Water . . . . .	790
		Albumen . . . . .	70
		Oxygen . . . . .	
		Nitrogen . . . . .	
		Carbonic acid . . . . .	
		Extractive matter . . . . .	
		Phosphorized fat . . . . .	
		Cholesterin . . . . .	
		Serolin . . . . .	
870 Serum	{	Oleic and margaric acids . . . . .	
		Chlorides of sodium and potassium . . . . .	
		Muriate of ammonia . . . . .	
		Carbonates of soda, lime, and magnesia . . . . .	
		Phosphates of soda, lime, and magnesia . . . . .	
		Sulphate of potash . . . . .	
		Lactate of soda . . . . .	
		Salts of the fatty acids . . . . .	
		Yellow colouring matter . . . . .	
1000			1000

570. *Analysis II. (Simon.)*

Water . . . . .	795.278
Fibrin . . . . .	2.104
Fat . . . . .	2.346
Albumen . . . . .	76.600
Globulin . . . . .	103.022
Hæmatin . . . . .	6.209
Extractive matter and salts . . . . .	12.012



571. *Analyses III & IV. (Becquerel and Rodier.)**Showing the mean composition of Male and Female Blood.*

	Male.	Female.
Density of defibrinated blood . . . . .	1060.00	1057.50
Density of serum . . . . .	1028.00	1027.40
Water . . . . .	779.00	791.10
Fibrin . . . . .	2.20	2.20
Fatty matters . . . . .	1.60	1.62
Serolin . . . . .	0.02	0.02
Phosphorized fat . . . . .	0.49	0.46
Cholesterin . . . . .	0.09	0.09
Saponified fat . . . . .	1.00	1.04
Albumen . . . . .	69.40	70.50
Blood-corpuscles . . . . .	141.10	127.20
Extractive matters and salts . . . . .	6.80	7.40
Chloride of sodium . . . . .	3.10	3.90
Other soluble salts . . . . .	2.50	2.90
Earthy phosphates . . . . .	0.33	0.35
Iron . . . . .	0.57	0.54

572. *Analysis V. (Lehmann.)*

	Blood-corpuscles.	Liquor sanguinis.
Water . . . . .	688.00	902.90
Solid constituents . . . . .	312.00	97.10
Specific gravity . . . . .	1.0885	1.028
Hæmatin . . . . .	16.75	Fibrin . 4.05
Hæmato-crystallin . . . . .	241.07	Albumen . 78.84
Cell-membranes . . . . .	41.15	
Fat . . . . .	2.31	1.72
Extractive matter . . . . .	2.60	3.94
Mineral substances (exclusive of iron) . . . . .	8.12	8.55
Chlorine . . . . .	1.686	3.644
Sulphuric acid . . . . .	0.066	0.115
Phosphoric acid . . . . .	1.134	0.191
Potassium . . . . .	3.328	0.323
Sodium . . . . .	1.052	3.341
Oxygen . . . . .	0.667	0.403
Phosphate of lime . . . . .	0.114	0.311
Phosphate of magnesia . . . . .	0.073	0.222



573. *Analysis VI. (Enderlin.)**Showing the Composition of the Ash of Human Blood.*

Tribasic phosphate of soda	22.100	83.746	{ Soluble salts.
( $3\text{NaO}, \text{PO}_5$ )	.		
Chloride of sodium	54.769		
Chloride of potassium	4.416		
Sulphate of potash	2.461	15.175	{ Insoluble salts.
Phosphate of lime	3.636		
Phosphate of magnesia	0.769		
Peroxide of iron and phosphate of iron	10.770		
	<hr/> 98.921 <hr/>		

Recent analyses have proved that, as would be expected, the quantitative composition of the blood varies with the part of the circulation from which it is drawn.

## CHAPTER III.

## MORBID BLOOD.

574. The chemistry of the blood in its pathological conditions, has, until within the last few years, occupied very little attention from the chemist or physician; the consequence of which has been, that much ignorance has always prevailed, and, it is to be feared, still prevails, among the great mass of the profession, respecting this important and interesting subject of inquiry. It is not unreasonable to anticipate that the fresh knowledge which we are now almost daily acquiring in this and other kindred branches of physiological and pathological chemistry, will gradually lead to highly important and beneficial practical results, in the more enlightened treatment of disease, and the more ready mitigation of suffering.

575. The variations which are found to occur in the chemical composition of morbid blood may be divided into two classes:

1st. Those in which, so far as we are aware, no abnormal



matter, not contained in healthy blood, is present ; but in which one or more of the normal constituents of healthy blood exists in a greater or less proportion than in the healthy fluid.

2nd. Those in which we can detect the presence of one or more abnormal matters which are not found in healthy blood.

576. To the first of these classes belong those cases in which we find an excess or deficiency of water, corpuscles, albumen, fibrin, fatty matters, cholesterin, urea, uric acid, or inorganic salts ; and to the second, those in which either sugar, biliary matter, pus, entozoa, or other abnormal matter, can be detected. I will briefly notice each of these morbid conditions of the blood, together with the mode of examination, whether chemical or microscopic, which will be found most readily applicable to each.

#### CLASS I.—*Morbid Blood in which no abnormal matter is present.*

##### SECTION I.

##### *Blood containing an excess or deficiency of Water.*

577. The proportion of water even in healthy blood appears to vary considerably, so that it is difficult to say what may be considered as the normal amount. The usual average, however, contained in human blood, seems to be from 790 to 800 in 1000 parts.

578. In some forms of disease, as, for example, anæmia and chlorosis, the proportion of water is usually much greater, and has been known to amount to upwards of 900 parts in 1000. In certain other pathological conditions, on the contrary, the blood is found to contain considerably less water than is present in the healthy fluid ; in cholera, for instance, where the blood is so rich in solid matter as almost to resemble jelly in appearance, it has been known to contain not more than 480 parts of water in 1000.

579. The proportion of water present in any specimen of blood may readily be ascertained, by evaporating a known weight of the fluid in a weighed or counterpoised capsule, on a chloride of calcium bath, heated to about 220° or 230°, until



it ceases to lose weight. The loss of weight during the evaporation will then represent the proportion of water in the quantity of blood employed, which may be reduced to 1000 parts, as follows :

$$\left\{ \begin{array}{c} \text{Weight of} \\ \text{blood} \\ \text{evaporated,} \end{array} \right\} : \left\{ \begin{array}{c} \text{Loss of weight} \\ \text{during evaporation.} \end{array} \right\} :: 1000 \left\{ \begin{array}{c} \text{Proportion of} \\ \text{water in 1000} \\ \text{parts of blood.} \end{array} \right\}$$

## SECTION II.

*Blood containing an excess or deficiency of Corpuscles.*

580. The average proportion of corpuscles contained in a healthy human blood appears to be from 120 to 130 parts in 1000. In disease, especially in some forms of fever, it sometimes increases considerably, and has been known to amount to 185 parts in 1000 ; while in anæmia, and certain other affections long known as being attended with great *poorness* of blood, the proportion of corpuscles frequently does not amount to more than 60 or 70, and has been known to be as low as 21, in 1000 parts.

581. The direct determination of the weight of the corpuscles is a matter of considerable difficulty, so that they are generally estimated by deducting the combined weights of the water, fibrin, and solid matters of the serum, which are easily determined experimentally, from that of the entire blood, in the manner described in paragraphs 518, 527, &c.

582. According to Figuier, their weight may be determined with considerable accuracy by mixing the blood, previously defibrinated by agitation with fragments of lead (507), and weighed, with about eight times its bulk of a saturated solution of sulphate of soda, filtering through a filter of known weight, and washing the corpuscles on the filter with a little more of the saline solution (456). When most of the liquid has drained through, the filter with its contents is dipped in boiling water, and allowed to remain in it some little time, in order to dissolve out the salt ; while the organic matter of the corpuscles is coagulated by the heat, and thus rendered insoluble. The filter, with the corpuscles, is then dried at 212°, weighed, and the weight of the dry filter, previously determined, being deducted, the difference will represent the weight of the corpuscles contained in the quantity of blood operated on.



583. The microscopic appearance of the corpuscles is also not unfrequently found to vary under the influence of disease, the modifications of form occurring occasionally in the living body, but more frequently after death. Most of these changes are due to the phenomena of endosmosis or exosmosis already referred to (456). Thus they are sometimes met with having a more or less globular form, owing to the entrance of fluid less dense than the serum of healthy blood; at other times they are found to have a wrinkled or indented outline, similar to that which the healthy corpuscle assumes when placed in contact with strong saline solutions of high specific gravity. (See Fig. 65.)

584. In examining the blood-corpuscles under the microscope with a view to detecting any abnormal appearance as a consequence of disease, it must be borne in mind that these and other analogous changes in the form of the corpuscle are artificially induced by the action of water or other liquids with which they may have been allowed to come in contact; such contact should therefore be carefully avoided. The wrinkled appearance is sometimes caused also by the concentration of the serous fluid, owing to spontaneous evaporation (456).

### SECTION III.

#### *Blood containing an excess or deficiency of Albumen.*

585. The average proportion of albumen in healthy blood appears to lie between 70 and 75 parts in 1000; while in disease it is occasionally (as in cholera) as high as 131, and (as in Bright's disease) as low as 55 parts in 1000.

586. The amount of albumen in any specimen of blood may be ascertained in the manner described in paragraph 547; or a weighed portion of serum may be carefully neutralized with dilute hydrochloric acid, diluted with an equal bulk of water, and boiled for about a quarter of an hour. The coagulum of albumen is then separated by filtration, dried at  $212^{\circ}$ , and treated with hot ether to remove the fat (545), and weighed before and after incineration; the difference between the two weighings being the weight of albumen in the quantity of serum used (548).

587. The quantitative estimation of the other constituents of the blood may, if necessary, be conducted as in the case of healthy blood (503, &c.).



## SECTION IV.

*Blood containing an excess or deficiency of Fibrin.*

588. Healthy human blood usually contains from two to three parts of fibrin in 1000; while in disease it has been found to vary from a mere trace, to upwards of ten parts in 1000; a considerable increase in the amount being usually found in most forms of inflammatory disease.

589. The peculiar appearance frequently to be seen after coagulation, in blood taken from the body during certain pathological conditions, long known as the *buffy coat*, is caused by the upper portion of the clot being composed almost entirely of fibrin, or of some modification of protein closely allied to it, unmixed with the red corpuscles. This may be owing either to the blood-corpuscles subsiding in the liquid more rapidly than in ordinary blood, or to the fibrin coagulating more slowly; in either case the upper portion of the coagulated fibrin would be more or less free from the corpuscles to which the red colour of the ordinary clot is due. The blood in which the buffy coat is found to occur is, in most cases, rather rich in fibrin, and it was formerly regarded as a sure sign of inflammation; an opinion which has since been proved to be altogether erroneous (452).

590. The proportion of fibrin may be readily determined either in coagulated or freshly drawn blood, in the manner already described. For freshly drawn blood, *see* paragraph 510, &c., and for coagulated blood, *see* paragraph 524, &c.

The quantitative estimation of the other ingredients may also, if necessary, be conducted in the same manner as in healthy blood (503, &c.).

## SECTION V.

*Blood containing an Excess of Fatty Matter.*

591. The average amount of fat in healthy blood appears to be something more than two parts in a thousand. The whole of the oily fat probably exists in combination with potash or soda, forming a kind of soap; so that in the healthy fluid no oil globules can be detected.

592. In certain pathological conditions, we occasionally meet with blood containing a considerable quantity of free fat,



which is held in suspension, in the form of minute globules, in the serum, giving that fluid a more or less opaque or milky appearance. In this form of

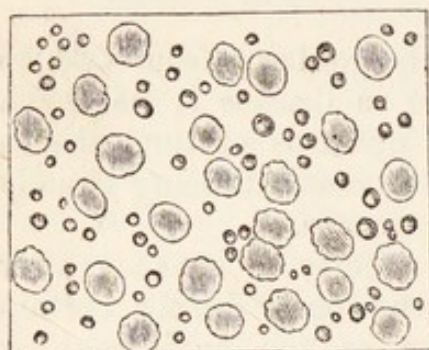


Fig. 67. Fat in Blood.

blood, which, from its peculiar appearance, has been called *milky blood*, may be seen, with the help of the microscope, innumerable fat globules, which may be readily distinguished by their bright centres, and black, well-defined outlines (Fig. 67). They may be separated by agitating the serum with a little ether, which will readily dissolve them.

593. The amount of fat in any specimen of blood may be determined by evaporating to dryness a known weight of the fluid, pounding the dry residue, and boiling it with successive small quantities of ether (545). The ethereal solution of the fat thus obtained is evaporated to dryness in a counterpoised capsule, and weighed; its weight representing the proportion of fat in the quantity of blood employed.

594. The quantitative determination of the other constituents of the blood may, if required, be effected in the same manner as in the healthy fluid (503, &c.)

## SECTION VI.

### *Blood containing an Excess of Cholesterin.*

595. Minute traces of cholesterin appear to be always present in healthy blood, though some observers have failed in their endeavours to detect it. The amount, however, in certain forms of disease not unfrequently rises as high as 0.15 to 0.20 in 1000 parts; and in one case of so-called *milky blood*, Lecanu found not less than 1.08 in 1000.

596. When an excess of cholesterin is suspected to be present in any specimen of blood, it may be separated and estimated with tolerable accuracy in the following manner. A known weight of the blood is evaporated to dryness on a water-bath, and the dry residue, after being reduced to fine powder in a mortar, is digested for a few hours in ether, the solvent action being assisted by occasional boiling (545). In this way the cholesterin, together with the other fatty matters,



is dissolved, and may be obtained by evaporating the ethereal solution on a water-bath. The residue is then deprived of the oily portion of the fat, by digestion with cold alcohol, which leaves undissolved the cholesterin, with the other solid fatty matters; the crystalline scales of cholesterin (Fig. 68), which are easily distinguishable from the rest, may then be, for the most part, mechanically separated with the point of a knife. Their weight may then, after drying, be ascertained if necessary.

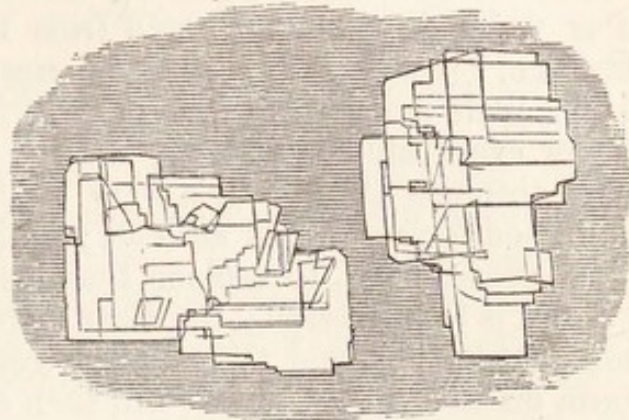


Fig. 68. Cholesterin.

597. The quantitative estimation of the other constituents may be conducted as in the case of healthy blood (503, &c.).

## SECTION VII.

### *Blood containing an Excess of Urea.*

598. Minute traces of urea are probably always present in healthy blood (484), though the amount is so small as to be incapable of determination, unless considerable quantities of blood are used. In some forms of disease, however, especially in Bright's disease, cholera,\* and certain other pathological conditions, in which the functions of the urinary organs are to any serious extent interfered with, the amount of urea is found to increase considerably, and may frequently be met with in a sufficiently large quantity to be weighed.

599. The detection and estimation of urea in the blood may be conducted in the following manner. A known weight of serum is first evaporated to dryness on a water-bath, at a *very gentle* heat, a precaution necessary to be observed, since a temperature of  $212^{\circ}$ , long continued, such as is required in this analysis, would probably cause the decomposition of some portion of the urea. The dry residue is reduced to fine powder in a mortar, and treated with distilled water, heated to

\* It has been stated that, in cholera, a peculiar ferment is present in the blood, which very rapidly converts the urea into carbonate of ammonia (11).



about  $200^{\circ}$ , the quantity of which may be about double the volume of the serum employed in the experiment. The mixture is allowed to digest for about half an hour at  $200^{\circ}$ , after which it may be filtered from the insoluble residue of albumen,\* which latter must be washed while on the filter with a little more warm water. The filtered aqueous solution is now evaporated to dryness, and the residue digested with a little absolute alcohol, at a very gentle heat, which may be continued for about half an hour; a little fresh alcohol being added occasionally, to replace that lost by evaporation. The mixture is then filtered; the clear alcoholic solution is evaporated to dryness, and the residue treated with a little lukewarm distilled water, which will then contain merely the urea, together with a small quantity of extractive matter.

600. The aqueous solution thus obtained is evaporated at a very gentle heat, to the consistence of a syrup, and then mixed with a few drops of pure and colourless nitric acid (16, 181), the mixture being kept cool by immersing the glass containing it in a little cold water, or, still better, in a freezing mixture composed of equal weights of crystallized nitrate of ammonia and water. If urea is present, delicate crystalline plates of nitrate of urea ( $C_2H_4N_2O_2, HO, NO_5$ ), will gradually appear (Fig. 2), which, if in sufficient quantity, may be dried by gentle pressure between folds of filtering paper, and weighed. From the weight thus obtained, that of the urea in the quantity of serum employed may be calculated as follows:—

$$\underbrace{\left\{ \begin{array}{c} \text{Atomic wt.} \\ \text{of nitrate} \\ \text{of urea.} \end{array} \right\}}_{123} : \underbrace{\left\{ \begin{array}{c} \text{Atomic} \\ \text{wt. of} \\ \text{urea.} \end{array} \right\}}_{60} :: \underbrace{\left\{ \begin{array}{c} \text{Wt. of nitrate} \\ \text{obtained.} \end{array} \right\}}_a : \underbrace{\left\{ \begin{array}{c} \text{Wt. of urea in} \\ \text{quantity of} \\ \text{serum employed.} \end{array} \right\}}_x$$

601. If no appearance of crystallization can be detected with the naked eye, a drop of the acid liquid, cooled by means of a freezing mixture, is to be examined under the microscope, by which means very small traces of urea may be detected (181).

602. The quantitative determination of the other constituents may be effected with a fresh portion of the blood, in the same manner as in the healthy fluid (503, &c.).

\* A more delicate process consists in drying the serum *in vacuo* over sulphuric acid, and extracting the powdered residue with alcohol, which dissolves the urea.



## SECTION VIII.

*Blood containing an Excess or Deficiency of Inorganic Saline Matter.*

603. The average proportion of inorganic saline matter in healthy blood, appears to be about seven parts in 1000. In scurvy, and some other pathological conditions, their amount has been found to increase, and has been known to amount to as much as eleven parts in 1000. In some other diseases, on the contrary, the amount falls below the healthy average.

The proportion of fixed saline matter in any specimen of morbid blood, may be determined as in the case of the healthy fluid—viz., by evaporating to dryness a known weight, and incinerating the residue until the ash becomes nearly colourless. The weight of the ash thus obtained represents the amount of salts in the quantity of blood employed. \*

604. The presence of uric acid (urate of soda) in the blood of gouty patients, may be shown by evaporating a little of the fluid to dryness on a water-bath, and, after washing the dry residue with alcohol, adding a slight excess of dilute hydrochloric or acetic acid to a strong aqueous solution of the extract which proved insoluble in the alcohol. After standing a day or two, minute crystals of uric acid, similar to those formed in the urine, are gradually deposited, and may be identified under the microscope (186, 194), or by their behaviour when treated with nitric acid and ammonia (23). Even in healthy blood, minute traces of uric acid may generally be detected.

A simpler test devised by Dr. Garrod, consists in treating the serum with acetic acid in a watch glass in which a fine thread is placed. After an hour or two, the thread is examined by the microscope, when crystals of uric acid are discernible upon it.

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CLASS II.—*Morbid Blood containing some Abnormal Ingredient.*

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## SECTION IX.

*Blood containing Sugar ( $C_{12}H_{14}O_{14}$ ).*

605. The blood of patients suffering from diabetes, appears

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\* See note to paragraph 63.



most commonly to contain a very sensible amount of sugar.\* This may usually be detected in the following manner:—

606. The portion of serum intended for examination is first evaporated to dryness, either in vacuo over sulphuric acid, or at a very gentle heat on a water-bath. The dry residue is then reduced to tolerably fine powder, and treated with a small quantity of boiling water, which will have the effect of coagulating the albumen, and dissolving out the sugar, together with the extractive matters and soluble salts. The mixture is then filtered, and the clear liquid examined for sugar, by means of Trommer's test, which may be thus applied:—

607. The liquid is treated with a drop or two of a solution of sulphate of copper, and then supersaturated with potash (123), the excess of which will probably, if sugar is present, redissolve the blue precipitate of hydrated oxide of copper at first thrown down. The mixture may now be gently boiled for a few minutes, when, if sugar is present, an orange-brown or ochre-coloured precipitate of suboxide of copper will be thrown down; while, if no sugar is contained in the mixture, the precipitate will be nearly black (124).

608. It is always more satisfactory, when practicable, even when Trommer's test affords tolerably decided indications of sugar, to confirm the result by applying also Böttger's test (127), the fermentation test (128), and examining under the microscope for the torula (132); since certain other organic matters besides sugar give rise to the formation of the suboxide.

609. When, after having proved the presence of sugar in the blood, it is required to determine its amount, the following method of insulating it is, perhaps, the best, though the results must not be regarded as by any means exact, but merely as an approximation to the truth. The fermentation process (128) cannot be here applied, since traces of carbonic acid may be evolved by some of the other constituents of the blood, when no sugar is present.

610. A known weight of serum is evaporated to dryness, either in vacuo over sulphuric acid, or at a very gentle heat on a water-bath. The dry residue is then finely comminuted and treated with boiling water, in which it may be allowed to digest for three or four hours, in order to insure the solution of the whole of the soluble matter. The aqueous solution

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\* See note to paragraph 484.



is separated from the albumen by filtration, and evaporated to dryness as before. The dry residue is now digested with alcohol, which leaves undissolved portions of the saline and extractive matters. The alcoholic solution is mixed with a little alcoholic solution of potash, and set aside for twenty-four hours, when a crystalline compound of sugar and potash will be deposited. The alcoholic solution may now be poured or filtered off, and the crystalline compound dissolved in water with a view to the determination of the quantity of sugar by means of the standard alkaline solution of copper (352).

When the freshly drawn blood can be obtained, the following process recommended by Figuier may be adopted. About six ounces of blood are defibrinated by stirring (477), and mixed with three volumes of a saturated solution of sulphate of soda. The blood globules are then filtered off, and the filtrate mixed with two volumes of alcohol, to coagulate the albumen and precipitate the sulphate of soda. These having been filtered off, the solution is evaporated to dryness on the water-bath, the residue extracted with water, and the sugar determined by the alkaline copper-solution (352).

611. The quantitative determination of the other constituents of blood containing sugar may be effected in the same manner as in the case of healthy blood, the weight of the sugar being deducted from the extractive matter (503, &c.).

## SECTION X.

### *Blood containing Biliary Matter.*

612. In jaundice, and some other affections in which the functions of the liver are interfered with, an accumulation of biliary matter is found to take place in the blood, giving the serum a more or less decided saffron or orange-brown colour, which is due to the peculiar colouring matter of the bile, called biliphœin (cholepyrrhin).

613. The presence of bile in the blood may be detected by adding to a little of the clear serum a few drops of nitric acid, which will throw down the albumen; the precipitate having, if biliary matter (biliphœin) is present, a decided greenish tint, while in healthy serum it would be white, or very nearly so.

614. If so small a quantity of bile is present as to fail in producing a perceptibly green colour with nitric acid, a little of the suspected serum may be first concentrated by evapora-



tion at a temperature not exceeding  $120^{\circ}$  or  $130^{\circ}$ , and then exhausted with alcohol or water, and the solution tested in the manner already described in the case of urine (149—152).

615. We have at present no means of estimating the quantity of biliary matter contained in blood, though the depth of colour of the serum furnishes some indication of the relative amount present. The quantitative determination of the other constituents of the blood may be made in the same manner as in the analysis of the healthy fluid (503, &c.).

## SECTION XI.

### *Blood containing Pus.*

616. The existence of pus in morbid blood is probably by no means a rare occurrence, especially in diseases which are attended with suppuration. Its detection, however, is far from easy, since we possess no characteristic chemical test by which it may be distinguished from the ordinary constituents of the blood; and in microscopic appearance, the pus granules very closely resemble the colourless corpuscles which are always present in the blood (464). The pus granules are in general somewhat larger than the white corpuscles of the blood, and when treated with dilute acetic acid, develop internal nuclei, which are usually from three to five in number, and more distinct than those in the white corpuscles of the blood. The pus granules, when present in blood, appear to have a tendency to adhere together in groups of five or six; while the colourless corpuscles of the blood always float detached from each other.

617. According to Heller, the granules of pus, when mixed with blood, subside much more slowly than the blood-corpuscles; so that when present, they may always be found in the uppermost layer of the coagulum.\* He recommends a thin slice to be taken from the upper surface of the latter, which, after being mixed with a little distilled water, should be filtered through muslin, in order to separate the fibrin. The blood-corpuscles are for the most part dissolved by the action of the water (458); and after allowing the filtered liquid to stand a short time in a tall glass, the pus granules will be found at the bottom of the liquid, and may be detected under the microscope.

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\* This remark also applies to the colourless corpuscles.



618. The action of ammonia upon pus has been proposed by Donné as a test for its presence in the blood. When blood, free from pus, is mixed with ammonia, it becomes clear; while if pus is present in any considerable quantity, the liquid becomes more or less gelatinous. If the amount of pus present is small, stringy flocculi only are formed, which subside to the bottom of the liquid.

## SECTION XII.

### *Blood containing Animalcules.*

619. Instances have occasionally been observed, in which minute thread-like animalcules have been present in considerable numbers in the blood. Those described by Dr. Goodfellow, which he detected in the blood of a patient suffering from fever, measured from  $\frac{1}{3000}$ th to  $\frac{1}{3000}$ th of an inch in length, and from  $\frac{1}{30000}$ th to  $\frac{1}{20000}$ th of an inch in diameter. The only method of detecting such entozoa in the blood, is to examine it carefully under the microscope, with as high a magnifying power as the observer has at his command.



## PART IV.

### MILK, BILE, MUCUS, PUS, BONE, &c.

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#### CHAPTER I.

#### MILK.

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##### SECTION I.

##### *General Characters of Milk.*

620. Milk, as is well known, is a watery liquid, having in solution a certain amount of casein, sugar of milk, or lactine and extractive matter, together with several inorganic salts, and holding in suspension myriads of extremely minute globules of fatty matter, plainly visible through the microscope, which give the fluid its peculiar white and opaque appearance. It has a pleasant and rather sweetish taste, and a slight agreeable smell, especially while warm. The specific gravity of milk varies considerably; that of woman being sometimes as low as 1020 (the average being 1032), while that of the sheep is as high as 1041.

621. Fresh milk is almost invariably slightly alkaline to test paper, but on exposure to the air, especially in warm weather, it rapidly becomes acid, owing to the conversion of the sugar of milk into lactic acid ( $2HO, C_{12}H_{10}O_{10}$ ), under the influence of the casein, which acts as a ferment (630). If the milk has been long retained in the mammary glands, this change occasionally takes place before being drawn; and in some morbid conditions also, the milk is found to have an acid reaction even when freshly drawn.

622. When allowed to stand for a few hours, the fatty globules, which have a somewhat lower specific gravity than



the fluid portion of the milk, gradually rise to the surface, carrying with them a portion of the caseous matter, forming a layer of cream, which is more or less thick and copious in proportion to the richness of the milk. \*

623. If a little acetic or lactic acid, rennet, or even sour milk, be added to hot milk, the casein of the latter is precipitated in the coagulated form; and the same effect is produced by warming milk or cream which has been allowed to turn sour; the sourness being due to the lactic acid, into which the sugar of milk has been converted. The solid and liquid portions into which the milk is thus divided, are commonly called curds and whey.

624. Before describing the mode of analysing milk, I will briefly notice the several constituents which we find contained in it—viz., casein, sugar of milk, fat globules, and saline matter.

## SECTION II.

### *Casein.*

625. Casein is one of the so-called protein-compounds † (472) peculiar to the milk, and constitutes the chief source of nourishment to the young animal; for which purpose it is admirably adapted, from the readiness with which it appears capable of being converted into the other bodies of the same class—viz., fibrin and albumen.

626. It may be obtained in a state of tolerable purity by evaporating a quantity of milk to dryness on a water-bath, and boiling the dry residue in successive portions of ether, in order to dissolve out the fat. The residue which remains insoluble in the ether is then dried, and digested in water, which will dissolve the casein and other soluble matters of the milk. On adding alcohol to the aqueous solution, a great part of the milk-sugar is thrown down in the form of a precipitate, leaving the casein in solution together with some milk-sugar and soluble salts.

It may be obtained in a purer condition by adding a little hydrochloric acid to skimmed milk, collecting the curd upon a linen strainer, and washing it first with water, then with

\* According to Müller, fresh milk, when allowed to stand, first undergoes a peculiar change, resulting in the dissolution of the membranes enclosing the fat globules, so that the proportion of fat which can be extracted by ether continues to increase for some time after the milk has been drawn.

† See note to 471.



water slightly acidified with hydrochloric acid, and finally with pure water. If it be then heated to  $110^{\circ}$  with a large volume of water, the greater portion will dissolve slowly, and may be reprecipitated from the filtered solution by neutralisation with carbonate of ammonia. After washing with water, and warming, first with alcohol, and afterwards with ether to remove all fatty matter, the casein is as pure as it can be obtained. It always leaves an ash of phosphate of lime when burnt.

627. It is most probable that pure casein is insoluble, or very sparingly soluble, in water, and owes its solubility in milk to the small quantity of alkali which is present. When dry, it closely resembles fibrin and albumen in appearance (479), and its behaviour with reagents is in most cases very similar; it differs from the latter chiefly in not coagulating when heated; and it is precipitated by acetic, and nearly all the acids, but redissolves in a considerable excess of most of them. Its solution in acetic acid is precipitated by dilute sulphuric acid. The ferrocyanide and ferridcyanide of potassium also cause precipitates in solutions of casein.

### SECTION III.

#### *Sugar of Milk or Lactine ( $C_{24}H_{24}O_{24}$ ).*

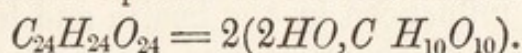
628. The sugar contained in milk may be prepared in the following manner:—The curd, including the greater part of the casein and fat globules, is first separated by the addition of a few drops of acid to hot milk, and the remaining traces of those substances are then removed by mixing a little well-beaten white-of-egg with the whey when cold, and afterwards boiling the mixture. The whey thus clarified by the coagulating albumen of the egg, is filtered from the precipitate by passing it through muslin or calico; and the clear liquid may then be evaporated to about one-fourth or one-fifth its bulk, and set aside in a cool place for a few days. The sugar will gradually separate from the liquid, in the form of minute hard crystals, which adhere to the surface of the containing vessel. These may be purified by dissolving them again in water, boiling the solution with animal charcoal, and recrystallizing.

629. This variety of sugar is less sweet than that obtained either from the cane or the grape (114); it is also harder, and less soluble in water, requiring as much as five or six times its weight of cold, and two and a half times its weight of hot, water to dissolve it. When mixed with a little hydro-



chloric or sulphuric acid, sugar of milk gradually becomes converted into grape sugar ( $C_{12}H_{22}O_{11}$ ), and this change takes place more rapidly if the solution is boiled.

630. Under the influence of the caseous matter of the milk, this form of sugar gradually passes into lactic acid ( $2HO, C_{12}H_{22}O_{11}$ ), a change easily accounted for, since the formula of the sugar is a multiple of that of the acid, one equivalent of the former being broken up into two of the latter.



#### SECTION IV.

##### *Fat Globules.*

631. The minute globules which are held suspended in milk, and to which the opacity and whiteness of the fluid are due, consist mainly of oily fat, which appears to be surrounded by a thin covering of insoluble matter, differing in its properties from fat, and probably composed of one of the protein compounds. It is for this reason that the fat globules cannot be removed from milk by agitation with ether, unless potash be previously added to dissolve the membranous envelopes.

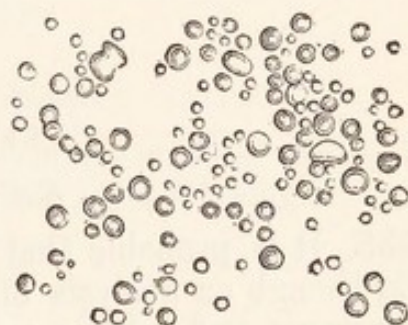


Fig. 69. Milk Globules.

632. The size of the globules in healthy milk varies from a mere point to about  $\frac{1}{2000}$ th of an inch in diameter, the average size being rather more than  $\frac{1}{4000}$ th (Fig. 69).

633. In the milk which is secreted during the first few days of lactation, called the colostrum, and which is always much richer in quality than ordinary milk, we find in addition to the common milk globules, numerous granular corpuscles of a pale yellowish colour, and considerably larger than the others, their diameter varying from  $\frac{1}{2000}$ th to  $\frac{1}{800}$ th of an inch (Fig. 70).

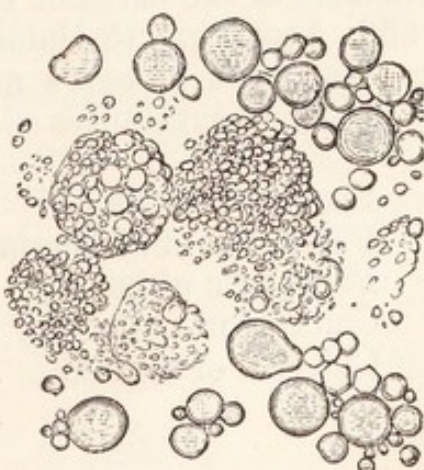


Fig. 70.  
Colostrum Corpuscles.



Similar corpuscles are also occasionally present in milk secreted during disease. They appear to be almost peculiar to human milk, being rarely met with in that of the cow and other animals.

634. The fatty matter of milk consists for the most part of a solid fat, called margarine ( $C_{108}H_{104}O_{12}$ ), mixed with a liquid fat or oil, called oleine ( $C_{114}H_{104}O_{12}$ ), together with small quantities of butyrine and other fats. The proportion in which these several fats are found mixed in milk, varies considerably, being influenced by the health and food of the individual, the season of the year, and other circumstances. A specimen of the fat contained in cow's milk, analysed by Bromeis, contained—

Margarine . . . . .	68
Oleine . . . . .	30
Butyric, caproic, and capric acids . . . . .	2
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#### SECTION V.

##### *Saline Matters.*

635. It is probable that the following salts are present in milk, though an analysis of the ash will not, of course, detect the organic and volatile compounds included in the list, since they are either decomposed or volatilized during the process of incineration:—the chlorides of potassium and sodium; the phosphates of potash, soda, lime, and magnesia, with traces of phosphate of the peroxide of iron.

636. According to Haidlen, the ash obtained by incinerating 1000 parts of cow's milk, consisted, in two instances, of the following substances:—

	I.	II.
Phosphate of lime . . . . .	2.31	3.44
Phosphate of magnesia . . . . .	0.42	0.64
Phosphate of peroxide of iron . . . . .	0.07	0.07
Chloride of potassium . . . . .	1.44	1.83
Chloride of sodium . . . . .	0.24	0.34
Soda . . . . .	0.42	0.45
	<hr/>	<hr/>
	4.90	6.77
	<hr/>	<hr/>

\* Heintz pronounces the margarine of butter to be a mixture of stearine ( $C_{114}H_{110}O_{12}$ ) and palmitine ( $C_{102}H_{98}O_{12}$ ). He has also obtained a new acid



637. The presence of these several salts may be proved by applying to a solution of the ash in water and hydrochloric acid, the tests mentioned in the chapters on the urine and the blood (41, 490, &c.).

## SECTION VI.

*Composition of Human Milk.*

638. In healthy human milk, the several constituents which I have now briefly described, are not always present in the same relative proportions; various circumstances, as those of age, temperament, and food of the mother, as well as the period of lactation, causing considerable variation in the composition of the secretion. The following examples will serve to show to what extent these variations usually occur. The proportions are calculated in 1000 parts of milk.

*Analysis I. (Simon.)*

*Showing the Mean of Fourteen Analyses made at different periods, with the Milk of the same Woman.*

Water	.	.	.	.	.	.	883.6
Solid constituents	.	.	.	.	.	.	116.4
Butter*	.	.	.	.	.	.	25.3
Casein	.	.	.	.	.	.	34.3
Sugar of milk and extractive matters	.	.	.	.	.	.	48.2
Fixed salts	.	.	.	.	.	.	2.3

*Analyses II, III, & IV, (Clemm.)*

	The fourth day after delivery.	The ninth day after delivery.	The twelfth day after delivery.
Water	879.848	885.818	905.809
Solid constituents	120.152	114.182	94.191
Butter	42.968	35.316	33.454
Casein	35.333	36.912	29.111
Sugar of milk and extractive matters.	41.135	42.979	31.537
Salts	2.095	1.691	1.939

called *butic* or *butinic acid* ( $\text{HO}, \text{C}_{40}\text{H}_{39}\text{O}_3$ ) from butter, as well as caprylic ( $\text{HO}, \text{C}_{16}\text{H}_{15}\text{O}_3$ ) and myristic ( $\text{HO}, \text{C}_{28}\text{H}_{27}\text{O}_3$ ) acids.

\* The portion of milk drawn at the commencement of a draught (whether from a woman or a cow) is not so rich in butter as that drawn at the conclusion.



*Analysis VII. (Chevallier and Henri).*

Water	.	.	.	.	.	.	879.8
Solid constituents	.	.	.	.	.	.	120.2
Butter	.	.	.	.	.	.	35.5
Casein	.	.	.	.	.	.	15.2
Sugar of milk	.	.	.	.	.	.	65.0
Salts	.	.	.	.	.	.	4.5

The recent analyses of MM. Vernois and Alfred Becquerel give the following as the composition of normal human milk:—

Water	.	.	.	.	.	.	889.08
Sugar	.	.	.	.	.	.	43.64
Casein and extractive	.	.	.	.	.	.	39.24
Butter	.	.	.	.	.	.	26.66
Salts (ash)	.	.	.	.	.	.	1.38
							<hr/>
							1000.00
							<hr/>

Specific gravity, 1032.67.

## SECTION VII.

*Composition of the Milk of other Animals.*

639. The proportion of the several constituents is found to differ considerably in the milk of different animals.\* The subjoined table, showing the composition of the milk of a few of the more important domestic animals, from the analyses of Chevallier and Henri, will serve to illustrate this:—

	Cow.	Ass.	Goat.	Ewe.
Casein . . .	4.48	1.82	4.08	4.50
Butter . . .	3.13	0.11	3.32	4.20
Sugar of milk .	4.77	6.08	5.28	5.00
Saline matter .	0.60	0.34	0.52	0.68
Water . . .	87.02	91.65	86.80	85.62
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	100.00	100.00	100.00	100.00
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639 a. According to the analysis of Morin† milk contains a

\* Schlossberger describes the milk of the carnivora as having usually an acid reaction. The milk of stall-fed cows, mares, and ewes kept on green food is also said to be frequently acid.

† 'Journ. Pharm.' (3), xxv, 71.



considerable quantity of a substance resembling gelatine, which he proposes to call *galactine*. Both this substance and the caseine of milk possess a specific power of emulsifying the fats, and he thus accounts for the minute state of division of the fatty matter in milk. It is also asserted that the coagulum obtained by boiling milk which has been curdled by acetic acid and filtered, consists of albumen. The results of Morin's analysis of cow's milk were :

Casein . . . . .	36.14
Soda, in combination with it . . . . .	0.48
Butter . . . . .	13.78
Albumen . . . . .	3.90
Sugar of milk . . . . .	36.00
Galactine (gelatigenous matter) . . . . .	3.82
Alcohol extractive . . . . .	5.42
Phosphate of lime . . . . .	2.56
Chloride of sodium . . . . .	0.56
Water . . . . .	897.34
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	1000.00
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## CHAPTER II.

### QUANTITATIVE ANALYSIS OF MILK.

640. Two portions of milk, one weighing about 100 grains, and the other about 400 grains, are to be accurately weighed, the first in a platinum crucible or capsule, and the second in a porcelain capsule; both the vessels having been previously weighed or counterpoised. The first portion, of 100 grains, we will call A, and the second, of 400 grains, we will call B.

641. *Treatment of the portion A.*—This portion, after being weighed, is to be evaporated to dryness on a water-bath, or, still better, on a chloride-of-calcium bath heated to about 220°, until, on being weighed at intervals of half an hour or an hour, it ceases to lose any further weight. The weight of the dry residue will then represent the amount of SOLID MATTER contained in the quantity of milk used, while the loss of weight during evaporation shows the amount of WATER.



642. In these and the other determinations, the proportion present in 1000 parts of the milk is calculated in the following manner:—

$$\left\{ \begin{array}{l} \text{Wt. of milk} \\ \text{used in the} \\ \text{experiment.} \end{array} \right\} : \left\{ \begin{array}{l} \text{Wt. of each} \\ \text{constituent} \\ \text{obtained.} \end{array} \right\} :: 1000 \left\{ \begin{array}{l} \text{Proportion of that} \\ \text{constituent in 1000} \\ \text{parts of the milk.} \end{array} \right\}$$

643. The weight of the dry residue having been noted, the crucible, with its contents, is to be placed over a lamp, and kept at a red heat until the whole of the charcoal is burnt away, and the ash becomes white or nearly so. The weight of the ash thus obtained will represent the amount of INORGANIC SALINE MATTER in the quantity of milk evaporated; from which the proportion in 1000 parts may be calculated as before (642).\*

644. *Treatment of the portion B.*—This portion, after being weighed, is to be mixed with about one-fourth its weight of finely-pounded hydrated sulphate of lime ( $\text{CaO}, \text{SO}_3 + 2\text{Aq}$ ), or unburnt gypsum, with which it is to be well stirred for a short time, and then raised to a temperature of  $212^\circ$ ; by which means the whole of the casein will become coagulated, and insoluble in water. The mixture is now to be evaporated to dryness on a water-bath, being occasionally stirred, in order that the solid residue of the milk may be pretty uniformly mixed with the sulphate of lime.

645. The mass, when dry, is then easily reduced to powder; after which it is to be digested with successive small quantities of ether (545), which will dissolve out the whole of the fatty matter. The ethereal solution is now evaporated to dryness on a water-bath, and the residue weighed; its weight representing the amount of FAT in the quantity of milk operated on; from which the proportion present in 1000 parts of milk may be calculated as before (642).

646. The portion of the residue which proved insoluble in ether (645), is now to be treated with hot, moderately strong alcohol, as long as anything dissolves. In this way, the whole of the sugar, together with a little saline matter and alcohol-extractive, is dissolved. The alcoholic solution is to be evaporated to dryness on a water or chloride-of-calcium bath, and the dry residue, having been accurately weighed, is incinerated; the difference between the weight before and after incineration will then represent the quantity of SUGAR, with a

\* See foot-note to paragraph 63.



little alcoholic extractive matter, in the portion of milk employed. The proportion contained in 1000 parts is then calculated as in former cases (642).

The sugar may be much more exactly determined by means of the alkaline copper-solution, as described at (352). 100 grs. of the milk are acidified with hydrochloric acid, heated to coagulate the casein, and filtered; after washing the coagulum once or twice with water, the filtrate and washings are boiled in a flask for about an hour, replacing the water which evaporates, in order to convert the milk-sugar into grape-sugar, since the former does not reduce the same proportion of oxide of copper. The volume of the liquid is then made up to 1000 grs., and the determination proceeded with as in (352).

647. The proportion of CASEIN may be estimated by adding together the amount of water, fat, sugar, and saline matter, already ascertained as being present in 1000 parts of the milk, and deducting the sum of them from 1000.

648. The caseine may also be approximately determined in another weighed portion of milk by acidulating with acetic acid, boiling, collecting the curd upon a weighed filter, washing two or three times and drying at  $212^{\circ}$ . From its weight that of the fatty matter contained in the milk (645) should be deducted.

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## CHAPTER III.

### MILK DURING DISEASE.

649. The milk which is secreted during disease is usually more or less modified in its composition; even slight derangements of the system, and any great mental anxiety or sudden emotion of fear, &c., not unfrequently have the effect of disturbing, in a remarkable manner, the natural character of the secretion. The exact nature of these changes is very imperfectly understood. They are probably sometimes merely variations in the relative proportions of the several constituents of the healthy fluid; at others, and perhaps more frequently, certain abnormal matters are formed.

650. With the assistance of the microscope, we are not unfrequently able, with great facility, to detect the presence of



certain morbid products which are not found in the healthy secretion. The peculiar form of milk called the colostrum, which is secreted during the first few days of lactation, has been already mentioned as differing very considerably in microscopic appearance from healthy milk, and as containing numerous granular corpuscles, much larger than the ordinary milk globules (633). The corpuscles of the colostrum also show a tendency to adhere to each other, while the globules of the healthy fluid usually float freely about. It occasionally happens that the milk, instead of changing, in the course of a few days, to its more natural condition, continues for a length of time to possess the characters peculiar to colostrum; and has even been observed to change back again to this condition, after being secreted for a time in a healthy state. The presence of the colostrum corpuscles (Fig. 70), and the slightly viscid appearance also characteristic of this condition, may at once be detected under the microscope.

651. The presence of pus, which during the formation of a mammary abscess often finds its way into the milk, may also be detected under the microscope, by the occurrence of the peculiar pus granules (Fig. 71). Blood-corpuscles, too (451), are also found, though more rarely than those of pus, owing, in most cases, to the rupture of some of the minute blood-vessels with which the mammary gland is permeated (Fig. 72.)

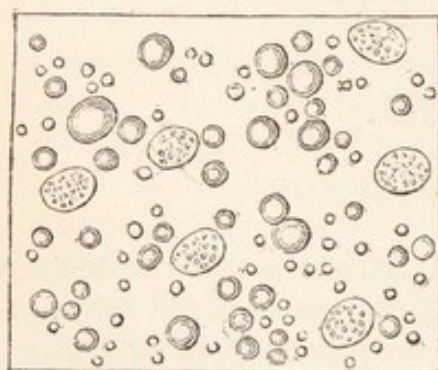


Fig. 71. Pus in Milk.

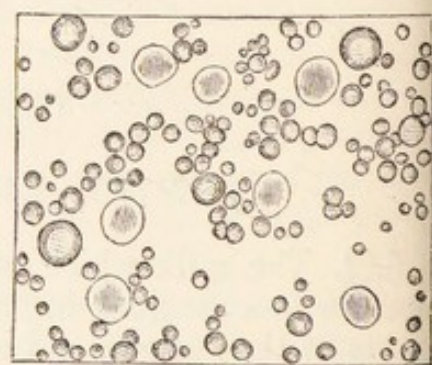


Fig. 72. Blood in Milk.

652. Urea is said to have been found in the milk of women affected by Bright's disease. In addition to the strictly morbid products, other substances, especially certain salts, which have been taken into the system either in the food or as medicine, appear occasionally to find their way into the milk, where they may sometimes be detected by the proper tests.



*Analysis of the Colostrum of a Woman, together with that of the Healthy Milk of the same individual. (Simon.)*

	Colostrum.	Healthy milk.
Water . . . . .	828.0	887.6
Solid constituents . . . . .	172.0	112.4
Fat . . . . .	50.0	25.3
Casein . . . . .	40.0	34.3
Sugar of milk . . . . .	70.0	48.2
Saline matter . . . . .	3.1	2.3

## CHAPTER IV.

## THE ADULTERATIONS OF MILK.

653. It is well known that much of the milk which is supplied in large towns is almost constantly more or less adulterated, and although the substances employed for the purpose are in most cases comparatively innoxious, it is much to be wished that some simple and efficient test of its genuineness and purity could be devised, capable of being applied by those who are unaccustomed to experiment.

654. The chief mode of adulteration practised in this country consists in diluting the milk with water, and at the same time occasionally removing the cream. To correct the bluish colour of the impoverished milk, it is said that a little mannatto is sometimes added. Milk has been occasionally found adulterated with gum, flour, and starch to conceal its diluted condition, and it is even asserted that the clumsy fraud of adding chalk and emulsion of sheep's brains has been detected.

655. On examining a little of the milk under the microscope,



Fig. 73. Starch Granules.



the peculiar granules of starch and flour may be readily seen (Fig. 73*a*), larger and more oval than the milk globules if either of those substances is present; and when examined with polarized light, each granule will be found to exhibit a dark cross, as shown at *b* in the figure. Should any doubt exist as to their real nature, the addition of a drop or two of a solution of iodine will impart to the farina granules a dark purple colour.

Gum may be detected by acidulating the milk with acetic acid, boiling, filtering off the coagulum, and mixing the filtrate with alcohol, when the gum is deposited and may be recognised by its behaviour with water. The presence of annatto would cause the milk to assume a brown colour on addition of carbonate of soda.

656. The microscope will also serve to show the presence of macerated brain, which may be recognised by the occurrence of fragments of nerve and other organised structures, not found in pure milk.

657. The presence of chalk may be still more easily discovered, since, owing to its specific weight, it soon subsides to the bottom of the liquid, where it may at once be recognised by its effervescing on the addition of a little dilute hydrochloric acid.

658. We have no chemical means of ascertaining whether water has been fraudulently added to milk, the only effect being to dilute it, and render it of poorer quality, which might arise from natural causes. A knowledge of the specific gravity will not even allow us to decide as to the richness of the milk, since the abstraction of a portion of the cream, which has a lower specific gravity than milk, may be made to neutralize the effect produced by the addition of water; the tendency of the removal of the cream being to raise the specific gravity of the fluid, and that of the addition of water, to lower it. A specimen of milk, therefore, which has been impoverished by the abstraction of its cream, and still further weakened by the addition of water, may be made to possess the same specific gravity as it had when taken pure from the udder.

For most practical purposes it is sufficient to compare the relative volumes of cream furnished by equal quantities of different specimens of milk. This may be readily effected by allowing the milk to stand in a graduated tube (*lactometer*) for twenty-four hours, at a moderate temperature, and measuring the number of divisions occupied by the cream.



Another method proposed by Daubrawa for the rapid determination of the quality of milk consists in mixing it with two volumes of alcohol of sp. gr. 0.833, filtering off the butter and casein (which may be dried and weighed), and taking the specific gravity of the filtrate. Every increase of .004 in the specific gravity above 0.905 (the sp. gr. of the mixture of alcohol with the water of the milk) indicates 1 per cent. of milk-sugar. For example, if the specific gravity of the filtrate be 0.922, there would be 4.25 per cent. of milk-sugar, for  $0.922 - 0.905 = .017$ , and  $.017 \div .004 = 4.25$ . The result may be controlled by evaporating the spirit, converting the milk-sugar into grape-sugar by boiling with a little dilute sulphuric acid, rendering the solution alkaline by potash, and determining the sugar by the standard copper solution (352).

659. It occasionally happens that the milk exposed for sale is the produce of an unhealthy animal. Such milk has usually some peculiarity of taste or smell, and also a slightly viscid and unnatural appearance; on being examined under the microscope, too, it will probably be found to contain pus or mucus corpuscles, or to present other appearances differing from those of the healthy secretion.

## CHAPTER V.

### BILE.

659*a*. The bile consists essentially of an aqueous solution of two salts, known as cholate (or glycocholate) and choleate (or taurocholate) of soda. It is generally, but not always, alkaline.

Bile is remarkable, among the secretions of the animal body, for the large proportion of carbon which it contains. Cholic acid ( $\text{HO}, \text{C}_{52}\text{H}_{42}\text{NO}_{11}$ ) contains 67 per cent., and choleic acid ( $\text{HO}, \text{C}_{52}\text{H}_{44}\text{NO}_{13}\text{S}_2$ ) 60.5 per cent. of carbon. There are also present in bile a quantity of mucus, to which it owes its viscosity, a peculiar colouring matter, and minute quantities of cholesterine, oleine, margarine, and lecithine (a phosphorised fat discovered in bile by Gobley,\* and also found in serum),

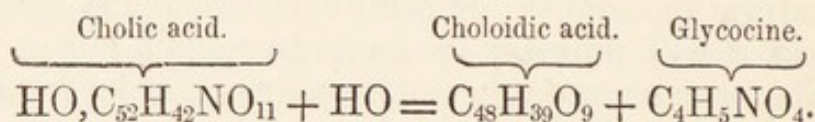
\* 'Journ. Pharm.', xxx, 241. Strecker has recently discovered lactic acid in bile, together with a new alkaline base—*Choline*  $\text{C}_{10}\text{H}_{13}\text{NO}_2$ .



together with choride of sodium, and alkaline and earthy phosphates.

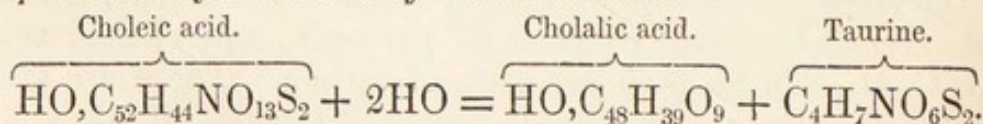
*Cholic acid.*—To isolate this acid, the bile is evaporated to dryness, the residue dried at 250° Fahr., and digested, in the cold, with absolute alcohol. The colouring matter is precipitated from the alcoholic solution by the gradual addition of ether, and the clear solution decanted from the deposit is mixed with more ether, when it gradually deposits tufts of needles consisting of the cholates of potash and soda. After having been washed with a mixture of absolute alcohol, with  $\frac{1}{10}$ th ether, the crystals are dried in vacuo, dissolved in a little water, and decomposed by dilute sulphuric acid, when cholic acid slowly separates in silky crystals, which are sparingly soluble in cold water, and in ether, but readily in alcohol.

When cholic acid is boiled with dilute hydrochloric acid, it assimilates the elements of water, and is decomposed into choloidic acid and glycocine (or sugar of gelatine):



*Choleic acid.*—This acid is not nearly so abundant in the bile as the preceding. In order to obtain it, the bile is mixed with water, and acetate of lead added to separate the mucus, the cholic, and the fatty acids. From the filtered liquid, the colour is removed by adding tribasic acetate of lead till the precipitate is white, and after filtration, the choleic acid is precipitated by adding more tribasic acetate of lead and ammonia. The lead salt is dissolved in alcohol, filtered, reprecipitated by water, and decomposed by sulphuretted hydrogen. The solution filtered from the sulphide of lead is evaporated, when it leaves the choleic acid.

When boiled with acids, choleic acid is decomposed, with the concurrence of the elements of water, into a new acid, and a peculiar crystalline body called taurine:



*Taurine*, which has also been found in the kidneys and in the lungs of the ox, is conspicuous among organic substances, for the large amount of sulphur which it contains (25.6 per cent.). It may be prepared in quantity by mixing bile with hydrochloric acid, filtering it from the mucus, and boiling for some



hours. The clear liquid having been poured off from the resinous deposit, is evaporated to a small bulk on the water-bath. The solution is drained from the crystals of chloride of sodium, and mixed with six volumes of alcohol. On standing, prismatic crystals of taurine are deposited, and may be recrystallized from water in order to purify them. It is insoluble in absolute alcohol and ether, and not very soluble in cold water. It may be identified by the odour of sulphurous acid when a crystal is heated on platinum foil.

*Colouring matter of bile.*—Two colouring matters have been obtained from bile, one of which, *biliverdine*, is easily soluble in alcohol, whilst *biliphæine* dissolves with difficulty. In order to separate them, Brücke recommends that the bile be well shaken with chloroform, which extracts the biliphæine. On evaporating the chloroform, and treating the residue with strong alcohol, the colouring matter is left in red crystals, which may be purified by washing with alcohol and ether. If biliphæine be dissolved in carbonate of soda, the solution oxidized by exposure to air, and neutralized with hydrochloric acid, a precipitate of biliverdine is obtained.

In Bright's disease, it is said that albumen and urea have been found in bile.

*Sugar-forming substance in the liver.*—If a fresh liver be cut into thin slices, heated with a small quantity of water, the solution filtered, evaporated to a small bulk, and mixed with a large excess of glacial acetic acid, a white flocculent precipitate is obtained which has the composition  $C_{12}H_{10}O_{10}$ \* (Kekulé). This substance, which resembles starch in some of its properties, as well as in its composition, has been called *animal amyloid*, *hepatine*, and *glycogene*. Like starch, it is converted into grape-sugar when boiled with dilute acids, but it gives a dark brown-red, instead of a blue colour, with iodine. It dissolves in water, giving a strongly opalescent fluid which is precipitated by alcohol. Sugar is also found in the decoction of liver, but it is doubtful whether it exists in the organ during life, or results from a *post-mortem* conversion of the glycogene. This substance has also been obtained in the milky fluid resulting from the injection of water into the liver in preparing it for the ordinary process of injection.

\* According to Pelouze,  $C_{12}H_{11}O_{11} + HO$ .



## CHAPTER VI.

## JUICE OF FLESH.

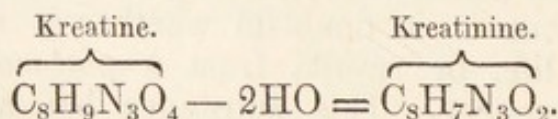
If a few pounds of finely divided flesh be digested for a short time in cold water, and afterwards well squeezed in a muslin bag, a reddish acid\* liquid is obtained, containing a little blood, together with the constituents of the *juice of flesh*, viz., albumen, kreatine, kreatinine, sarcine, inosite; lactic acid, butyric acid,† phosphoric acid, in combination with potash, lime, and magnesia; and chloride of potassium with a little chloride of sodium.

*Kreatine* ( $C_8H_9N_3O_4, 2Aq.$ ). To extract this substance, the above infusion is heated in a water-bath until the whole of the albumen is coagulated; it is then strained, and mixed with baryta-water until it is alkaline to tumeric paper. The precipitate (phosphates of baryta, lime, and magnesia) is filtered off, and the solution evaporated to a syrup. After standing for a few days, it will deposit prismatic crystals of kreatine, which may be purified by recrystallization, with the use of a little animal charcoal.

1000 parts of beef yield about 0.7 of kreatine, 1000 parts of cod-fish, 1.3 parts, and 1000 of fowl about 3 parts. Human flesh is said to be particularly rich in kreatine.

Kreatine has no alkaline reaction, but is capable of forming crystalline salts with acids. It dissolves in 75 parts of cold, and in much less boiling water. It is very slightly soluble in alcohol, and insoluble in ether.

A pure solution of kreatine will not give any precipitate with solution of chloride of zinc, but if the solution be previously boiled for some time, kreatinine is formed, which yields, with chloride of zinc, a granular crystalline precipitate:



The conversion is much accelerated by the addition of a little hydrochloric or sulphuric acid.

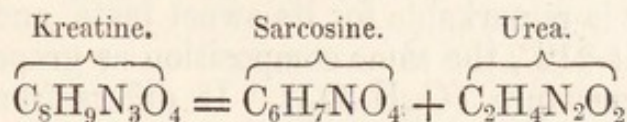
If kreatine be dissolved in water and boiled with 10 parts s

\* According to Du Bois Reymond, the juice of flesh is naturally alkaline, but it becomes acid very speedily after death.

† Scherer has also found formic and acetic acids in the juice of flesh.



of crystallized baryta, as long as any ammonia is disengaged, it yields a new base, *sarcosine*, which may be obtained in prismatic crystals by saturating the solution with carbonic acid gas, boiling to precipitate the excess of baryta, and evaporating the filtered liquid to a small bulk :



The urea, which is the other product of this decomposition of kreatine, is decomposed by the ebullition with baryta into carbonic acid and ammonia.

*Kreatinine*. If the liquid from which the crystals of kreatine were deposited be mixed with a strong solution of chloride of zinc, and set aside, crystals of the compound of kreatinine with chloride of zinc will be separated. The preparation and properties of kreatinine have been described under the head of urine (30a).

*Sarcine* ( $\text{C}_{10}\text{H}_{12}\text{N}_4\text{O}_2$ ). To obtain this base, the mother-liquor from the crystals of kreatine is diluted, and mixed with a dilute solution of acetate of copper. The precipitate is washed, suspended in water, and decomposed by sulphuretted hydrogen; after filtering from the sulphide of copper, the solution is gently heated on a water-bath to expel the excess of sulphuretted hydrogen, and boiled with hydrated oxide of lead to remove the colouring matter. After another filtration, sulphuretted hydrogen is again passed through the solution, the sulphide of lead filtered off, and the filtrate evaporated to a small bulk, when it deposits crystals of sarcine.

This substance, like kreatine, is a weak base, forming salts with acids. It is much less soluble in water than kreatine, requiring 300 parts of cold and 78 parts of boiling water. It is much more sparingly soluble in alcohol.

The composition of sarcine is the same as that of *hypoxanthine*, another basic substance which has been found in the spleen.

*Inosite* ( $\text{C}_{12}\text{H}_{12}\text{O}_{12}, 4\text{Aq}$ ). This substance, as well as *inosic acid* ( $\text{HO}, \text{C}_{10}\text{H}_6\text{N}_2\text{O}_{10}$ ) does not appear to be so invariably present in the juice of flesh as kreatine is.\* In order to extract it, if present, from the mother-liquor after the separation of the kreatine, dilute sulphuric acid is added, in quantity exactly sufficient to precipitate the baryta, and the filtered liquid

\* It was originally obtained from the heart, but has recently been found in the kidneys, liver, spleen, and lungs of the ox.



is well shaken with ether, which removes the lactic acid. To the aqueous liquid, alcohol is then added in successive portions; the first addition causes a precipitation of sulphate of potash and other salts, and after separating these and a further addition of alcohol, small crystals of inosite are deposited.

This body is remarkable for its sweet taste, and for having, when dried at  $212^{\circ}$ , the same composition as grape-sugar dried at that temperature ( $C_{12}H_{12}O_{12}$ ). It differs from that substance, however, in not reducing the oxide of copper in alkaline solution to the state of suboxide, and in not giving a brown solution when boiled with potash. Neither can it be made to undergo the vinous fermentation. Inosite is readily dissolved by water, but is insoluble in absolute alcohol and in ether. It is said to exist to the extent of  $\frac{3}{4}$  per cent. in unripe beans.

*Lactic acid* ( $2HO, C_{12}H_{10}O_{10}$ ). The lactic acid extracted, as described above, by ether, from the juice of flesh, is commonly called *sarco-lactic acid*, to distinguish it from ordinary lactic acid obtained by the fermentation of milk-sugar (621). Although the properties of both varieties of lactic acid in the free state are similar, their salts are not precisely so. The sarco-lactate of zinc has the formula,  $2ZnO, C_{12}H_{10}O_{10}, 4Aq.$ , whilst the ordinary lactate of zinc contains 6Aq. Again, the sarco-lactate of lime is  $2CaO, C_{12}H_{10}O_{10}, 8Aq.$  and the ordinary lactate, which is also the more soluble in water, contains 10Aq.

The lactic acid is obtained by evaporating the ethereal solution, as a syrupy acid liquid, which does not crystallize, and is best characterised by boiling it with a few zinc-filings, when the lactate of zinc will be formed, which deposits in sparingly soluble crystalline crusts.

*Butyric acid* ( $HO, C_4H_7O_2$ ). By acidulating the mother-liquor from the kreatine with sulphuric or hydrochloric acid, and distilling, a very dilute solution of butyric acid is obtained. The acid may be identified by its odour of rancid butter, and in order to obtain it in a pure state, the acid distillate from a large quantity of flesh must be neutralized with baryta and evaporated, when butyrate of baryta crystallizes out. By dissolving this in a little water and adding just enough sulphuric acid to precipitate the baryta, a concentrated solution of the acid may be obtained. By introducing into this, in a tube, fragments of fused chloride of calcium, the butyric acid is separated and rises, as an oily layer, to the surface, whence it may be drawn off, and purified by distillation with a little more chloride of calcium.



## CHAPTER VII.

## MUCUS.

## SECTION I.

*General Characters of Mucus.*

660. Healthy mucus, which is secreted by the mucous membrane with which the internal surfaces of the several parts of the body are covered, is a semi-fluid viscid substance, the general appearance of which is well known. It is sometimes so thin and limpid as almost to resemble water in appearance; while at others, and more commonly, it is tough and extremely tenacious, becoming stringy when attempted to be drawn out. When thin and watery, it is nearly transparent and colourless; the more viscid forms, however, being turbid or opaque, and usually of a pale yellowish or greyish colour. It is generally alkaline to test paper, insoluble in water, and somewhat heavier than that fluid; so that when placed in water it gradually sinks to the bottom, unless it is buoyed up by entangled air-bubbles. The mucus obtained from the several parts of the body differs considerably in appearance, and probably also in chemical composition. When dry it is hard and friable, resembling horn in appearance; the dry mass, on being digested in water, gradually swells up, and partially reassumes its former appearance.

661. When mucus is examined under the microscope, with a power of about 200 diameters, it is found to contain numerous round or oval granular corpuscles, together with epithelial scales (Fig. 5), entangled in a more or less viscid fluid, to which latter the peculiar tenacious character of mucus appears to be due. Mucus, therefore, consists of two distinct portions: the solid corpuscles with epithelial scales, and the fluid with which they are surrounded. Under favorable circumstances, and with a high magnifying power, the fluid portion appears to be filled with extremely minute molecular particles, the nature of which is not clearly understood.

662. The size of the mucus corpuscles varies considerably, the average diameter being about  $\frac{1}{2000}$ th of an inch. Their



surfaces are granular, similar to those of pus; and when treated with dilute acetic acid, the exterior covering loses its granular appearance, and becomes transparent, rendering visible from one to five internal nuclei. The same effect is produced by dilute oxalic and tartaric acids; but the dilute mineral acids cause little or no change.

663. Mucus appears to contain in its composition the following substances:—mucus corpuscles, epithelial scales, mucin, traces of extractive matters and fat, sometimes a small trace of albumen, and saline matters; which latter consist of alkaline chlorides and lactates, phosphate of lime, and traces of carbonate of soda. The *mucin*, to which the peculiar tenacious character of mucus appears to be due, is insoluble in pure water, and is probably held in solution in the fluid portion of the mucus, by the small excess of alkali usually present; it separates in the form of a white coagulum when mucus is treated with water, and still more completely when neutralized with dilute acetic acid. The minute traces of fat found in mucus probably exist in the corpuscles, though the exact chemical nature of these is by no means clearly ascertained.

## SECTION II.

### *Quantitative Analysis of Mucus.*

664. The quantitative determination of the principal constituents of mucus may be made in the following manner. The mucus intended for analysis is first divided into two portions, A and B; the first, A, being about one quarter, and the second, B, about three quarters, of the whole. Both portions are to be weighed in counterpoised capsules, that containing A being of platinum, and evaporated to dryness on a chloride-of-calcium bath, at a temperature of about  $220^{\circ}$ .

665. *Treatment of the portion A.*—This portion, after being dried until it ceases to lose weight, is to be accurately weighed. The weight of the dry residue gives the amount of SOLID MATTER in the quantity of mucus evaporated, while the loss represents the amount of WATER.

666. The proportion of these and the other ingredients, contained in 1000 parts of the mucus, may in each case be estimated by the following calculation:—



$$\left\{ \begin{array}{c} \text{Weight of} \\ \text{mucus} \\ \text{before eva-} \\ \text{poration.} \end{array} \right\} : \left\{ \begin{array}{c} \text{Wt. of each con-} \\ \text{stituent contained} \\ \text{in the quantity of} \\ \text{mucus employed.} \end{array} \right\} :: 1000 : \left\{ \begin{array}{c} \text{Proportion of that} \\ \text{constituent con-} \\ \text{tained in 1000 parts} \\ \text{of mucus.} \end{array} \right\}$$

667. The dry residue is then to be incinerated at a low red heat, until the ash becomes white, or nearly so. The weight of the ash will then represent the amount of SALINE MATTER in the quantity of mucus used; from which the proportion present in 1000 parts may be calculated as before (666).

668. *Treatment of the portion B.*—The dry residue left after evaporation (664), is to be removed from the capsule, and reduced to fine powder in a mortar. It is then boiled with successive small portions of ether, which will dissolve out the fat (545). The ethereal solution is evaporated to dryness on a water-bath, when the weight of the residue will indicate the amount of FAT in the quantity of mucus employed; from which the proportion in 1000 parts may be estimated as before (666).

669. The residue which proved insoluble in the ether (668) is to be boiled with a little alcohol, after which the alcoholic solution is to be evaporated to dryness, and the dry residue weighed. This is then incinerated, and the weight of the ash, deducted from that of the dry extract, will give the amount of ALCOHOL EXTRACTIVE, with the lactic acid of the lactates, in the quantity of mucus used; which may be corrected, as before, for 1000 parts (666).

670. The portion of the residue which proved insoluble in the alcohol (669) is to be dried and weighed; the weight indicating the amount of MUCIN, together with cellular matter, and probably traces of albumen, in the quantity of mucus employed; from which the proportion present in 1000 parts of mucus may be calculated, as in the former cases (666).

671. According to Nasse, the composition of fresh pulmonary mucus is as follows:

Water . . . . .	955.520
Solid constituents . . . . .	44.480
Mucin, with a little albumen . . . . .	23.754
Water extract . . . . .	8.006
Alcohol extract . . . . .	1.810
Fat . . . . .	2.887
Chloride of sodium . . . . .	5.825
Sulphate of soda . . . . .	0.400
Carbonate of soda . . . . .	0.198



Phosphate of soda . . . . .	0·080
Phosphate of potash, with traces of iron . . . . .	0·974
Carbonate of potash . . . . .	0·291
Silica, and sulphate of potash . . . . .	0·255

## SECTION III.

*Morbid Mucus.*

672. The characters of mucus secreted during disease are usually more or less different from those of the normal secretion, and an admixture of foreign matters frequently alters its appearance considerably. Pus, for instance, when mixed with it, diminishes its tenacity, owing to the mucin being present in smaller proportion (663); and when the liquid portion of mucus containing an admixture of pus is tested for albumen (254, 677), a considerable amount of that substance may usually be detected; since the *liquor puris*, or liquid portion of pus, contains a comparatively large quantity of albumen, but no mucin. Our means of detecting the presence of minute traces of pus in mucus are very imperfect; the decided presence of albumen in the purulent secretion is, indeed, almost the only test, since the microscopic characters of the corpuscles appear to be very similar (249).

673. The morbid mucus expectorated in pulmonary disease frequently contains, besides pus, red blood corpuscles, minute globules of fat, fragments of tuberculous matter, and other abnormal substances, most of which may generally be detected without difficulty under the microscope. The indications afforded by a careful microscopic examination of such expectorations, indeed, may often lead to results in diagnosis, of great importance to the practical physician.



## CHAPTER VIII.

## PUS.

## SECTION I.

*General Characters of Pus.*

674. Pus is the peculiar semi-fluid matter which is formed in abscesses, and in other kinds of wounds. In common language, a considerable variety of substances, more or less resembling each other in appearance, though differing in many respects, are included under the name of pus; and hence it has been found necessary to distinguish the normal secretion by the name of *true*, or *genuine pus*; the other substances being called *spurious*, or *false pus*.

675. Normal pus is a thick creamy-looking fluid, perfectly opaque, and usually of a pale yellow or greenish colour. It possesses little or no tenacity, and may consequently be poured in separate drops; in which respect it differs essentially from mucus, which, in colour and general appearance, it often much resembles. Its specific gravity is usually about 1030 or 1033, so that it sinks in water; and if shaken up with that fluid, it mixes uniformly with it. The mixture, after standing a short time, gradually deposits a sediment, consisting of pus-corpuscles (678). It is most commonly neutral to test paper, but is also occasionally met with slightly acid or alkaline.

676. Like mucus, pus consists of a clear fluid portion or serum, in which float innumerable minute granular corpuscles, which latter appear to be almost precisely the same as those contained in mucus, and when examined under the microscope, exhibit the same granular appearance. The liquid portion of pus, or *liquor puris*, however, differs essentially from that of mucus, and contains the following substances in solution, which, it will be seen, are nearly the same as those held in solution in the serum of the blood (568)—viz., albumen, together with a peculiar compound called pyin, or tritoxide of protein (which is soluble in water, and precipitated by acetic acid), fat, cholesterin, extractive matters, and inorganic salts.\*

\* Leucine ( $C_{12}H_{13}NO_4$ ) has also been found in pus.



These latter consist, for the most part, of chloride of sodium, with small quantities of phosphate, sulphate, and carbonate of soda; the chlorides of potassium and calcium; phosphates and carbonates of lime and magnesia; and traces of peroxide of iron.

677. The presence of these matters in the *liquor puris* may be shown by placing some pus in a tall narrow glass, and allowing it to stand, in order to give the corpuscles time to subside; after which, a little of the clear liquid may be drawn off with a pipette. On boiling a few drops of this in a test-tube, the albumen becomes coagulated, and separates from the liquid; after which the pyin may be thrown down in the form of a white flocculent precipitate, by adding a little acetic acid.\* The liquid may then, if necessary, be tested for the several inorganic salts above enumerated (676, 490).

678. The pus-corpuscles, though quite invisible to the naked eye, may be distinguished under the microscope with a magnifying power of from fifty to one hundred diameters; a con-

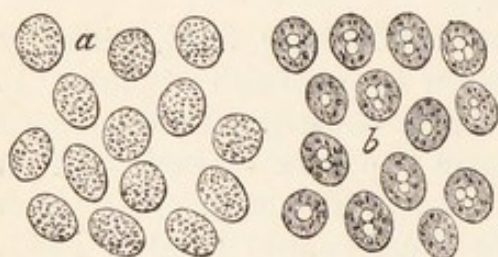


Fig. 74. Pus-corpuscles, magnified 400 diameters.

siderably higher power, however, is required for exhibiting their peculiar granular structure (Fig. 74, *a*). The size of these corpuscles varies considerably, being commonly about  $\frac{1}{2000}$ th of an inch in diameter. They are nearly spherical; and have a very pale yellowish colour,

which is scarcely perceptible, unless several of them are aggregated together. Being slightly heavier than the liquor puris with which they are surrounded, they gradually subside to the bottom, leaving the fluid portion nearly clear. Minute globules of fat may usually be detected, mixed with the corpuscles.

679. The pus-corpuscles, when treated with liquids of different densities, exhibit the phenomena of endosmosis and exosmosis, somewhat similar to those already described as taking place in the corpuscles of the blood (456); increasing in size when the external liquid, such as pure water, is of lower density, and collapsing when it is of higher density, than the

\* Pyin is precipitated by chloride of mercury and by acetate of lead, which is not the case with mucin.



fluid contained in them. When treated with dilute acetic acid, the external covering becomes transparent, and exhibits one or more internal nuclei (Fig. 74, *b*).

680. When mixed with a solution of ammonia or potash, pus loses its fluidity, and assumes a jelly-like appearance, which is highly characteristic, and is employed to distinguish it from mucus, which becomes less tenacious than before when treated with alkalies. A somewhat similar effect is produced also by the alkaline carbonates, and certain other salts.

681. Although the general appearance and characters of pus are usually sufficiently marked to enable us to identify it, it is always advisable, in cases where any doubt exists, to submit it to microscopical examination; since occasionally we meet with fluids containing a large quantity of epithelium and other products, which, in appearance, closely resemble pus, though differing entirely in composition from that substance, and containing no trace of the characteristic pus-corpuscles (678). The form of these corpuscles is found to vary considerably under certain pathological conditions; but there may generally be traced sufficient resemblance to the normal corpuscles to enable us to distinguish them from other matters. The modes of distinguishing between pus and mucus, have been already noticed in paragraphs 248, &c.

681a. *Blue Pus*.—In certain rare cases, the bandages upon which pus has been discharged assume a blue colour. By treating them with water, and agitating the aqueous solution with chloroform, Fordos has extracted a blue crystalline colouring matter, which he calls *pyocyamine*. It is soluble in water, alcohol, and ether; its colour is changed to red by acids, but the blue is restored by alkalies.

## SECTION II.

### *Quantitative Analysis of Pus.*

682. The quantitative analysis of pus may be made in the following manner:—Two portions of the fluid are to be weighed out; the first, A, in a small counterpoised flask; and the second, B, in a counterpoised or weighed evaporating dish.

683. *Treatment of the portion A*.—The portion A, after being weighed in a flask, is to be boiled with successive small quantities of strong or absolute alcohol, which must be separated while hot, either by filtration or decantation, from the insoluble



portion. The alcoholic solution is then set aside to cool, and allowed to stand a few hours, in order that the fat may, for the most part, crystallize out. The cold alcoholic liquid is then poured off, and the solid matter dried and weighed; when the weight thus obtained will represent the amount of FAT in the quantity of pus employed in the experiment.

684. The cold alcoholic liquid (683) is now to be evaporated to dryness, on the water-bath, in a counterpoised platinum capsule, and the dry residue, after being weighed, is incinerated. The weight of the ash is then ascertained, when the difference between the weight before and after incineration will represent the quantity of EXTRACTIVE MATTER (together with traces of fat which had not separated from the cold alcohol), in the portion of pus employed.

685. The residue which proved insoluble in the boiling alcohol (683), is to be dried on a water-bath, and then boiled with a little water, which will dissolve out the pyin, and at the same time cause the coagulation of the albumen. The aqueous solution thus obtained is to be separated from the insoluble portion; evaporated to dryness in a platinum capsule on a water-bath; and the weight of the dry residue having been noted, it is to be incinerated. The difference between the weight of the dry residue previous to incineration, and that of the inorganic ash, represents the amount of PYIN in the portion of pus used in the experiment.

686. The matter which remained insoluble in the hot water (685), is now to be dried and weighed. The dry residue is incinerated; and the loss of weight which it experiences during incineration will show the amount of ALBUMEN AND CORPUSCLES in the quantity of pus operated on.

687. *Treatment of the portion B.*—The weight of this portion having been noted, it is to be evaporated to dryness on a chloride-of-calcium bath, at a temperature of about 220°, the heat being continued until it ceases to lose weight on being weighed at intervals of half an hour or an hour. The loss of weight during the evaporation will then represent the proportion of WATER in the quantity of pus employed; while the weight of the dry residue shows the amount of SOLID MATTER.

688. The dry residue is now to be incinerated in a platinum capsule or crucible, until the ash becomes white or pale grey. The weight of the ash will then show the amount of inorganic SALINE MATTER in the quantity of pus used in the experiment.



689. The proportion of the several constituents contained in 1000 parts of pus, may then be estimated from the numbers obtained in the above experiments, by the following calculation :—

$$\left\{ \begin{array}{l} \text{Wt. of pus} \\ \text{used in the} \\ \text{experiment.} \end{array} \right\} : \left\{ \begin{array}{l} \text{Wt. of each} \\ \text{constituent} \\ \text{obtained.} \end{array} \right\} :: 1000 : \left\{ \begin{array}{l} \text{Proportion of that} \\ \text{constituent in} \\ \text{1000 parts of pus.} \end{array} \right\}$$

690. From the analysis of Dr. Wright, the composition of pus appears to be as follows :—

	Pus from a vomica.	Pus from a psoas abscess.	Pus from a mammary abscess.
Water . . .	894.4	885.2	879.4
Fatty matter . . .	17.5	28.8	26.5
Cholesterin . . .	5.4		
Mucus . . .	11.2	6.1	
Albumen . . .	68.5	63.7	83.6
Lactates, carbonates, and phosphates of soda, potash, and lime	9.7	13.5	8.9
Iron . . .	a trace.		
Loss . . .	3.3	2.7	1.6
	<u>1000.0</u>	<u>1000.0</u>	<u>1000.0</u>

## CHAPTER IX.

### BONE.

#### SECTION I.

#### *General Characters of Bone.*

691. The colour, texture, specific gravity, and general characters of bone, differ very much in different parts of the body ; and the proportions of the several chemical ingredients are also found to vary considerably. The two principal con-



stituents of bone are cartilage\* and phosphate of lime ( $3\text{CaO}, \text{PO}_5$ ); the proportion of the former being usually about 29 to 34 per cent., and that of the latter from 50 to 60 per cent. of the entire bone. The other substances, which are present in smaller quantity, are, carbonate of lime ( $\text{CaO}, \text{CO}_2$ ); phosphate of magnesia ( $3\text{MgO}, \text{PO}_5$ ); fluoride of calcium ( $\text{CaF}$ ); soluble soda salts, chiefly chloride of sodium; traces of the oxides of iron and manganese; and fat; which latter, however, does not belong strictly to the bone, but to the marrow contained in it. The presence of these several substances may be demonstrated by the following experiments.

692. The cartilaginous matter of bone may be obtained almost entirely free from the saline and other ingredients, by digesting a bone for a day or two, at a temperature not higher than about  $50^\circ$ , in dilute hydrochloric acid, composed of about one part of the strong acid and five parts of water. The earthy and saline matters gradually dissolve in the acid, leaving the cartilage unaffected, and still retaining the exact form of the bone. In this state it is soft and elastic; becoming, when dried, hard, somewhat brittle, and horny in appearance.

693. If the cartilage be boiled for some time in water, it will almost wholly dissolve, being converted into gelatine (the *glutin* of some writers), leaving undissolved nothing more than a delicate network of vessels. The aqueous solution thus obtained becomes, unless very dilute, gelatinous on cooling.

694. The fat may be separated by boiling a few fragments of crushed bone with ether, and evaporating the ethereal solution; when the fat will be left behind as a residue.

695. The phosphate of lime and phosphate of magnesia may be isolated by dissolving a fragment of calcined bone† in dilute hydrochloric acid, and supersaturating the acid solution with ammonia; which will throw down a white gelatinous precipitate of the mixed earthy phosphates. If this precipitate be examined under the microscope, it will be found to be chiefly composed of amorphous particles of phosphate of lime, mixed with a small quantity of the crystalline double phosphate of ammonia and magnesia ( $2\text{MgO}, \text{NH}_4\text{O}, \text{PO}_5 + 12\text{Aq}$ ), showing the presence of phosphate of magnesia.

696. The presence of carbonic acid (carbonate of lime) may

\* Called *osséine* by Frémy, who states that it contains the same proportions of carbon, hydrogen, nitrogen, and oxygen as gelatine, into which it is convertible with a facility which is inversely as the age of the animal.

† A large piece of bone may be burnt in a clear fire till perfectly white.



be proved by the effervescence which ensues when a fragment of uncalcined bone is moistened with dilute hydrochloric acid. If the solution, filtered from the precipitate of earthy phosphates (695), be tested with oxalate of ammonia, it will be found still to contain a considerable amount of lime, which existed in the bone as carbonate; since that portion only of the lime was precipitated by the ammonia, which was in combination with phosphoric acid.

697. If calcined bone, reduced to powder, be boiled for some little time in a test tube or glass flask, with a little rather dilute sulphuric acid, consisting of about equal parts of the strong acid and water, the inner surface of the glass will generally be found to be slightly corroded, owing to the disengagement of hydrofluoric acid (HF) by the action of the sulphuric acid on the fluoride of calcium.  $\text{CaF} + \text{H}_2\text{SO}_4 = \text{CaO} + \text{SO}_2 + \text{HF}$ . This substance, however, does not appear to be invariably present in bone, and some observers have been unable to detect it.

698. The presence of chloride of sodium may be shown by boiling a little calcined bone reduced to powder with water, filtering from the insoluble earthy portion, and testing a few drops of the aqueous solution with nitrate of silver, which will give an abundant precipitate of the chloride (AgCl). By concentrating the rest of the solution to a small bulk and testing it on platinum wire in the blowpipe flame, a yellow colour will appear, showing the presence of soda.

699. A little sulphate of soda may also be detected, by means of chloride of barium, in the soluble portion of calcined bone, though no trace of sulphuric acid is to be found in it previous to calcination; being produced, during ignition, by the oxidation of the sulphur contained in the tissues.

## SECTION II.

### *Quantitative Analysis of Bone.*

700. About three hundred grains of the bone intended for analysis should be first cleaned from adhering fat, periosteum, and other impurities, and then reduced to tolerably small fragments either by crushing or rasping.

701. *Treatment of the first portion*—One hundred grains of the bone are to be dried in a counterpoised platinum capsule or crucible, on a chloride-of-calcium bath, at a temperature of about  $250^\circ$ , until it ceases to lose weight on being weighed at



intervals of half an hour or an hour. The loss of weight which it experiences during desiccation represents the per-centage of **WATER**.

702. The dry mass is now to be incinerated in the capsule at a low red heat, until the whole of the organic matter is burnt away, and the ash becomes throughout perfectly white. The weight of this ash gives the per-centage of **INORGANIC MATTER** contained in the bone; while the loss during incineration represents the per-centage of **ORGANIC MATTER**. The inorganic residue may then be digested in dilute hydrochloric acid, and retained for subsequent examination (706).

703. *Treatment of the second portion.*—A second portion of the crushed or rasped bone, weighing one hundred grains, is to be digested for a day or two, in cold dilute hydrochloric acid, containing one part of the strong acid to five or six of water; the whole being kept at a temperature not higher than about  $50^{\circ}$ , as otherwise some traces of the animal matter of the bone would be acted upon by the acid. The whole, or at least by far the greater portion of the inorganic matter is thus dissolved, and when the acid liquid has been well washed out of the insoluble residue by means of cold water, little will remain but the cartilaginous matter of the bone.

704. The cartilaginous residue is to be dried on a water-bath, and then boiled with a little ether, which must be poured off, and renewed if necessary, until all the fat is dissolved. The ethereal solution is then evaporated to dryness in a counterpoised capsule on a water-bath; when the weight of residue will give the per-centage of **FAT** in the bone.

705. The matter which proved insoluble in the ether (704), consisting chiefly of cartilage, with traces of inorganic matter, is now to be dried on a chloride-of-calcium bath, at a temperature of about  $250^{\circ}$ , weighed and incinerated. The difference between the weight of the dry residue before and after incineration, will then represent the per-centage of **CARTILAGE** in the bone.

706. The ash left after the incineration of the first hundred grains of bone (702), is now to be dissolved in moderately dilute hydrochloric acid; a gentle heat being applied if necessary. The acid solution is then slightly supersaturated with ammonia, which will throw down the phosphate of lime, together with the small quantity of phosphate of magnesia and fluoride of calcium; as well as any traces of peroxide of iron and oxide of manganese that may be present. The precipitate



is to be well washed, filtered, dried, and ignited; after which its weight will represent the amount of BONE EARTH, consisting of PHOSPHATE OF LIME with PHOSPHATE OF MAGNESIA, and FLUORIDE OF CALCIUM, in one hundred parts of the bone.

707. If it is required to determine separately the proportion of phosphate of magnesia, the ignited precipitate (706), after being weighed, is to be redissolved in dilute hydrochloric acid; the acid solution is then mixed with an excess of perchloride of iron ( $Fe_2Cl_3$ ), and supersaturated with ammonia. The phosphoric acid of the earthy phosphates is thus precipitated in combination with peroxide of iron, together with any excess of uncombined peroxide of iron, leaving in solution the chlorides of calcium and magnesium.\* The lime (chloride of calcium) is first precipitated by adding oxalate of ammonia ( $NH_4O, C_2O_3$ ) as long as it causes a precipitate, boiling the mixture, and filtering. The filtered solution is then concentrated by evaporation, and the magnesia thrown down by adding phosphate of soda ( $2NaO, HO, PO_5$ ) and a decided excess of ammonia. The mixture is allowed to stand for some hours, after which the precipitated double phosphate of ammonia and magnesia ( $2MgO, NH_4O, PO_5 + 12Aq$ ) is to be filtered, dried, and ignited, by which it is converted into phosphate of magnesia ( $2MgO, PO_5$ ), and weighed. This weight will represent the amount of PHOSPHATE OF MAGNESIA in the 100 grains of bone; which, when deducted from the whole of the earthy phosphates (706), will give the per-centage of PHOSPHATE OF LIME.

707a. *General method of determining phosphoric acid.*—Since phosphoric acid is a constant component of the inorganic part of the solids and fluids of the body, it is necessary to be able to determine it directly in all cases. The opportunity may be taken of applying the following process (Chancel) to the determination of phosphoric acid in bone. Ten grains of the bone-ash are dissolved in nitric acid, with the aid of heat. The solution is diluted and a little nitrate of baryta added, to remove sulphuric acid; nitrate of silver is then added, without previous filtration, to precipitate the chlorine. The filtered solution is treated with sulphuretted hydrogen, and the liquid filtered from the sulphide of silver is heated until no more sulphuretted hydrogen is perceptible. A solution of nitrate of bismuth ( $BiO_3, 3NO_5$ ) is then added, and the precipitate of

\* The process described in paragraphs 68-71 will give more exact results.



phosphate of bismuth ( $\text{BiO}_3, \text{PO}_5$ ) is collected upon a filter, washed with boiling water, ignited in a porcelain crucible (the filter being burnt separately) and weighed. The following proportion then gives the phosphoric acid :

$$\begin{array}{ccc} \text{Atc. wt. of} & & \text{Atc. wt. of} \\ \text{phosphate of} & & \text{phosphoric} \\ \text{bismuth.} & & \text{acid.} \\ \hline 305 & : & 71 \end{array} :: \text{Weight of precipitate} : x$$

708. The solution filtered from the precipitate of earthy phosphates (706), containing the portion of lime which existed in the bone as carbonate, is now to be treated with oxalate of ammonia as long as any precipitate is produced. The whole of the lime is thus separated as oxalate ( $\text{CaO}, \text{C}_2\text{O}_3 + 2\text{Aq}$ ), which, after boiling the mixture, is filtered, dried, and ignited.\* The oxalate is converted, during the ignition, into carbonate ( $\text{CaO}, \text{CO}_2$ ), so that the weight of the ignited precipitate will represent the amount of CARBONATE OF LIME in the hundred grains of bone.

709. As a check upon the estimation of the carbonate of lime, the amount of carbonic acid in the bone may be deter-

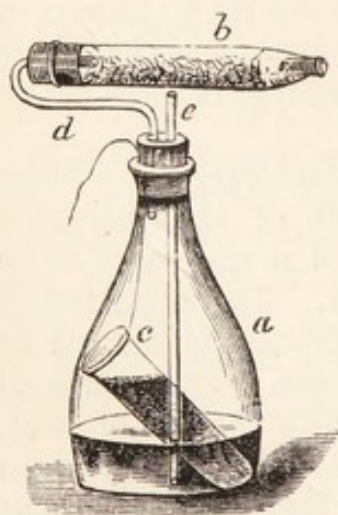


Fig. 75.

mined by placing 100 grains of the unburnt bone in fine powder, in a flask *a*, provided with a desiccating tube *b*, containing fragments of chloride of calcium (Fig. 75). A test-tube (*c*) containing hydrochloric acid is then placed in the flask, and the whole apparatus is weighed; after which the acid is allowed to flow gradually upon the powder, from which it will expel the CARBONIC ACID. The amount of the latter which, being gaseous, escapes in a dry state through the chloride of calcium tube *d*, is then represented by the loss of weight which the apparatus with its contents

experiences during the experiment (337). It will probably be found that the carbonic acid thus determined, bears to the carbonate of lime (708) the proportion of 22 to 50, those

\* See paragraph 69.



being the atomic weights of carbonic acid and carbonate of lime respectively.\*

710. The solution filtered from the oxalate of lime (708), which contains the soluble salts (chiefly chloride of sodium), together with the excess of oxalate of ammonia employed to precipitate the lime, is now to be evaporated to dryness, and the residue ignited in order to expel the ammoniacal salts. The weight of the residue after ignition will then represent the per-centage of SOLUBLE SALINE MATTER.

711. The following analyses will serve to illustrate the per-centage composition of bone both of man and some of the lower animals.

*Analysis I (Von Bibra).*

*Showing the Composition of the Bones of a Child two months old.*

	Tibia.	Ulna.
Phosphate of lime, with a little fluoride of calcium . . . . .	57.54	56.35
Carbonate of lime . . . . .	6.02	6.07
Phosphate of magnesia . . . . .	1.03	1.00
Soluble salts . . . . .	0.73	1.65
Cartilage† . . . . .	33.86	34.92
Fat . . . . .	0.82	1.01

*Analysis II (Von Bibra).*

*Composition of the Bones of a Middle-aged Man.*

	Femur.	Tibia.	Humerus.	Costa.
Phosphate of lime with a little fluoride of calcium . . . . .	59.63	58.95	59.87	55.66

To obtain an exact result by this process, the flask should be furnished with a narrow tube *e*, dipping into the liquid, kept closed during the evolution of gas; at the conclusion of the experiment the tube should be opened and air slowly sucked through the drying-tube *b* as long as the weight of the apparatus diminishes.

According to Frémy, the per-centage composition of this portion of bone is

Carbon . . . . .	49.21
Hydrogen . . . . .	6.50
Nitrogen . . . . .	17.86
Oxygen . . . . .	25.14



	Femur.	Tibia.	Humerus.	Costa.
Carbonate of lime . }	7.33	7.08	7.76	6.644
Phosphate of magnesia . }	1.32	1.30	1.09	1.077
Soluble salts .	0.69	0.70	0.72	0.629
Cartilage .	29.70	30.42	29.28	33.977
Fat . .	1.33	1.55	1.28	2.044

*Analysis III (Berzelius).*

*Composition of Human Bone.*

Phosphate of lime . . . . .	51.04
Fluoride of calcium . . . . .	2.00
Carbonate of lime . . . . .	11.36
Phosphate of magnesia . . . . .	1.16
Soda, with a little chloride of sodium . . . . .	1.20
Cartilage . . . . .	32.12
Vessels . . . . .	1.13

*Analysis IV (Von Bibra).*

*Composition of the Bones of the Lower Animals.*

	Femur of sheep aged 4 years.	Femur of bull aged 4 years.	Femur of horse aged 6 years.	Humerus of cat aged 6 years.
Phosphate of lime with a little fluoride of calcium . }	55.94	54.07	54.37	59.30
Carbonate of lime . }	12.18	12.71	12.00	10.69
Phosphate of magnesia . }	1.00	1.42	1.83	1.70
Soluble salts .	0.50	0.80	0.70	0.40
Cartilage .	29.68	29.09	27.99	27.21
Fat . .	0.70	1.91	3.11	0.70



	Vertebræ of dolphin.	Humerus of thrush.	Vertebræ of snake.	Vertebræ of salmon.
Phosphate of lime with a little fluoride of calcium.	52.51	62.65	59.41	36.64
Carbonate of lime.	9.37	6.05	7.82	1.01
Phosphate of magnesia.	0.98	0.90	1.00	0.70
Soluble salts.	1.24	0.84	0.73	0.83
Cartilage.	33.97	28.02	24.93	21.80
Fat.	.	1.54	6.11	38.82

## SECTION III.

*Morbid Bone.*

712. Certain diseases are found to be always accompanied by remarkable changes in the chemical composition of the bones; the earthy matters being sometimes so deficient, that they no longer possess the rigidity and strength necessary for sustaining the weight of the body. Other variations also are occasionally met with, a few examples of which are subjoined, the composition being calculated upon 100 parts.

*Analyses of the Tibiæ of three Rachitic Children. (Lehmann.)*

	I.	II.	III.
Phosphate of lime	32.04	26.94	28.13
Carbonate of lime	4.01	4.88	3.75
Phosphate of magnesia	0.98	0.81	0.87
Chloride of sodium	0.21	0.27	0.28
Soda	0.54	0.81	0.73
Cartilage	54.14	60.14	58.77
Fat	5.84	6.22	6.94

*Analyses of Bone in Osteomalacia. (Prösch.)*

	Vertebra.	Costa.
Phosphate of lime	13.25	33.66
Carbonate of lime	5.95	4.60
Sulphate of lime and phosphate of soda	0.90	0.40
Cartilage	74.64	49.77
Fat	5.26	11.63



*Analyses of Carious Bone. (Valentin.)*

	Vertebrae of a man aged 20.	
Phosphate of lime . . . .	33.914	34.3833
Carbonate of lime . . . .	7.602	6.6366
Phosphate of magnesia . . .	0.389	1.1822
Chloride of sodium . . . .	3.157	1.9199
Carbonate of soda . . . .	0.118	
Organic constituents . . . .	54.830	55.8800

*Analysis of Necrotic Bone of a Middle-aged Man.  
(Von Bibra.)*

Phosphate of lime with a little fluoride of calcium . . . .	72.633
Carbonate of lime . . . . .	4.033
Phosphate of magnesia . . . . .	1.933
Soluble salts . . . . .	0.611
Cartilage . . . . .	19.588
Fat . . . . .	1.222

## CHAPTER X.

## EXAMINATION OF MIXED ANIMAL FLUIDS.

713. On account of the great number and variety of organic substances which may enter into the composition of such a mixture as we are now considering, it is altogether impossible to lay down any general and consecutive scheme of experiments, which shall comprise all even of the more commonly occurring organic compounds. All that I shall attempt, therefore, in the present chapter, is to describe very briefly the methods of detecting the presence of a few of the substances which are most frequently met with in organic liquids, and which are of the most practical importance to the pathologists and the physician.

714. The colour, consistence, and general appearance of the fluid, should be first carefully observed, as the presence of many substances, such as blood, mucus, fat, fibre, &c., may often be readily detected, even with the naked eye. Should any solid or semi-solid matter be held in suspension in the



liquid, or be found as a sediment at the bottom, it should be separated, either by decantation, or by filtering through fine muslin or paper.

715. The matters thus separated from the fluid may be reserved for examination under the microscope, and also, if necessary, with other tests. The following substances, among others, may in this way be readily detected :—muscular fibre and other organized tissues; epithelium (328); mucus and pus granules (329); fat and milk globules (325, 632, 633); infusoria of several kinds; besides various amorphous and crystalline substances, many of which may at once be recognised by their peculiar form and appearance (315—332, &c.).

716. The liquid may first be tested with litmus and turmeric paper, since the behaviour of several of the substances about to be noticed, with reagents, will be found to vary according as the liquid containing them is acid, alkaline, or neutral.

717. The specific gravity may also be ascertained, when it can conveniently be done, as a knowledge of the density of the fluid will serve to furnish some indication of the amount of solid matter held in solution (279).

### *Fibrin.*

718. When fibrin, in the soluble state, is contained in a liquid, it gradually undergoes spontaneous coagulation, and separates from the solution, forming a more or less firm coagulum or jelly: and if other matters are held in suspension in the liquid previous to the coagulation, they are usually entangled in it,—a familiar instance of which is afforded by the coagulation of blood (473). The more important peculiarities of fibrin have been already noticed in paragraphs 472 to 481.

### *Albumen.*

719. When albumen is suspected to be present in solution, the clear liquid is to be gently boiled for a few minutes; if coagulation takes place, and if the precipitate thus occasioned does not disappear on the addition of a few drops of nitric acid, albumen is present. If the mixture is alkaline, it should be neutralized with nitric acid previous to boiling, since any excess of alkali would tend to retain the albumen in solution, and thus



prevent the coagulation. For further particulars respecting albumen, and its behaviour with reagents, *see* paragraphs 133, 235, 466, &c.

### *Casein.*

720. Casein may be recognised by its forming a white curdy precipitate, when the solution containing it is neutralized or very slightly supersaturated with acetic acid. It redissolves, however, if the acid be added in decided excess. If the liquid is slightly acid to test paper, casein hardly need be looked for, since it is not soluble in acid solutions, unless the acid is present in considerable excess. It may be distinguished from albumen by not coagulating when heated; it forms, however, a thin insoluble pellicle on the surface when exposed to the air while hot—of which a familiar example is afforded in the *skin* of boiled milk. If casein be dissolved in acetic or any other acid, it is precipitated on the addition of ferrocyanide of potassium, thus resembling the other modifications of protein (625).

### *Pyin.*

721. This substance, which appears to be identical with the so-called tritoxide of protein,\* and is consequently closely allied to the other protein compounds (472), may be recognised by its throwing down a precipitate with acetic acid, which does not redissolve in an excess of the acid. A solution of alum also causes a white precipitate, insoluble in excess; in which respect pyin differs from gluten and chondrin (725, 726). Unlike most of the protein compounds, it is not precipitated by ferrocyanide of potassium.

### *Pus.*

722. When pyin has been detected in a liquid, it is not improbable that, on examination with the microscope, the peculiar pus granules (678) will also be found to be present, since pyin is one of the characteristic constituents of the fluid portion of pus (676). The principal characters of this substance, together with the modes of its detection, have been already described in paragraphs 153, 247, 674, &c.

\* This name was conferred by Mulder upon the soluble substance obtained by boiling any of the protein compounds for several hours with water.



*Mucus.*

723. If much mucus is present, it gives to the mixture a more or less tenacious and ropy consistence, which is very characteristic. Under the microscope the peculiar mucus corpuscles, as well as the fragments of epithelium which usually accompany them, will also probably be apparent (Fig. 5); and these, in conjunction with the ropiness above alluded to, are generally sufficient evidence of the existence of mucus. When present only in minute quantity, and especially when mixed with pus, it is often extremely difficult, if not impossible, to identify it with any degree of certainty. (See also paragraphs 31, 99, 210, 660, &c.)

*Gelatine ; Chondrin.*

724. These substances, which are formed by boiling the cartilaginous tissues in water, closely resemble each other in many respects; and their hot aqueous solutions become gelatinous on cooling. Glue, isinglass, and the several varieties of gelatine, met with in commerce, are all modifications of these principles. Both gelatine and chondrin are immediately precipitated, even in very dilute solutions, by a solution of tannin. They are not precipitated by ferrocyanide of potassium; in which respect they differ from the protein compounds. They are thrown down from their strong solutions by alcohol, in the form of a white tenacious precipitate; and creosote causes their solutions to become turbid and gelatinous.

725. *Gelatine*, which is obtained by boiling in water for some hours the cartilage of bone, tendons, skin, &c., is characterised by giving with acetic acid a very slight precipitate, which readily redissolves in an excess of the acid. A solution of alum gives with gelatine no precipitate; or if a slight opalescence is occasioned, it disappears on the addition of a further quantity of the precipitant.

726. *Chondrin*, on the other hand, which is formed by boiling in water any of the permanent cartilages, as those of the larynx, ribs, &c., is immediately precipitated by acetic acid, and an excess of the acid does not redissolve it. Alum, too, causes a precipitate, which, however, readily dissolves when the salt is added in excess. The solubility of chondrin in a solution of alum serves to distinguish it from pyin (721).



*Blood.*

727. The colour which it imparts to any liquid with which it is mixed, is usually almost sufficient evidence of the presence of blood, unless the quantity is very small. The red corpuscles may also, in most cases, be detected under the microscope, more or less altered in form and size by the action of the fluid in which they float (456, 583). When blood is present, albumen also will be found dissolved in the liquid, unless it has been previously coagulated by heat or otherwise; it may be detected by the application of heat, and nitric acid, in the manner described in paragraphs 235, 236, &c.

*Biliary Matter.*

728. Biliary matter, if present in any considerable quantity, generally communicates a more or less decided brown or yellowish colour to the liquid, and also a peculiar bitter taste. It may be identified by means of Heller's and Pettenkofer's tests, described in paragraphs 149 & 151. If these fail to detect it in the fluid, a little of the latter may be evaporated nearly to dryness on a water-bath, and a strong aqueous solution of the residue tested as before.

*Urea.*

729. This substance may be detected in organic liquids in the following manner:—The portion of the organic mixture intended for the examination, is evaporated to dryness at a gentle heat on a water-bath, and the dry residue treated with alcohol, which will dissolve out any urea that may be present, together, probably, with some other of the matters with which it is associated. The alcoholic solution is then evaporated to dryness, and the dry extract digested with a very small quantity of moderately warm water; which will readily dissolve out the urea. The aqueous solution thus obtained is then mixed, after filtering, with pure nitric acid, in the manner described in paragraph 16, and then cooled by means of a freezing mixture; when, if urea is present, delicate crystals of the nitrate (Fig. 2) will gradually appear.\* When the quantity of urea is very small, the microscope may be employed to detect any traces of

\* Urea may be detected in this manner in blood, chyle, and lymph, as well as in urine.



the crystalline nitrate, and some other precautions must be observed, which have been described in paragraphs 181, 184, 341, &c.

### *Kreatine.*

729*a*. For the detection of kreatine in an organic mixture, the solid portion should be divided as finely as possible, the whole dried upon the water-bath, and digested with hot alcohol. The alcoholic solution having been pressed out, is evaporated to dryness on the water-bath, the residue treated with water, the solution precipitated by acetate of lead (not added in excess) and filtered. The filtrate is saturated with sulphuretted hydrogen, to precipitate the lead, and the solution separated from the sulphide of lead is evaporated to a syrup and set aside for some days, when kreatine will crystallize out and may be recognised by the characters described above, especially by converting it into kreatinine and obtaining the crystalline precipitate with chloride of zinc.

### *Inosite.*

729*b*. The presence of inosite may be ascertained (in an aqueous infusion of bullock's lung, for example), by acidulating with acetic acid, coagulating the albumen by heat, and precipitating the filtered liquid with acetate of lead. The filtrate from this precipitate is mixed with tribasic acetate of lead, which will throw down any inosite if present. The precipitate is suspended in water, decomposed by sulphuretted hydrogen, the liquid filtered from the sulphide of lead, evaporated to a very small bulk, mixed with four volumes of boiling alcohol, filtered if necessary, and set aside for twenty-four hours. If no crystals of inosite have then been deposited, ether is added by degrees until a permanent milkiness is produced on agitation. The liquid is again set aside, when inosite will crystallize out, and may be identified by treating it with ammonia and chloride of calcium, and slowly evaporating to dryness, when a rose-colour will be produced.

### *Fat.*

730. When any considerable amount of fatty matter is present in an aqueous mixture, it may be readily detected with the naked eye, and still more delicately under the micro-



scope, by the appearance of oily or fatty globules floating on the surface. When, however, the quantity is very small, or, owing to other circumstances, no appearance of fat is to be seen; a little of the mixture suspected to contain it, is to be evaporated nearly to dryness on a water-bath, and the residue digested with a little warm ether, which will readily dissolve any traces of fatty matter that may be present. On evaporating the ethereal solution on a water-bath, the oil or fat will be left as a residue, and may be identified by its possessing the well-known physical characters of fatty matters (158).

731. The saponifiable fats most commonly met with in animal fluids are, oleine ( $C_{114}H_{104}O_{12}$ ), stearine ( $C_{114}H_{110}O_{12}$ ), margarine ( $C_{108}H_{104}O_{12}$ ), and butyrine. The degree of hardness or of oiliness, and the temperature to which the fatty matter requires to be raised before it melts, serve to furnish some indication as to the relative amounts of the solid stearine and the oily oleine. Butyrine may generally be detected by the peculiar smell which it gradually acquires, resembling that of rancid butter.

#### *Cholesterin and Serolin.*

732. If either of these substances are present, they will have been dissolved by the ether (730), together with any other fatty matters that may be contained in the liquid. They may be separated from the other fats by digestion with a solution of potash, which will dissolve out the saponifiable fats, and leave the cholesterin and serolin unaffected (596). These may be distinguished from each other by their different fusing points, that of cholesterin being  $293^{\circ}$ , while that of serolin is as low as  $97^{\circ}$ .

#### *Milk.*

733. The well-known physical characters of milk are generally sufficiently apparent to lead to its detection, unless largely diluted with other matters. When any doubt exists as to its presence, a drop of the liquid may be examined under the microscope for the milk globules (632); and the clear liquid, after filtration, may be tested with acetic acid for casein (623); the existence of which, in any fluid, is strong evidence of the presence of milk. The residue left by evaporating the liquid to dryness, may be tested for fat also, by



digestion with warm ether, and evaporating the ethereal solution on a water-bath (730).

### *Sugar.*

734. The most convenient test for the presence of sugar is that known as Trommer's, which has already been fully described in paragraphs 122 to 124. Maumené's (125), and the fermentation test (128), may also, in many cases, be employed with advantage; and, indeed, it is always more satisfactory to confirm the results of Trommer's experiment, by applying also the fermentation test; since the suboxide of copper may be sometimes produced by certain other organic substances, even when no sugar is present. If the sugar is present only in very minute quantity, it may be advisable to evaporate the liquid to dryness on a water-bath, and re-dissolve the soluble portion of the residue, including the sugar, in a small quantity of hot water, in the manner described in the process for detecting sugar in the blood (606). The strong aqueous solution may then be examined by Trommer's, Maumené's, and the other tests.

735. When cane sugar is suspected to be present, the solution should first be boiled for a few minutes with dilute sulphuric acid before the application of Trommer's test, in order to convert it into grape sugar; since the cane variety does not otherwise produce the same characteristic results.

### *Ammonia.*

736. This substance, which is so constantly to be met with in animal fluids, as one of the results of the decomposition of nitrogenous compounds, may be readily detected, even when present in very small quantities. A portion of the liquid is mixed in a test tube with a little caustic potash, or, still better, with caustic baryta (note to 38), and warmed. The ammonia, if present, is thus disengaged, and may be detected by the smell, or, still more delicately, by holding at the mouth of the tube a glass rod moistened with dilute hydrochloric acid, when white fumes of chloride of ammonium will be distinctly visible.

737. If the ammonia is present only in minute quantity, a little of the suspected liquid may be mixed with a few drops of dilute sulphuric acid, in order to fix the ammonia, and then concentrated by evaporation at a gentle heat on a water-bath; the concentrated liquid may then be supersaturated with potash or baryta, and examined in the manner above described.



*Uric (or Lithic) Acid.*

738. When an organic mixture is suspected to contain uric acid, it may, if free from albuminous matter, be acidified with a few drops of hydrochloric acid, and allowed to stand a short time. The uric acid will gradually separate in the form of minute crystals (20), which may be examined under the microscope, and also tested with nitric acid and ammonia, in the manner described in paragraph 23. If any albuminous matter is mixed with the liquid, the latter is to be evaporated to dryness on a water-bath, and the residue digested with a dilute solution of caustic potash. The alkaline solution is then supersaturated with a decided excess of hydrochloric acid, which will throw down the uric acid in the form of a crystalline precipitate. If the quantity is small, a drop of the liquid may be mixed with the acid on a strip of glass, and examined for the characteristic crystals under the microscope (318).

738*a*. Another process, which is sometimes more convenient than the above, consists in mixing the solution with acetate of lead as long as it yields a fresh precipitate, filtering, and adding tribasic acetate of lead. On standing, urate of lead will be precipitated, which must be washed, suspended in water, and decomposed by sulphuretted hydrogen. The solution is boiled, filtered from the sulphide of lead, and evaporated to a small bulk. On cooling, crystals of uric acid are deposited.

739. The principal characters of uric acid, and the methods of detecting and estimating it in the urine, have been already noticed in the several chapters of Part I.

*Systematic Examination of Mixed Fluids for the Proximate Constituents of Animal Bodies.*

Some of the proximate constituents of the solids and fluids of the body, such as fibrine and gelatine, are recognisable by their characteristic external appearance, but the greater number give no indication of their existence unless specially sought for, so that it is desirable to follow some systematic plan of examination in order that they may not be overlooked. The solid organs must be divided as minutely as possible, and digested for some time in tepid water.

The reaction of the liquid to test-papers having been recorded, it is acidified, if necessary, with acetic acid, and heated on a water-bath in order to coagulate *albumen*, which is then filtered off. On allowing the hot filtrate to stand for



some time, *uric acid* will crystallize out as it cools. The solution is then very nearly neutralized with potash, and acetate of lead added, to precipitate sulphuric and phosphoric acids, as well as colouring and extractive matters; the filtered liquid is mixed with an excess of tribasic acetate of lead. The precipitate, which may contain *inosite*, is decomposed by sulphuretted hydrogen, and the solution evaporated till a portion becomes permanently turbid when mixed with alcohol, the whole is then mixed with an equal volume of alcohol, heated till the turbidity disappears, and set aside for a day or two that the inosite may crystallize out.

The filtrate from the precipitate produced by tribasic acetate of lead is now mixed with a slight excess of ammonia, and any precipitate is collected, decomposed by sulphuretted hydrogen, and the solution, after evaporation, examined for *sugar*.

The liquid filtered from the precipitate produced by ammonia is treated with sulphuretted hydrogen, filtered from the sulphide of lead, evaporated to a small bulk upon the water-bath, and tested, with oxalic acid (14), for *urea*.

The solution is then evaporated to a syrup, and mixed with moderately strong alcohol to precipitate the oxalates of the alkalies. The alcohol is evaporated, the excess of oxalic acid removed by the careful addition of lime-water, and the filtered solution evaporated till an equal volume of absolute alcohol renders it permanently turbid. It is then mixed with alcohol and set aside for a day or two, when *taurine* will be deposited in crystals.

The alcoholic solution is again evaporated on the water-bath, and if the residue is much coloured, it may be dissolved in a little water and boiled with hydrated oxide of lead, the filtered solution being afterwards treated with hydrosulphuric acid to remove the lead, and evaporated to a syrup. On allowing this to stand for some time, *kreatine* and *leucine*\* may crystallize out. They may be washed with cold absolute alcohol, and identified, the kreatine by its convertibility into kreatinine, and the leucine by the woolly sublimate which it furnishes when heated in a tube. The mother liquor from these may contain *kreatinine*, recognisable by its behaviour with chloride of zinc.

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\* Leucine ( $C_{12}H_{13}NO_4$ ) and tyrosine ( $C_{13}H_{11}NO_6$ ) have been detected in the spleen, pancreas, and liver, and the former also in the lungs, and both have been observed in the urine in disease. Tyrosine is identified by digesting it with sulphuric acid, diluting with water, heating the solution with carbonate of lime, filtering, and adding perchloride of iron, which produces a violet colour. Proto-nitrate of mercury gives a red precipitate and a pink liquid when added to a solution of tyrosine.



# PART V.

## THE DETECTION OF POISONS IN ORGANIC MIXTURES, &c.

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### CHAPTER I.

#### ARSENIC.

It is impossible to insist too strongly on the necessity for the most careful examination into the purity of all the substances employed in the detection of poisons. No evidence with respect to the presence of a poisonous substance can be regarded as *perfectly* conclusive, unless the reagents employed have been tested, in the same quantities, and by the same processes, as were employed in detecting the poison, without affording any indication of its presence.

Moreover, if any particular substance, though not a poison, be detected in the course of the examination, with all the bearings of which upon the tests employed the investigator is not perfectly familiar, the same series of operations should be conducted with that substance, to ascertain whether it might lead to error.

740. Although all, or nearly all, the compounds of arsenic appear to be more or less intensely poisonous, I shall here allude especially to the detection of arsenious acid ( $\text{AsO}_3$ ); since in the vast majority of cases in which arsenic is taken, whether criminally or accidentally, it is in the form of arsenious acid, or, as it is often called, oxide of arsenic, or white arsenic. The experiments which I am about to describe will serve, however, for the most part, equally well for identifying the presence of arsenic in other forms of combination than that of arsenious acid; so that, if the processes are carefully conducted, the risk of any traces of the metal escaping detection is very small.



741. When the presence of the sulphide (or sulphuret) of arsenic ( $\text{AsS}_3$ ) is suspected, the substance supposed to contain it may be first examined for any particles of yellow powder; which, if present, should be mixed, when dry, with a little black flux, or with a mixture of dry carbonate of soda and charcoal, and heated in a small German glass tube, closed at one end; when, if it consists of sulphide of arsenic, a crust of the metal will appear in the upper part of the tube (743). If no yellow powder can be detected, the mass in which it is suspected to be present is to be treated according to the directions given hereafter.

#### SECTION I.

##### *Identification of Arsenious Acid when unmixed with other substances.*

742. Place a little of the white powder in a small tube of German glass, closed at one end, and heat it gradually in the flame of a spirit lamp, taking care to warm the upper part of the tube slightly before heating the arsenious acid. If it is arsenious acid, it will sublime, and condense in the upper part of the tube, forming a colourless crystalline sublimate, which, when examined with a good lens or microscope, will be found to consist of beautiful sparkling octohedral crystals (Fig 76). The size and regularity of the crystals appear to depend on the slowness with which the vapour is condensed. If the surface of the glass on which the condensation takes place is quite cold, the sublimate is often amorphous, as may be seen by holding a piece of cold glass in the fumes given off by a little arsenious acid heated on charcoal.\*

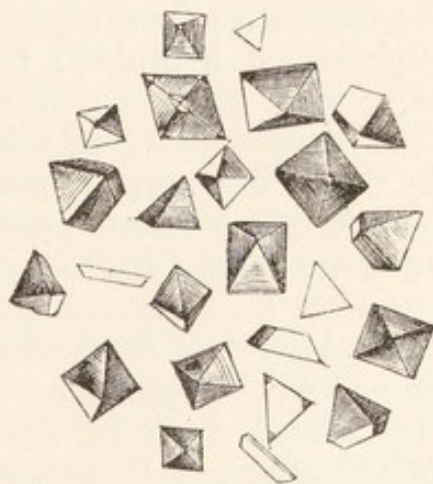


Fig. 76. Arsenious Acid.

\* For much valuable information with respect to the detection of arsenious acid with the aid of the microscope, we are indebted to Dr. Guy's minute investigations, an abstract of which is given in his 'Principles of Forensic Medicine,' 1st edition.





Fig. 77.

743. Mix a little of the suspected powder with a large proportion of black flux, or of carbonate of soda and charcoal, which for this purpose should be perfectly dry, and heat the mixture in a small tube of German glass before the blowpipe. If arsenic is present, it will be reduced to the metallic state, and sublime into the upper part of the tube, forming a shining metallic crust (*a*, Fig. 77). The part of the tube which contains the crust may then be filed off, wrapped in a piece of strong paper and broken, the fragments of the crust being placed in another tube, and again heated. The reduced metal will in this way be reconverted into arsenious acid, crystals of which will condense in the cool part of the tube (742).

744. Make a solution of some of the powder by boiling it for some minutes with water, in which arsenious acid is sparingly soluble, and apply to separate portions of the solution the following tests. (See also 745 & 749.)

(*a*) Acidify a portion of the solution with a drop or two of hydrochloric acid, and pass a current of hydrosulphuric acid gas (sulphuretted hydrogen) through the liquid, until it smells distinctly of the gas. If arsenious acid is present, a bright yellow precipitate of sulphide ( $\text{AsS}_3$ ) will be thrown down, very easily dissolved by ammonia.

(*b*) Add to a second portion of the solution a few drops of ammonio-nitrate ( $\text{AgO}, \text{NO}_3, 2\text{NH}_3$ ) of silver. If arsenious acid is present, a canary-coloured precipitate of arsenite of silver ( $3\text{AgO}, \text{AsO}_3$ ) will be thrown down, which is soluble in nitric acid and also in ammonia.

(*c*) Test a little of the solution with ammonio-sulphate ( $\text{CuO}, \text{SO}_3, 2\text{NH}_3, \text{HO}$ ) of copper. This will cause, with arsenious acid, a pale-green precipitate of arsenite of copper ( $2\text{CuO}, \text{HO}, \text{AsO}_3$ ).

#### *Marsh's Test.*

745. Arrange a wide-mouthed bottle, of six or eight ounces' capacity, with tubes as shown in the annexed figure; the tube *d* being of hard German glass. Place in it a few fragments of zinc, and add a little dilute sulphuric acid, consisting of one part of the strong acid to six or eight of water. When the



hydrogen has been coming off about five minutes,\* apply a light to the gas as it issues from the aperture at *e*, and hold over it, or rather *in* it, a clean porcelain crucible lid, in order to prove whether any traces of arsenic are contained in the zinc or acid employed, in which case a more or less distinct arsenical stain would be produced. If the materials are thus found to be pure, a little of the solution of the supposed arsenic is to be introduced through the tube *b*.

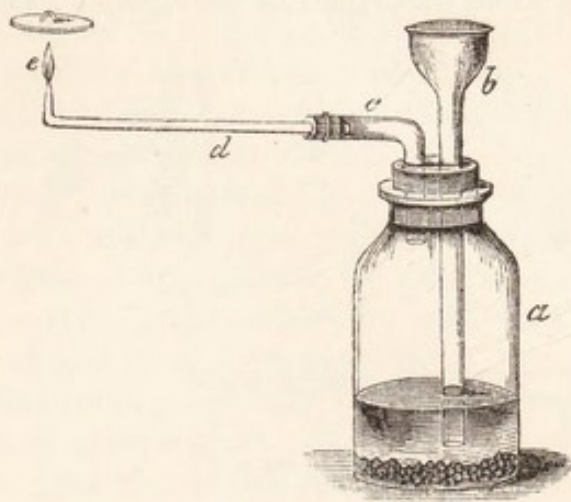


Fig. 78.

746. Again apply a light to the jet of gas at *e*, and hold in the flame a clean porcelain crucible lid. If arsenic is present, dark spots of the metal will be deposited on the surface of the porcelain, wherever it has been allowed to enter the flame. A few of these stains may be prepared and tested in the following manner, in order to prove whether they really consist of arsenic, and not of antimony; which latter, if present, would produce stains very similar in appearance to those of arsenic.

(a) Apply the heat of a spirit lamp to one of the spots. If arsenic, it will readily volatilize, and a slight smell, resembling garlic, will probably be perceptible.

(b) Moisten one of the spots with a drop of yellow hydrosulphate of ammonia, containing an excess of sulphur. If it consists of arsenic it will remain undissolved for some considerable time; while, if it were antimony, it would immediately dissolve.

(c) Add a drop or two of a solution of chloride of lime ( $\text{CaOCl}$ ) to one of the stains. If it consists of arsenic it will immediately dissolve.

747. Hold *over* the flame a short wide test tube (Fig. 79), so as to collect the fumes of arsenious acid formed during the combustion of the arseniuretted hydrogen. The arsenical

\* This interval must be allowed to elapse, in order that the whole of the common air in the apparatus may be expelled before the light is applied; since a mixture of hydrogen and common air is highly explosive.



sublimate may be dissolved in hot water, and the solution tested as described in paragraph 744, *a*, *b*, & *c*. (See also 749.)

The sublimate formed in the tube by antimony, under the same circumstances, would, on the contrary, prove quite insoluble in water.

748. Apply the heat of a spirit lamp to the tube at the point *c* (Fig. 78), and observe the formation of a dark

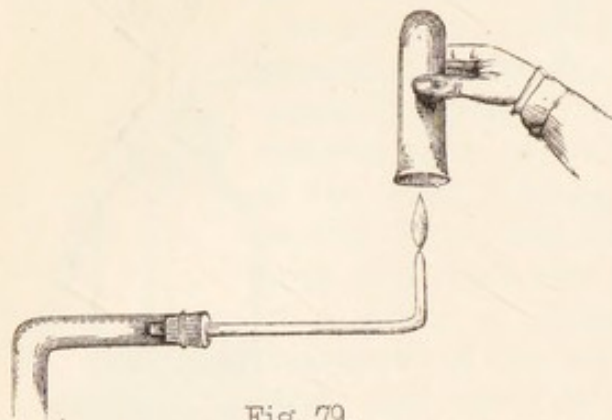


Fig. 79.

ring of metallic arsenic\* inside the tube, a little in advance of the heated point. The arsenic thus deposited may be volatilized backwards and forwards in the tube, by applying the heat of a spirit lamp (765, *a*). If the tube be then disconnected from the bottle, and the arsenic volatilized in it while filled with atmospheric air, the metal will gradually become oxidized and converted into arsenious acid, crystals of which will appear in the cool part of the tube.

For testing very small quantities of a substance suspected to contain arsenic, it is advisable to employ a much smaller apparatus (Fig. 80) than that above described. A two-ounce

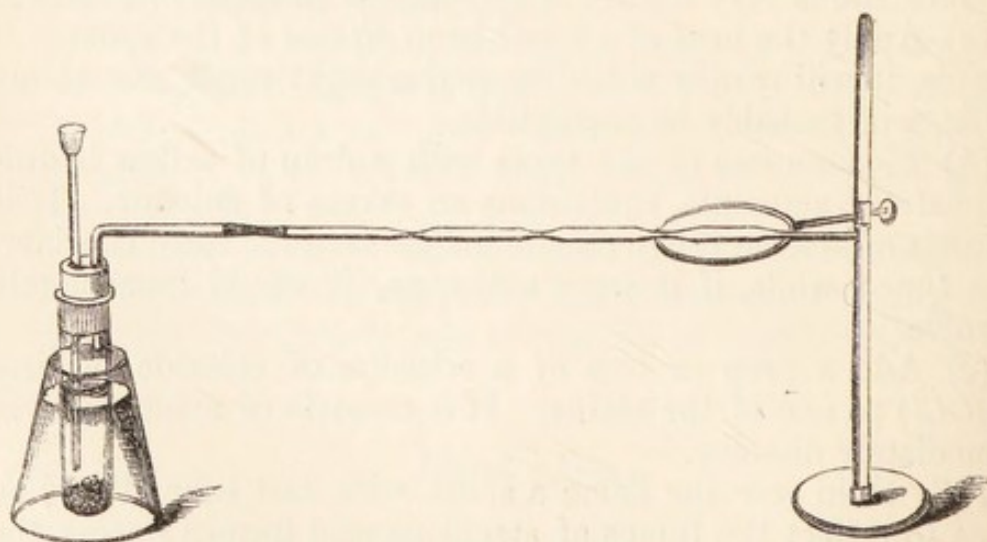


Fig. 80.

\* In consequence of a secondary decomposition between the zinc and sulphuric acid (Fordos and Gélis) a little sulphuretted hydrogen is sometimes formed, which gives rise to a yellow ring of sulphide of arsenic by the side of the metallic ring.



bottle may be used, or even a short wide test-tube capable of containing about half an ounce of liquid. The tube through which the gas escapes should be drawn out at the extremity, so as to form a very narrow tube about two inches long. In such cases it is not advisable to run the risk of allowing any arsenic to escape, on which account the shoulder of the tube should be heated with a spirit-lamp before the suspected solution is introduced, when a metallic deposit will be formed in the narrow portion of the tube if any arsenic be present. By employing a long tube constricted at intervals, and heating each of the wide portions of the tube, several metallic deposits may be produced at the same time, and may be made the subjects of various confirmatory experiments. When a very small apparatus is employed, the zinc should be free, not only from arsenic, but from all other metals, so that the evolution of hydrogen may be slow and uniform.

If Marsh's apparatus be immersed in a considerable volume of cold water whilst in use, to prevent a great rise of temperature, it is not necessary, even in very minute testing, to dry the gas, as is sometimes recommended.

The metallic crusts obtained in the tube should be examined by the following confirmatory tests.

(a) Place the piece of tube with the deposit in a small hard glass tube, and apply the heat of a spirit-lamp; the crust will gradually disappear, and crystals of arsenious acid (742) will be deposited on the cooler part of the tube.

(b) Place the tube and deposit in a little water contained in a small test-tube, and add two or three drops of yellow sulphide of ammonium; an arsenical crust should not dissolve even on shaking for some little time. Pour off the liquid, wash the tube several times with distilled water, and add some clear solution of chloride of lime (bleaching-powder), by which the crust should be dissolved on agitation.

(c) Boil the tube containing the metallic deposit in a few drops of pure nitric acid, in a small test-tube; when the crust is dissolved, rinse the solution into a very small porcelain dish, and evaporate just to dryness over the lamp. The residue of arsenic acid ( $\text{AsO}_5$ ) which should be left, would become moist on exposure to the air for a few minutes, would dissolve easily in water, and the solution would give a brown-red precipitate of arseniate of silver ( $3\text{AgO}, \text{AsO}_5$ ) on adding nitrate or ammonio-nitrate of silver. Should no precipitate be obtained, perhaps a little free nitric acid may have been left, in which case, a



glass rod dipped in dilute ammonia will produce the precipitate.

*Reinsch's Test.*

749. Before applying this test, the simplest by which minute quantities of arsenic can be detected, great care is required to ensure the absence of that metal in the hydrochloric acid and the copper necessary for its execution. As it is not easy to procure ordinary copper not containing a little arsenic, it is better to employ the metal deposited by the electrotyping process from a solution of sulphate of copper which has been carefully purified by re-crystallization. In order to test the copper and hydrochloric acid for arsenic, the following experiments should be made.

(1) Mix one ounce of the hydrochloric acid with four ounces of water, boil it in a small flask and introduce a small strip of the copper; boil for fifteen or twenty minutes, and if the copper remain perfectly bright, the hydrochloric acid may be deemed sufficiently pure.

(2) Cut a square inch of the copper into small strips and heat them in a small tube of hard glass, first with the flame, and afterwards with the blowpipe, to see if any crystals of arsenious acid are deposited.

(3) Cut two square inches of the copper into strips, place them in a shallow dish, moisten them with the hydrochloric acid, and leave them for some time exposed to the air; add a little more hydrochloric acid from time to time, until all the copper is dissolved (which will require many hours' exposure); pour the solution into a small retort, add about half its bulk of hydrochloric acid, and distil over about two thirds at a moderate heat, condensing the vapours by passing them through a tube kept cool by wet filtering-paper. Dilute the distilled liquid with four times its bulk of water, and boil it with a strip of copper, which should remain untarnished if no arsenic be present.

In this test the arsenical copper is converted into subchloride of copper, by favour of the oxygen of the air,  $\text{Cu}_2 + \text{HCl} + \text{O} = \text{Cu}_2\text{Cl} + \text{HO}$ .

The arsenic passes into solution as terchloride ( $\text{AsCl}_3$ ) together with the subchloride of copper which is soluble in hydrochloric acid. On distilling, the terchloride of arsenic passes over with the acid.



The hydrochloric acid employed in Reinsch's test should also be free from sulphurous acid, which may be detected by dissolving a little zinc in the dilute acid and conducting the gas into solution of acetate of lead in which a black precipitate of sulphide of lead ( $\text{PbS}$ ) would be produced if any sulphurous acid were present.

In order to apply this test, acidify a little of the aqueous solution of the substance suspected to contain arsenic, with a few drops of pure hydrochloric acid, and boil in it two or three strips of clean copper foil. If arsenic is present, it will be deposited in the metallic state\* on the surface of the copper, and may be proved to be arsenic in the following manner.

(a) Wash the copper strips and dry them by gentle pressure between folds of filtering-paper, or by warming them on a water-bath; when dry, place them in a small clean and dry tube of German glass, closed at one end, and apply heat, first with the flame alone, and afterwards with the blow-pipe. The arsenic will volatilize; and becoming oxidized while in contact with the air, arsenious acid will condense in the upper part of the tube, forming a crystalline sublimate, which may be examined with a lens (742).

(b) Dissolve the sublimate obtained in a in a little hot water, and apply to the solution the tests described in paragraph 744. If it be very minute, it may be dissolved by boiling with a few drops of nitric acid, and tested as in 748c.†

## SECTION II.

### *Detection of Arsenic in the presence of Organic Matter.*

750. The cases in which arsenic has to be detected in the presence of organic matter may be classed under three heads, the first including pretty clear and homogeneous liquids, such as beer, milk, and urine; the second, such thick heterogeneous mixtures as gruel, pudding, and the contents of the stomach or intestines; and the third, the solid organs of the body, such as the liver, in which arsenic is generally to be detected after death by poisoning.

\* Lippert has shown that this deposit is not pure metallic arsenic, but an alloy containing 32 per cent. of arsenic and 68 of copper ( $\text{Cu}_5\text{As}$ ).

† Arsenic is much more difficult to detect by Reinsch's test when in the state of arsenic acid. It then requires long boiling with copper in the presence of a very large excess of hydrochloric acid.



*Detection of Arsenic in Organic Liquids which are pretty clear and homogeneous.*

751. When the investigation has any judicial interest, the quantity of the liquid should be carefully recorded, and every portion which is taken for a separate experiment must be measured, so that, if the poison be detected, the analyst may be able to give an opinion respecting its quantity.

A portion of the liquid may then be acidified with a little pure hydrochloric acid (the purity of which has been previously ascertained), and then boiled with two or three small strips of tested copper foil. If arsenic is present it will probably be deposited, in the course of a few minutes, upon the surface of the copper, and must be treated in the manner presently to be described. It must not, however, be considered certain that no arsenic is contained in the liquid, until after boiling the mixture for half an hour, or even longer; when, if no stain is produced which, on examination, gives indications of arsenic, it may safely be concluded that no trace of the metal is present.

It occasionally happens that a little fatty animal matter is deposited on the surface of the copper during the boiling. When this is the case, the copper should be boiled with a little ether or alcohol, in order to dissolve it, before being exposed to heat in the tube.

752. The copper strips must now be heated in a small clean and dry tube, closed at one end; when, if any arsenic has been deposited upon them, a crystalline sublimate of arsenious acid will appear in the upper part of the tube. If, on examination with a lens the sublimate is found to exhibit the characteristic crystalline form and appearance of arsenious acid (742), there can scarcely be a doubt of the existence of arsenic. Proceed further as in 749 (b).

753. In most cases, when no arsenic has been thus reduced upon the copper, it may be inferred that no appreciable amount of that metal is present, but should arsenic be detected, the analyst should, if possible, proceed to obtain further evidence of its presence by other methods.

If the liquid be not very viscid and liable to froth, it may be acidulated with a little pure sulphuric acid, filtered if necessary, and subjected to Marsh's test (745). If frothing should take place to any inconvenient extent, a drachm or two of alcohol may be poured down the funnel-tube in order to arrest it.



In the case of viscid liquids to which Marsh's test cannot be applied, it is recommended to evaporate them to a small bulk upon a water-bath, to acidify strongly with pure hydrochloric acid, and to add chlorate of potash, in small quantities, until the liquid becomes so limpid as to admit of filtration. The heat of the water-bath is continued until the smell of chlorine has in great measure disappeared, and the liquid, if necessary, filtered. The clear filtrate may be tested, either by Marsh's process (745), or by electrolysis.

*Electrolytic test for Arsenic.*—This test depends upon the circumstance that when a pretty powerful galvanic current is made to traverse an acid liquid containing arsenic, arseniuretted hydrogen is evolved at the negative terminal, together with the hydrogen of the decomposed water.

To construct the apparatus (Fig. 81) for this test, a two ounce bottle, with a moderately wide mouth, is selected, and a file-mark made at a little distance from the bottom, so that it may be extended into a crack by placing a red-hot wire against it, and led round the bottle so as to cut off the bottom. The edges having been smoothed with a file, or by grinding on a wet stone, a pretty deep groove is filed round the bottle at about half an inch from the edge. A piece of wet parchment-paper\* is doubled down over the bottom of the bottle, and secured by a piece of stout platinum wire, for which the groove has

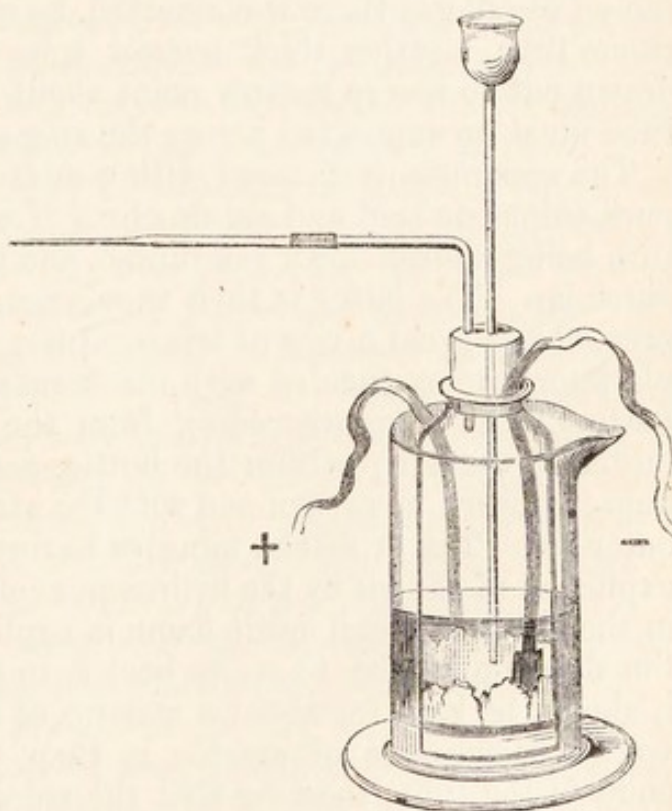


Fig. 81.

\* Parchment-paper is now commonly sold. It may be made by immersing white blotting-paper for an instant in a cold mixture of the strongest oil of vitriol with half its volume of water, and rapidly washing.



been cut in the glass. A perforated cork is inserted in the mouth of the bottle, provided with a narrow funnel-tube somewhat drawn out at the extremity, which should nearly touch the paper diaphragm, and with a small tube bent at right angles, for the exit of the gas. A strip of platinum foil cut into the shape of a spade is secured between the cork and the neck of the bottle, so that its broad end, hanging down within the bottle, may nearly touch the diaphragm, the other end projecting an inch or two beyond the neck of the bottle. The narrow part of this strip may be about a quarter of an inch wide, and the broader part an inch square. The bottle is placed in a cylindrical jar not much wider than itself, and not so flat at the bottom as perfectly to close the bottom of the bottle; between the side of the jar and the bottle, a second strip of platinum foil similar to the above is suspended, so as nearly to touch the bottom of the jar. With the bent tube provided for the egress of gas there is connected, by means of a small caoutchouc tube, a rather thick narrow tube of very infusible glass drawn out to a very narrow point about two inches long; this tube must be supported across the ring of a retort-stand.

The apparatus is charged with a mixture of two drachms of pure sulphuric acid and six drachms of water, half of the mixture being poured down the funnel, and the remainder into the outer jar. The latter is then immersed in a basin of water to prevent too great a rise of temperature, and the two strips of platinum are connected with the terminal wires of a galvanic battery,\* the wire proceeding from the zinc extremity being united to the strip within the bottle, and that from the platinum-, copper-, or carbon-end with the strip which dips into the outer jar. Ten or fifteen minutes having been allowed for the expulsion of the air by the hydrogen evolved from the platinum in the bottle, a small spirit flame is applied to the shoulder of the drawn-out tube so as to heat it to dull redness, at which it should be kept for about a quarter of an hour; if no ring of arsenic or sulphide of arsenic is then visible in the narrow point of the tube, showing that the sulphuric acid is pure, the solution to be tested for arsenic† is poured slowly down the funnel-tube, care being taken to avoid the introduction of air-bubbles. If much froth should now make its appearance, a

\* Five or six Grove's cells are very suitable for the purpose, but of course any battery which decomposes acidulated water pretty briskly will answer.

† If this solution contains very much free hydrochloric acid, it is advisable to dilute it with twice its volume of water.



drachm or two of alcohol may be poured down the tube. If no deposit of arsenic or of (yellow) sulphide of arsenic be visible after ten minutes, half a drachm of a strong solution of washed sulphurous acid or of sulphuretted hydrogen is poured in, and the experiment continued for another quarter or half hour. If any arsenic be present, either the metallic ring of arsenic or a greenish-yellow iridescent ring of sulphide of arsenic, or both, will be deposited in the narrow part of the tube.\* The lamp is then removed, the tube allowed to cool, and that part which contains the deposit cut off by a file, and gently warmed in a small test-tube with a little solution of carbonate of ammonia in which the yellow sulphide of arsenic will slowly dissolve. The tube with the metallic portion of the crust is then washed, and tested according to the directions in 748 *a*, *b* and *c*.

The liquid which has been subjected to electrolysis is seldom exhausted of arsenic unless a very minute quantity is present. In order to extract the remainder, the solution may be mixed with a large excess of a saturated solution of sulphuretted hydrogen, and heated in a covered beaker for an hour or two.

753*a*. The precipitate of sulphide of arsenic mixed with organic matter, is collected on a filter, well washed, dried, and thrown, in small portions into a little nitre fused in a porcelain crucible, the filter being cut in pieces and thrown in also. The fused mass (containing the arsenic as arseniate of potash), when cool, is dissolved in a little water, the solution mixed with chloride of ammonium and ammonia, filtered if necessary, and well stirred with solution of sulphate of magnesia. After standing for some time, a highly crystalline precipitate of arseniate of magnesia and ammonia ( $2\text{MgO}, \text{NH}_4\text{O}, \text{AsO}_5$ ) will separate, which may be collected on a filter and washed.

The arseniate of magnesia and ammonia may be easily distinguished from the phosphate which much resembles it, by moistening it with nitrate of silver, when it assumes the characteristic red colour of arseniate of silver, whilst the phosphate becomes bright yellow. Of course it might also be dissolved (even after treating it with nitrate of silver) in hydrochloric acid, and tested by boiling with hydrosulphuric acid, or by Marsh's test.

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\* If no odour of sulphuretted hydrogen be perceptible at the orifice of the tube at the end of the experiment, a little more sulphurous acid or sulphuretted hydrogen should be poured in, lest the liberated chlorine should have traversed the diaphragm and prevented the evolution of arseniuretted hydrogen.



## SECTION III.

*Detection of Arsenic in Organic Mixtures containing both Liquid and Solid Matters; such as the contents of a Stomach, vomited matters, &c.*

754. When the liquid and solid portions of the mixture are found capable of ready separation, either by subsidence or filtration, it is generally better to examine each of them separately. When this is not the case, *see* paragraph 758.

755. *Examination of the liquid portion.*—The clear liquid after the removal of the solid matter, either by filtration or otherwise, is to be examined in the manner described in paragraph 751.

756. *Examination of the solid portion.\**—This should first be examined for any small lumps of arsenious acid, which, in cases of poisoning, are often to be found adhering to the coats of the stomach. These should be carefully picked out, and tested according to the directions given in paragraphs 742—744.

757. The solid or semi-solid organic matter is then to be heated on the water-bath with dilute hydrochloric acid, containing about one tenth of the strong acid. A part of the acid solution may then be boiled with copper strips, which are to be dried, and examined for arsenic in the manner before described (749). The remainder of the acid solution may be examined by 753, and the undissolved solid portions by 761.

758. When the organic matter is viscid, and incapable of ready separation into solid and liquid portions (754), it may be mixed with a little dilute hydrochloric acid, well stirred together, and boiled; if the solution thus obtained be sufficiently thin, it may be filtered, and dealt with as in 757, but otherwise, it must be treated according to 759.

## SECTION IV.

*Detection of Arsenic in the Tissues, and in other solid Organic Matters.*

759. In medico-legal investigations as to the presence of arsenic, it is absolutely necessary, in case none of the poisonous

\* In some cases it may be of importance to note the weight of the solid matter.



can be detected in the stomach and its contents, to examine the various tissues of the body; since the poison, when introduced into the stomach during life, becomes gradually absorbed, and diffused through the whole system, and may be found in the blood, urine, muscles, and viscera, especially the liver. It is therefore advisable to examine each of these for the poison; and it should never be concluded, that because it cannot be detected in the stomach and its contents, none is to be found in other parts of the body. Should the patient, however, survive during several days after swallowing the poison, it is possible that the whole of it may be eliminated from the body; in which case no trace of it will afterwards be detected.

760. If the solid matters to be examined have undergone putrefaction, the arsenious acid may have been partly converted into sulphide of arsenic, which is sometimes perceived in bright yellow patches.

761. The solid matter intended for examination\* is to be cut up and heated on the water-bath for an hour or two with hydrochloric acid, consisting of one part of the strong acid to eight or ten of water. The mixture is then filtered through fine muslin, in order to separate the more solid matters; and the clear liquid thus obtained is concentrated to about half its bulk, by evaporation on the water-bath, and treated as in 751. The undissolved solid portions are heated in a porcelain dish, placed upon a water-bath, with a mixture of six measures of water and one of hydrochloric acid, to which chlorate of potash is added in small portions, with constant stirring, until the solid has disintegrated, and the liquid is fit for filtration. The further treatment is then conducted as in 753.

If a galvanic battery be not procurable, the solution obtained by means of hydrochloric acid and chlorate of potash is evaporated to a small bulk, mixed with a strong solution of washed sulphurous acid,† until it has a decided odour of the gas, heated in a flask placed in a water-bath, until the odour of sulphurous acid has disappeared, mixed with a large excess of a saturated solution of sulphuretted hydrogen, and digested for some time at a moderate heat. The precipitate of sulphide of arsenic, which is never pure yellow, but dingy, from the pre-

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\* The part of the body in which the poison is most likely to be found is the liver, which should always be preferred for these experiments. The pancreas, kidneys, and urine, should also, if possible, be examined, before deciding on the absence of arsenic.

† To reduce the arsenic acid ( $\text{AsO}_5$ ) to arsenious acid ( $\text{AsO}_3$ ).



sence of organic matter, is collected upon a small filter, washed, and treated as in 753a.

The solution obtained by treatment with hydrochloric acid and chlorate of potash might also be tested by Marsh's or by Reinsch's test, but, if the solution is at all viscous, the frothing is a serious obstacle to the application of the former, and the introduction of copper in the latter test unfits the solution for further examination if it should be necessary.

Should it be required to examine for arsenic the organic matter left undissolved by hydrochloric acid and chlorate of potash, it may be dried, mixed with five or six times its weight of pure nitre, and thrown by degrees into a red-hot dish or crucible. The deflagrated mass, which would contain arseniate of potash, is dissolved in as little water as possible, acidulated with hydrochloric acid, gently heated, and if necessary, filtered. The clear liquid is mixed with a few drops of solution of sulphate of magnesia and a considerable excess of ammonia, and after being well stirred, is set aside for twenty-four hours. The precipitate will contain the arsenic in the form of arseniate of magnesia and ammonia ( $2\text{MgO}, \text{NH}_4\text{O}, \text{AsO}_5$ ), together with earthy phosphates derived from the organic matter. It is collected upon a filter, washed with dilute ammonia (pure water dissolves it), and dissolved in as little dilute hydrochloric acid as possible. The solution may then be tested either by Marsh's or the electrolytic test.

761a. Another method which has been found very useful for separating arsenic from organic matters, consists in drying them upon a water-bath, digesting in a covered vessel with moderately concentrated hydrochloric acid, separating the solid matters by filtration, and distilling, the acid vapour being condensed in a flask containing water. The arsenic is found in the distillate, having passed over as terchloride of arsenic ( $\text{AsCl}_3$ ) and may be detected by the ordinary tests.

761b. *Detection of arsenite of copper in paper-hangings and other fabrics.* A very ready method of effecting this consists in soaking the fabric to be tested in a little solution of ammonia; the blue solution which will be found if copper is present, is acidulated with hydrochloric acid and boiled with clean copper (749).



## SECTION V.

*Quantitative Determination of Arsenic.*

762. The exact determination of the quantity of arsenic present in a mixture containing much organic matter is attended with great difficulty. In general, a sufficiently accurate estimate of the amount may be found by comparing the crusts obtained by Marsh's test with those furnished by known quantities of arsenious acid, but should a direct determination be necessary, it may be effected by a process of which an outline is here given, though without the minute details of manipulation requisite for perfect accuracy.

After having obtained the arsenic in a state of solution by heating the organic matter with hydrochloric acid and chlorate of potash, as described above, the clear liquid is mixed, in a flask, with a strong solution of bisulphite of soda until it smells very strongly of sulphurous acid, even after being heated for some minutes in a water-bath to reduce the arsenic acid ( $\text{AsO}_5$ ) to the state of arsenious acid ( $\text{AsO}_3$ ). The application of heat is continued till the smell of sulphurous acid has disappeared, and the solution is thoroughly saturated with sulphuretted hydrogen, after which the flask is corked and set aside in a warm place for some hours. The precipitate, containing sulphide of arsenic mixed with organic matter, is collected upon a filter, washed, and dried. The dry precipitate is mixed with about six parts of nitre and six parts of dry carbonate of soda, and projected, by degrees, together with the fragments of the filter, into a red-hot porcelain crucible. The deflagrated mass, containing arseniate of potash, is dissolved in a little water, the solution filtered if necessary, and mixed with chloride of ammonium, ammonia,\* and sulphate of magnesia. After being well stirred, it is set aside for twenty-four hours, when the arseniate of magnesia and ammonia will have separated as a crystalline precipitate, which must be collected on a weighed filter, washed with dilute ammonia, dried at  $212^\circ$  and weighed (753a). The quantity of arsenious acid is calculated by the proportion,

$$\underbrace{2\text{MgO}, \text{NH}_4\text{O}, \text{AsO}_5}_{+ \text{HO.}} \quad \underbrace{\text{AsO}_3.}_{\quad} \quad 190 \quad : \quad 99 \quad :: \quad \text{Weight of the precipitate} : x.$$

\* Should any precipitate be caused by the ammonia, it must be filtered off.



## CHAPTER II.

## ANTIMONY.

763. The form in which antimony is generally met with in medico-legal investigations, is the double tartrate of antimony and potash ( $\text{KO}, \text{SbO}_3, \text{C}_8\text{H}_4\text{O}_{10} + \text{Aq}$ ), commonly called tartar-emetie or tartarized antimony, which is often taken medicinally, and occasionally as a poison. It may be recognised by applying to its solution the following tests.

(a) Hydrochloric acid, which gives a white precipitate of teroxide of antimony ( $\text{SbO}_3$ ) soluble in excess.

(b) Hydrosulphuric acid, in the solution form (a), gives a bright orange precipitate.

(c) Hydrochloric acid and metallic copper (749), on the application of heat, will give a purplish black deposit of metallic antimony, upon the surface of the copper. The latter, when dried and heated in a tube, will not yield a crystalline sublimate like arsenic, but may give a white amorphous deposit on strongly heating. The deposit is dissolved by boiling the strip of copper in a dilute solution of potash, and if the solution be treated with sulphuretted hydrogen, filtered from any precipitate, and acidulated with hydrochloric acid, the orange sulphide of antimony is precipitated.

(d) Marsh's test (745) will yield metallic crusts of antimony, as with arsenic, but the antimonial crusts are readily distinguished by the following characters. (1) They are much less volatile than those of arsenic, and are therefore deposited very much nearer to the heated portion of the tube. (2) They give no crystalline sublimate when heated in a tube, but, with a strong heat, an amorphous sublimate. (3) Antimonial crusts will not dissolve easily in solution of chloride of lime; but (4) they do so at once in yellow sulphide of ammonium, and if the solution be slowly evaporated, it leaves an orange residue of sulphide of antimony ( $\text{SbS}_3$ ). (5) When oxidized by nitric acid and evaporated, the antimonial crust gives a residue which refuses to dissolve in water and produces a dirty grey precipitate with nitrate of silver (compare 748 a, b, c).

(e) The electrolytic test evolves antimonietted hydrogen far less readily than arsenietted hydrogen, most of the antimony being deposited as a black coating upon the platinum plate



connected with the zinc end of the battery. If the plate be washed, and gently heated with a little yellow sulphide of ammonium, it dissolves the antimony and leaves the orange residue when evaporated. By pouring a strong solution of hydrosulphuric acid down the funnel-tube at the commencement of the electrolysis, it will entirely prevent the evolution of antimonietted hydrogen, whilst it promotes that of arsenietted hydrogen.

(f) If a solution of antimony, acidified with hydrochloric acid be poured into a platinum capsule, or placed upon platinum foil (or even upon a gold or silver coin), and a strip of zinc (or iron, such as a knife-blade) be placed in it, a black deposit of antimony will be found around the point of contact of the two metals. The deposit may be tested by placing a drop of yellow sulphide of ammonium upon it, and evaporating to obtain the orange residue, which should be compared with the residue left by a drop of sulphide of ammonium alone upon the uncoated metallic surface.\*

## SECTION I.

### *Detection of Antimony in the presence of Organic Matter.*

764. For the detection of antimony in organic liquids and solids, the same methods of proceeding are adopted as in the case of arsenic (751—761), the antimony being recognised by the characters described in 763.

765. Should both metals be present, they may still be easily detected, unless there was a very great disproportion between them, by the behaviour of the crusts obtained in Marsh's test.

766. Even if one of the metals largely predominates, the electrolytic test permits the detection of both; it is only necessary to allow the electrolysis to proceed for five or ten minutes in order to deposit the antimony on the negative plate before adding the hydrosulphuric acid, which will at once arrest the evolution of the antimonietted hydrogen, and if the reduction tube be changed, a deposit of pure arsenic or sulphide of arsenic will be obtained.

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\* Of course a surface of silver would be blackened by the sulphide.



## SECTION II.

*Quantitative Determination of Antimony.*

767. The remarks in paragraph 762, upon the determination of the quantity of arsenic, will apply also in the case of antimony, and precisely the same steps must be taken in order to ascertain the amount of the latter metal, except that dried nitrate of soda should be used instead of nitrate of potash, for the deflagration of the sulphide. On digesting the fused mass, in the cold, with a little water, almost the whole of the antimony will be left in the form of antimoniate of soda ( $\text{NaO}, \text{SbO}_5$ ), which must be collected upon a filter, washed with cold water, dried, and fused with five or six times its weight of cyanide of potassium in a porcelain crucible, until the antimony has collected into a globule at the bottom of the fused mass. The latter is dissolved, when cold, in water, and the metallic button weighed. Each grain of antimony corresponds to 2.73 grains of crystallized tartar-emetic.

## CHAPTER III.

## MERCURY.

768. The most common preparation of mercury, by which life has been sacrificed or endangered, is the chloride ( $\text{HgCl}$ ), commonly called corrosive sublimate; the subchloride, or calomel ( $\text{Hg}_2\text{Cl}$ ), the red oxide ( $\text{HgO}$ ), and some of the other compounds, are also sometimes administered, either criminally or accidentally, with fatal effect, and may consequently have to be looked for by the medical jurist. In the process I am about to describe, however, any of these compounds will be brought into a state of solution; after which the mercury contained in them may readily be detected by the proper tests.

## SECTION I.

*Detection of Mercury in Organic Mixtures.*

769. When the presence of mercury is suspected in an organic mixture, such as the contents of a stomach, the solid and liquid portions of the matter to be examined may be sepa-



rated from each other, either by filtration or decantation, provided the separation takes place readily; or if this is not the case, the whole of the mixture may be treated with acid, and examined in the manner described in paragraphs 774—776.

770. *Examination of the liquid portion.*—The liquid portion may be first examined. Acidify it with a few drops of hydrochloric acid, and boil the mixture for a quarter or half an hour, with two or three strips of clean copper foil. If any mercury is present in the liquid, it will in this way be entirely separated from the solution and deposited on the surface of the copper. The latter is then removed from the acid liquid, and washed with a little dilute solution of ammonia, in order to remove from the surface any adhering oxide or subsalt of copper. The strips are then dried by gentle pressure between folds of bibulous paper, and placed in a small and perfectly dry German glass tube, three or four inches long, closed at one end.

771. The heat of a spirit lamp is then applied to the bottom of the tube containing the copper strips; when, if any mercury has been deposited upon them, it will be volatilized by the heat, and condensed in the cooler part of the tube, forming a delicate and dew-like ring of minute globules of metallic mercury; the real nature of which may be at once seen with the assistance of a common lens, if not with the naked eye.

771 *a.* If the sublimate is so minute that the globules are not distinguishable, it may be gently rubbed with a glass rod to unite the small globules. The strip of copper may be shaken out of the tube, a very minute particle of iodine introduced, and a gentle heat applied to vaporise it. The mercurial sublimate will thus be converted into iodide of mercury, which is yellow at first, and becomes scarlet when rubbed with a glass rod (Lassaigne).

772. If, in the experiment above described (771), the appearance of metallic globules is distinctly visible, it will scarcely be necessary to apply any further tests to prove the presence of mercury, since no other substance is capable of producing such a sublimate. If, however, any doubt exists as to the nature of the sublimate, the following experiments may be made:

773. Remove the copper from the tube, and dissolve the sublimate in nitrohydrochloric acid; by which the mercury, if present, will be converted into the soluble chloride ( $HgCl$ ). Expel the excess of acid by evaporation at a gentle heat; and



apply to an aqueous solution of the residue, the following tests:

(a) Solution of iodide of potassium (*KI*) gives a brilliant red precipitate of iodide of mercury ( $\text{HgI}$ ), which is very soluble in excess of the iodide of potassium.

(b) Solution of potash gives a yellow precipitate of hydrated oxide of mercury, which is insoluble in an excess of the precipitant.

(c) A solution of hydrosulphuric acid (sulphuretted hydrogen), or a drop or two of hydrosulphate of ammonia, forms at first a white precipitate, consisting of a double compound of chloride and sulphide ( $2\text{HgS}, \text{HgCl}$ ), which, unless the precipitant be added very sparingly, almost immediately becomes darker, owing to the admixture of the black sulphide ( $\text{HgS}$ ). If the precipitant be added in excess, the whole of the precipitate becomes black.

(d) The dry mercurial salt, when mixed with carbonate of soda, and heated in a narrow tube before the blowpipe, yields a sublimate of minute globules of metallic mercury.

774. *Examination of the solid portion.*—The solid portion of the mixture may contain mercury in combination with certain animal matters, besides particles of calomel, oxide, or some other mercurial compound. It may first be examined for any visible fragments of these, which, if detected, may be picked out, and tested for mercury, by mixing them, when dry, with carbonate of soda, and heating the mixture in a small tube before the blowpipe; when the mercury will be sublimed into the cooler part of the tube (773 d).

775. The rest of the solid matter may now be warmed with a little hydrochloric acid and chlorate of potash, as directed in 761; one part of the filtered solution may be tested by boiling with copper, and the other may be subjected to electrolysis according to the directions given in 753.

If mercury be present in the solution, it will be deposited upon the negative plate (connected with the zinc end of the battery), and may be readily detected by boiling the plate in a few drops of nitric acid, expelling the greater excess of the latter by evaporation, rendering the solution alkaline with ammonia (because the free nitric acid would dissolve the copper), acidifying very slightly with hydrochloric acid, and boiling with a slip of clean copper foil, which may afterwards be washed and heated in a tube. If the negative plate be of gold (or of platinum gilt by immersing it whilst attached to the



battery, in a solution of cyanide of gold in cyanide of potassium), the mercury will of course be easily recognised by the silvery appearance of the deposit.

In the absence of a galvanic battery, the solution obtained by means of hydrochloric acid and chlorate of potash may be mixed with excess of hydrosulphuric acid and heated for some time, when the black sulphide of mercury will be deposited, and may be collected on a filter and washed; it will be found nearly insoluble in hot nitric acid, but readily soluble in a mixture of nitric and hydrochloric acids. The presence of mercury in the solution may be proved by evaporating it to a small bulk, adding ammonia in excess, then hydrochloric acid in excess, and boiling with metallic copper (770).

776. A very small quantity of mercury may also be detected by placing the solution upon a gold or copper coin, and touching the gold, through the liquid, with a piece of zinc or the blade of a knife, when a silvery deposit will be formed around the point of contact, and will disappear when the washed coin is gently heated.

## SECTION II.

### *Detection of Mercury in the Tissues.*

777. When the presence of mercury is suspected in the viscera or other tissues of the body, the part intended for examination should first be cut into thin slices, and heated with hydrochloric acid and chlorate of potash (761); by which means any mercury that may be present will be converted into the bichloride, and thus brought into a state of solution. The undissolved matter is then separated by filtration or decantation, and the liquid portion evaporated to a small bulk on a water-bath, and treated as in 775.

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## CHAPTER IV.

### LEAD.

778. Although instances of criminal poisoning with compounds of lead are of comparatively rare occurrence, still the accidental admission of it into the system, either in the form of the solid carbonate (white lead) so extensively employed in the



arts, or through the medium of water impregnated with it, very frequently leads to serious and even fatal results; so that its detection is often a matter of grave importance.

779. In testing for minute quantities of lead, it must be borne in mind that several of the test solutions employed in analysis, when kept even for a few weeks in bottles of flint glass, dissolve out very perceptible traces of the metal from the glass, in which it is present in considerable quantity; so that unless the experimenter is on his guard, he may be led to suppose that he has detected the metal in the liquid which he is examining, while, in fact, he has himself introduced it in one of his reagents. Solutions of potash and soda, and their carbonates, are especially liable to become in this way impregnated with lead; and several other saline solutions also, under peculiar circumstances, do the same, though more slowly, and in a less degree. On this account it is always advisable to test each of the reagents employed with solution of hydrosulphuric acid in large excess, which will, if any traces of lead are present, give the liquid a more or less decided brown tint; or even cause a black precipitate, if the quantity of metal is at all considerable.

#### SECTION I.

##### *Examination of Water suspected to be impregnated with Lead.*

780. The water intended for examination (which should always be tested as soon as possible after being taken from the cistern or pipe in which it has been standing) is placed in a beaker or bottle of German or green glass, free from lead, the surface of which should be washed perfectly clean with distilled water. A stream of hydrosulphuric acid (sulphuretted hydrogen) gas is then transmitted through the water, until the latter smells distinctly of the gas. When lead is present, the liquid will generally assume a brown tint almost immediately, unless the quantity of lead is extremely small; but before deciding that the water is pure, it should be set aside for a few hours, after being saturated with the gas, during which time the sulphur will be partially precipitated, owing to the decomposition of the hydrosulphuric acid by the oxygen of the air mixed, if any trace of lead is present, with a little sulphide of lead ( $PbS$ ), which will give the sediment a more or less decided brown or fawn colour. If, on the contrary, the water continues



colourless, and the precipitated sulphur is white, or of a very pale sulphur colour, it may be concluded that no perceptible trace of lead is contained in the water.

781. The most satisfactory method of applying this test for lead in waters is the following. Two test-tubes of clear white glass, free from lead, about nine inches long, and half an inch in diameter, are nearly filled, one with pure distilled water, the other with the water under examination, to which a few drops of pure acetic acid are added to prevent the precipitation of any sulphide of iron. A similar quantity of acetic acid having been added to the distilled water, for the purpose of comparison, the contents of both tubes are tested either with a clear solution of hydrosulphuric acid, or with a few bubbles of the gas. By holding both tubes, after shaking, over a sheet of white paper, so that the eye may look along the axis of each tube, the slightest trace of lead may be discovered by the dark colour of the liquid, and all fallacy is excluded by the comparison with the pure water. It is evident that by comparing the result with those furnished by solutions containing known quantities of lead, a close approximation to the amount of the latter may be arrived at.

782. Lead may also be detected in waters by evaporating a quart to dryness, heating the residue with strong hydrochloric acid, and mixing the hydrochloric solution with a large excess of strong solution of hydrosulphuric acid, when the purplish or black precipitate, or colour, of sulphide of lead will be obtained.

## SECTION II.

### *Detection of Lead in Organic Mixtures.*

783. If the organic matter to be examined is a mixture of solid and liquid, such as the contents of a stomach, the two portions should, if practicable, be separated by filtration through paper or muslin; having been previously diluted, if necessary, with a little water, and gently heated with a drachm or two of pure acetic acid, which will cause the liquid to pass more readily through the pores of the filter. The liquid portion may be first tested; and in case none of the metal can be detected in it, the solid or semi-solid matter may be afterwards examined (788).

784. *Examination of the liquid portion.*—A current of hydrosulphuric acid gas is passed through the liquid for about a



quarter of an hour, by which means any lead that may be dissolved will be precipitated as the black sulphide.\* This is to be separated by filtration, and the greater part of it digested, with the aid of a gentle heat, in moderately dilute nitric acid; a small portion being retained for examination with the blow-pipe (787).

785. When the sulphide is for the most part decomposed by the nitric acid (which may be known by the undissolved residue, consisting chiefly of sulphur, becoming nearly white), the clear solution is to be filtered from the insoluble matter, and tested in the following manner (786); the undissolved residue being also retained, in case it may be required for subsequent examination (787). The digestion in warm acid should not be continued longer than necessary, since the prolonged action of the nitric acid might have the effect of oxidizing the sulphur as well as the lead, forming sulphuric acid, which would combine with the oxide of lead, and precipitate it from the solution in the form of the insoluble sulphate ( $\text{PbO}, \text{SO}_3$ ).

786. The clear solution (785) is now to be evaporated to dryness on a water-bath, in order to expel the excess of nitric acid; after which the residue is to be redissolved in warm water, and tested in the following manner; or, if the quantity is small, the tests *b*, *c*, & *d* only need be applied.

(*a*) Hydrosulphuric acid, and hydrosulphate of ammonia cause a black precipitate of sulphide of lead ( $\text{PbS}$ ).

(*b*) Dilute sulphuric acid, or a solution of sulphate of soda, gives a white precipitate of sulphate of lead ( $\text{PbO}, \text{SO}_3$ ), which is insoluble, or nearly so, in acids, but gradually dissolves in a solution of caustic potash.

(*c*) The sulphate formed in *b*, after being washed with distilled water, is instantly blackened when moistened with hydrosulphate of ammonia or a solution of hydrosulphuric acid, owing to the formation of the black sulphide. The sulphate of lead may in this way be readily distinguished from the sulphates of baryta and strontia, which it resembles in many respects.

(*d*) A solution of iodide of potassium (*KI*) throws down a bright yellow precipitate of iodide of lead ( $\text{PbI}$ ), which is soluble in hot water, and, on cooling, separates from the solution in the form of brilliant crystalline scales of great beauty.

\* Minute quantities of lead escape precipitation in liquids containing organic matter, and can only be detected by evaporating to dryness and incinerating the residue (788).



(e) Hydrochloric acid, or a solution of chloride of sodium, causes, if the solution is not very dilute, a white crystalline precipitate of chloride of lead ( $\text{PbCl}$ ), which dissolves when the mixture is heated, and crystallizes in the form of delicate needles as the solution cools.

(f) Chromate of potash ( $\text{KO}, \text{CrO}_3$ ) gives a rich yellow precipitate of chromate of lead ( $\text{PbO}, \text{CrO}_3$ ), which is soluble in potash, and insoluble in dilute acids.

(g) If any of the precipitates formed in the above experiments be dried, and heated on charcoal, with or without a little dried carbonate of soda, in the inner flame of the blow-pipe, minute metallic beads will be obtained; which may be recognised as lead by their softness and malleability; a yellow incrustation of oxide of lead appears upon the charcoal.

787. If no decided indication of lead can be obtained from the nitric acid solution, the other portion of sulphide (784), and also the residue which proved insoluble in the acid (785), may be dried, mixed with carbonate of soda, and heated in the inner flame of the blowpipe; when, if any lead is present, it will be speedily reduced to the metallic state, forming minute malleable beads.

788. *Examination of the solid portion.*—If the examination of the liquid portion should fail in proving the presence of lead, the poison may still be sought for in the solid or semi-solid matters left on the filter (783), since it may exist in combination with animal matter, or in some other insoluble form. The mixture is evaporated to dryness, and the dry mass incinerated in a clean fire-clay or porcelain crucible. The grey ash is reduced to powder and treated with boiling water as long as a drop of the washings leaves any residue when evaporated on a slip of glass. The solution thus obtained may possibly contain lead, and should therefore be tested with sulphuretted hydrogen (784).

789. But the greater part of the metal would remain in the residue, which must now be boiled with a mixture of equal measures of nitric acid and water for a few minutes. The filtered solution is examined for lead as in 786.

790. The residue left undissolved by nitric acid may still contain lead in the form of sulphate: by boiling it with a little acetate of ammonia (prepared by mixing ammonia with acetic acid in slight excess), this may be extracted, and the lead detected in the solution by hydrosulphuric acid. It may still be advisable to dry the residue, mix it with carbonate of soda



and charcoal, and fuse it at a bright red heat, either before the blowpipe, or in a covered crucible, in order to obtain if possible a bead of metallic lead.

### SECTION III.

#### *Detection of Lead in the Tissues.*

791. When, in a suspected case of poisoning by lead, no trace of the metal can be detected in the contents of the stomach, &c., it is necessary, before deciding upon the absence of the poison, to examine the tissues of the stomach, intestines, and especially the liver; since it may be often found absorbed in these tissues, even when no trace is to be met with elsewhere.

792. The portion of the body intended for examination is to be cut into thin slices, dried, incinerated as completely as possible at a moderate heat, and examined as in 788—790.

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## CHAPTER V.

### COPPER.

793. Like lead, copper is not often employed for the purpose of criminally destroying life; but is not unfrequently taken accidentally, dissolved in articles of food, with serious, and sometimes fatal, results. The chief cause of such accidents is the employment of untinned copper vessels for culinary purposes; and although such vessels, when perfectly clean, may be used in the preparation of certain articles of food without risk of impregnation, still the number of alimentary substances capable of acting upon and dissolving small quantities of the metal, is so great, that it is far safer to avoid the use of untinned copper vessels in all culinary operations. Acid and fatty substances especially, and liquids containing common salt and other saline matters in solution, should never be boiled in such vessels; since the quantity of copper dissolved by them is sometimes so considerable, as to impart a green or bluish colour to the mixture.



## SECTION I.

*Detection of Copper in Organic Mixtures.*

794. Copper may exist in such mixtures either in a state of solution, or in combination with certain organic or other substances, forming compounds which are more or less insoluble in water. On this account, when the mixture to be examined consists of both liquid and solid matters, it should first be warmed with a little hydrochloric or acetic acid, by which means the copper will be brought into solution. The solution may then be filtered from the insoluble portion, which latter should be retained, in case it may be required for subsequent examination.

795. The clear liquid, slightly acidified with a few drops of hydrochloric acid, is now to be tested for copper, by placing in it a piece of clean iron, free from rust, such as a needle or knife-blade. If copper is present in the liquid, it will in a short time be deposited in the metallic state on the surface of the iron, giving it all the appearance of copper; while the iron is at the same time dissolved in atomic proportion. The colour of the freshly-deposited copper is so peculiar and characteristic, that it can hardly be mistaken after being once seen; so that this experiment is generally sufficient of itself to prove the presence of the metal. If, however, any doubt exists as to its presence, the following tests may be applied, either to a portion of the liquid from which the copper has not been removed by means of the iron, or to a solution of the precipitated copper, scraped off the iron, in dilute nitric acid.

796. (a) Hydrosulphuric acid and hydrosulphate of ammonia throw down a black precipitate of sulphide of copper ( $\text{CuS}$ ).

(b) Ammonia, when added in small quantity, throws down a pale blue precipitate, which, if the ammonia be added in excess, redissolves, forming a beautiful blue solution.

(c) Potash throws down in the cold solution a pale blue precipitate of hydrated oxide ( $\text{CuO}, \text{HO}$ ); which, on boiling the mixture, becomes black, owing to the formation of the anhydrous oxide ( $\text{CuO}$ ). The potash must here be added slightly in excess, as otherwise the precipitate would consist of a basic salt, which would not become black when boiled.

(d) Ferrocyanide of potassium causes, even in very dilute acid or neutral solutions, a mahogany-coloured precipitate of



ferrocyanide of copper ( $\text{Cu}_2\text{FeCy}_2$ ), which is insoluble in dilute acids.

797. In case no copper can be detected in the liquid portion, it is advisable, before deciding that the metal is altogether absent, to examine the residue which proved insoluble in the dilute acid (794). For this purpose, it is to be evaporated to dryness, and ignited in a covered Berlin porcelain crucible. The incinerated residue is then warmed with a little dilute nitric acid, which will dissolve any traces of copper that may be present. The acid solution is evaporated nearly to dryness, in order to expel most of the excess of acid, and filtered; after which the solution may be tested with a piece of clean iron (795), and also, if necessary, with the other reagents above enumerated (796).

798. When the solution containing copper, even in the presence of much organic matter, is subjected to electrolysis (753), the metal is deposited upon the negative plate, and may be recognised by its colour, and by the appropriate tests (796) applied to the solution obtained by boiling the negative plate with nitric acid.

## SECTION II.

### *Detection of Copper in the Tissues.*

799. Like the other metallic poisons, copper is readily absorbed by the tissues, where it may frequently be found in cases where no trace can be detected in the contents of the stomach and intestines. On this account, it is necessary, before concluding that no copper can be found, to examine the liver and other viscera, which may be done in the following manner.

800. The part intended for examination is to be cut into thin slices, and treated with hydrochloric acid and chlorate of potash, as directed in 761. The acid solution, after filtering, is evaporated nearly to dryness; after which it may be tested with a piece of clean iron (795), and, if necessary, by paragraphs 796, 798.

If no copper can be detected in this solution, the insoluble organic matter may be incinerated and examined according to 797.



## SECTION III.

*Quantitative Determination of Copper.*

801. The quantity of copper present in any organic mixture may be ascertained by incinerating the dried organic matter, boiling the ash with dilute nitric acid, saturating the liquid (after filtering) with hydrosulphuric acid gas, which will throw down the whole of the copper as sulphide. The precipitate is to be dissolved in hot nitric acid, and the copper thrown down as oxide, by supersaturating the hot solution of the nitrate with potash. The black oxide thus precipitated is to be washed with a large quantity of hot water, filtered, dried, ignited, and weighed. From the weight of the oxide, that of the metallic copper may be calculated as follows:

Atc. wt. of oxide of copper.		Atc. wt. of copper.		Wt. of oxide obtained.		Wt. of copper in the quantity of mixture employed.
<u>40</u>	:	<u>32</u>	::	<u>a</u>	:	<u>x</u>

## CHAPTER VI.

## ZINC.

*Detection of Zinc in Organic Mixtures and in the Tissues.*

802. Zinc has occasionally to be looked for in organic mixtures and in the tissues, the sulphate being often administered as an antidote in cases of poisoning. It may be detected by boiling the suspected matters in a finely divided state, with a little dilute hydrochloric acid and chlorate of potash, and filtering, if necessary, from any insoluble residue. The clear solution thus obtained may then be supersaturated with ammonia, and filtered; after which the clear ammoniacal solution may be tested with a current of hydrosulphuric acid gas (sulphuretted hydrogen), which, if zinc is present, will throw down a white precipitate of sulphide ( $\text{ZnS}$ ). The sulphide thus formed may be separated from the liquid by filtration, and dissolved in a little nitric acid; the excess of acid



employed being afterwards expelled by evaporating the solution to dryness.\*

803. The residue should be boiled with water and a little nitric acid, the solution filtered, if necessary, and examined by the following tests :

(a) Add ammonia in excess ; filter the solution from any precipitate which may be formed, and add hydrosulphate of ammonia ; if zinc be present, a white flaky precipitate of sulphide of zinc should be produced.†

(b) Add ammonia in excess ; filter if necessary, acidify the filtered liquid with acetic acid, and add ferrocyanide of potassium ; a white precipitate of ferrocyanide of zinc should be obtained.

(c) Mix the remainder of the solution with carbonate of soda in slight excess and boil for a few minutes ; collect the precipitate (basic carbonate of zinc) upon a filter, wash it, and incinerate the filter, when dry, in a small porcelain crucible. If zinc be present, the ash will be yellow while hot and white on cooling. Place it in a small cavity on a piece of charcoal, moisten it with nitrate of cobalt and heat strongly before the blowpipe ; a green mass should be produced if zinc be present.

804. If it be desired to examine the zinc in the residue left by the hydrochloric acid and chlorate of potash (802), it must be washed, dried, and calcined at a low red heat in an open crucible. The mass may then be boiled with hydrochloric acid, the filtered solution evaporated to dryness and further tested as directed above (803).

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\* If this residue is carbonized when further heated, it is advisable to continue the heat as long as any vapours are evolved, to boil the charred mass with hydrochloric acid, and to evaporate the filtered liquid to dryness.

† If ammonia had not previously been added in excess, the hydrosulphate of ammonia would have given a white precipitate of sulphur, even if no zinc were present ; but this precipitate would simply impart a milky appearance to the liquid instead of separating in distinct flakes. The precipitate of sulphide of zinc is usually dirty white, from the presence of a little sulphide of iron.



## CHAPTER VII.

## IODINE.

## SECTION I.

*Detection of Uncombined Iodine in Organic Mixtures, &c.*

805. When iodine is present in an organic mixture, it may be detected in the following manner; which will also serve to identify it after having been absorbed by the tissues of the stomach, liver, or other organ, such organ having been first carefully cut into thin slices, and macerated with a little water. The characteristic smell of iodine is generally perceptible in liquids containing it; and it usually imparts to organic mixtures a yellow or greenish colour.

806. The mixture may first be examined for any particles of iodine that may be present in the solid state; which, if found, may be at once identified as such by the beautiful violet-coloured vapour which they form when gently heated in a small glass tube.\*

807. If no solid iodine can be found, the liquid may be tested with a solution of starch; or a strip of cotton or paper, impregnated with starch, may be moistened with it. If iodine is present in the solution, it will immediately strike a more or less decided purple colour, the intensity of the tint varying from almost black to a pale shade of pink or lilac, according to the quantity of iodine dissolved in the liquid.

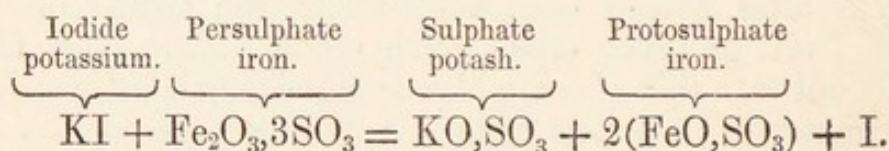
808. Should the quantity of iodine in the solution be so minute as to fail in producing a sufficiently decided result, the liquid may be well shaken with a little chloroform, which will dissolve the iodine and acquire a beautiful violet colour. The mixture is allowed to stand in order that the chloroform may collect at the bottom of the vessel, and the aqueous liquid is drawn off by a syphon or pipette. The chloroform is poured into a watch glass, and allowed to evaporate spontaneously, when the iodine will be left and may be recognised either by

\* Since indigo also gives violet vapours, the next test should always be applied.



heating it in a tube (806), or by stirring it with water, and adding a little thin starch-paste (807).

809. If the iodine be present in the solution as an iodide, it must be liberated previously to the application of the above tests. For this purpose the organic mixture should be slightly acidulated with hydrochloric acid, heated for some time upon a water-bath, and, if necessary, filtered. The cold solution is then mixed with a little sesquichloride of iron to which enough dilute sulphuric acid has been added to render it colourless, and agitated with chloroform, when the liberated iodine will be recognised as in (808). In this process, the persalt of iron is reduced to the condition of a protosalt, by imparting an atom of oxygen to the hydrogen or metal in combination with the iodine, which is thus set free :



Chlorine and chloride of lime are sometimes employed to liberate the iodine, but it is not easy to avoid the addition of an excess, which converts the iodine into chloride of iodine.\*

## CHAPTER VIII.

### SULPHURIC ACID ( $\text{HO}, \text{SO}_3$ ).

#### SECTION I.

##### *Detection of Sulphuric Acid in Organic Mixtures.*

810. Sulphuric acid may be readily detected, even when mixed with a large quantity of foreign matter. Should the substance to be examined be viscid or semi-solid, it may be diluted with a little water, and boiled; after which, if any solid matter remains in suspension, it may be filtered through muslin or paper.

\* The introduction of a little sulphuric acid and a piece of zinc, however, will restore the blue colour when destroyed by an excess of chlorine.



811. If the liquid contains free sulphuric acid, it will of course strongly redden blue litmus paper.

812. Mix the liquid to be tested with a little hydrochloric acid, and add a solution of chloride of barium or nitrate of baryta. If sulphuric acid is present, a copious white precipitate of sulphate of baryta will be produced, which will not dissolve on boiling the acidified mixture, nor yet on diluting it with a considerable quantity of water.

813. Since traces of sulphuric acid may be contained in the nitric acid used in acidifying the mixture (814), a little of the nitric acid employed should be diluted with three or four times its bulk of water, and tested with chloride of barium.

814. It is possible, also, that the substance under examination may contain some soluble sulphates in solution, as sulphate of magnesia, sulphate of zinc, &c., which would cause the precipitation of sulphate of baryta with chloride of barium, even when no free sulphuric acid is present. To remove this source of error, a little of the suspected fluid may be evaporated nearly to dryness at a gentle heat, when any saline matter that may be present will crystallize out; while the free sulphuric acid will continue liquid, and may be identified by the proper tests.

815. It is not often, however, that any serious uncertainty can exist as to whether the sulphuric acid found mixed with organic matter was or was not uncombined, especially in cases of suspected poisoning; since the corrosive effects of the acid upon the parts with which it has been in contact, or other corroborative circumstances, will generally of themselves furnish evidence sufficiently conclusive.

816. A very simple and delicate test for free sulphuric acid consists in dipping a piece of white linen or paper into the solution, and drying it before the fire. As the water evaporates, the acid will carbonize the fabric. Solutions containing organic matter as well as free sulphuric acid yield a carbonaceous mass when evaporated nearly to dryness.

## SECTION II.

### *Detection of Sulphuric Acid in Stains on Clothing.*

817. The stains formed by sulphuric acid on articles of clothing are usually moist to the touch, and most commonly of a brown or red colour, varying, however, with the nature of



the material and of the dye. The acid may be detected in them by boiling the stained part with water, and testing the solution as in the preceding section.\*

### SECTION III.

#### *Detection of Sulphate of Indigo in Organic Mixtures, &c.*

818. The solution of indigo in sulphuric acid, commonly called sulphate of indigo, which is occasionally either employed as a poison, or criminally thrown upon the person, may be detected in the same manner as the simple acid. It has a deep blue colour, which may be destroyed by boiling with nitric acid previous to the application of the tests; after which the sulphuric acid may be identified either in organic mixtures or on articles of clothing, by the experiments described in paragraphs 812—817.

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## CHAPTER IX.

### HYDROCHLORIC ACID ( $HCl$ ).

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#### SECTION I.

#### *Detection of Hydrochloric Acid in Organic Mixtures.*

819. When free hydrochloric acid is present in an organic mixture, it may be detected in the following manner. If solid or semi-solid matter is mixed with the liquid, it should be first boiled, and filtered through muslin; and when the mixture is thick and viscid, a little water should be mixed with it before boiling. The liquid is then treated with a tolerably strong infusion of galls, as long as it causes a precipitate, in order to throw down most of the dissolved animal matter, which would otherwise tend to prevent the acid distilling over. The pre-

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\* When the stained fabric is dried before the fire, it becomes charred where the acid had touched it.



precipitate is then separated from the clear liquid, either by again filtering through muslin, or by decantation.

820. A few drops of the solution, thus purified from the greater portion of the organic matter, may now be tested with nitrate of silver. If this causes a white precipitate, soluble in ammonia, and insoluble in nitric acid, the liquid will have to be further examined (821); since the precipitate *may* be owing to the presence of chloride of sodium or some other soluble chloride. But if no such precipitate is occasioned by the silver salt, the absence of hydrochloric acid may be relied on; unless, indeed, the solution is ammoniacal, in which case it should first be neutralized or slightly supersaturated with nitric acid.

821. In order to prove whether the precipitate caused by nitrate of silver is owing to the presence of free hydrochloric acid, or of some soluble chloride, the liquid is to be distilled to dryness in a retort. The neck of the retort is to be attached by means of a perforated cork to a quilled receiver, the quill of which should be allowed to dip just below the surface of a little pure water placed in the flask or bottle intended for its reception. The bulb of the retort is to be heated in a chloride-of-calcium bath, the heat of which may be raised, towards the end of the distillation, to about  $230^{\circ}$ .

822. When the whole of the liquid has distilled over, the contents of the receiver are to be examined, first with blue litmus paper, which, if any free acid is present, will become reddened; and also with nitrate of silver, which will give a copious white precipitate of chloride, soluble in ammonia, and insoluble in hot nitric acid, in case any free hydrochloric acid was present in the mixture, since such acid would distil over with the water.

823. A little of the distilled liquid may also be mixed with a few drops of pure nitric acid, and boiled for a few minutes with a small fragment of gold leaf. If the latter dissolves, it is an additional proof that the acid is hydrochloric.

824. In examining the contents of a stomach, it must be borne in mind that minute quantities of free hydrochloric acid are probably always present as one of the normal constituents of the gastric juice; so that the distilled liquid may always be expected to contain some traces of it. The amount of the acid derived from this source is, however, so small, that it may readily be distinguished from the comparatively



large quantity usually to be found when the acid has been swallowed.\*

## SECTION II.

*Quantitative Determination of Hydrochloric Acid.*

825. The chloride of silver ( $\text{AgCl}$ ) obtained by adding nitrate of silver to the distilled acid liquid (822), is to be washed on a filter, dried, and heated to dull redness in a counterpoised porcelain crucible, until it begins to fuse. From the weight of the chloride thus obtained, that of the hydrochloric acid present in the mixture may be calculated as follows:—

Atc. weight of chloride of silver.	Atc. wt. of hydro- chloric acid.	Wt. of chloride obtained.	Wt. of hydrochloric acid in the quantity of mixture employed.
$\underbrace{\hspace{1.5cm}}$ 143.5	$\underbrace{\hspace{1.5cm}}$ 36.5	$\underbrace{\hspace{1.5cm}}$ $a$	$\underbrace{\hspace{1.5cm}}$ $x$
:	:	:	:

## CHAPTER X.

NITRIC ACID ( $\text{HO}, \text{NO}_5$ ).

## SECTION I.

*Detection of Nitric Acid in Organic Mixtures.*

826. If any solid or semi-solid organic matter is present in the mixture, it should be separated by filtering through muslin, having first heated it on a water-bath in order to effect the separation of the greater part of the acid present, from the solid matters which may be more or less impregnated with it. Should the liquid be thick and viscid, it may be first diluted with a little water.

827. If free nitric acid is present in any considerable quantity

\* It must be remembered that free hydrochloric acid would distil over from mixtures containing chlorides together with free sulphuric, phosphoric, oxalic, or lactic acid.



in the liquid, it will probably be recognised by its peculiar smell; and the characteristic yellow stain of the tissues with which it has been in contact is in most cases perceptible. The want of smell, however, is no proof of the absence of the acid; which may still be present in considerable quantity, either diluted with a comparatively large amount of liquid, or even more or less completely neutralized by magnesia, or some other alkaline substance that may have been administered as an antidote. In the latter case, the liquid may be neutral, or nearly so, to test paper.

828. In order to detect nitric acid, the liquid, after filtration, may, if acid, be neutralized with carbonate of potash, and evaporated to dryness at a gentle heat. The nitric acid will thus be obtained in combination with potash, forming nitrate of potash, which will be deposited in needle-shaped crystals when most of the water is expelled; unless, indeed, the crystallization is prevented by the admixture of much animal or other matters.

829. The greater part of the saline residue thus obtained, is to be dissolved in as small a quantity of water as possible, and the solution placed in four test tubes, for the following experiments:—

(a) The first portion is mixed in a small test tube, with a few drops of strong sulphuric acid; after which a clean strip or two of copper, or a little roll of copper wire, is dropped in, and, if necessary, a gentle heat applied. If nitric acid is present, orange fumes of nitrous acid will be given off, the smell of which may generally be recognised, even when in too small quantity to be apparent to the eye.

(b) To the second portion add a few drops of hydrochloric acid, and put a small fragment or two of gold leaf into the mixture. If nitric acid is present, the gold leaf will be partially or wholly dissolved; and the presence of gold in the solution may be proved by protochloride of tin causing with it a purple precipitate.

(c) The third portion is to be acidified with a few drops of strong sulphuric acid, and as soon as the mixture is cool, a small crystal of protosulphate of iron is dropped in; when, if nitric acid is present, the liquid round the crystal will assume a brown colour, which disappears on boiling the mixture.

(d) Mix the remaining portion of the solution with sulphuric acid, and add a drop of dilute sulphate of indigo, sufficient to



give the liquid a pale blue colour. If nitric acid is present, the colour of the indigo will disappear, especially on warming the mixture.

## SECTION II.

### *Detection of Nitric Acid in Stains on Clothing.*

830. Stains occasioned by the action of nitric acid on woollen cloth are usually of a brown or yellowish colour, and, unlike those caused by sulphuric acid (817), become in a short time dry and extremely rotten. The yellow colour of the nitric acid stain is changed to orange by potash or ammonia, which will generally restore the original colour of fabrics stained by hydrochloric or sulphuric acid. If recent, the acid may generally be detected in them, by boiling the stained part with a little water, neutralizing with potash, and applying the tests mentioned in paragraph 829, but if any considerable time has elapsed since the production of the stain, it is probable that all traces of the acid will have disappeared, partly by evaporation, and partly by decomposition occasioned by contact with the organic matter.

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## CHAPTER XI.

### OXALIC ACID ( $HO, C_2O_3$ ).

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## SECTION I.

### *Detection of Oxalic Acid in Organic Mixtures.*

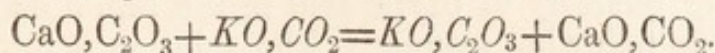
831. Before proceeding to apply the several tests for oxalic acid in the contents of a stomach, vomited matters, or other mixtures containing organic matter, it is advisable first to separate the latter, since its presence might interfere with the action of some of the reagents.\* If lime or magnesia has been used as an antidote, the oxalic acid, if present, will be either wholly or in part in the form of an insoluble oxalate; so that, in that

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\* Separation by dialysis will be found of the greatest use in the case of oxalic acid.



case, it is necessary to boil the sediment with a solution of carbonate of potash, whereby the acid will be brought into solution as oxalate of potash ( $KO, C_2O_3$ ); an insoluble carbonate of the earth being at the same time formed.



832. The suspected mixture is first boiled, to ensure the solution of the whole of the acid contained in it, and filtered, if necessary, from any solid residue. A solution of acetate of lead is then added as long as it causes any precipitate, which will throw down the oxalic acid in the form of the insoluble oxalate of lead ( $PbO, C_2O_3$ ), together with the greater part of the soluble organic matter. The precipitate thus formed is digested for an hour or two in dilute hydrosulphate of ammonia, and the mixture then evaporated to dryness on a water-bath. The lead is in this way separated, in the form of the insoluble black sulphide, from the acid; which, in combination with the ammonia (oxalate of ammonia), may be dissolved out with water, leaving the sulphide undissolved, together with the greater part of the organic matter.

833. The solution thus obtained is then filtered, and examined in the following manner for oxalic acid:

(a) A solution of sulphate of lime, or a very dilute solution of chloride of calcium, added to a portion of the solution, gives, if any oxalic acid is present, an immediate white precipitate of oxalate of lime ( $CaO, C_2O_3 + 2Aq$ ), which readily dissolves in dilute nitric or hydrochloric acid, but is insoluble in acetic or tartaric acid.

(b) If the oxalate of lime formed in *a*, be gently ignited on platinum foil, it will be converted into carbonate, with little or no blackening. The carbonate of lime thus produced will be found to effervesce when treated with dilute hydrochloric acid, and if a little of it be strongly ignited for a short time, it will be still further decomposed, and the carbonic acid expelled; after which the residue of caustic lime will, when placed on a piece of moistened turmeric paper, change the yellow colour to brown.

(c) Test another portion of the solution with nitrate of silver. If oxalic acid is present, a white precipitate of oxalate of silver ( $AgO, C_2O_3$ ) will be produced, which is soluble both in nitric acid and ammonia. If the precipitate be dried, and gently heated on platinum foil, it will be decomposed with a slight puff, leaving a residue of metallic silver.



## SECTION II.

*Quantitative Determination of Oxalic Acid.*

834. The quantity of oxalic acid in the liquid may be estimated in the following manner. The solution is first acidified with a little acetic acid, in order to decompose any soluble carbonate that may be present. A solution of chloride of calcium is then to be added as long as it causes any precipitate; and the mixture is boiled and filtered. The precipitate, after being washed on the filter, is dried, and gently ignited in a counterpoised crucible. It is then, after cooling, moistened with a solution of carbonate of ammonia, and again heated a little below redness, in order to expel the excess of the ammoniacal salt, which was added for the purpose of supplying carbonic acid to any lime that may have been rendered caustic during the first ignition (833*b*).

835. The oxalate of lime is thus wholly converted into carbonate; which is to be weighed, and from its weight that of the oxalic acid may be calculated as follows:

Atc. wt. of carbonate of lime.	Atc. wt of crystallized oxalic acid.	Wt. of carb. lime obtained.	Wt. of oxalic acid in the quantity of liquid employed.
$\underbrace{\hspace{1.5cm}}$ 50	$\underbrace{\hspace{1.5cm}}$ 63	$\underbrace{\hspace{1.5cm}}$ $a$	$\underbrace{\hspace{1.5cm}}$ $x$
:	:	:	:

## CHAPTER XII.

HYDROCYANIC (OR PRUSSIC) ACID ( $H\text{Cy}$ ).

836. The presence of hydrocyanic acid, even when largely diluted, may usually be detected by its peculiar and characteristic odour, somewhat resembling that of oil of bitter almonds. Great caution is necessary not to inhale more than the smallest quantity of the vapour, since headache and other unpleasant symptoms may be occasioned by merely smelling it, even when in a highly diluted state.

837. It must be remembered, in cases of suspected poisoning with this acid, that no time should be lost in applying the tests for its presence; since it rapidly volatilizes, and, unless



carefully protected from the air, disappears entirely in the course of a few days. The facility with which the acid is decomposed when in contact with putrefying organic matter also prevents its detection after some time has elapsed.

## SECTION I.

### *Detection of Hydrocyanic Acid in Organic Mixtures.*

#### *I. Detection of the acid in the state of vapour.*

838. Very small traces of the acid may be detected by one or other of the following tests, which may be readily applied to any liquid or mixture suspected to contain it. There is also this advantage in being able to identify it without going through the process of distillation at a higher temperature—viz., that while the tests for the vapour which I am about to describe are equally, or even more delicate than those for the liquid after distillation, the chances in favour of the spontaneous formation of the acid by the decomposition of the organic matter are greatly diminished.

839. A little of the mixture suspected to contain the poison, slightly acidified, if neutral or alkaline, with dilute sulphuric acid, may be placed in a watch-glass,\* over which another similar watch-glass is to be inverted, having been previously moistened with a drop or two of a solution of nitrate of silver, care being taken that none of the latter is allowed to run into the lower glass. The glass containing the suspected solution is then very gently warmed by holding it in the hand; when, if any hydrocyanic acid is present, it will volatilize into the upper glass; where, on coming in contact with the silver solution, it will form a white film of cyanide of silver ( $\text{AgCy}$ ). This test is very delicate: but as a somewhat similar effect might be produced by hydrochloric acid, it is always advisable to confirm the result by the following experiments:

840. A little of the suspected mixture, previously acidified, if necessary, with a little dilute sulphuric acid, is put into a watch-glass, over which is placed another glass moistened with a drop or two of solution of potash. The hydrocyanic acid, if

\* Since, in medico-legal investigations, it is not desirable to transfer the liquid from one vessel to another if it can be avoided, it is better to invert the watch-glasses moistened with the reagents over the mouth of the bottle in which the contents of the stomach, &c., have been placed as soon as removed from the body. Of course, pieces of glass might be used instead of watch-glasses.



present, gradually evaporates into the upper glass, where it combines with the potash, forming in solution a little cyanide of potassium. This is then mixed, first with a drop of a solution of protosulphate of iron (which should have been exposed to the air for a short time, so as to have become partially converted into the persulphate),\* and afterwards with a drop or two of dilute hydrochloric acid, which should be added in slight excess. Should any hydrocyanic acid have been present in the mixture, a blue precipitate of Prussian blue will be immediately formed, the appearance of which may be considered as a sure proof of the existence of the acid. This experiment is commonly known as *Scheele's*, or the *iron test*.

If the hydrocyanic acid be present in small quantity, no precipitate of Prussian blue will be seen at first, but a green liquid will be formed which will deposit flakes of Prussian blue on standing. It is always desirable to compare the result with that obtained by mixing together in another watch-glass, the potash, iron-salts, and hydrochloric acid in the same proportions as were employed in the test.

841. The following test, commonly known as *Liebig's test*, which is perhaps the most delicate of all, may also be applied. A little of the suspected fluid, slightly acidified, if necessary, is put into a watch-glass as before, and over this another watch-glass is inverted, containing a drop of hydrosulphate of ammonia, which for this purpose should contain an excess of sulphur, and consequently have a yellow colour. The glasses may be allowed to remain together for about a quarter or half an hour; after which the upper one is removed, and placed on a water-bath, until the hydrosulphate of ammonia is evaporated to dryness. Should any hydrocyanic acid have been present in the liquid, some of its vapour will have combined with the hydrosulphate, with which it would form sulphocyanide of ammonium. The residue left after the evaporation of the drop is now to be moistened with a dilute solution of persulphate or perchloride of iron; which, in case any sulphocyanide of ammonium had been formed, or, in other words, in case any hydrocyanic acid had been present in the suspected mixture, will immediately produce a blood-red colour, owing to the formation of sulphocyanide of iron. This colour is bleached by perchloride of mercury (corrosive sublimate), and is thus distinguished from that caused by acetate or meconate of iron.

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\* Or a drop of perchloride of iron may be added as well as the protosulphate.



## DETECTION OF HYDROCYANIC ACID.

### II. *Detection of Hydrocyanic Acid in solution.*

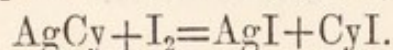
842. The mixture suspected to contain the poison is to be distilled in a retort heated on a water-bath, the receiver being kept cool by immersion in cold water, or in a freezing mixture composed of ice and salt, or of equal weights of nitrate of ammonia and water. When about one-eighth part of the liquid has passed over into the receiver, the distillation may be stopped. Should the mixture, previous to distillation, be neutral, or at all alkaline to test paper, it should be slightly acidified with dilute sulphuric acid, in order to disengage the hydrocyanic acid from the ammonia, or other bases which may be present, and which would tend to prevent the distillation of the acid at the low temperature employed. The presence of hydrocyanic acid in the distilled liquid may be ascertained by the following peculiarities:

843. Unless the quantity of acid be very minute, the peculiar odour, resembling that of oil of bitter almonds, will probably be apparent.

844. Test a little of the distilled liquid with a solution of nitrate of silver. If hydrocyanic acid is present, a white precipitate of cyanide of silver is produced, which is soluble in ammonia and in hot nitric acid, but insoluble in the cold acid.

845. The cyanide of silver should be collected upon a filter, washed as long as the washings are acid, and dried at a gentle heat. It is then to be subjected to the following tests:

846\*. Place a very small fragment of iodine at the bottom of a small tube closed at one end, and above it as much as can be spared of the supposed cyanide of silver. Apply a very gentle heat, by holding the tube at some distance above a flame, when iodide of cyanogen will be formed, and will condense on the cool part of the tube in very fine white needles.



(If very little cyanide of silver be used, it is well to cover it with a layer of carbonate of soda, to retain any excess of iodine.)

847. File off that portion of the tube which contains the sublimate, and warm it in a test tube, with a little dilute yellow hydrosulphate of ammonia in which it will dissolve. Evaporate the solution on the water-bath, and test the residue with perchloride of iron (841).

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\* For this excellent test we are indebted to MM. Henry and Humbert.



If a sufficient quantity of the cyanide of silver can be obtained, another sublimate of iodide of cyanogen may be made and tested by dissolving in potash and applying the Prussian blue test (840).

848. Boil a little of the cyanide of silver with yellow hydrosulphate of ammonia, filter, evaporate on a water-bath, and apply Liebig's test (841). A very small particle of cyanide of silver may be tested in a watch-glass, by moistening with a drop of yellow sulphide of ammonium, evaporating on the water-bath, and testing with perchloride of iron.

849. Add to a little of the distilled liquid in a test tube, first a little solution of potash; then a drop or two of a solution of protosulphate of iron, containing also a little persulphate (840); and lastly, a slight excess of dilute hydrochloric acid. If the liquid contains hydrocyanic acid, a precipitate of Prussian blue will be immediately produced; or if only a small trace is present, a few hours may elapse before it becomes apparent.

850. Mix another portion of the distilled liquid with a few drops of yellow hydrosulphate of ammonia, evaporate the mixture to dryness on a water-bath, and test as in 841.

851. The evaporation to dryness in this experiment is necessary, in order to expel the excess of hydrosulphate of ammonia; which would otherwise form with the iron solution a black precipitate of sulphide, and thus obscure the appearance of the characteristic red colour.\* During the evaporation the heat must be kept very moderate, lest any of the sulphocyanide of ammonium that may be formed by the action of the hydrocyanic acid on the hydrosulphate should be decomposed.

## SECTION II.

### *Quantitative Determination of Hydrocyanic Acid.*

852. The quantity of hydrocyanic acid contained in an organic mixture may be ascertained with sufficient accuracy for most purposes by distilling the acid (842), and precipitating the distilled liquid by means of nitrate of silver. The precipitated cyanide of silver is washed and dried in a hot water oven until it ceases to lose weight. From the weight of the

\* Which may still be rendered evident by filtering the liquid.



cyanide thus obtained, that of the anhydrous hydrocyanic acid (HCy) may be calculated as follows :

Atc. wt. of cyanide of silver.		Atc. wt. of hydrocyanic acid.		Wt. of cyanide of silver obtained.		Wt. of hydrocyanic acid in the quantity of mixture employed.
134	:	27	::	$a$	:	$x$

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### CHAPTER XIII.

#### OPIUM.

853. Of the several compounds contained in, and peculiar to opium, two only, morphia ( $C_{34}H_{19}NO_6$ ) and meconic acid ( $3HO, C_{14}HO_{11}$ ), are possessed of sufficiently characteristic properties to enable us to identify them when mixed with other matters; the tests for these substances, moreover, are not particularly delicate, so that it is difficult, and not unfrequently impossible, to detect small traces of them. In cases of poisoning with opium it is seldom that any traces of it can be found in the contents of the stomach; so that the tissues of the stomach itself, the intestines, and also any vomited matters, ought to be carefully examined for the poison.

#### *Detection of Opium in Organic Mixtures, Tissues, &c.*

854. If the substance to be examined is liquid or semi-fluid, it should first be evaporated to dryness, or nearly so, on a water-bath. If solid, the suspected substance may be cut into thin slices. The residue left after evaporation, or the sliced solid matter, as the case may be, is then to be digested for an hour or two, with the aid of a gentle heat, in a flask or dish placed on a water-bath, with a small quantity of water containing a little acetic acid. The mixture is filtered, and the clear liquid, containing a slight excess of acetic acid, is treated with a solution of acetate of lead ( $PbO, C_4H_3O_3$ ) as long as any precipitate is produced. The meconic acid, if present, is thus thrown down in combination with oxide of lead, forming meconate of lead ( $3PbO, C_{14}HO_{11}$ ); while the morphia remains in solution in combination with acetic acid (acetate of morphia,



together with any excess of acetate of lead that may have been employed. The mixture is warmed (not boiled, since by boiling some of the meconic acid might become decomposed) and, when again cold, is filtered.

855. The clear solution may first be examined for morphia; reserving the precipitate for subsequent examination.

856. A current of hydrosulphuric acid (sulphuretted hydrogen) is passed through the solution, until the latter smells distinctly of the gas, in order to decompose the excess of acetate of lead. The precipitated sulphide of lead is separated by filtration from the solution; which latter, after boiling, and if necessary concentrated by evaporation, is to be examined for morphia by means of the following tests:

857. Place a drop or two of the concentrated solution on a strip of glass, and add a drop of ammonia. The morphia will be precipitated in the form of minute needle-shaped crystals, which may be examined under the microscope.

858. Mix a small quantity of the solution in a test-tube or watch-glass, with enough solution of carbonate of soda to give it a decided alkaline reaction, and stir the mixture with a glass rod, rubbing the sides of the vessel. A crystalline precipitate of morphia will be deposited on standing, especially upon the lines of friction. Should the carbonate of soda produce an immediate flocculent precipitate (of the phosphate or carbonate of lime, for example), this should be filtered off immediately, before the solution is stirred with the glass rod, and should be reserved for further examination.

859. If any crystalline precipitate has been produced by the carbonate of soda, it must be collected upon a small filter (reserving the filtered liquid) and washed with very small quantities of cold water as long as the washings are decidedly alkaline (these washings are to be mixed with the filtered liquid). The filter is then carefully spread out upon a glass plate, and a drop of solution of perchloride of iron placed, with a glass rod, upon a part of the precipitate; if morphia be present, an indigo-blue colour will be produced. Another particle of the precipitate may be touched with a drop of strong nitric acid, which tinges morphia of an orange red colour.\*

860. The solution† in which carbonate of soda has not

\* Concentrated sulphuric acid, to which 0.004 per cent. of nitric acid ( $\text{NO}_5$ ) has been added, gives a violet purple colour when gently heated with morphia or the hydrochlorate (Erdmann).

† If possible this solution should be set aside for some hours in order to allow the morphia to crystallize out.



caused any crystalline precipitate, or which has been filtered from the precipitate, is now evaporated to dryness on a water-bath, and heated with alcohol, which would dissolve the morphia; on filtering the solution and evaporating to dryness on the water-bath, the morphia will be left, and may be tested as in 859.

861. The flocculent precipitate produced by carbonate of soda (858) may be examined by washing it with a little hot alcohol to dissolve the morphia which may then be separated by evaporation and tested as in 859.

862. The precipitate, supposed to contain meconate of lead (854), is now to be mixed with water in a beaker glass; and while suspended in the liquid, treated with a current of hydrosulphuric acid, the mixture being stirred occasionally. The meconate of lead is thus decomposed; the black sulphide of lead being precipitated, while the meconic acid, if present, remains in solution. The mixture is filtered to separate the sulphide of lead, and the clear liquid is gently warmed (not boiled (854),) in order to expel the excess of hydrosulphuric acid; and, if necessary, concentrated by evaporation on a water-bath. The meconic acid, if present in sufficient quantity, may then be detected by the following tests:

863. A solution of perchloride of iron gives the liquid, in case meconic acid is present, a bright red colour, owing to the formation of meconate of iron. The colour closely resembles that caused in solutions of iron by the sulphocyanides, from which it may be distinguished by not being decolorized by a solution of bichloride of mercury (841). It is, however, destroyed by boiling nitric acid, chloride of tin, and the caustic alkalies.

864. Solutions of acetate of lead, chloride of barium, and nitrate of silver, produce white precipitates of meconates, which are all soluble in an excess of nitric acid.

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## CHAPTER XIV.

### STRYCHNIA.

865. Strychnia ( $C_{42}H_{22}N_2O_4$ ) is the poisonous alkaloid contained in *nux vomica* and other plants of the *strychnos* tribe. Although a very minute quantity of strychnia is sufficient to cause death, the symptoms to which it gives rise are usually so



well marked, and the tests by which it may be recognised are so characteristic and delicate, that less difficulty is experienced in deciding upon the question of its administration than in the case of most other vegetable poisons.

*Detection of Strychnia in Organic Mixtures, Tissues, &c.*

866. The matter under examination, which, if solid, should be cut into shreds or slices, is digested, for about an hour, in a dish placed upon a water-bath, with dilute hydrochloric acid (1 part of the strong acid with 10 parts of water). The solution is then rendered pretty clear by filtration through muslin, or paper, evaporated on a water-bath to as small a bulk as convenient, and rendered strongly alkaline by solution of potash. The alkaline solution is poured into a narrow stoppered bottle, and shaken with about an equal volume of ether for several minutes; on standing, the ether will rise to the surface, holding the strychnine in solution. The ethereal layer is drawn off with a small syphon or pipette, and evaporated to dryness in a small capsule placed on the water-bath. Very minute particles of the solid residue may then be taken on the point of a knife, and examined for strychnia by the following tests:

867. The extremely bitter taste of strychnia is perceptible even when a very minute quantity is examined.

868. A particle of the suspected substance is placed upon a piece of white porcelain, and beside it, but not touching it, a drop of strong sulphuric acid is placed with a glass rod. A trace of solution of chromate or bichromate of potash is introduced, by a glass rod, into the drop of sulphuric acid, and the particle supposed to contain strychnia is then pushed into it. A fine violet blue colour should then be produced, which soon changes into red.\*

869. Another particle of the substance is moistened with strong nitric acid, and a very minute quantity of the peroxide (brown oxide) of lead is added. Violet streaks should arise from the particles of the peroxide, and should gradually pervade the liquid, ultimately changing to red.

870. If the results obtained by these tests should not be satisfactory, in consequence of some fatty or other organic

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\* Although the presence of any considerable proportion of morphia interferes with the detection of strychnia, the separation of the two alkaloids is so easily effected by ether or benzole, which will readily dissolve strychnia, but scarcely morphia, that there is no danger of strychnia being overlooked from this cause.



matter having also been dissolved by the ether and left with the strychnine on evaporation, the remainder of the suspected residue (866) in the capsule, should be moistened with strong sulphuric acid, and heated for some time upon the water-bath, when the extraneous organic matters will be carbonized, and the strychnia converted into a sulphate. The carbonaceous mass is treated with water, filtered, the clear solution mixed with a slight excess of ammonia, and shaken with about one-sixth of its volume of chloroform (for which ether may, in case of need, be substituted).\* When the chloroform, holding the strychnia in solution, has fallen to the bottom of the liquid, the latter is drawn off and the chloroform evaporated upon a water-bath, when the strychnia will be left and may be identified by the tests described in 868, 869.

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## CHAPTER XV.

### NICOTIA.

871. The poisonous oily alkaloid of tobacco, nicotia, or nicotine ( $C_{10}H_7N$ ), has occasionally been employed with criminal intention, but it is much less likely to be met with than morphia or strychnia, on account of its very powerful and characteristic odour.

#### *Detection of Nicotia in Organic Mixtures.*

872. This alkaloid may be extracted by a process similar to that followed in the case of strychnia (866), viz., by digesting the mixture with dilute hydrochloric acid, liberating the alkaloid by means of potash, and removing it from the aqueous solution with ether. On allowing a few drops of the ethereal solution to evaporate spontaneously, impure nicotia will be left and may be recognised by its pungent odour recalling that of tobacco. In order to obtain the nicotia in a pure state, the ethereal solution must be shaken with water mixed with about one-fifth of sulphuric acid, which dissolves the nicotia in the

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\* Benzole, which is lighter than water, and less volatile as well as cheaper than chloroform or ether, may also be employed for the extraction of strychnia, though it is not so good a solvent for it as chloroform.



form of sulphate. The ether is then poured off and the aqueous solution of sulphate of nicotia is shaken with a little more ether to remove any fatty matters which may be present. It is then rendered alkaline by potash and again shaken with ether, the ethereal layer containing the nicotia being afterwards allowed to evaporate spontaneously, when oily drops of nicotia are left and may be recognised by the smell, especially when gently heated. If ammonia be present, it may be removed by exposing the nicotia under an exhausted receiver containing a dish of strong sulphuric acid.

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## CHAPTER XVI.

### DETECTION OF PHOSPHORUS IN CASES OF POISONING.

873. The comparative frequency of accidental poisoning by phosphorus matches in the hands of children, together with the circumstance that a phosphorus poison for vermin is now in very common use, render it necessary that the medical chemist should be able to obtain evidence of its presence in organic mixtures. If possible the vomited matters should be especially examined, as they have been found to contain the largest proportion of phosphorus.

874. If the substance to be examined has not been long exposed to the air, it may still contain phosphorus in the free state, but in most cases oxidation will have converted the phosphorus into phosphorous acid ( $\text{PO}_3$ ), or possibly even into phosphoric acid ( $\text{PO}_5$ ). Since this last is a normal constituent of the body and of the food, it would afford no evidence of the administration of phosphorus, but fortunately, unless after prolonged exposure, either phosphorous acid or phosphorus itself can be detected whenever this element has been taken into the system.

875. The organic matter should first be examined as to its odour, that of phosphorus being very characteristic, and as to its luminosity in the dark. It should also be carefully looked over to ascertain if any solid particles of the phosphorized composition can be detected.

876. The suspected matter is then placed in a flask, and



acidulated with dilute sulphuric acid, a little water being also added if necessary. The flask is connected by means of a cork with a bent glass tube passing through a wide tube (having a perforated cork at each end) through which a stream of cold water can be kept trickling. On applying a moderate heat to the flask, in a darkened room, the condensing vapours should exhibit a phosphorescent appearance. The liquid which distils over may possibly contain white finely divided phosphorus. If it be at all turbid it should be shaken, in a tall vessel, with bisulphide of carbon, which will dissolve the phosphorus and sink to the bottom. On allowing the bisulphide of carbon to evaporate spontaneously, it will leave the phosphorus, often in a spontaneously inflammable condition. If no free phosphorus has been detected, both the distilled liquid and that remaining in the flask must be examined for phosphorous acid by the following process :

877. The liquid to be tested having been acidulated with sulphuric acid, is placed in a flask, together with a few fragments of pure zinc. A cork with a bent tube is then attached so that the evolved hydrogen may be passed through a solution of nitrate of silver. A strip of filter-paper moistened with nitrate of silver may also be suspended some distance above the surface of the liquid in the flask. Should any inconvenience be caused by the frothing, a little alcohol may be poured upon the surface of the liquid. If any phosphorous acid be present, phosphuretted hydrogen ( $\text{PH}_3$ ) will be evolved, and will produce a dark precipitate in the solution of nitrate of silver, and a mingled grey and yellow stain upon the filter paper, which will become bright yellow when dipped into a little strong nitric acid. (Sulphuretted hydrogen would merely blacken the nitrate of silver, and the stain would be dissolved by the acid without turning yellow.) The dark precipitate (phosphide of silver,  $\text{Ag}_3\text{P}$ ) is collected upon a small filter, washed, dissolved in a little hot nitric acid, and the solution evaporated to dryness. On adding a little water, and a trace of ammonia upon the end of a glass rod, the yellow precipitate of phosphate of silver ( $3\text{AgO}, \text{PO}_5$ ) will be produced. The characteristic odour of phosphuretted hydrogen should also be looked for in this experiment.



## CHAPTER XVII.

## DETECTION OF ALCOHOL IN ORGANIC MIXTURES.

878. In cases where alcohol ( $C_4H_6O_2$ ) in any form has been taken shortly before death, it may generally be detected in the contents of the stomach. The odour having been carefully noticed, the organic matter, mixed with a little water if necessary, should, if acid, be exactly neutralized with potash, and distilled, by the heat of a water-bath, in a flask or retort connected with a tube carefully cooled in order to condense the vapours. When a sufficient quantity of the liquid has distilled over, it may be examined for alcohol by the following tests:

879. Observe whether it has the smell and taste of alcohol. Dip a glass rod into it and see if it will burn.

880. Mix a small portion with dilute sulphuric or hydrochloric acid, and add a drop of solution of bichromate of potash ( $KO, 2CrO_3$ ). On applying heat to the mixture, the presence of alcohol would be indicated by the change of the orange colour of chromic acid ( $CrO_3$ ) into the green of oxide of chromium ( $Cr_2O_3$ ), and by the odour of aldehyde ( $C_4H_4O_2$ ) at the mouth of the tube.

881. The remainder of the alcoholic distillate may be placed in a tube, and dry powdered carbonate of potash added to it, in small portions as long as it is dissolved. The water will be thus taken up by the carbonate of potash, and the alcohol will rise to the surface, and will now be found to be inflammable, although it may have been too much diluted when previously tried.

## CHAPTER XVIII.

GENERAL SYSTEMATIC COURSE FOR THE DETECTION  
OF POISONS IN ORGANIC MIXTURES.

882. Although it rarely happens that an organic mixture is submitted to examination without some clue which enables the chemist to limit the inquiry to a very few poisons, still such a case should be provided for, and it is here proposed to give an outline of a systematic course for the detection of the leading



poisons, in three divisions, referring to cases where (1) the poison is believed to be metallic, (2) it is believed to be an organic poison, and (3) nothing whatever is known about it. Alcohol, phosphorus, opium, and hydrocyanic acid will not be included in this course, since they at once afford indications of their presence, and may be detected according to the directions already given.

#### THE POISON IS BELIEVED TO BE METALLIC.

*Examination for Arsenic, Antimony, Mercury, Copper, Lead, Zinc, Barium, Silver, Bismuth.*

883. The solid portions of the mixture having been as finely divided as possible, with a sharp knife or scissors, the whole is heated (for about an hour) in a porcelain dish placed upon a water-bath, with a mixture of six measures of water and one of hydrochloric acid,\* to which chlorate of potash is added by degrees, with constant stirring, until the solid has disintegrated, and the liquid is sufficiently thin for filtration. It is then filtered, the insoluble part is washed several times with water (the washings being mixed with the filtrate), and is then set aside for further examination (892).

884. The filtered liquid is evaporated, on a water-bath, to a small bulk, and subjected to electrolysis as directed in 753. After the passage of the current has been continued for about an hour (during which the exit tube has been heated in order to detect arsenic and antimony (753, 766), about half a drachm of a strong solution of washed sulphurous acid is poured down the funnel-tube; the exit tube having been first changed if any deposit should have been formed in it.

885. After the current has passed for another half hour, the electronegative platinum plate is removed from the decomposing cell, without suspending the current, and washed with a little distilled water. The liquid in the cell is reserved for subsequent examination (888).

886. The appearance of the deposit upon the electronegative plate having been carefully observed, it is boiled with a little dilute yellow hydrosulphate of ammonia, which will dissolve the antimony, to be detected in the solution according to 763 (e).

887. The plate is again well washed, and boiled with dilute

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\* If there be much liquid, less water must be added so that the acid may form about a sixth of the whole.



nitric acid. The solution is boiled down (in a test-tube) to a very small bulk, and mixed with an excess of ammonia. The presence of copper will be indicated by the blue colour. Hydrochloric acid is very carefully added till the solution is slightly acid, and it is then largely diluted with water. A milkiness will be produced if bismuth be present. The liquid is again evaporated to a small bulk, rendered clear by adding a little hydrochloric acid, and boiled with a strip of bright copper, to detect mercury (770, 771). In case any mercury should have been left upon the platinum in the form of sulphide, the plate may be again boiled with dilute nitric acid and a drop of hydrochloric acid (taking care not to dissolve much of the platinum). The solution is boiled down to a small bulk, mixed with excess of ammonia, then with hydrochloric acid in excess, and boiled with copper.

888. The liquid which has been subjected to electrolysis (885) is now evaporated, at a moderate heat, until the organic matter is so far carbonized that a small portion, when diluted with water, and filtered, yields a nearly colourless solution. The whole is then mixed with a few drops of hydrochloric and nitric acids, heated to boiling, again diluted and filtered. The carbonaceous residue is washed and reserved for further examination (891).

889. The filtrate and washings are saturated with sulphuretted hydrogen, boiled, again saturated with sulphuretted hydrogen and set aside in a warm place, in a covered vessel, for several hours. Any precipitate, except sulphur, which may have been deposited is filtered off, washed, and examined for metals according to the general method followed in qualitative analysis.

890. The solution filtered from this precipitate is evaporated to a small bulk, and mixed with an excess of ammonia. If this should produce a precipitate (generally consisting of earthy phosphates or peroxide of iron) it is filtered off, and the solution mixed with a little hydrosulphate of ammonia to precipitate any zinc as sulphide, which may be tested by the blow-pipe (803c).\*

891. The carbonaceous residue from 888 is dried,† incine-

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\* If this precipitate is very dark and impure, it must be dissolved in a mixture of hydrochloric and nitric acids, the solution evaporated to dryness, the residue heated till all organic matter is destroyed, and dissolved in nitric acid. The solution is mixed with excess of ammonia, filtered, if necessary, and tested with hydrosulphate of ammonia.

† A small charcoal fire will be found very convenient in these incinerations.



rated, and examined together with the other incinerated residue from 883.

892. The residue left undissolved by hydrochloric acid and chlorate of potash in 883 is dried and incinerated at a low red heat. The ash is mixed with that obtained in 891, and boiled with nitric acid diluted with two volumes of water. The solution is filtered and examined for metals as usual in qualitative analysis.

893. Any portion of ash left undissolved by the nitric acid is washed, dried, and, if still retaining any carbon, again incinerated. The residue, which might possibly contain barium (as sulphate), silver (as chloride), and lead (as sulphate), is fused with three or four parts of carbonate of soda in a porcelain crucible. The fused mass having been boiled with water, the residue is washed till the washings leave nothing on evaporation, and dissolved in hot dilute nitric acid. The nitric solution is tested (1) for silver with hydrochloric acid, which would give a white precipitate, soluble in ammonia (2) for barium with sulphate of lime (white precipitate\*) and hydrofluosilicic acid (white crystalline precipitate), (3) for lead with dilute sulphuric acid (white granular precipitate), and hydrosulphuric acid (purplish black precipitate).

#### THE POISON IS BELIEVED TO BE ORGANIC.

##### *Examination for Oxalic Acid, Morphia, Strychnia, Nicotia, and Conia.*

893a. The organic mixture, of which the solid portions have been as finely divided as possible, is digested in water acidulated with hydrochloric acid, for an hour or two, in a dish placed upon a water-bath. The solution is then filtered, first through muslin, and afterwards through paper, and evaporated upon the water-bath to a small bulk.

894. A small portion of this solution is tested for oxalic acid by adding a little chloride of calcium, then ammonia in excess, and finally, acetic acid in excess. If any white crystalline precipitate of oxalate of lime be left undissolved by the acetic acid, the larger portion of the solution must be examined for oxalic acid according to Chapter XI.

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\* If this precipitate be collected on a filter, well washed, and heated on a platinum wire moistened with hydrochloric acid, in the inner blow-pipe flame, it will tinge the outer flame bright green.



895. The remainder of the solution from 893*a* is rendered pretty strongly alkaline by potash, and shaken with four or five times its volume of ether.\* After standing for some time, the ethereal layer is poured off or drawn off with a small syphon or pipette.

896. The ethereal solution is poured into a dish and allowed to evaporate spontaneously. If either nicotia or conia be present, it will be left in oily drops which will evolve the powerful odour of the base when gently heated on the water-bath. If any solid residue be left by the ether, it must be examined for strychnia and morphia. For this purpose it is dissolved in a very little dilute hydrochloric acid, filtered, if necessary, and the solution rendered alkaline with carbonate of soda, briskly stirred, and set aside for an hour or two. If any crystalline precipitate is formed, it must be examined for strychnia and morphia according to 868, 869, 859. The solution in which carbonate of soda has failed to produce a precipitate, or which has been filtered from the precipitate, is evaporated to dryness on a water-bath, and the residue gently heated with absolute alcohol. The alcoholic solution is evaporated to dryness, and the residue tested for strychnia and morphia.

897. The aqueous layer separated in 895 is slightly acidified with hydrochloric acid, and evaporated to dryness on the water-bath. The residue is heated with strong alcohol for some time. The alcoholic solution is filtered and evaporated to dryness on the water-bath. The residue is redissolved in a very little water, the solution briskly stirred with a slight excess of carbonate of soda,† and the further examination conducted precisely as in 896.

#### NOTHING IS KNOWN OF THE NATURE OF THE POISON.

898. In this case, the first part of the process must be conducted on the supposition that the poison is organic (893*a*); the residues left undissolved by alcohol in 896, 897, as well as so much of the precipitate produced by carbonate of soda as is not

\* Uslar and Erdmann have recommended amylic alcohol as a solvent for the poisonous alkaloids, but the advantage with which its employment in certain cases may be attended is in great measure counterbalanced by the very disagreeable and injurious character of its vapour.

† If oxalic acid be present, it might go down in this precipitate as oxalate of lime. The remainder of the oxalic acid will be found as oxalate of soda in the residue left by absolute alcohol in extracting the morphia.



consumed in testing for alkaloids, must be dissolved in hydrochloric acid, mixed with the original organic matter left undissolved by the hydrochloric acid (893*a*), and the examination for metallic poisons proceeded with as in 883.

## CHAPTER XIX.

### SEPARATION OF POISONS FROM ORGANIC MIXTURES BY DIALYSIS.

899. The important observation made by Mr. Graham, that crystallizable bodies will pass in a state of solution through membranous and other diaphragms which will not permit the passage of the amorphous substances composing the bulk of most organic mixtures, has been applied by him to the separation of poisons. As the process is very simple, easy of execution, and does not involve any operations which would interfere with the subsequent application to the same mixture of any other process for the separation of the poison, it will probably come into very general use in medico-legal investigations.

This process is, indeed, a refined filtration, and is applicable instead of that operation, in all the steps of the separation of poisons from organic mixtures, with this very great advantage, that it removes not only substances mechanically suspended in the liquid, as is the case with filtration, but also colouring matters, albuminous substances, &c., which so interfere with the application of tests to the liquid obtained by filtration.

900. A circular piece of parchment-paper \* (see note to p. 241) is folded, as in preparing a filter, into a cone, which should be at least twice as large as is necessary to contain the mixture under examination. This cone is placed in the mouth of a cylindrical jar (Fig. 82), (a beaker or common tumbler), filled nearly to the brim with distilled water, the volume of which should be about eight times that of the organic mixture.

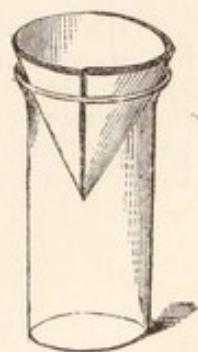


Fig. 82.

901. The solid portion of the organic matter having been

\* This should have been well soaked in distilled water, and dried before use.



cut up, and, if it be thought necessary, a little water having been added to thin the mixture, it may be poured at once upon the cone of parchment-paper arranged as above directed. The whole may then be covered with a bell-glass, or placed in a secure cupboard, and left for as long a period as can be conveniently allowed to elapse, if possible, for at least forty-eight hours. The *diffusate*, as the liquid in the glass is termed, may then be evaporated to a small bulk and examined for the poison by the appropriate methods, whilst the organic mixture remaining upon the cone or *dialyser* may be subjected to the ordinary processes for the separation of poisons from organic matter.

902. Of course, in cases where there is reason to suspect the presence of hydrocyanic acid, alcohol, or phosphorus, it would not be prudent to subject the mixture to dialysis until at least a portion of it had been examined for those poisons.

903. If time could be spared, it would evidently be desirable to dialyse the organic mixture at first without any addition (except water), since not only would all question of impurity in the reagents be avoided, but a knowledge of the state of the poison, whether soluble or not, would be thus gained, which might, in many cases, prove of great service. The mixture might then be dialysed a second time after digestion with the proper solvent, such as hydrochloric acid, or that acid with the addition of chlorate of potash.\*

---

\* The Editor has obtained most satisfactory results by this process in separating arsenious acid, strychnine, morphine, opium, and oxalic acid (as oxalate of lime). The arsenious acid was separated in some cases by simply dialysing the organic mixture, in others by digesting with hydrochloric acid and dialysing, and in others by first digesting with hydrochloric acid and chlorate of potash. In all cases the diffusate was colourless.



# APPENDIX.

## WEIGHTS AND MEASURES.

### *Troy or Apothecaries' Weight.*

Pound.	Ounces.	Drachms.	Scruples.	Grains.	French Grammes.
1	= 12	= 96	= 288	= 5760	= 372.96
	1	= 8	= 24	= 480	= 31.08
		1	= 3	= 60	= 3.885
			1	= 20	= 1.295
				1	= 0.0647

### *Avoirdupois Weight.*

Pound.	Ounces.	Drachms.	Grains.	French Grammes.
1	= 16	= 256	= 7000	= 453.25
	1	= 16	= 437.5	= 28.328
		1	= 27.343	= 1.77

### *Imperial Measure.*

Gallon.	Pints.	Fluid Ounces.	Fluid Drachms.	Minims.
1	= 8	= 160	= 1280	= 76800
	1	= 20	= 160	= 9600
		1	= 8	= 480
			1	= 60

### *Weight of Water at 62°, contained in the Imperial Gallon, &c.*

					Grains.
1	Imperial Gallon	.	.	=	70000
1	" Pint	.	.	=	8750
1	" Fluid Ounce	.	.	=	437.5
1	" Fluid Drachm	.	.	=	54.7
1	" Minim	.	.	=	0.91



*Cubic Inches contained in the Imperial Gallon, &c.*

				Cubic Inches.
1	Imperial Gallon	.	.	277·276
1	„	Pint	.	34·659
1	„	Fluid Ounce	.	1·732
1	„	Fluid Drachm	.	0·2166
1	„	Minim	.	0·0036

## FRENCH WEIGHTS AND MEASURES.

*Measures of Length.*

		English Inches.					
Millimetre	=	·03937					
Centimetre	=	·39371					
Decimetre	=	3·93710					
Metre	=	39·37100	Mil.	Fur.	Yds.	Feet.	In.
Decametre	=	393·71000	= 0	0	10	2	9·7
Hecatometre	=	3937·10000	= 0	0	109	1	1
Kilometre	=	39371·00000	= 0	4	213	1	10·2
Myriometre	=	393710·00000	= 6	1	156	0	6

*Measures of Capacity.*

		Cubic Inches.			English Imperial Measure.		
			Gal.	Pints.	F. oz.	F. drms.	Min.
Cubic Centimetre } or Millilitre }	=	·06102	= 0	0	0	0	16·3
Centilitre	=	·61028	= 0	0	0	2	43
Decilitre	=	6·10280	= 0	0	3	3	2
Litre	=	61·02800	= 0	1	15	1	43
Decalitre	=	610·28000	= 2	1	12	1	16
Hecatolitre	=	6102·80000	= 22	0	1	4	40
Kilolitre	=	61028·00000	= 220	0	12	6	24
Myriolitre	=	610280·00000	= 2200	7	13	4	48

*Measures of Weight.*

		English Grains.			
Milligramme	=	·0154			
Centigramme	=	·1543			
Decigramme	=	1·5432			
Gramme	=	15·4323			
Decagramme	=	154·3234	=	Poun.	Oun.
Hecatogramme	=	1543·2348	=	0	0
Kilogramme	=	15432·348	=	0	3
Myriogramme	=	154323·480	=	2	3
					Drm.
					5·65
					8·5
					5
					2

*Avoirdupois.*



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