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THE SCIENCE OF HYGIENE

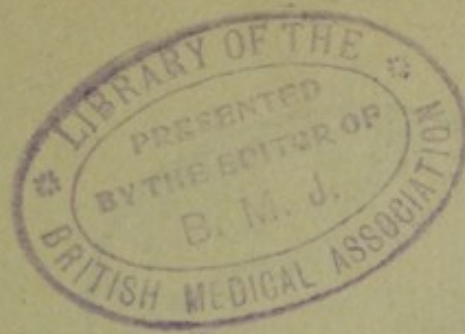
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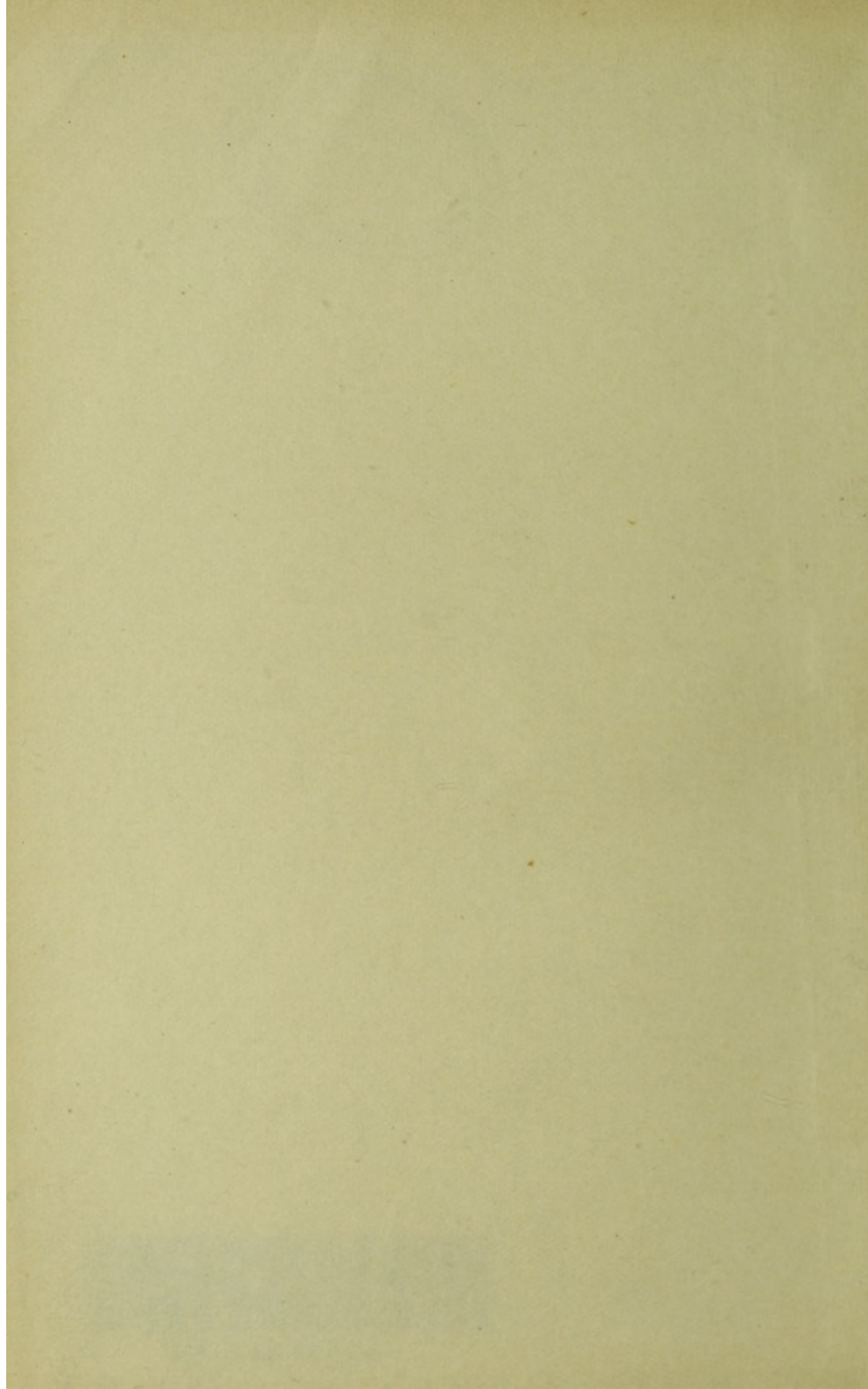


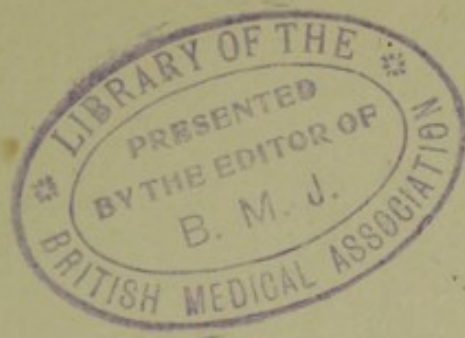
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THE SCIENCE OF MEDICINE





THE SCIENCE OF HYGIENE



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THE SCIENCE OF HYGIENE

A TEXT-BOOK OF LABORATORY PRACTICE
FOR PUBLIC HEALTH STUDENTS

BY

WALTER C. C. PAKES

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NEW EDITION

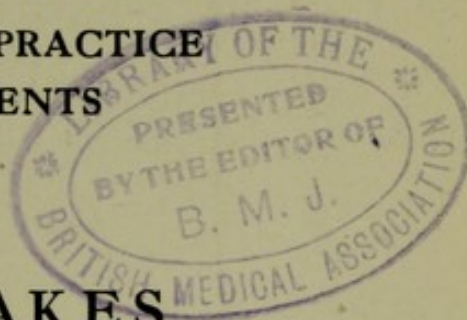
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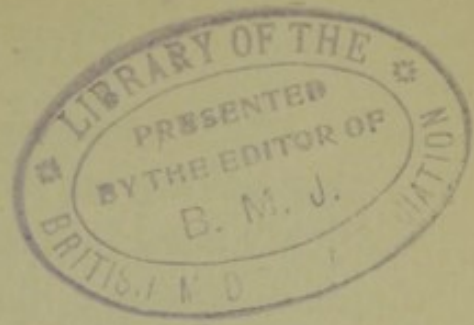
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PREFACE TO THE REVISED EDITION

THIS book is intended for the use of students who are working for a diploma in public health; and it is hoped that they will find it useful not only before their examinations, but afterwards, should they happen to work again in public health laboratories.

When I was working for my D.P.H. I found the first edition of Dr. Pakes' book more valuable than any other text-books on laboratory work: the arrangement of the subject matter was simple and concise, there was no unnecessary overlapping, and what the Author had to say he said plainly and directly. I have tried, in this new edition, to attain the same standard of excellence.

All the practical laboratory work, apart from bacteriological methods, required by D.P.H. students, is included in this volume; and no effort has been spared to make the book complete. Many modern text-books have been consulted; but nothing has been merely transferred from these.

The first edition of this book contained chapters on physics, bacteriology, and vital statistics. It was felt, however, that such subjects were better treated in other, and necessarily larger, books; and that much of these sciences could hardly be considered as laboratory work. For these reasons, no part of this volume is devoted to these subjects.

On the other hand, many additions have been made, and much of the text has been re-written.

The illustrations in this volume, which appeared also in the first edition, are the admirable work of Dr. T. G. Stevens.

My thanks are especially due to Dr. A. J. Malcolm, my fellow-demonstrator in the Public Health Laboratories of King's College, for his help and suggestions.

A. T. N.

KING'S COLLEGE

UNIVERSITY OF LONDON



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THE SCIENCE OF HYGIENE

WATER ANALYSIS

COLLECTION OF SAMPLES

IN collecting samples of water for chemical analysis, attention should be paid to two points: first, to the cleanliness of the vessel in which the water is to be collected; and, secondly, to the actual sample itself. With regard to the first, a glass-stoppered Winchester quart bottle is by far the best for a collecting vessel: this bottle should be thoroughly cleansed. It should be well rinsed with strong sulphuric acid, subsequently with distilled water until all trace of acid has disappeared, and finally with ammonia-free water. The stopper should be replaced and tied down but *not sealed*. In actually collecting the samples the bottle should first be filled with the water to be collected and emptied, and this should be done a second time. After this a true sample should be allowed to run in the bottle if collected from a tap, or the bottle should be sunk into the water so that the mouth is about two inches below the surface if collected from a reservoir or river. The water should not fill the bottle, but should come just above the shoulder. In sending the water to a laboratory for examination, it is convenient, after having replaced the glass stopper and tied it down, to place the bottle in the well-known baskets made for that purpose.

With regard to the second point, the actual collection of the sample, it should be borne in mind that the sample should really be one to test the potentiality for evil of the water in question. For instance, if the water is suspected of containing lead, it would be obviously unfair to send for analysis the water which has been standing for some hours in lead pipes, since the consumers do not drink this water as a matter of common practice.

In other words, in order to collect a water sample for examination, that sample should be such as is ordinarily consumed.

If the water be one from a reservoir or tank, it should be collected as it is flowing from such reservoir or tank into the mains.

In the case of a well water the same would hold, that is to say, the pipe should not be emptied of the water if this is not done as a matter of ordinary practice.

It sometimes happens that owing either to a leaden or leaky pipe, the water from a well may be quite good as it comes from the well, but bad as it is delivered. In this case it may be necessary to examine samples of the water collected from both ends of the pipe. Again, the tap is sometimes contaminated, and it may be necessary to clean that thoroughly before collecting the samples.

In the case of a river it is obvious that the water may be taken from either above or below any sources of contamination or pollution that may flow into the river. If therefore we wish to analyse such a water, it must be taken into consideration whence the water from the river is usually removed for drinking purposes. For example, if the intake of a company is at a certain point in the river the sample should be collected at a point as near the actual intake as possible.

The water should be examined as soon after collection as possible, as there is no doubt that changes do occur after a lapse of a short time only, in the water kept even in a well-stoppered bottle.

This is especially the case with unstable waters, such as sewage effluents, or superficial wells liable to pollution. If the water therefore has to be transmitted a considerable distance it is advisable, wherever possible, to surround the Winchester quart with ice. By doing so the changes in the water are retarded, and the water as analysed is practically the water as collected.

The bottle should have a label attached to it. The label should bear the following particulars:—

1. The name and address of sender.
2. The date of collection of the sample.
3. Source of water (river, well, etc.).

If the water is from a well, further details should be added to the above:—

1. The Depth of the well.
2. The nature of the Geological formation from which the water is obtained.

3. Proximity to Sea or Tidal River.
4. Proximity of Drains, Cess-pools, Manured Lands, or collections of decaying organic matter.
5. Any other details which may seem to bear upon the purity of the sample.

WATER REPORT

A chemical water report should contain a statement of the salts and organic material present in the water. These figures, while they afford evidence as to possible pollution, either recent or remote, enable us to judge of the general fitness of the water for drinking or domestic purposes.

A considerable amount of confusion has been caused by the fact that different analysts return their results in different forms. It will be understood that the amount of these materials must, in a sample of water, be extremely small. In order therefore to state the results without necessitating the use of fractions of a milligram, it is usual to express results as parts per 100,000, per 1,000,000, per 100,000,000, or in grains per gallon. It may be stated that there are advantages in each of these methods. In England a fairly large number of analysts still return their results in grains per gallon. On the Continent, however, it is universally the case that they are reported either in parts per 100,000 or in parts per 1,000,000. It will be seen therefore that reports expressed in parts per 100,000 can be compared with the Continental reports without previously converting.

It is sometimes the custom to express the ammonia in parts per 1,000,000 or in parts per 100,000,000, while the rest of the report is expressed in parts per 100,000. This method is adopted in order that the ammonia should be returned in units and not in decimals, but the disadvantage of having two forms of return in the same report would appear to be greater than that of having the ammonia expressed in decimals. If one accustoms oneself to the quantities expressed in parts per 100,000, it is merely a matter of remembering that 0.005 is the limit instead of 5. In the following pages, therefore, the results will all be tabulated in parts per 100,000.

When a report is expressed in grains per gallon and it is necessary to convert these figures into parts per 100,000, it is obviously only necessary to multiply these results by $\frac{10}{7}$. If, on the other hand, we require to convert parts per 100,000 into

grains per gallon, it is necessary only to multiply by 0.7. The reasons for this, of course, are that there are 100,000 grammes in 100,000 c.c. and 70,000 grains in a gallon.

PHYSICAL CHARACTERS

Before proceeding to the analysis of the water by the various chemical methods which are adopted, it is generally necessary to make some preliminary observations on the physical characters of the sample.

1. **Turbidity.** Waters vary somewhat in the amount of turbidity they show, although as a rule the best waters are perfectly clear. It is of course possible that a perfectly safe drinking water might have been rendered turbid through the shaking up with it of some mineral matter, but, speaking generally, a water that shows any marked turbidity will prove, upon further examination, to be unfit for drinking purposes. One expresses the degrees of turbidity as "clear," "slightly turbid," or "very turbid," the last applying more especially to sewage or sewage effluents.

2. The next point to be considered is **the colour.**

In order to determine the colour it is necessary first of all to allow any sediment to deposit. The clear supernatant water is then poured into what is known as a two-foot tube. This consists of a glass cylinder, at each end of which a piece of plate glass is screwed. When the water is poured in, the plate which has been removed in order to allow of this should be screwed down, and the whole apparatus carefully wiped. The eye should then be placed at one end, and at the other a white porcelain slip or piece of paper. Good waters are generally slightly blue when seen through this tube; if there is any yellowish or brownish colour, there will be some suspicion of sewage contamination unless the water happens to have been collected from a peaty soil. Occasionally the water possesses a decidedly green colour owing to the presence of the green algæ, which are of course, in themselves, harmless.

3. **Taste.** This is by no means a valuable test; the pleasant taste of a water is generally due to the solution of gases, and even the best of waters when not aerated are insipid. When it is remembered that a smaller quantity of sodium chloride than 75 grains to the gallon or 100 grammes to the 100,000 cannot be

tasted, it will be seen of what little worth the taste is. Some algæ, however, when they decompose liberate volatile oils, of which even a trace may cause a water to taste and smell unpleasantly. Such algae are found chiefly in reservoirs and filter beds: their decomposition products, although they make the water objectionable, do not seem to make it, in any way, dangerous to health.

4. **Smell.** A good drinking water should, of course, be absolutely inodorous. If there is a marked degree of contamination, or the water has been collected from a peaty soil or from the neighbourhood of some dye or chemical works, it is possible that the water may possess some odour. It by no means follows, however, that a water which is so contaminated as to be utterly unfit to drink possesses any odour at all.

In order to detect the odour, the most convenient method is to place about 250 c.c. in a glass-stoppered bottle, which is then placed in a water bath or oven at about 30° C. for a few minutes. The stopper should then be removed and the nose applied to the bottle at once. This is necessary, since the odour is extremely evanescent.

5. As has been before stated, a palatable water is **well aerated**. In order to test this properly it is generally only necessary to pour some of the water into an open beaker, and notice the evolution of small bubbles of gas. This test is of no value as regards the fitness of the water for drinking purposes, but is merely evidence of its palatability.

6. **Reaction.** The reaction of most drinking waters is alkaline. Occasionally a drinking water is found to be acid, and is then held not to be suitable for a water-supply because of the facility with which it takes up lead. This has been found to be the great objection to the Yorkshire moor supply for Sheffield, etc. Such a water derived from peaty soil contains humic and ulmic acids. The alkalinity or acidity of a water in itself is no criterion of the pollution of the water by sewage, since most waters which are highly contaminated by sewage still retain their alkalinity. The waters drawn from the neighbourhood of dye or chemical works are sometimes acid, but other criteria of their unsuitability for drinking purposes will be found on further analysis.

7. **The Sediment.** As has been before stated, a good water should contain no sediment, but the mere presence of a slight sediment will not necessarily condemn the water. The sediment may consist either wholly of mineral matter, or of vegetable

matter, or of both. Its nature is generally discovered by microscopical examination, and will be treated under that head.

TOTAL SOLIDS

Apparatus required

1. A platinum dish.
2. A flask graduated at 200 c.c.
3. A water bath.
4. A water oven.
5. A desiccating chamber.
6. A balance which will turn to $\frac{1}{5}$ of a milligram.

The Process

In a clear water with no sediment it is obvious that the solids will be those in solution. In turbid waters the total solids may be either those in solution plus those in suspension, or those in solution alone. Most analysts determine the total solids in solution; if, therefore, the water is turbid, it must be allowed to sediment, or must be filtered before proceeding with the determination.

1. 200 c.c. of the clear water should be measured out into the graduated flask.
2. The platinum dish should be thoroughly well cleansed with HCl, and subsequently with distilled water; then heated in the Bunsen flame and placed in the desiccator over H_2SO_4 until it is cold. It must then be weighed, and a note of its weight taken.
3. Into the dish as much of the 200 c.c. as it will conveniently hold is poured. The dish is then placed over a water bath and covered with an inverted funnel in order to protect the contents from the dust. From time to time more water is added until the whole of the 200 c.c. has been evaporated to dryness.
4. The dish with the evaporated contents is now removed from the water bath and placed in the water oven and kept at 100°C . for from 20 minutes to half an hour.
5. Now remove the dish from the oven and place it in the desiccator over H_2SO_4 until it is cool.
6. Now weigh it carefully two or three times, replacing it in the desiccator for 5 or 10 minutes between each weighing.

Instead of heating the dish in the water oven at 100°C . some analysts heat it in an air oven at 120°C . This latter method ensures the disappearance of the water of crystallization from any of the salts which may be present with their water of crystallization in the residue evaporated at 100°C .

EXAMPLE

Weight of dish	29.7834
„ of residue of 200 c.c. of water + dish	30.0030
<hr/>	
Weight of residue	0.2196
\therefore Total solids per 100,000 = $0.2196 \times 500 = 109.800$	

Notes

If it is necessary to determine the total solids in a short time, a less quantity of water than 200 c.c. may be taken; but as the residue is sometimes only very small the error of experiment must necessarily be greater. In a sample of water which is known to be very hard, or to be very saline, 100 c.c. will be ample.

LOSS ON IGNITION

After the total solids have been ascertained, the platinum dish containing the residue is heated over the flame of a Bunsen burner to a white heat for some time, allowed to cool and re-weighed. The loss in weight is then recorded as Loss on Ignition.

Whilst the solids are being ignited, blackening will take place if much organic matter is present, and this fact should be noted.

The loss on ignition represents to a large extent the organic matter present, since the ignition causes this to be oxidized, and CO_2 and H_2O to be given off. It is not, however, altogether a measure of the amount of organic matter, since the water of crystallization and the ammonium salts are also driven off.

EXAMPLE

Weight of dish + total solids	30.0030 grammes
„ „ + total solids (after ignition)	29.9939 „
<hr/>	
Loss on ignition (200 c.c.)	0.0091 gramme
\therefore Loss on ignition per 100,000 = 4.55 grammes.	

ESTIMATION OF CHLORIDES

Apparatus, etc., required

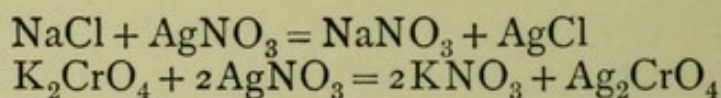
1. A white porcelain evaporating dish.
2. A small glass stirring rod.
3. A 50 c.c. pipette.
4. A burette graduated in 0.1 c.c.
5. Potassium chromate solution.
6. Standard silver nitrate solution.

The Process

1. Fill the burette with the standard silver solution.
2. Measure 50 c.c. of the water to be examined in the graduated pipette and run it into the white evaporating dish.
3. Add one or two drops of the solution of K_2CrO_4 to the water and stir.
4. Allow the silver solution to run into the water drop by drop until the evanescent brown colour remains.
5. Read off the height of the solution in the burette and note it.

Explanation of the Process

When a solution of silver nitrate is added to an alkaline solution of a chromate a reddish brown precipitate of silver chromate is formed. If, however, any chloride is present, the silver will combine with this chloride before it combines with the chromate. The interactions taking place are expressed by the following equations:—



When, therefore, the permanent brown precipitate is formed, all the chloride will have combined with the silver, and the amount of the silver used at this juncture will be a measure of the chlorides present in the water.

EXAMPLE

1st reading of burette	.	.	.	6.6 c.c.
2nd ,, ,,	.	.	.	8.9 ,,
				—
Amount of silver used	.	.	.	2.3 ,,

Now 1 c.c. of silver nitrate solution = 1 milligram of chlorine

$\therefore 2.3$ " " = 2.3 " "

But this quantity was present in 50 c.c. of the water

\therefore in 100 c.c. there will be 4.6 milligrams of chlorine

i.e. there will be 4.6 parts of chlorine in 100,000 parts of water.

The quantity of chlorine is frequently expressed in terms of NaCl as well as in terms of Cl itself. In order to express the Cl in terms of NaCl it is only necessary to multiply the weight of Cl

by $\frac{58.5}{35.5}$. In our example we found that 4.6 parts of Cl were

present. Expressed as NaCl this will be $1.648 \times 4.6 = 7.5$ parts.

Our report should read :—

Chlorine	.	.	.	4.6 parts per 100,000
Expressed in NaCl	.	.	.	7.5 " "

Notes

The estimation should always be repeated. If we know when to expect the "end point," we are able to estimate more easily the exact point when the colour changes.

Before filling the burette it must be scrupulously clean. A new burette should be washed out with strong sulphuric acid and subsequently with distilled water until all traces of the acid have disappeared. After it is clean, a few c.c. of the solution should be poured in and (holding it horizontally) allowed to run over the whole of the surface. The burette must be emptied and filled with the solution. A few drops must now be allowed to run out, in order to fill the nozzle, and any drop which hangs on the nozzle must be removed. The height of the fluid in the burette must be carefully read and noted. In the case of colourless fluids it is customary to read off the division of the scale which corresponds to the bottom of the meniscus. In the case of indigo, however, it is convenient to read off the division of the scale which corresponds to the top of the meniscus.

TOTAL HARDNESS

Apparatus, etc., required

1. A small glass-stoppered bottle (about 125 c.c. capacity).
2. A burette graduated in tenths of a c.c.
3. An Erlenmeyer flask.
4. A 50 c.c. pipette.
5. A 100 c.c. measure graduated in c.c.
6. Standard soap solution.

The Process

1. Fill the burette with the standard soap solution, and read off the height.
2. Measure 50 c.c. of recently boiled distilled water into the bottle.
3. Add 0.5 c.c. of soap, replace the stopper, and shake the bottle well. Add more soap, a few drops at a time, until a permanent lather is formed after brisk shaking.
4. Read off the height of the soap in the burette.

The difference between the two readings will be the amount of soap required to form a lather with perfectly soft water.

(*Note.*—It is well for a beginner to do this several times before he begins to try to determine the hardness, so as to familiarize himself with the appearance of a permanent lather, and in order that he may find for himself the amount of soap which must subsequently be deducted from that used to form a lather with the water under examination.)

After having done this, proceed to the examination of the water.

1. By means of the 50 c.c. pipette, run 50 c.c. of the water into the bottle.
2. Add 1 c.c. or less of the standard soap at a time to the water and shake well. When a certain amount of the soap has been added, the lather, which is at first very transient, begins to remain for a short time. When this point has been reached, the soap must be added only a few drops at a time, in order not to overshoot the mark.
3. When sufficient soap has been added the bubbles on the surface of the water will break very slowly; the bottle should then be laid on its side for five minutes. If at the end of this time the lather is still present, even if it is diminished in thickness, the estimation is done, and it only remains to read off the height of the soap in the burette, and deduct from it the height observed before beginning the estimation. The difference will be the amount of soap required, and from this is calculated the hardness.

Explanation

A soap is a salt, the base of which is a metal and the acid one of the fatty acids. Some of these salts, as those of sodium and potassium, are soluble in water, and when rubbed up with or

shaken up in water cause a lather. Others, such as those of calcium and magnesium, are insoluble in water, and therefore are precipitated; being precipitated, they are incapable of forming a lather. If a soluble calcium salt is present in water and a solution of a sodium soap is added, the insoluble calcium soap is formed.

The cause of hardness in water is the presence in it of soluble salts of calcium and magnesium. So long as either of these salts exists unprecipitated in the water, it will be impossible to form a lather. Directly they have been precipitated only a slight addition of soap is required to form a lather. The amount of soap required to form a lather is therefore the measure of the amount of the salts of calcium and magnesium present in the water.

EXAMPLE

It was found that it took 0.8 c.c. of the soap solution to form a lather with 50 c.c. of distilled water.

The sample of water to be examined took 7.6 c.c. of soap to form a lather with 50 c.c.

50 c.c. sample required .	.	.	7.6 c.c. soap
50 c.c. distilled „	.	.	0.8 „ „

Soap solution required to precipitate Ca and Mg salts = 6.8 c.c.

But 1 c.c. of the Standard soap solution = 1 milligram of CaCO_3 .

\therefore 50 c.c. of the water contain 6.8 milligrams of Ca and Mg (expressed as CaCO_3).

\therefore 100 c.c. of the water contain 13.6 milligrams.

In other words, the sample has a hardness of 13.6 parts per 100,000.

Notes

It will have been noticed that the hardness is expressed in terms of CaCO_3 , whether it is due to calcium or magnesium. This is merely for convenience, so that one need only have one standard solution. As a matter of fact, more soap is required to form a lather with a certain amount of magnesium than is required for an equivalent quantity of calcium.

The soap solution does not remain permanently of the same strength. After the lapse of a certain time it undergoes certain changes and becomes weaker. Unfortunately this change does not show any degree of constancy. The solution may remain up to standard for weeks, and then suddenly change.

In order, therefore, to ascertain the hardness of any sample

of water with great accuracy, three determinations should be made:—

1. The soap required to lather 50 c.c. of distilled water.
2. The soap required to lather 50 c.c. of distilled water containing 6 c.c. of the standard calcium solution.
3. The soap required to lather 50 c.c. of the sample.

EXAMPLE

No. 1 required . . . 1.0 c.c.
 No. 2 „ . . . 8.4 „
 No. 3 „ . . . 15.6 „ (diluted 1 in 2 took 7.8 c.c.)

6 c.c. Ca solution required . . . $8.4 - 1 = 7.4$ c.c.

Hardness in sample „ . . . $15.6 - 1 = 14.6$ „

But 7.4 c.c. of soap solution = 6 milligrams of CaCO_3 .

$\therefore 14.6$ c.c. „ „ = $\frac{14.6}{7.4}$ of 6 milligrams of CaCO_3
 = 11.8 milligrams of CaCO_3 .

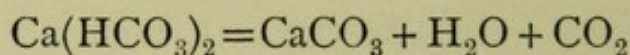
Thus 50 c.c. of the sample contain 11.8 „ „

and 100 c.c. „ „ 23.6 „ „

\therefore 100,000 parts contain 23.6 parts of hardness, expressed as CaCO_3 .

PERMANENT HARDNESS

Hardness is spoken of as either temporary or permanent. The former consists of calcium bicarbonate and is held in solution by the carbonic acid in the water. When the carbonic acid is expelled from the water as by boiling, the bicarbonate is converted into the carbonate and is at the same time precipitated, as it is insoluble in water.



The permanent hardness is due to the sulphates, chlorides, and nitrates of calcium and magnesium, and these are unaffected by boiling.

The Process

1. Measure 100 c.c. of the water to be examined in a graduated cylinder and pour it into an Erlenmeyer flask.
2. Boil over a piece of wire gauze until the bulk is reduced to about one-half.
3. Allow it to cool and filter through a hard white filter-paper which has been well washed with distilled water.

4. Make up the volume to 100 c.c. with distilled water.
5. Take 50 c.c. of this and estimate the hardness as before.

This estimation is also expressed in terms of CaCO_3 .

Having estimated the total and the permanent hardness, the difference will obviously be the temporary hardness.

Notes

The hardness is expressed in grains per gallon (degrees), or in parts per 100,000 of CaCO_3 . Although this does not represent the actual truth, since other salts have their share in producing this hardness, it is an expression for the factor of practical importance, i.e. the soap-destroying power of the water.

In laundry work the whole work of the water used exercises its soap-destroying power, and it is only when the soap is in excess of this, that the detergent action of the latter begins.

It will occur to the reader that in washing one's hands, only the small quantity of water adhering to the hands need affect the soap, before a lather is obtained. Therefore it is possible to wash the hands with comfort in a much harder water than can be used in the laundry without a great waste of soap.

FREE OR SALINE AMMONIA

Apparatus, etc., required

1. A 32-ounce glass retort, or a long-necked two-litre glass flask.
2. A condenser.
3. Clamps and burner.
4. 12 Nessler glasses to hold 100 c.c. with a graduation at 50 c.c.
5. White glazed porcelain slab.
6. 50 c.c. burette graduated in tenths of a c.c.
7. 200 c.c. Erlenmeyer flask.
8. A 2 c.c. pipette.
9. Nessler's reagent.
10. Standard solution of Ammonia (1 c.c. = 0.01 milligram of NH_3).

Before beginning the process for the estimation of ammonia it is advisable to practise the method first on known quantities of ammonia added to water, and secondly on unknown quantities which are also added to the water.

1. In order to do this add 1 c.c., 2, 3, etc., up to 10 and 15 c.c. of the standard ammonia solution to each of ten of the Nessler glasses, fill each up to the 50 c.c. mark with distilled ammonia-free water, and add 2 c.c. of Nessler's solution. Allow this to stand for two minutes after well shaking.
2. Compare these tints carefully in order to get an idea of the depth of tint produced by the varying quantities of ammonia. After having become familiar with these tints, add a small unknown quantity of ammonia to the 50 c.c. of distilled water in a Nessler glass and estimate the amount added.
3. Repeat this several times in order to get an idea of the approximate amount of ammonia which a certain tint indicates.

Suppose that we find that 5 c.c. of the standard ammonia solution gives a darker tint than the unknown quantity, we add 3 c.c. and find this too little; next we add 4 c.c. and also find this too little; the addition of 4.5 exactly matches, that is to say, our unknown quantity contains the equivalent of 4.5 c.c. of the standard ammonia solution. As the standard ammonia contains 0.01 milligram per c.c. it is evident that the 50 c.c. of water contains $4.5 \times 0.01 = 0.45$ milligrams. There is another method of determining the amount present in a Nessler glass when the tint cannot be exactly matched by any of the standard solutions we have made. For example, the unknown quantity is found to be less than the equivalent of 5 c.c. of the standard ammonia. By means of a clean pipette remove some of the fluid in the standard solution until, when both glasses are looked at from above over the white slab, the tints are exactly alike; next measure the fluids left. Suppose 35 c.c. are left of the standard solution, and match 50 c.c. of the test solution; then the ammonia in the test-glass equals $\frac{35}{50}$ of the standard. But the standard contains 5 c.c. of the ammonia solution; therefore the test-glass contains $\frac{35}{50}$ of 0.05 milligrams of NH_3 , and this equals 0.035 milligrams of ammonia.

If the fluid contained in the test-glass is deeper than that in the standard glass with the 5 c.c. of the ammonia solution, some of the fluid from the test-glass must now be removed until the tints are exactly alike. Suppose 10 c.c. have to be removed, then 40 of the test equal 50 of the standard; therefore 50 of the sample equal $\frac{50}{40}$ of 50 c.c. of the standard. But the standard contains 0.05 milligrams of ammonia; therefore our test contains 0.0625 milligrams of ammonia.

The Process

1. Rinse out the retort or flask with strong HCl, and subsequently with good tap water, until all traces of the acid have disappeared, then rinse out well with two or three lots of distilled ammonia-free water and empty.
2. Fix the retort or flask in a good clamp and attach the condenser, which is also supported by a clamp.
3. When the retort and condenser are fitted together, connect the outside case of the condenser with the water tap, remembering to connect it in such a way that the water runs in from below upwards. Do not turn on the tap yet.
4. Pour about 500 c.c. of ammonia-free distilled water into the flask or retort, add a few grains of pure sodium carbonate and a few pieces of broken pumice. The latter will prevent "bumping." Light the burner and put it under the flask, the bottom of which should be protected with wire gauze.
5. Distil until the steam has issued from the lower end of the condenser for several minutes, and then turn on the water through the outer casing of the condenser.
6. Collect about 200 c.c. of the water, and as each 50 c.c. comes over add 2 c.c. of Nessler. If the condenser and retort are perfectly clean there will be no colour in the third or fourth distillates.
7. After thus cleaning the condenser and flask, add 500 c.c. of the water to be examined. The small amount of Na_2CO_3 present in the flask will neutralize any acid in the sample water, and leave the ammonia free to be distilled off. Replace the stopper or cork and begin to distil.
8. Distil over 50 c.c. into a Nessler glass. When this is done collect the next distillate in a fresh Nessler glass and add 2 c.c. of Nessler to the first distillate. If a colour is developed upon the addition of the Nessler, determine the amount of ammonia present as explained above. When the second Nessler glass is full remove that and place the third. Add 2 c.c. of Nessler to the second distillate and if there is still a colour produced determine the amount of ammonia in the second distillate. Continue to do this until the addition of Nessler to the distillate produces no colour. (It is generally found that the whole of the saline ammonia comes off in the first 150 c.c. of the distillate, so that one always expects to find no ammonia in the fourth 50 c.c. distilled over.)

9. Having determined the quantity of ammonia present in each of the distillates, add these quantities together and it will represent the quantity of saline ammonia present in 500 c.c. of the water. From this the amount present can be readily expressed in parts per 100,000.

EXAMPLE

The first distillate of 50 c.c. was matched by 1.5 c.c. of standard NH_3 .

The second distillate of 50 c.c. was matched by 1.0 c.c. of standard NH_3 .

The third distillate of 50 c.c. was matched by 0.5 c.c. of standard NH_3 .

The fourth distillate gave no colour with Nessler's solution.

\therefore the three distillates, or all the free and saline ammonia in the 500 c.c., were matched by 3 c.c. of the standard NH_3 solution.

In other words, 500 c.c. contained 0.03 milligrams of ammonia.

\therefore 100 " " 0.006 " " "

Or there were .006 parts of free and saline ammonia in 100,000 parts of the sample.

ALBUMINOID AMMONIA

The Process

1. While the saline ammonia is being distilled off, 50 c.c. of the alkaline permanganate solution should be poured into an Erlenmeyer flask and about 150 c.c. of ammonia-free water added. This must now be boiled until the bulk is reduced to 100 c.c.
2. When all the free and saline ammonia has been distilled off and the distillation stopped, the alkaline permanganate is poured carefully into the flask or retort, the stopper is replaced, and the distillation begun again.
3. As the water comes off it should be collected in the Nessler glasses as before, and the Nessler reagent added.
4. The quantity of ammonia must be estimated as in the case of the saline ammonia.

Explanation

The nitrogenous matter in the water is reduced by boiling with alkaline permanganate, and converted into ammonia, which is distilled off.

Notes

The preliminary boiling of the permanganate ensures the absence of both ammonia and organic matter, and therefore all the ammonia distilled over must come from the water under examination.

The organic matter is not all reduced instantaneously, but more or less gradually. The ammonia does not therefore come off necessarily in the first 150 c.c. of the distillate as does the saline ammonia.

Every 50 c.c. of water as it comes off must therefore be tested with Nessler reagent until there is no reaction.

If a sample of water contains a considerable quantity of organic matter, it sometimes happens that there is a danger of the retort boiling almost dry. When this occurs 100 or 200 c.c. of organically pure ammonia-free water must be added to the retort, and the distillation continued.

TESTS FOR THE PRESENCE OF NITRITES

BY POTASSIUM IODIDE AND STARCH

Apparatus, etc., required

1. Two Nessler glasses.
 2. Potassium iodide solution.
 3. Starch solution.
 4. Dilute sulphuric acid (about 10%).
1. Pour 50 c.c. of the water to be tested in a Nessler glass and 50 c.c. of distilled water into another.
 2. Add a few drops of the KI solution to each of the Nessler glasses and then a few drops of the starch solution.
 3. Add a few drops of dilute sulphuric acid to each tube.

If nitrites are present, a blue colour will be immediately formed, the depth of the colour depending on the amount of nitrites present.

Explanation

The sulphuric acid liberates nitrous acid from the nitrites. The free nitrous acid then liberates iodine from the potassium iodide, and this iodine combines with the starch and gives the blue colour

This is an extremely easy method of determining the presence

of nitrites, but care must be taken to make the observations at once, since nitrates may give the same reaction after the lapse of a short interval.

BY METAPHENYLENE-DIAMINE-HYDROCHLORIDE

GRIESS'S METHOD

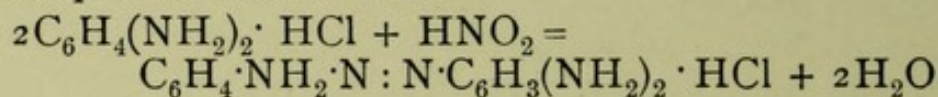
Apparatus required

1. Two Nessler glasses.
2. Solution of metaphenylene-diamine-hydrochloride.
1. Pour 50 c.c. of the water to be tested into a Nessler glass, and 50 c.c. of distilled water into another.
2. Add to each glass 1 c.c. of the solution of metaphenylene-diamine-hydrochloride, and a few drops of hydrochloric acid.

If nitrites are present a brown colour will be formed in the sample, due to the production of Bismarck Brown.

Explanation

Metaphenylene-diamine-hydrochloride is a colourless solution which in the presence of nitrous acid gives rise to triamido-azobenzol or Bismarck Brown, and hence colours the solution. The hydrochloric acid is added to liberate nitrous acid from the Nitrites present in the water.



Notes

Metaphenylene-diamine-hydrochloride, when dissolved in water, tends to become dark in colour, and this interferes with the delicacy of the test. The solution is better when made fresh; failing this it must be filtered through animal charcoal until it is colourless.

QUANTITATIVE ESTIMATION OF NITRITES

GRIESS'S METHOD

Apparatus required

1. Nessler glasses.
2. Solution of metaphenylene-diamine-hydrochloride.
3. Standard nitrite solution.

The Process

1. Into each of ten Nessler glasses put varying amounts of the standard nitrite solution—1 c.c., 2 c.c., etc., to 10 c.c.
2. Fill each glass up to the 50 c.c. mark with distilled water. Add to each a few drops of HCl.
3. Add 50 c.c. of the sample water to another Nessler glass, together with a few drops of HCl.
4. Add 1 c.c. of metaphenylene-diamine-hydrochloride solution to each glass.
5. Compare the various tints, and see with which of the ten glasses the sample water matches.
6. If the sample is too dark, dilute with twice or more times the volume of distilled water and repeat the process from the beginning.

EXAMPLE

It was found that the Nessler glass containing the sample water matched the Nessler glass in which there were 5 c.c. of the standard nitrite solution.

Now 1 c.c. of the standard nitrite solution = 0.01 milligrams of N_2 as nitrite.

\therefore 5 c.c. of the standard nitrite solution = 0.05 milligrams of N_2 as nitrite.

Thus in 50 c.c. of the water under examination there were 0.05 milligrams of N_2 as nitrite.

So in 100 c.c. there were 0.1 milligrams of N_2 as nitrite, or, in other words, 0.1 part per 100,000.

Notes

Griess's method estimates nitrites alone.

The principle involved is the same as in Nessler's method of estimating ammonia, and the student will find that when he has mastered the one method the other will come easily to him.

TESTS FOR THE PRESENCE OF NITRATES

BRUCINE METHOD

1. 2 c.c. of the suspected water are placed in a perfectly clean white porcelain dish and evaporated to dryness.
2. A drop of pure H_2SO_4 is allowed to drop on the residue.
3. A minute crystal of Brucine is now added.

If nitrates are present, a pink colour will appear. This test is an extremely delicate one, a reaction being obtained when the nitrate is present in the proportion of only 1 in 10,000,000.

DIPHENYLAMINE TEST

1. A few crystals of diphenylamine are put into a porcelain dish.
2. 1 c.c. of pure H_2SO_4 is added.
3. A little of the suspected water is poured into the dish. If nitrates are present a blue colour will appear. Nitrites give no colour with this test.

BY POTASSIUM IODIDE AND STARCH

The method is the same as in testing for nitrites, and is of no use for detecting the presence of nitrates if the nitrites are also there. If, however, no colour comes at once, but does come after the lapse of a few minutes, it may be inferred that nitrates, and not nitrites, are present in the water. The test, however, is not so satisfactory as those already mentioned.

QUANTITATIVE ESTIMATION OF NITRATES

PHENOL-SULPHONIC ACID METHOD

Apparatus, etc., required

1. Evaporating dishes.
2. Nessler glasses.
3. Phenol-sulphonic acid solution.
4. Standard nitrate solution.

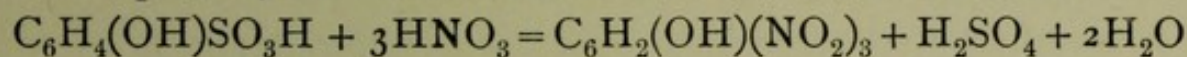
The Process

1. Place 10 c.c. of water in the dish and evaporate to dryness on the water bath.
2. Place in another dish 1 c.c. of the standard nitrate solution and evaporate to dryness in the same manner.
3. To each of the dried residues add 1 c.c. of phenol-sulphonic acid, 1 c.c. of distilled water, and 2 or 3 drops of sulphuric acid.
4. Warm gently over the water bath.
5. Distilled water is now added and excess of ammonia, and the bulk of each is made up to 100 c.c.

6. 50 c.c. of the solutions are pipetted into two Nessler glasses.
7. Compare the tints and pipette off from the darker tinted glass as much of the water as is necessary to make the tints alike when viewed from above.
8. Calculate from the quantities remaining in the glasses the amount of nitrate present.

Explanation

The nitrates present in the water convert the phenol-sulphonic acid into a mixture of nitro compounds, the chief of which is trinitrophenol, the ammonium salts of which are coloured.



The depth of the colour is then a measure of the amount of these compounds present, and so of the nitrates originally present in the water.

Notes

The phenol-sulphonic acid method estimates nitrates alone. It cannot be used for the estimation of nitrates if the water under examination contains much organic matter (as, for example, does a sewage effluent), because such organic matter gives much charring when the water is evaporated, and so interferes with the colour of the trinitrophenol. In such a case one of the following methods which estimate both nitrites and nitrates must be employed, and when the result is obtained the nitrate value of the water is determined by subtracting from the result the figure for nitrites obtained by Griess's method, the remainder, of course, being the amount of nitrates present.

QUANTITATIVE ESTIMATION OF NITRITES AND NITRATES

I. INDIGO METHOD

Apparatus, etc., required

1. A small beaker holding about 60 c.c.
2. A small glass stirring rod.
3. A 20 c.c. pipette.
4. A graduated glass cylinder to hold 20 c.c.
5. A burette graduated in 0.1 c.c.
6. Pure redistilled nitrate-free concentrated sulphuric acid.
7. Standard indigo solution.

The Process

1. Fill the burette with the standard indigo, and make a note of the height of the indigo, reading the top of the meniscus.
2. Pipette 20 c.c. of the water into the beaker, taking care to drain the pipette.
3. Measure 20 c.c. of H_2SO_4 in the cylinder.
4. Pour the H_2SO_4 into the beaker, stirring briskly, and do not wait for the last drops of the acid to drain.
5. At once allow the indigo to run into the now hot mixture drop by drop, until the greenish evanescent colour becomes permanent.
6. Read off the height of the indigo in the burette.

The estimation must now be repeated, in a slightly different order, as follows:—

1. Pipette the 20 c.c. of water into the beaker.
2. Allow about 0.5 c.c. less of the indigo than was required in the first estimation to run into the beaker.
3. Now pour in the H_2SO_4 , and stir briskly.
4. Immediately the colour is discharged, add more indigo drop by drop, until the colour is once more permanent.
5. Now read off the height of the indigo.

Explanation

The concentrated H_2SO_4 decomposes the nitrites and nitrates present and liberates free nitric acid. This then oxidizes the indigo, forming isatin, which is colourless. Directly all the nitric acid has been used up a further addition of indigo will not be oxidized and will therefore retain its colour. The amount of indigo required to give a permanent colour will indicate the quantity of nitrates and nitrites present in the 20 c.c. of water.

EXAMPLE

A second or control estimation gave the following:—

1st reading of burette	.	.	.	10.3 c.c.
2nd ,, ,,	.	.	.	12.5 ,,
				<hr/>
Amount of indigo used	.	.	.	2.2 ,,
1 c.c. of indigo = 0.086 milligrams of N_2				
\therefore 2.2 c.c. ,, = 0.1936 ,, ,,				

But this amount was contained in 20 c.c. of the water.
∴ in 100 c.c. there will be 0.968 milligrams of N_2
or 0.968 parts per 100,000 of the water.

Notes

It is common practice to standardize the indigo for the amount of water that is to be used. The same quantity should therefore always be used. 20 c.c. has been found to be convenient.

If the nitrates are present in considerable amount, the solution becomes so dark that it is very difficult to determine the "end point" with accuracy. In these cases it becomes necessary to dilute the water, generally from 1 to 4 times. Very occasionally it becomes necessary to dilute 10 or even 20 times. If it is necessary to dilute 4 times, 10 c.c. should be thoroughly well mixed with 30 c.c. of distilled water, and 20 c.c. of the diluted water pipetted into the beaker and treated as in the case of the undiluted water.

When 20 c.c. of the concentrated H_2SO_4 are added, the right amount of heat is generated to allow the oxidation of the indigo by the liberated nitric acid. The estimation must therefore be done as quickly as possible. It is in order that this temperature may be maintained up to the end point that the second order of procedure given above is adopted.

II. COPPER-ZINC COUPLE METHOD

Apparatus, etc., required

1. Zinc foil, about 9 sq. inches.
2. 3% solution of $CuSO_4$.
3. Wide-mouthed, glass-stoppered bottle of about 8 oz. capacity.
4. 100 c.c. graduated measuring-glass.
5. 10 c.c. pipette.
6. Apparatus as in the estimation of free and saline ammonia.

The Process

1. Immerse the zinc foil in the copper sulphate solution until the surface of the zinc is coated with metallic copper.
2. Wash the foil in ammonia-free water.
3. Place the foil in the glass-stoppered bottle.
4. Put about 120 c.c. of the water to be examined into the bottle.
5. Stopper tightly, and put the bottle away in a warm dark place until the following day.

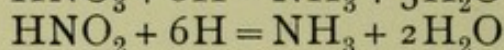
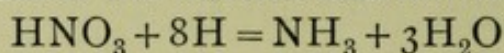
6. Then remove 10 c.c. of the water and test for the presence of nitrites by Griess's method.
7. If nitrites are present, restopper the bottle, put it away again for 12 hours and then test again.
8. If nitrites are absent, measure out 100 c.c. of the water, transfer it to a flask or retort, and estimate the ammonia in the ordinary way.

From the amount of ammonia obtained, the nitrogen as nitrites and nitrates is readily calculated.

It will be evident that by this process any ammonia which is present in the water originally, will be present after the conversion of the nitrates and nitrites into ammonia, and therefore the ammonia found will represent the ammonia reduced from the nitrates plus the original ammonia. The amount of original ammonia must be deducted in order to find the true amount of nitrates and nitrites.

Explanation

The zinc copper couple liberates nascent hydrogen from the water, and this reduces the nitrates and nitrites to ammonia.



OXYGEN ABSORBED

TIDY'S PROCESS

Apparatus, etc., required

1. Two glass-stoppered bottles holding about 350 c.c.
2. A water bath or incubator regulated at 27° C.
3. A 50 c.c. burette graduated in tenths of a c.c.
4. Organically pure ammonia-free water.
5. Thiosulphate solution.
6. Starch solution.
7. Potassium iodide solution.
8. 25% sulphuric acid.
9. Standard permanganate solution.

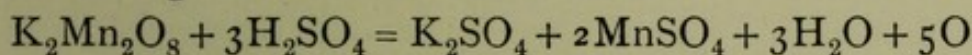
The Process

1. Measure 250 c.c. of good distilled water into one bottle (the control), and 250 c.c. of the water under examination into the other.

2. Into each of these measure 10 c.c. of the standard potassium permanganate solution and 10 c.c. of the specially prepared, organically pure 25% H_2SO_4 .
3. Shake the bottles up and place in the water bath at 27°C . for four hours.
4. At the end of this time add a few drops of the KI solution to each bottle. The pink colour will disappear and be replaced by a yellow one.
5. Fill the burette with thiosulphate solution, and carefully read the height of the fluid.
6. Run the thiosulphate into the control bottle until the yellow colour is almost discharged and then add a few drops of starch solution. The colour will now turn blue. Add the thiosulphate cautiously until all the colour is discharged, and read off the height of the fluid in the burette.
7. Repeat this with the bottle containing the sample of water.

Explanation

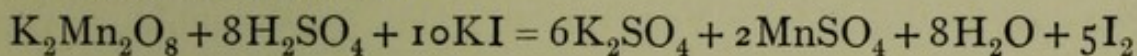
The potassium permanganate in the presence of sulphuric acid oxidizes the organic matter in the water.



The amount of this organic matter is to a certain extent gauged by the quantity of permanganate used up.

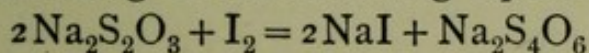
The control bottle, containing only distilled water and therefore no organic matter, will use up no oxygen, and so will contain permanganate at the end of the four hours, the equivalent of 1 milligram of oxygen.

The potassium iodide is decomposed by the permanganate, and iodine is liberated.



The amount of iodine liberated will be in proportion to the amount of permanganate in solution (i.e. not used up). In this case it will be the equivalent of 1 milligram of oxygen.

The amount of iodine liberated is now measured by the quantity of thiosulphate required to decolourize the solution, which it does according to the following equation:—



The starch solution is added towards the end of the operation in order the more easily to determine the actual end point. It performs no other function.

EXAMPLE

Control (C) took 15.2 c.c. of the thiosulphate to decolourize the iodine.

The sample of water took 13.4 c.c. to decolourize it.

15.2 c.c. of thiosulphate therefore represent 10 c.c. of the permanganate solution, and this equals 1 milligram of available oxygen.

The sample of water required 13.4 c.c. The difference between this and 15.2 c.c. represents the amount of oxygen absorbed by the water. This difference is 1.8 c.c.

Now as 15.2 c.c. of thiosulphate = 1 milligram of oxygen, 1.8 c.c. will be $\frac{1.8}{15.2}$ of 1 milligram, which equals 0.118 milligrams.

But 250 c.c. of the water was taken; therefore 100 c.c. will absorb 0.047 milligrams; or, in other words, the oxygen absorbed during four hours at 27° C. is equal to 0.047 parts per 100,000.

Notes

The results obtained by this method merely give an indication as to whether or not there is much oxidizable organic matter in the water. Various analysts take various times and temperatures in performing this experiment. For examination purposes the student will find that three hours at room temperature will be the most convenient conditions under which to perform the experiment.

One of the chief reasons for making a control examination for each sample tested, is that the solution of thiosulphate undergoes changes and gradually becomes weaker. The trouble of constantly having a standard solution, therefore, would be found in practice far greater than making a control each time. The purpose in allowing the control to stand along with the sample is to eliminate the possibilities of error due to the destruction of the permanganate by any condition other than the organic matter present in the sample.

If, as sometimes happens with a bad water such as sewage, the 10 c.c. of permanganate is totally decolourized before the expiry of the four hours, a further 10 c.c. should be added, and if necessary a third 10 c.c., and the amount of thiosulphate used to decolourize the iodine liberated deducted from twice or three

times that required to decolourize the control, in order to obtain the equivalent of oxygen absorbed, in terms of thiosulphate.

This method of ascertaining the amount of organic matter present in a sample of water is useless in the presence of iron in the ferrous state, since this takes up the permanganate with great avidity.

POISONOUS METALS

A. LEAD

1. Qualitative Tests

a. *With Sulphuretted Hydrogen*

Pipette about 100 c.c. of the water into a Nessler glass, add two or three drops of acetic acid, and then several drops of a saturated aqueous solution of H_2S . Stir the mixture well. If there is an appreciable amount of lead in the water, there will be a brown colour developed directly.

b. *With Potassium Chromate*

If the H_2S gives a decided colour with the sample of water, add a few drops of $KCrO_4$ to a fresh 100 c.c. If lead is present, a yellow precipitate will be formed.

If only a very faint darkening is observed with the H_2S it is necessary to concentrate the water. This is done by evaporating about 250 c.c. in an evaporating dish to 20 c.c. and then adding the $KCrO_4$. By this means the precipitate is much more easily seen.

c. *With Sulphuric Acid*

Add a few drops of H_2SO_4 to 100 c.c. of the water and allow it to stand for some time. A white precipitate of lead sulphate will be formed.

2. Quantitative Estimation

Having now determined that lead is present in the water, it remains to find the quantity.

1. Measure 100 c.c. of the water into a Nessler glass, add a few drops of acetic acid and several drops of sulphuretted hydrogen.
2. Run 100 c.c. of distilled water into another Nessler glass, add a few drops of acetic acid and several of sulphuretted hydrogen.

3. Allow the standard solution of lead (1 c.c. = 0.1 milligram of Pb) to run into this, drop by drop, stirring constantly until the depth of colour is the same in both glasses.
4. Read off the quantity of lead solution used, and calculate the quantity of lead in the sample.

EXAMPLE

100 c.c. of the water were found to contain as much lead as was present in 2 c.c. of the standard lead solution.

1 c.c. = 0.1 milligrams of lead \therefore 2 c.c. = 0.2 milligrams of lead

But this was present in 100 c.c., i.e. there are 0.2 parts of lead in 100,000 of the water.

Notes

If a very small quantity of lead is present in the water, and it is necessary to know the exact amount, 250 or 500 c.c. of the water acidified with acetic acid should be evaporated almost to dryness. The residue should be poured on to a small filter-paper, and the dish and filter-paper washed well with small quantities of distilled water acidified with acetic acid. The washing should be collected in a Nessler glass and made up to 50 c.c.

The determination is now proceeded with, and the quantity found will be that present in 250 c.c. of the original water.

If copper is present as well as lead, the above method will estimate both these metals. The copper must be estimated separately, as will be seen later, and the amount thus obtained deducted from that obtained by the H_2S method.

The difference will be the quantity of lead.

B. COPPER

1. Qualitative Tests

a. *With Sulphuretted Hydrogen*

This test is performed in exactly the same manner as was described under lead.

b. *With Potassium Ferrocyanide*

1. Place about 100 c.c. of the water in a Nessler glass and acidulate with a little dilute hydrochloric acid.

2. Add a few drops of potassium ferrocyanide and stir up well. A brown or chocolate colour will be developed if copper is present, owing to the formation of copper ferrocyanide.

c. By the Platinum-Steel Couple

This is an extremely delicate test, and is very simple.

Half fill a platinum dish with the water and acidulate with HCl. Then lay a large polished steel needle in the dish so that one end rests on the bottom, and the other on the edge. Part will thus be immersed and part dry. After being in this position for about half an hour the needle is withdrawn and examined. If copper is present in the water there will be a deposit on the needle. This deposit can be made more conspicuous, if necessary, by allowing it to come in contact with bromine vapour for a few seconds.

2. Quantitative Estimation

Having determined that copper is present, it remains to estimate the amount.

1. Measure 100 c.c. of the water into a Nessler glass, add a few drops of dilute HCl, and then sufficient $K_4Fe(CN)_6$ to produce the maximum colour.
2. Measure 100 c.c. of distilled water into another Nessler glass, add the same quantities of HCl and $K_4Fe(CN)_6$ as were added to the sample.
3. Allow the standard solution of copper to run into this drop by drop until the colours match. When this occurs, read off the quantity used, and calculate the amount of copper present per 100,000.

Notes

In testing for copper, controls should always be made in a similar manner to those described for the determination of the presence of lead.

C. IRON

1. Qualitative Tests

a. With Ammonium Sulphide

Take about 100 c.c. of the water in a Nessler glass, add a few drops of ammonia solution and also a few drops of solution of ammonium chloride. Now add about 2 c.c. of a solution of ammonium sulphide. If iron is present, there will be a brown colour developed.

b. *With Potassium Ferrocyanide and Ferricyanide*

To 100 c.c. of the water add a few drops of dilute HCl and a few drops of $K_4Fe(CN)_6$ and $K_3Fe(CN)_6$. If a blue colour develops, iron is present either as a ferrous or a ferric salt.

c. *With Potassium Sulphocyanide*

To about 100 c.c. of the water add a few drops of pure dilute HNO_3 and a crystal of KCNS. If iron is present a red colour will be formed.

2. Quantitative Estimation

1. Measure 100 c.c. of the water into a Nessler glass, and add a few drops of nitric acid and a few drops of a solution of potassium ferrocyanide until the blue colour produced reaches the maximum.
2. Measure 100 c.c. of distilled water into another Nessler glass, add a few drops of potassium ferrocyanide solution, and then add the standard ferric chloride solution until the blue colour developed matches that in the first glass.
3. Measure the amount of standard iron solution used, and thus calculate the amount of iron present in the sample water.

Another method is to use potassium sulphocyanide instead of the ferrocyanide as an indicator.

D. ZINC

1. Qualitative Tests

a. *With Ammonium Sulphide*

Take about 100 c.c. of the water in a Nessler glass, add a few drops of NH_4HO , and also a few drops of NH_4Cl solution. Now add about 2 c.c. of a solution of fresh $(NH_4)_2S$. If zinc is present in any but the minutest traces, a white precipitate of zinc sulphide will be produced.

b. *With Potassium Ferrocyanide*

Acidulate 100 c.c. of the water with a few drops of HCl and add a few drops of $K_4Fe(CN)_6$. If zinc is present in any but the smallest traces, a white precipitate will be formed.

If the presence of zinc is suspected, and no precipitate is

obtained with either of the above reagents, half a litre of the water should be evaporated to a small bulk and tested in the above manner.

2. Quantitative Estimation

1. Having determined that neither lead nor copper is present in the water, measure 250 c.c. into an evaporating basin and evaporate over the water bath until the bulk is about 50 c.c.
2. Add solution of NH_4HO and filter any precipitated hydrated oxide of iron. To the filtrate add $(\text{NH}_4)_2\text{S}$ in slight excess.
3. Filter the water through an ashless filter-paper, and wash the precipitate with dilute $(\text{NH}_4)_2\text{S}$ solution.
4. Dry the precipitate in the water oven, transfer it together with the filter-paper to a tared porcelain crucible, and carefully ignite over a Bunsen burner.
5. Allow it to cool, and reweigh. The gain in weight represents the weight of oxide of zinc, and from this the amount of zinc present is calculated.

Notes on Poisonous Metals in Drinking Waters

Copper and zinc are rarely found in drinking waters, and when they are present they are derived from copper and zinc storage vessels. Iron is present naturally in some waters, especially in those collected from the greensand strata. Lead, the metal occurring most frequently in waters, is derived from the action of the water on the lead pipes through which the supply is distributed to the consumers.

The kinds of water which have a marked solvent action on lead are:—

1. Soft waters.
2. Waters not aerated.
3. Waters containing excess of nitrate.
4. Acid waters as from peaty soils.

From the hygienic point of view, the presence of lead in water is one of very great importance, and therefore the estimation of the lead when it is present, or even the detection of the lead, is a very important point. The consumption of water containing lead gives rise, as is well known, to symptoms of chronic lead-poisoning, and it is laid down by many authorities that not more than $\frac{1}{80}$ th grain per gallon (0.025 parts per 100,000) should ever be present in a water intended for human consumption. Other

authorities who are not quite so rigid lay down that $\frac{1}{30}$ th grain is the limit, but it follows from what has been said above that when two sources of water are available, that which contains no lead, or will take up no lead from the pipes, should always be chosen in preference to one which would take up lead.

GASES IN WATER

The detection and estimation of the gases contained in various samples of water are interesting, but are of questionable hygienic import. One gas is, however, of some importance, namely oxygen, and a brief account of the estimation will be given. Should the reader require to estimate other gases, he can find all the details in several text-books more pretentious than this.

OXYGEN IN WATER

WINKLER'S METHOD

Apparatus, etc., required

1. Two glass-stoppered bottles of 350 c.c. capacity.
2. Solution of MnCl_2 (40 grammes to 100 c.c.).
3. Solution of KOH (33%) and KI (10%) in water.
4. 100 c.c. burette, graduated in 0.1 c.c.
5. Freshly prepared starch solution.
6. Sodium thiosulphate solution (1 c.c. = 0.25 milligrams of oxygen).
7. Two large porcelain dishes.
8. Pure H_2SO_4 .

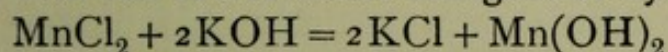
The Process

1. Fill one of the bottles with distilled water, shaking it well so that as much oxygen as possible shall be dissolved.
2. Fill the other bottle by means of a syphon with the water under examination. Do not splash the water or shake the bottle.
3. To each bottle add 1 c.c. of the strong solution of manganous chloride.
4. To each bottle add 2 c.c. of the solution containing KOH and KI.
5. Replace the stoppers, taking care that each bottle is quite full and no air bubbles are included.

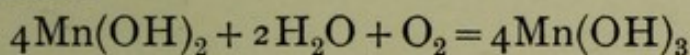
6. Invert both bottles several times, so as to mix the solutions in them.
7. Put the bottles away in a dark cupboard for fifteen minutes.
8. Remove the bottles from the cupboard, and pour their contents carefully, and without splashing, into the two porcelain dishes.
9. Label the dishes.
10. To each dish add 3 c.c. of H_2SO_4 . The brown colour that appears is due to free iodine.
11. From the burette run into the dishes enough of the thiosulphate solution to discharge the brown colour. Use the starch solution, as in Tidy's process, to estimate the end point.
12. Read the burette, and note the amounts of thiosulphate solution used in both the dishes.

The Explanation

The MnCl_2 with the KOH forms manganous hydrate :—

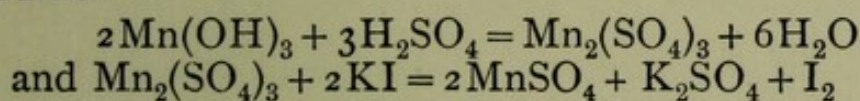


And when this $\text{Mn}(\text{OH})_2$ is in contact with water containing dissolved oxygen, it takes up the oxygen and is oxidized to manganic hydrate :—



The amount of $\text{Mn}(\text{OH})_3$ formed is an index, therefore, of the amount of dissolved oxygen in the water.

On addition of H_2SO_4 , the $\text{Mn}(\text{OH})_3$ is converted into manganic sulphate, which latter reacts with the KI and liberates free iodine.



The amount of iodine liberated is proportionate to the amount of $\text{Mn}_2(\text{SO}_4)_3$, and so proportionate to the amount of $\text{Mn}(\text{OH})_3$, and to the oxygen dissolved in the water.

EXAMPLE

The dish containing the distilled water shaken with air was found to require 14.4 c.c. of sodium thiosulphate solution, in order that the colour of the iodine might be discharged.

Now 1 c.c. of thiosulphate solution = 0.25 milligrams of oxygen

$$\therefore 14.4 \quad \text{,,} \quad \text{,,} \quad = 3.60 \quad \text{,,} \quad \text{,,}$$

\therefore in 350 c.c. of distilled water there were 3.6 milligrams of oxygen

\therefore in 100 c.c. of distilled water there were 1.02 milligrams of oxygen

or 1.02 parts of oxygen per 100,000 of the water.

Again, it was found that the dish containing the water under examination required 12.6 c.c. of sodium thiosulphate solution, in order that the brown colour of the iodine might be discharged.

And since 1 c.c. of thiosulphate solution = 0.25 milligrams of oxygen

\therefore 12.6 c.c. of thiosulphate solution = 3.15 milligrams of oxygen

\therefore in 350 c.c. of the sample water there were 3.15 milligrams of oxygen

\therefore in 100 c.c. of the sample water there were 0.90 milligrams of oxygen

or 0.9 parts of oxygen per 100,000 of the water.

But we have seen that a fully saturated water takes up 1.02 parts of oxygen; and if this represents 100 per cent, then the water under consideration will give a percentage saturation with oxygen of $\frac{0.9 \times 100}{1.02} = 88.2\%$.

The result may be returned either as a percentage or in terms of the actual amount of oxygen present in the water. Both results are given in this example.

Notes

The estimation of the amount of oxygen dissolved in water is of some value, as the oxygen becomes much diminished in the presence of organic matter. A low figure for dissolved oxygen is, at least, confirmatory evidence in condemning a water of which suspicion is entertained.

When decolourizing with thiosulphate solution, it often happens that the colour returns two or three times. The estimation, therefore, should not be considered as completed until the water remains colourless for four or five minutes.

Although the thiosulphate is made up to a definite strength, it will not keep constant; so it is advisable to standardize it occasionally against standard potassium permanganate.

THE INTERPRETATION OF AN ANALYSIS OF WATER

The attention of the Public Health Student is especially directed to the importance of grasping the fundamental principles laid down in this chapter; and he is asked to give very careful attention to the analyses of various waters, with which the lessons of this chapter are illustrated.

Every water sample must be treated on its own merits, and local circumstances and the history of the water must be taken into consideration before the analyst gives a favourable or an adverse judgment. It is easy to say when a water is good; equally easy to determine when it is very bad; but often extremely difficult to decide on many waters which are neither excellent nor foul. It is in judgment on these indifferent waters that a student will be most troubled; and it is here that some knowledge of geology, and of the composition of waters from various strata, will be of use to him in deciding whether or not the sample under his consideration deviates from the type to which it should conform.

In the first place it must be remembered that no water must be passed or condemned, unless it contain poisonous doses of lead or copper, on any single figure in the analysis. All the figures must be taken together; this will be seen by the following notes upon the individual estimations.

Physical Characters

Almost all waters used for drinking purposes have excellent physical characters. The colour is faintly blue as seen in the two-foot tube, the water is bright and free from opacity, and has no smell nor taste. Any odour or opacity should at once arouse suspicion about the sample. If the water is brown in colour this may be due to salts of iron, or to the colouring matter extracted by the water from peat and upland surfaces. There should be no sediment, or very little, and in a good water this sediment should contain neither epithelium nor hairs nor matter of obvious animal origin.

Total Solids

These vary very much in different samples of water. Rain water may have about 3 parts per 100,000 of total solids, and a water derived from the greensand as much as 110 parts per 100,000. A well, polluted by sea water, may, of course,

show still higher figures. Generally speaking, a good drinking water should not contain more than 100 parts per 100,000 of total solids.

On ignition some of these total solids are removed, and the proportion removed is of some importance. Salts, except for their water of crystallization, are not affected by ignition; but organic matter present is driven off. If, therefore, there is much loss on ignition, and if the residue shows signs of blackening, the sample should be regarded as suspicious.

REACTION OF THE WATER

Most drinking waters are alkaline. Some, however, derived from upland surfaces, may be acid, and even markedly so. Now an acid water will dissolve lead pipes and take lead into solution, greatly to the detriment of the consumer: so, if a water is acid, search should be made for lead, and the water condemned if this is found in any amount exceeding 0.025 parts per 100,000. In itself, and without the lead, an acid water is of no harm.

HARDNESS

The hardness of a water is not of very great hygienic importance. It is maintained by many medical men that water which is excessively hard, say above 30 parts per 100,000, causes a certain amount of dyspepsia, and a water containing such a degree of hardness could not be recommended for drinking purposes. The chief objection to having a hard water is a purely economical one. It is said that since Glasgow has been supplied with water from Loch Katrine the saving to the city of Glasgow in soap per annum has been about £30,000. Speaking generally, the hardest waters are those which are derived from superficial wells. The softest is of course pure rain water. The average hardness of the London water is about 16 parts per 100,000, and this is looked upon generally as the limit of hardness for a water for drinking purposes.

CHLORIDES

The sources of chlorides in water are various. Rain water, especially when collected near the sea, always contains traces of salt. Certain geological formations also contain considerable quantities of chlorides. This being so, the purest water would be expected to contain certain traces of chlorides, and this is found to be invariably the case. The water drawn from districts

in which there are manufactories, such as alkali works, mines, etc., is also found to be rich in chlorine. Urine contains about 1% of chlorides, and therefore the addition of sewage to any drinking water would probably raise the quantity of chlorine. In considering the percentage of chloride which is to be allowed, it is necessary in the first place to know something about the geological stratum from which the water is drawn.

If the chlorine content of a water is found to be uniform throughout the year, any deviation from the usual figure may well give rise to suspicion that the water has been contaminated. Well waters from the chalk and limestone generally have a chlorine figure not over 3 parts per 100,000; from the greensand and some of the marls, and in the neighbourhood of salt mines or the sea, the chlorine figure may be very high. Rain water collected in or near towns contains more chlorine than that collected in the open country.

FREE AND SALINE AMMONIA

Waters from all sources contain ammonia. Even rain water from the country shows some traces. Many very pure waters from deep wells have large quantities of ammonia, which is derived from the reduction of nitrates and nitrites: the greensand waters are examples of this. Speaking generally, free and saline ammonia should not exceed 0.005 parts per 100,000. If it exceeds this, the albuminoid ammonia figure should be low. Water which is derived from rivers, sewage, or known polluted sources contains large quantities of ammonia.

ALBUMINOID AMMONIA

It will be understood from what has been said in the sections on the actual analysis of water that the albuminoid ammonia does not exist as such in the water, but is simply a laboratory product from the organic matter present in the water. From this it will be seen that even where albuminoid ammonia exists in considerable quantities, there is no method of determining whether the organic material present in the water is derived from vegetable or animal sources. Nor is the amount of albuminoid ammonia found in the water a very accurate test of the amount of organic material present in the water. Waters which are derived from deep wells, or even from surface wells where there is no possibility of contamination, do not, however, yield much albuminoid ammonia. Indeed, many deep well waters, that is

waters derived from beneath the impervious layers, contain practically no albuminoid ammonia at all. The consideration, however, of the quantities of free ammonia and albuminoid ammonia together, often gives a good clue to the presence or no of contamination.

As a rule the figure for albuminoid ammonia should not exceed 0.005 parts per 100,000. If it exceeds this, the figure for free and saline ammonia should be low. In other words, the free ammonia and the albuminoid ammonia should not both be high in a water; if the one is raised, the other should not also exceed the standard.

The albuminoid ammonia figure is high in waters that have been polluted with animal matter; and is also raised in upland surface waters, in which latter case the free ammonia figure is invariably low.

NITRITES

Nitrites may be found in waters that have been polluted by sewage or excrement. Lower greensand waters also may contain nitrites from the reduction of nitrates by the ferrous salts of the stratum. Unless it can be shown that nitrites are present in a drinking water in consequence of such reduction of nitrates, the water, if it contains even traces of any nitrites, must be considered as highly suspicious.

NITRATES

Nitrates may be regarded as one of the end products of the oxidation of organic matter, and their presence in water is therefore an index of past organic contamination. Certainly, the organic contamination may be very remote, and the nitrate may have been produced many years before the water comes to be analysed. If the other figures of the analysis are good, and the nitrate figure is high, this excess over the average may be accounted for by the nitrates in the stratum, and is of little consequence; but if the rest of the analysis throws doubt upon the water, a raised nitrate figure (together, possibly, with increased chlorine) will turn the balance against the water under consideration. Some geological strata contain nitrates in considerable degree, but a large proportion of nitrates is by no means so common as in the case of chlorine. Peaty upland waters and waters derived from the Liassic strata may be rich in nitrates; but, speaking as a rule, to which, of course, there are notable exceptions, a good drinking water ought not to contain more than 0.5 parts per 100,000 of nitric nitrogen.

Oxygen Absorbed from Permanganate

Waters which contain a considerable amount of organic material absorb an amount of oxygen which to some degree corresponds to the organic material present. Organic material animal in origin absorbs oxygen more readily than does that derived from vegetable sources. If this were all, the oxygen process would be an extremely valuable one, but unfortunately this is not the case. Certain other matters, particularly the proto-salts of iron, etc., will absorb oxygen when in a water containing absolutely no organic material. If, therefore, any of these salts are present, a considerable allowance must be made for the amount of oxygen absorbed, and if in the water any sediment is present which consists of these, the water must be carefully filtered before being submitted to the process.

Frankland and Tidy give the following table, which may be useful as forming some standard to which waters should comply in regard to the oxygen absorbed.

AMOUNTS OF OXYGEN ABSORBED BY 100,000 PARTS OF WATER

	Water from Upland Surfaces.	Water from Sources other than Upland Surfaces.
Water of great organic purity	Not more than 0.1	Not more than 0.05
Water of medium purity	„ „ 0.3	„ „ 0.15
Water of double purity	„ „ 0.4	„ „ 0.2
Polluted water . . .	More than 0.4	More than 0.2

Examples of Waters from various Sources

The first two analyses given are those of a very good and a very bad water respectively ; the remainder are analyses of waters from different sources and from some of the main water-bearing geological strata in this country.

Analyses 3-16 inclusive show uncontaminated waters ; 17-22 show some river water analyses ; and 23-30 are analyses of waters that are contaminated. In the light of what has already been said in this chapter, the careful consideration of these

WATER ANALYSIS

various analyses should do much to help the student to appreciate the significance of the analytical figures.

	No. 1 A very good water.	No. 2 A very bad water.
Physical characters .	Excellent	Excellent
Reaction	Slightly alkaline	Very alkaline
Total solids . . .	16'4	42'8
Volatile	3'8	20'3
Appearance on ignition	Nil	Marked blackening
Hardness	9'2	30
Chlorine	1'3	6'2
Free and saline ammonia	0'0008	0'030
Albuminoid ammonia .	0'001	0'018
Nitrites	Nil	A trace
Nitrates	0'01	0'96
Oxygen absorbed in 4 hours at 27° C. . .	0'01	0'34

Notes on 1 and 2

All figures represent parts per 100,000. These two waters are good examples of the extremes of which it is easy to judge.

	No. 3 Rain water—country.	No. 4 Rain water—town.
Physical characters .	Excellent	Good
Reaction	Faintly alkaline	Slightly acid
Total Solids . . .	3'2	5'1
Volatile	1'7	2'7
Appearance on ignition	Nil	Nil
Hardness	0'5	0'7
Chlorine	0'31	1'2
Free and saline ammonia	0'042	0'058
Albuminoid ammonia .	0'003	0'005
Nitrites	Nil	Nil
Nitrates	0'01	0'04
Oxygen absorbed in 4 hours at 27° C. . .	0'004	0'022

Notes on 3 and 4

Both these waters show the low total solids and the low figure for hardness characteristic of rain water. The free ammonia figure is high in each; but the albuminoid ammonia and the oxygen absorbed are low, showing the freedom of these waters from much organic matter. Obviously, No. 3 is a better water than No. 4.

	No. 5 Upland surface water, not peaty.	No. 6 Upland surface water, peaty.
Physical characters .	Good	Brown
Reaction	Neutral	Acid
Total solids . . .	5'0	10'0
Volatile	1'7	7'6
Appearance on igni- tion	Nil	Blackening
Hardness	2'9	3'4
Chlorine	0'9	0'9
Free and saline ammonia	0'003	0'001
Albuminoid ammonia .	0'004	0'020
Nitrites	Nil	Nil
Nitrates	0'06	0'04
Oxygen absorbed at 27° C. in 4 hours . . .	0'05	0'14

Notes on 5 and 6

Both these waters show low figures for hardness and for total solids, although No. 6 gives slightly higher readings in these respects than No. 5. No. 5 really is very much like the rain water analyses shown, except that the ammonia figure is less. No. 6 is typical of peaty waters, being acid and having a high figure for volatile solids: these latter it is noticed show charring on ignition, and are composed of organic matter derived from the peat. The high figure for albuminoid ammonia is due to this vegetable organic matter. Note that in No. 6 the figure for free ammonia is low. Both waters were free from contamination with animal organic matter.

	No. 7 Subsoil water. Shallow well in Sand.	No. 8 Subsoil water. Shallow well in Gravel.
Physical characters .	Excellent	Excellent
Reaction . . .	Alkaline	Alkaline
Total solids . . .	8'5	32'2
Volatile . . .	2'7	10'1
Appearance on ignition	Nil	Nil
Hardness . . .	5'7	25'5
Chlorine . . .	2'0	2'0
Free and saline ammonia	Trace	Trace
Albuminoid ammonia .	0'005	0'0005
Nitrites . . .	Nil	Nil
Nitrates . . .	0'01	0'51
Oxygen absorbed at 27°C. in 4 hours . . .	0'07	0'01

Notes on 7 and 8

Both these are very good waters. They show low ammonia figures, and low figures for the oxygen absorbed from permanganate. No. 7 does not show so much hardness as most shallow well waters; No. 8, however, is very hard. The nitrate figure of No. 8 is high, but of little import since the water is, in other respects, so excellent: probably these nitrates are from some very old organic pollution of the gravel from which the water was drawn. Shallow well waters are not usually so pure as these, and have frequently much larger quantities of ammonia, both free and albuminoid.

	No. 9 Deep well in the Chalk.	No. 10 Deep well in the Chalk.
Physical characters .	Excellent	Excellent
Reaction . . .	Alkaline	Alkaline
Total solids . . .	26'5	35'7
Volatile . . .	9'0	10'4
Appearance on ignition	Nil	Nil
Hardness . . .	20'4	21'6
Chlorine . . .	1'5	1'7
Free and saline ammonia	0'0005	0'0015
Albuminoid ammonia .	0'0005	0'001
Nitrites . . .	Nil	Nil
Nitrates . . .	0'41	0'27
Oxygen absorbed in 4 hours at 27° C. .	0'01	0'03

Notes on 9 and 10

Both of these are excellent chalk waters in which the hardness is fairly high: about half this hardness is temporary and can be removed by appropriate softening processes. The chlorides and nitrates are, of course, derived from the strata through which the water has passed. The ammonia figures and the oxygen absorbed are low. Some chalk waters contain much more total solids than do these.

	No. 11 Upper Greensand.	No. 12 Lower Greensand.
Physical characters . . .	Excellent	Good
Reaction	Alkaline	Alkaline
Total solids	13'1	106'0
Volatile	3'0	21'4
Appearance on igni- tion	Nil	Nil
Hardness	5'8	18'0
Chlorine	2'0	11'7
Free and saline ammonia	Trace	0'038
Albuminoid ammonia . .	0'001	0'001
Nitrites	Nil	Trace
Nitrates	0'25	0'30
Oxygen absorbed at 27° C. in 4 hours . .	Nil	0'36

Notes on 11 and 12

No. 11 is obviously an excellent water, and otherwise has no special characteristics. No. 12, at first sight, might seem to be much polluted: the free ammonia figure is high, and the chlorides are excessive, moreover nitrates are present. This water on analysis showed a trace of iron, and to the reducing properties of this metal the figures on which I have commented are due. The nitrites are formed by the reduction of nitrates, and so is the free ammonia: the high chlorine figure is typical of some green-sand waters. Note the small amount of albuminoid ammonia, which points to the absence of organic pollution.

	No. 13 Oolite.	No. 14 New Red Sandstone.
Physical characters .	Excellent	Excellent
Reaction	Alkaline	Alkaline
Total solids . . .	74·6	33·5
Volatile	18·2	9·1
Appearance on ignition	Nil	Nil
Hardness	2·7	22·8
Chlorine	2·2	2·8
Free and saline ammonia	0·0004	Trace
Albuminoid ammonia .	Trace	Trace
Nitrites	Nil	Nil
Nitrates	0·01	0·25
Oxygen absorbed at 27° C. in 4 hours .	0·008	Nil

Notes on 13 and 14

Both these are very good waters. No. 13 from the Oolite is very similar in composition to a chalk water. No. 14 is better than most sandstone waters: these vary greatly in their composition, depending on the nature of red sandstone deposit, which may be pure or impure, soft or hard. The total solids and hardness in these waters may in consequence be sometimes very high.

	No. 15 Coal Measures.	No. 16 Carboniferous Limestone.
Physical characters .	Excellent	Excellent
Reaction	Alkaline	Alkaline
Total solids	55·9	30·4
Volatile	14·7	8·7
Appearance on ignition	Nil	Nil
Hardness	35·7	23
Chlorine	1·2	1·7
Free and saline ammonia	0·006	0·002
Albuminoid ammonia .	0·002	0·001
Nitrites	Nil	Nil
Nitrates	0·004	0·27
Oxygen absorbed at 27° C. in 4 hours .	0·4	0·056

Notes on 15 and 16

Both these waters are of good quality, as far as absence of organic pollution is concerned. No. 15 is exceptionally hard, and for that reason would make the water unfitted for trade purposes, unless some softening process were adopted.

River Waters

River waters may be divided into two classes—(a) those affected by the tide and (b) those not so affected.

The first class will obviously show the variations of the second class, but will have an additional variant in the greater or less amount of sea water they contain at any moment.

The non-tidal river waters will vary with the sources from which they are derived. In the upper reaches the rivulets may consist of either upland surface, subsoil or deep well water, the two latter appearing as springs. In their lower reaches most probably all rivers consist of a mixture of all three, in different proportions in different rivers. All rivers drain the subsoil water of their basins.

In the following table these differences are well seen:—

RIVER WATERS

No.	SOURCE.	PARTS PER 100,000							
		Total Solids.	Total Hardness.	Chlorine.	Saline NH ₃ .	Albuminoid NH ₃ .	Nitrites.	Nitric N.	Oxygen Absorbed.
17	Scotland . .	4'1	2'4	1'0	Trace	'005	Nil	Nil	'18
18	Westmorland .	4'3	2'4	1'0	'008	'015	Nil	Trace	'15
19	Devon . .	5'0	2'7	1'1	Trace	'005	Nil	Trace	'05
20	Worcester . .	23'1	11'0	5'8	'013	'022	Nil	'07	'44
21	Wilts . .	54'5	38'8	2'4	Trace	'076	Nil	Trace	'13
22	„ . .	70'2	51'4	2'2	'004	'01	Nil	'09	'14

Notes on 17-22

No. 17 is a pure water. No. 18 shows evidence of organic contamination, which is possibly vegetable in origin. No. 19 is a pure water. No. 20 is very foul. Nos. 21 and 22 show much hardness, and high figures for the albuminoid ammonia.

The following eight analyses are those of contaminated waters :—

	No. 23 Upland surface, peaty water.	No. 24 Shallow well— Sand.
Physical characters .	Brown	Excellent
Reaction . . .	Acid	Alkaline
Total solids . . .	16·2	13·1
Volatile . . .	12·1	8·2
Appearance on igni- tion . . .	Blackening	Slight blackening
Hardness . . .	2·8	5·0
Chlorine . . .	1·5	2·2
Free and saline ammonia	0·008	0·013
Albuminoid ammonia .	0·026	0·033
Nitrites . . .	Nil	Nil
Nitrates . . .	0·35	0·47
Oxygen absorbed at 27° C. in 4 hours .	0·46	0·53

Notes on 23 and 24

Both these waters are bad. Each shows high ammonia figures, increased nitrates and an excessive amount of oxygen absorbed from permanganate. Moreover, No. 23 shows a chlorine figure that is considerably above what is usually found in upland surface waters. Note that both these waters show some blackening when their total solids are ignited.

	No. 25 Shallow well in gravel.	No. 26 Shallow well in gravel.
Physical characters .	Slightly turbid	Excellent
Reaction . . .	Alkaline	Alkaline
Total solids . . .	24·6	46·7
Volatile . . .	10·6	16·9
Appearance on igni- tion . . .	Marked blackening	Nil
Hardness . . .	13·6	28·5
Chlorine . . .	2·7	5·9
Free and saline ammonia	0·010	0·001
Albuminoid ammonia .	0·022	0·002
Nitrites . . .	Nil	Nil
Nitrates . . .	0·91	1·95
Oxygen absorbed at 27° C. in 4 hours .	0·20	0·18

Notes on 25 and 26

No. 25 is an analysis of a badly contaminated water, as shown by the blackening of the total solids on ignition and by the raised figures for the ammonia and nitrates. No. 26 shows an analysis of a well water that has been heavily polluted in the past; but which has oxidized almost all the organic matter into nitrates. The chlorine figure for this well is also high, and the nitrates, of course, excessive. This water is potentially very dangerous: it was badly contaminated once, and there is no reason why this should not happen again.

	No. 27 Shallow well in Chalk.	No. 28 Deep well in Chalk.
Physical characters . . .	Excellent	Excellent
Reaction	Alkaline	Alkaline
Total solids	60·0	72·8
Volatile	22·1	26·4
Appearance on ignition	Slight blackening	Nil
Hardness	23·4	20·0
Chlorine	2·8	2·1
Free and saline ammonia	0·007	0·006
Albuminoid ammonia	0·008	0·005
Nitrites	Nil	Nil
Nitrates	0·84	0·52
Oxygen absorbed at 27° C. in 4 hours	0·093	0·07

Notes on 27 and 28

The shallow well water, No. 27, shows increased ammonia figures and high nitrates. The well was near a heap of manure, and water percolated from this through the subsoil and so into the well, the sides of which were not protected. No. 28 shows the analysis of a deep well chalk water, which was liable to intermittent contamination through defects in the stratum. Although the figures for No. 28 are not in themselves very excessive, they are greatly above the average for this well, in the waters of which albuminoid ammonia usually occurs only as a trace. The normal chlorine figure for this well is 1·8.

	No. 29 Deep well in Chalk.	No. 30 Deep well in Chalk.
Physical characters .	Excellent	Brackish taste
Reaction	Alkaline	Alkaline
Total solids	40'4	272'4
Volatile	12'7	36'0
Appearance on ignition	Faint blackening	Faint blackening
Hardness	20'5	50
Chlorine	1'7	120'4
Free and saline ammonia	0'002	0'005
Albuminoid ammonia .	0'003	0'002
Nitrites	Nil	Nil
Nitrates	0'32	1'07
Oxygen absorbed at 27° C. in 4 hours .	0'05	0'071

Notes on 29 and 30

No. 29 looks like the analysis of a good water. Normally, however, the chlorine content of this water is 1'5, and the nitrate figure 0'22. Usually there are only traces of albuminoid ammonia, the saline ammonia is less than 0'001, and no oxygen is absorbed from permanganate. With the knowledge of this past history, we can condemn water No. 29 as showing organic contamination. The analysis No. 30 is of a deep well water near a tidal river. In times of flood and high tides some of the salt water finds its way into the well, as is shown by the analysis.

In an examination, or where the student is required quickly to complete a water analysis, it is satisfactory to have a **plan of work**, and to adhere rigorously to this; by so doing, labour is saved and the worker is spared from having to improvise on a sudden some scheme of his own.

The following order of performing the various analyses of a water sample can be recommended: it is put forward as a useful scheme, but not necessarily the best. Whether the student adopts this or some other, let him adhere to one only; for by so doing he will save himself much confusion, and be able the better to co-ordinate his work.

1. Examine the physical properties of the water. Start the water evaporating for the estimation of the total solids; start some more boiling for the estimation of permanent hardness.

Set up and start the apparatus for estimating free and saline ammonia. Set some water boiling to concentrate for testing for poisonous metals. Start Tidy's process.

2. Test for nitrites and nitrates.
3. If nitrates are present start the phenol-sulphonic acid method of their estimation.

Note

All the lengthy processes have now been begun. The student can now perform various smaller manipulations until the free ammonia begins to come over.

4. Estimate the chlorine. Estimate the nitrites, if such are present. Estimate the total hardness. Using the water that has been boiling, estimate the permanent hardness. Calculate the temporary hardness, and make notes of all your results.

5. The free and saline ammonia should have distilled over by now. Start the distillation of the albuminoid ammonia.

6. Estimate the free and saline ammonia.

7. The phenol-sulphonic acid method should now be ready for completion. Finish this.

8. Test the concentrated water for the poisonous metals. If present, estimate them.

9. The albuminoid ammonia will now be distilled off. Estimate it.

10. Weigh the total solids. Ignite and weigh the residue of non-volatile solids.

11. Finish off Tidy's method.

Note

The wise student takes to examinations with him porcelain or platinum dishes of known weight. By so doing he saves much time and trouble. All results should be noted as they are obtained, and the last hour of the examination should be devoted to careful revision of the results and to the writing of the report.

In all practical examinations it is impossible to over-estimate the importance of the written part, for it is by this, very largely, that the examiner judges of the candidate's grasp of the practical work. In the case of a water examination, the report should be written and arranged as if it were issued from the laboratory of a public analyst, and should be headed after the following manner: "Report on sample of water marked X, received on

such a date and examined on such a date." Then should follow details of the source of the water, if this is known.

Workers in laboratories should aim at order and cleanliness in their methods. A messy and untidy bench does not impress an examiner favourably.

SEWAGE ANALYSIS

THE composition of sewage varies within wide limits. Some towns have abundance of water to dilute their sewage; others have little; others, again, discharge trade refuse of every conceivable description into the sewers. There is no such thing as a standard sewage, for no two are alike. Even in one town or village the sewage varies in composition from day to day and from hour to hour.

In making a chemical analysis of sewage, the procedure described under water is adopted, but with slight modifications.

TOTAL SOLIDS

These may be estimated in two ways. The total solids both in suspension and solution may be estimated, or a portion of the sewage may be filtered. The residue is dried, weighed, and subsequently ignited, and the loss on ignition noted; the filtrate is then evaporated to dryness, dried, weighed, and subsequently ignited and weighed again.

Chlorides

The chlorides of a sewage will be found to vary considerably. They are estimated in the way already described.

Saline Ammonia

As this is usually very high, often as high as 1 to 2 parts per 100,000, and sometimes much higher, it is obvious that it will be useless to distil 500 c.c., since we should require no less than 500 c.c. of standard ammonia to match the distillates

Before determining what quantity to use for distillation it is useful to filter a little of the sewage. Take 10 c.c. of the filtrate and dilute it with 40 c.c. of ammonia-free water in a Nessler glass. Then add 2 c.c. of Nessler and match this with the standard ammonia. If the ammonia required is less than 10 c.c., use 20 c.c., and if more, use 10 c.c.

Suppose we decide to use 20 c.c. Pipette 20 c.c. of the sewage into the retort or flask and add half a litre of organically pure ammonia-free water.

At this stage the reaction of the sewage should be determined, since in some places where the refuse matter from chemical works accompanies the domestic sewage, it may be neutral or even slightly acid. If it is not found to be distinctly alkaline, a very small quantity of freshly fused Na_2CO_3 should be added, just sufficient to render it faintly alkaline.

The distillation is now proceeded with as in the case of ordinary water.

Instead of estimating the amount of ammonia in each 50 c.c., the first 200 c.c. may be collected in a flask and well shaken. 20 c.c. of this may then be estimated and the result multiplied by 10.

Albuminoid Ammonia

When all the saline ammonia has come over, the alkaline permanganate solution is added, and the distillation proceeded with in the ordinary manner.

Instead of merely adding 50 c.c. of alkaline permanganate it is advisable to dilute this with as much as 200 c.c. or 250 c.c. of distilled water and boil the mixture well for a few minutes. By doing this there is much less chance of the flask boiling dry, since it often happens that 12 or 14 Nessler glasses are collected before the yield of ammonia ceases.

It is better to estimate the organic nitrogen by Kjeldahl's method, as is described later.

Nitrites

It is generally found that the reaction for nitrites is either not given or only very faintly. This is probably due to the fact that as fast as they are formed by reduction of the nitrates they are entirely reduced to ammonia.

Nitrates

These salts are present only in minute traces. The reducing power of sewage is very great, so whatever nitrates are present are reduced rapidly to ammonia. The indigo method of estimating the nitrates should be used, and there is seldom any need previously to dilute the sewage.

Oxygen Absorbed

20 c.c. is a very convenient quantity to take for this examination. 180 c.c. of organically pure ammonia-free water are added and the usual 10 c.c. of standard permanganate. Even with so small a quantity as 20 c.c. it sometimes happens that a further 10 c.c. of permanganate are required.

Dissolved Oxygen

The dissolved oxygen figure for sewage and for sewage effluents can be determined by the method of Winkler, which has already been described.

The analysis of sewage effluents should be carried out on the same lines as laid down for crude sewage. The solids in suspension should be estimated; also the chlorides, nitrites, nitrates, and free ammonia. The albuminoid ammonia should be determined by Kjeldahl's method, as the ordinary means do not give reliable results.

Total Nitrogen (exclusive of nitrites and nitrates)

KJELDAHL'S METHOD

Apparatus, etc., required

1. 200 c.c. flask of Jena glass.
2. 750 c.c. distilling flask.
3. Glass tube bent to a convenient angle with a bulb blown on the vertical arm.
4. Ordinary flask.
5. Burette.
6. Concentrated nitrogen-free H_2SO_4 .
7. Nordhausen (or fuming) H_2SO_4 .
8. Crystals of pure KMnO_4 .

9. KOH solution 50%.
10. $\frac{N}{50}$ HCl or H_2SO_4 and $\frac{N}{50}$ NaOH.
11. Phenolphthalein or rosolic acid.

The Process

1. Pipette 10 c.c. of the sewage into the small flask, add about 1 c.c. of H_2SO_4 , mix well, and evaporate slowly over a small flame guarded by wire gauze.
2. When the fluid is reduced to rather less than half, add 20 c.c. of H_2SO_4 and 3 c.c. of Nordhausen H_2SO_4 and continue to heat slowly.
3. From time to time add a small crystal of $KMnO_4$ until the colour disappears very slowly, and continue the heating for about 3 hours.
4. Allow the contents of the flask to cool, and carefully transfer them to the distilling flask. Wash the flask with a small quantity of distilled water two or three times and pour the washings into the distilling flask.
5. Add 70 c.c. of the KOH solution and allow the mixture to cool. After cooling, add 30 c.c. more of the KOH solution, drop in two or three pieces of clean granulated zinc and carefully insert an indiarubber stopper. Connect the flask by means of rubber tubing with the bent glass tube so that the free end dips down to the bottom of a clean Erlenmeyer flask containing 50 c.c. of $\frac{N}{50} H_2SO_4$, the bulb being a little distance above the mouth of the flask.
6. Carefully distil until the residue is rather less than half the amount of the original fluid.
7. Remove the flask containing the distillate, add a drop of rosolic acid, and add the $\frac{N}{50}$ NaOH carefully until the reaction is neutral. Read off the amount of alkali required and calculate the amount of NH_3 therefrom. $\frac{1.4}{17}$ of the ammonia calculated will represent the nitrogen.

Explanation

Upon boiling nitrogenous organic matter with H_2SO_4 under the conditions above described, the nitrogen is slowly converted

into ammonium sulphate. When the process of conversion is completed, the H_2SO_4 is over-neutralized with KOH , and free NH_3 will then distil over.

EXAMPLE

After the distillation was finished it was found that the 50 c.c. of $\frac{\text{N}}{50}\text{H}_2\text{SO}_4$ required 46.2 c.c. of $\frac{\text{N}}{50}\text{NaOH}$.

\therefore 3.8 c.c. of $\frac{\text{N}}{50}\text{H}_2\text{SO}_4$ had been neutralized by the NH_3 distilled over

$$\therefore \text{N}_2 \text{ present} = \frac{14}{17} \times \frac{3.8}{1000} \times 0.34 \text{ gramme} \\ = 1.06 \text{ milligrams.}$$

But this was present in 10 c.c. of the sewage effluent.

\therefore the sewage effluent contains 10.6 parts per 100,000 of nitrogen.

The distillation of the sewage effluent showed that there were 10.9 parts of saline ammonia per 100,000. This is equivalent to 8.97 parts of nitrogen.

Therefore the organic nitrogen in the sample of sewage effluent is $10.6 - 8.97 = 1.63$ parts per 100,000.

Notes

Great care must be exercised in heating the distilling flask at first, since there is a great tendency for the fluid to "bump." After a little time it will boil quite quietly until the ammonia has all been driven off, when it will once more begin to bump.

General Considerations

From what has been said of the varying composition of sewage, it may be inferred that sewage effluents also show much variation: and this is indeed the case. It follows from this that a set standard of purity is neither obtainable nor desirable. Each sewage must be treated on its own merits, and the local circumstances taken into consideration before judgment is passed on an effluent.

Whether or not an effluent causes harm to a stream depends mainly on two factors: firstly, on the quantity and concentration of the effluent; secondly, on the size and volume of the stream. It is obvious that an effluent, approaching crude sewage in composition, would do much more harm to a small than to a large

river ; and, again, a small amount of a sewage effluent would do less damage than a larger amount. Doubtless it would be desirable to have local standards of purification for sewage, which standards should take into consideration such circumstances as these ; and whatever standards of purity were established they should have for their main object the protection of rivers from contamination and harm.

The injury caused to a river by the inflow of sewage or a bad effluent may be considerable. The water may be deprived of its oxygen, and the fish of the river may die : trade refuse may cause a like mortality. Organic matter, deposited from the sewage, may stink, and, ultimately, silt up the river : sewage fungus may grow and decay : the river may be discoloured. Finally, the water may be poisoned by intestinal organisms, and be made unfit for drinking purposes to the detriment of cattle and man. An effluent should not be discharged into a stream if it is likely to harm the water in any of these ways.

Satisfactory Effluents

The Royal Commission on Sewage Disposal, which was appointed in 1898, consider that the effect which an effluent has on a stream does not depend so much on the amount of organic matter present, as on the condition of this organic matter—whether or not it is easily putrescible, and is likely to take up oxygen from the water.

The Commissioners conclude that an effluent would generally be satisfactory if it fulfilled the following conditions :—

1. That it should not contain more than 3 parts per 100,000 of suspended matter.
2. That after being passed through filter-paper it should not absorb more than—
 - (a) 0·5 part by weight per 100,000 of dissolved or atmospheric oxygen in twenty-four hours ; or
 - (b) 1·0 part by weight per 100,000 of dissolved or atmospheric oxygen in forty-eight hours ; or
 - (c) 1·5 parts by weight per 100,000 of dissolved or atmospheric oxygen in five days.

An effluent which fulfils these conditions will probably not be putrescible.

It may be laid down as a physical standard for all sewage effluents that they should not smell nor be offensive after

incubation for three days at 27° C. Effluents also should show little opacity.

In contradiction to the Sewage Commissioners some authorities consider the organic ammonia figure to be the best index of a satisfactory effluent. A limit of 0.1 parts per 100,000 has been advocated by some chemists; 0.15 parts, and 0.2 parts by others. Some have taken the oxygen absorbed figure as a standard, and said that no sewage effluent ought to absorb more than 1.4 parts of oxygen per 100,000 from permanganate.

It will be seen, therefore, that really there is no chemical standard for a sewage effluent; and the physical standards, combined with the incubation tests, are the best means at the present for arriving at a decision as to the suitability of an effluent for discharge into a stream.

PREPARATION OF REAGENTS

Ammonia-free Water

THE following process is most usually adopted:—

Distil from a large glass retort (or better, from a copper or tin vessel holding 15–20 litres) ordinary distilled water which has been rendered distinctly alkaline by the addition of sodic carbonate. A glass Liebig's condenser or a clean tin worm should be used to condense the vapour; it should be connected to the still by a short indiarubber joint. Test the distillate from time to time with Nessler's solution (which is described below), and when free from ammonia collect the remainder for use. The collection of water must be stopped when at least 2 litres (in such a sized still) remain.

Ammonia-free Water—Organically Pure

Distilled water, to which 1 gramme of potassium hydrate and 0.2 gramme of potassium permanganate per litre have been added, is boiled gently for about 24 hours in a similar vessel to that used in preparing water free from ammonia, a redux condenser being fitted on to the top of the flask in order to return the condensed water. At the end of that time the condenser is adjusted in the usual way, and the water carefully distilled, the distillate being

tested at intervals for ammonia, as in preparing the ordinary ammonia-free water. When ammonia is no longer found, the remainder of the distillate may be collected, taking care to stop well short of dryness. The neck of the retort or still should point slightly upwards, so that the joint which connects it with the condenser is the highest point. Any particles carried up mechanically will then run back to the still, and not contaminate the distillate. The water thus obtained should be rendered slightly acid with sulphuric acid, and redistilled from a clean vessel, again stopping short of dryness.

Alkaline Permanganate Solution

Dissolve 8 grammes of KMnO_4 and 200 grammes of NaOH in 1100 c.c. of distilled water. Boil until the bulk is reduced to 1000 c.c.

The object of the boiling is to drive off as ammonia any organic matter that may be present, either in the permanganate or the water.

Metaphenylene-diamine-hydrochloride

1 gramme of the base is dissolved in 200 c.c. of distilled water, and slightly acidulated with HCl .

Nessler Solution

1. Dissolve 35 grammes of KI in 100 c.c. of NH_3 -free H_2O .
2. " 17 " " HgCl_2 " 300 " " "
3. " 200 " " NaOH " 1000 " " "

The HgCl_2 dissolves more quickly on heating, but it must be subsequently cooled.

4. Pour the HgCl_2 solution into the KI solution until a permanent precipitate of HgI_2 is formed.
5. Dilute this mixture to 1000 c.c. with the NaOH solution. The precipitate will be re-dissolved.
6. Add more of the HgCl_2 solution until the permanent precipitate is again formed.
7. Allow the mixture to stand in a clean glass-stoppered bottle for 24 hours.
8. Pipette off the clear fluid from time to time as required.

When the Nessler is sensitive, it has a slight yellow colour. If it is colourless it will not be sensitive, and a little more HgCl_2 solution must be added and allowed to settle in order to saturate

the solution with HgCl_2 , since the sensitiveness of the Nessler depends upon this saturation.

Phenol-Sulphonic Acid

32 c.c. of concentrated H_2SO_4 are added to 4 c.c. of pure phenol. These are well mixed and heated to 100°C . for two or three hours. 110 c.c. of distilled water are then added and the solution is ready for use.

Potassium Chromate Solution

A strong solution of pure neutral K_2CrO_4 free from chlorine is required.

Dissolve some crystals of pure K_2CrO_4 in pure distilled water, and when the solution is made, add a drop or two of the standard solution of AgNO_3 until a permanent red precipitate is formed. This ensures the absence of any chlorine in the solution. After the precipitate has settled, syphon or decant the clear yellow fluid into a small clean bottle.

Potassium Ferrocyanide Solution

1 gramme of $\text{K}_4\text{Fe}(\text{CN})_6$ is dissolved in 100 c.c. of distilled water.

Potassium Iodide Solution

This solution should be made as it is required, by adding a crystal of KI into a test-tube, and half filling it with distilled water.

Before being used, a little starch solution should be added to a few drops diluted with water, in order to ensure the absence of free iodine.

A solution of zinc iodide is frequently used instead of potassium iodide, since the former does not liberate free iodine on keeping, as does the latter.

Starch Solution

This solution must be made up on each occasion as it is required.

As much starch as will go on to an ordinary bacteriological platinum loop is dropped into a clean test-tube, and the test-tube is three-parts filled with distilled water and well shaken. It is then well boiled until the liquid becomes quite clear, and allowed to cool, when it is ready for use.

Sodium Thiosulphate Solution

Dissolve 2 grammes of the thiosulphate in 1000 c.c. of distilled water.

The solution undergoes changes and becomes weaker, so that in practice it is standardized every time it is used, by making control or blank experiments side by side with the sample.

Sulphuretted Hydrogen Water

This is made by acting on FeS with dilute HCl, passing the gas through a small quantity of water, and then into distilled water until no more H_2S is dissolved by the water.

The H_2S solution must be kept in a well-stoppered bottle, and preferably in the dark.

STANDARD SOLUTIONS

General Considerations. Standard solutions are made to contain either a known or definite amount of a substance in a certain measure, or an amount sufficient to neutralize or precipitate a definite weight of another substance. Thus the standard ammonia is a solution of ammonium chloride of such a strength that each c.c. contains 0.001 gramme of ammonia. The standard silver nitrate, on the other hand, contains that amount of silver nitrate in each c.c. which exactly precipitates 0.001 gramme of chlorine.

In making the standard NH_4Cl solution we require 1 gramme of NH_3 per litre. The combining weight of the former is 53.5, and of the latter 17. We require, therefore, $\frac{53.5}{17}$ grammes (i.e. 3.147 grammes) of NH_4Cl in a litre.

In a similar manner we require sufficient $AgNO_3$ dissolved in a litre of water to precipitate 1 gramme of Cl. The c.w. of $AgNO_3$ is 169.7, and that of Cl 35.5. The amount of $AgNO_3$ necessary is therefore $\frac{169.7}{35.5}$ grammes; i.e. 4.780. Thus it remains to weigh 4.780 grammes of $AgNO_3$ and dissolve them in 1 litre of water.

The labour involved in weighing out exactly 3.147 or 4.780 grammes is, for those who are not very adept at balance work, very great, and a simpler method is to weigh out a certain quantity exactly and to calculate the amount of water required.

Great accuracy can be obtained in measuring water with graduated flasks and pipettes. For instance, if 970 c.c. was the quantity required, a 1000 c.c. flask would be filled to the graduation mark, and 20 c.c. and 10 c.c. could be removed with pipettes graduated for these quantities. If 1270 c.c. were required, 1000 c.c. and 250 c.c. flasks could be filled and drained well into one holding 1500 or 2000 c.c. and 20 c.c. added by means of a 20 c.c. pipette.

All standard solutions should, of course, be always kept in glass-stoppered bottles.

Standard Ammonium Chloride

NH_4Cl , *m.w.* 53.5 ; NH_3 , *m.w.* 17

A. Strong Solution

Weigh out as nearly as possible 3.15 grammes of dry anhydrous NH_4Cl and dissolve it in $\frac{w}{3.15} \times 1$ litre of distilled ammonia-free water.

1 c.c. = 0.001 gramme NH_3

B. Weak Solution

Measure 10 c.c. of "A" very accurately by means of a 10 c.c. pipette, and add 990 c.c. of distilled ammonia-free water.

1 c.c. = 0.00001 gramme NH_3

Standard Calcium

Each c.c. of the standard must contain an amount of CaCl_2 having the same weight of Ca as 0.001 gramme of CaCO_3 .

Weigh as nearly 1 gramme of pure crystalline calcite as possible, and dissolve it in the least quantity of dilute HCl which will dissolve it, taking care to cover the vessel in which the solution is being made with a watch or clock glass to prevent the loss of calcium by the spitting. Evaporate to dryness over a water bath, dissolve again in water, and evaporate to dryness a second time; in order to ensure the absence of HCl it is advisable to repeat this a third time. Now dissolve the CaCl_2 in the proper quantity of freshly boiled distilled water, i.e. $\frac{w}{1} \times 1000$ c.c. where w is the weight of CaCO_3 taken.

Standard Copper Solution¹

$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$, *m.w.* 249 ; Cu, *c.w.* 63

1 gramme of copper is contained in 3.95 grammes of copper sulphate. Weigh out as nearly as possible 3.95 grammes of the crystals of CuSO_4 , and dissolve them in $\frac{w}{3.95} \times 1000$ c.c. of distilled water.

1 cc. = 0.001 gram Cu

Standard Indigo Solution

Weigh out approximately 2 grammes of indigo carmine. Digest this with 10 grammes of Nordhausen sulphuric acid for 24 hours. Add 30 c.c. of concentrated sulphuric acid and mix well. Pour this carefully into about 500 c.c. of distilled nitrate-free water. Wash the indigo out of the vessel with distilled water, a few c.c. at a time, until all the indigo has disappeared. Filter and make up the solution to 1 litre.

In order to standardize the indigo solution, a burette graduated in $\frac{1}{10}$ th c.c. is filled with the solution. 2 c.c. of the standard nitrate solution are mixed with 18 c.c. of distilled water and poured into a small wide-mouthed flask. 20 c.c. of strong H_2SO_4 are quickly run into the flask and well mixed. The indigo is now run in, a few drops at a time, and the flask well shaken. After a certain quantity of indigo has been run in, the greenish colour, which quickly disappeared at first, becomes permanent. The amount of indigo required is next read off. Suppose the quantity required was found to be 2.2 c.c.

Then 2.2 c.c. of indigo = 2 c.c. of standard nitrate ; but

1 c.c. of nitrate = 0.000014 gramme of nitrogen

1 c.c. of indigo = $\frac{1}{2.2}$ of 0.000014 \times 2 grammes of
nitrogen = 0.0000127 gramme

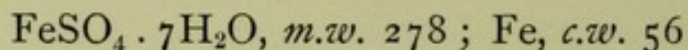
Several experiments must be made to ensure the correctness of these figures, and when the true figures have been found, the indigo should be labelled

“ 1 c.c. = 0.0000127 gramme N ”

¹ The standard solutions of lead, copper, and iron are made up so that 1 c.c. = 0.001 gramme. When using it is convenient to dilute at least ten times.

Standard Iron Solution

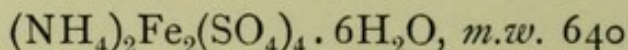
A.



1 gramme of iron is contained in 4.96 grammes of ferrous sulphate. Weigh out as nearly as possible 4.96 grammes of the crystals of pure FeSO_4 , and dissolve them in $\frac{w}{4.96} \times 1000$ c.c. of distilled water.

$$1 \text{ c.c.} = 0.001 \text{ gramme Fe}$$

B.



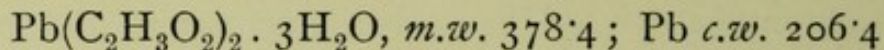
Owing to the fact that absolutely pure FeSO_4 is difficult to procure, it is better to make up a standard solution with ammonium iron alum, which has the advantage that it is stable.

1 gramme of iron is contained in 5.714 grammes of ammonium iron alum.

Weigh out as nearly as possible this quantity and dissolve it in $\frac{w}{5.714} \times 1000$ c.c. of distilled water.

$$1 \text{ c.c.} = 0.0001 \text{ gramme Fe}$$

Standard Lead Solution

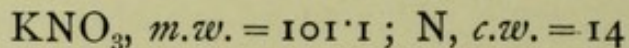


1 gramme of lead is contained in 1.83 grammes of lead acetate. Weigh out as nearly as possible 1.83 grammes of pure lead acetate and add it to about 500 c.c. of distilled water, and then add sufficient acetic acid to render the solution clear.

Add water to make up the quantity to $\frac{w}{1.83} \times 1000$ c.c.

$$1 \text{ c.c.} = 0.001 \text{ gramme Pb}$$

Standard Nitrate Solution



Weigh out as nearly as possible 1.011 grammes of KNO_3 . Dissolve this in $\frac{w}{1.011} \times 1000$ c.c. of distilled water. Each c.c. of the solution will contain 0.001 gramme of KNO_3 , or be equivalent to 0.000014 gramme of N.

Standard Nitrite Solution

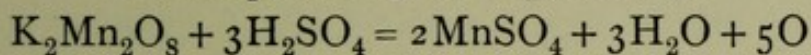
AgNO_2 , *m.w.* 153.7 ; *Ag*, *c.w.* 107.7

Weigh out as nearly as possible 0.406 gramme of pure silver nitrite. Dissolve it in boiling distilled water, and add sufficient sodium chloride to precipitate the whole of the silver. Make the solution up to $\frac{w}{0.406}$ of 1000 c.c. with distilled water, and allow the silver chloride to settle. Remove 100 c.c. of the clear solution and dilute it to 1 litre with distilled water.

1 c.c. = 0.00001 gramme NO_2

Standard Potassium Permanganate Solution

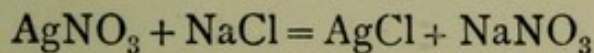
KMnO_4 , *m.w.* 316 ; O_2 , *m.w.* 32



i.e. 316 grammes of KMnO_4 yield 80 grammes of oxygen. The solution has been found most convenient when 10 c.c. of it yield 0.001 gramme of O_2 . 1 litre of the solution must contain 0.1 gramme of available oxygen. The amount of permanganate required to allow this is $\frac{316}{80} \times 0.1$ grammes in a litre ; i.e. 0.395 gramme. Weigh out therefore as nearly as possible 0.395 gramme of pure potassium permanganate and dissolve it in $\frac{w}{0.395} \times 1000$ c.c. of pure distilled water (*w* being the weight of the permanganate actually taken). The solution is then to be labelled

“ 10 c.c. = 0.001 gramme O_2 ”

Standard Silver Nitrate



107.7 + 14 + 48 grammes of AgNO_3 are required to precipitate 35.5 grammes of Cl. Therefore $\frac{169.7}{35.5}$ (i.e. 4.780) grammes of AgNO_3 will be necessary to precipitate 1 gramme of Cl.

As the solution must contain enough silver nitrate in each c.c. to precipitate 0.001 gramme of Cl, it is evident that every litre should contain 4.780 grammes of the nitrate.

Weigh out accurately, as nearly as possible 4.780 grammes of pure recrystallized silver nitrate. Dissolve in about 500 c.c. of water in a perfectly clean chlorine-free flask; add more distilled water, so that the total bulk of the water shall equal $\frac{w}{4.780} \times 1000$ c.c. where w is the actual weight taken. When the dilution is completed the solution should be poured into a suitable glass-stoppered bottle and kept in the dark.

Suppose that w was found to be 5.124 grammes, then the total must be $\frac{5.124}{4.780}$ of 1000 c.c. = 1071.9 c.c.

Standard Soap Solution

Each c.c. of the soap solution must contain an amount of soap which will precipitate 0.001 gramme of CaCO_3 .

Weigh out 10 grammes of sodium oleate or the *Sapo durus* of the B.P. and mix it with a litre of equal parts of methylated spirit and water.¹

Shake well from time to time and set aside in a cool place for 24 hours. Filter into a flask which has been thoroughly well washed out with distilled water.

In order to standardize the soap, it is necessary in the first place to find the amount of the soap which is required to make a permanent lather with 50 c.c. of distilled water. Suppose this is 0.6 c.c.

Into a 6-oz. bottle run 6 c.c. of the standard calcium solution, and add 44 c.c. of distilled water. Now find the quantity of the soap which is required to form a lather. Suppose that 5.3 c.c. are necessary. Then we have—

$$\begin{array}{llll} \text{Soap for } 0.006 \text{ gramme } \text{CaCO}_3 + 50 \text{ c.c. water} & = & 5.3 \text{ c.c.} \\ \text{,, } 50 \text{ c.c. water} & & & = 0.6 \text{ c.c.} \\ \therefore \text{,, } 0.006 \text{ gramme } \text{CaCO}_3 & & & = 4.7 \text{ c.c.} \end{array}$$

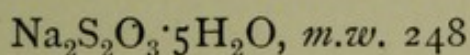
But the soap must be of such a strength that 6 c.c. are required. Therefore the volume of the standardized soap must be $\frac{6}{4.7}$ of what it is unstandardized. Suppose that in this case

the volume is 940 c.c. Then the total volume must be $\frac{6}{4.7}$ of

¹ For the solution and subsequent dilution, exactly equal parts of methylated spirit and water should be mixed together and allowed to cool. The quantities of the cold mixture should be measured.

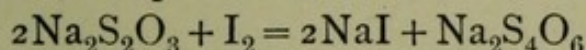
940 c.c. = 1200 c.c. That means that 260 c.c. of spirit and water must be added so that 1 c.c. of the soap shall exactly precipitate 1 c.c. of the standard calcium solution.

Standard Thiosulphate Solution



This solution is made so that 1 c.c. = 0.00025 gramme of oxygen. On referring to Winkler's method for estimating the oxygen dissolved in water it is seen that, according to the equations, 16 grammes of oxygen will liberate 254 grammes of iodine.

On referring to the oxygen absorbed process it will be seen that thiosulphate takes up iodine, thus



that is, 316 grammes of thiosulphate combine with 254 of iodine. Thus since 16 grammes O liberate 254 grammes I, and 254 grammes I are converted into NaI by 316 grammes of $\text{Na}_2\text{S}_2\text{O}_3$, 16 grammes O are equivalent to 316 grammes $\text{Na}_2\text{S}_2\text{O}_3$.

$\text{Na}_2\text{S}_2\text{O}_3$ has 5 molecules of water of crystallization ;

$$\therefore 316 \text{ grammes } \text{Na}_2\text{S}_2\text{O}_3 = 496 \text{ grammes } \text{Na}_2\text{S}_2\text{O}_3 \cdot 5\text{H}_2\text{O}$$

1 c.c. of our standard solution must therefore contain $\frac{0.00025}{16}$

of 496 grammes of thiosulphate, i.e. 0.00775 ; or in other words, 7.75 grammes of crystalline sodium thiosulphate are dissolved in 1 litre of water.

ANALYSIS OF MILK

Cow's milk contains proteins, carbohydrates, fats, salts and water.

The average composition is :—

Water 87.2%

Solids 12.8%, consisting of

{	Sugar, 4.8% Fat, 3.7% Proteins, 3.6% Ash, 0.7%
---	---------------------------------------------------------

Cow's milk, from the point of view of the hygienist, is the only kind of any importance. The results of a large number of analyses have shown that the milk of a healthy cow contains certain proportions of the various constituents. Although the

composition varies with the time of year, the breed of the cow, the interval since calving, the lactation after first or subsequent parturition, etc., it has been found that no milk from a healthy cow is worse than a certain standard. If, therefore, a sample of milk is examined and found to fall below this standard, in some manner or other, the conclusion is arrived at that cream has been extracted, water added, etc.

It is therefore necessary to know what these standards are, in order to be able to say whether the sample of milk under examination is normal or no.

The determinations upon which the quality of the milk is decided are:—

1. The specific gravity.
2. The total solids.
3. The fat.
4. The total solids not fat.
5. The total ash.
6. The quality of the ash.

THE SPECIFIC GRAVITY

Apparatus required

1. A specific gravity bottle.
2. A good balance (sensitive to 0.001 gramme).

The Process

1. A specific gravity bottle holding 25 c.c. is thoroughly cleaned with strong HCl and washed with hot water. It is then dried outside, and rinsed with alcohol and finally with ether. The ether is expelled by blowing into the bottle with the nozzle of a blow-pipe.

The perforated stopper should be treated in the same way.

2. About 50 c.c. of the milk and a like quantity of distilled water should be allowed to stand in glass-stoppered bottles in the laboratory for an hour or so, in order that the temperature of each may be that of the laboratory.
3. The bottle is carefully weighed with the stopper and the weight noted.
4. The specific gravity bottle is now filled with the water to the top of the neck. The stopper is then carefully inserted so that no bubbles of air are contained, and the water fills the hole in the stopper. The bottle is carefully dried outside with a towel, care being taken not to heat the bottle with the hand in the operation.

5. The bottle containing the water is now weighed and the weight noted.
6. The bottle is next emptied, dried, and filled with the milk, taking all the precautions as before.
7. It is again weighed, with the milk, and the weight noted.

EXAMPLE

Weight of bottle	.	18.306 grammes
„	„ + water	43.314 grammes
		<hr/>
Weight of water	.	25.008 grammes
		<hr/>
Weight of bottle + milk	.	44.056 grammes
		<hr/>
Weight of milk	.	25.750 grammes
		<hr/>

$$\begin{aligned}\text{Sp. gr. of milk} &= \frac{25.750}{25.008} \times 1000 \\ &= 1029.6\end{aligned}$$

Notes

It will be observed that the temperature of both water and milk is that of the laboratory. This is, of course, not the theoretical manner of taking the specific gravity, but it is done in practice, and the error is very small. If it were called the relative density instead of the specific gravity, perhaps no objection could be raised either to the method or title.

A simpler method is to take the sp. gr. with a lactometer, or hydrometer, but this is rarely done by analysts of repute, as the error is greater than that obtained by weighing, owing to a variety of circumstances, including the temperature, the inaccuracy of the instrument, etc.

Before taking the sp. gr. the milk must be well shaken, so as to obtain a fair sample. This statement applies to all the other determinations.

THE TOTAL SOLIDS

This determination, as in the case of water, aims at finding the percentage of solid constituents in the liquid, by evaporating the milk and weighing the residue left.

The Process

1. Having thoroughly cleaned and dried a platinum dish, weigh it.
2. Weigh out in the tared dish 5 grammes of the milk after well shaking it.
3. Evaporate to dryness over a water bath, inverting a glass funnel over the dish to prevent any dust from getting into the dish. The drying takes about two hours.
4. Heat in the water oven for half an hour, and weigh the dish after it is cooled.
5. Replace in the oven for a further interval and again weigh. If there is no decrease in the weight, this is accepted. If there is a decrease, the dish is placed in the oven again for a short time and again weighed, until two successive weighings give no difference.

EXAMPLE

Platinum dish	.	.	.	= 10.324 grammes
„ „ + milk	.	.	.	= 15.324 „
„ „ + solids	.	.	.	= 10.964 „
Total solids	.	.	.	= 10.964 - 10.324 grammes
				= 0.64 gramme

But this is the quantity in 5 grammes ;

$$\therefore \text{total solids \%} = 0.64 \times 20 = 12.8\%$$

Notes

The skin that forms on the surface of the milk delays the drying. The formation of this skin may be prevented, and the process therefore hastened, by the addition to the milk, before evaporation, of a few drops of a mixture of one part of acetic acid with nine of methylated spirits.

In order to weigh out exactly 5 grammes of milk, the dish is tared and 5 grammes are added to the weights already counterpoising the dish. Now pipette the milk in a 5 c.c. pipette and allow 4.7 c.c. to run into the dish on the scale of the balance, taking care to have the balance down. Allow the milk to run into the dish after this drop by drop, until upon raising the beam it is found that there is just too much milk. With a clean, small-pointed, glass rod remove a trace of the milk and again weigh. Wipe the rod on a clean towel and continue to remove traces until the milk weighs exactly 5 grammes. A little practice will enable the experimenter to perform the weighing both quickly and accurately.

THE ASH

Having weighed the total solids, the dish is heated to dull redness over a Bunsen, or preferably over an Argand burner, until the whole of the organic matter is burnt.

The process is expedited by breaking up the masses of dried milk with a somewhat stiff platinum wire from time to time.

When the whole of the contents are of a greyish white appearance, the dish is removed to a desiccator to cool and afterwards weighed.

EXAMPLE

Weight of dish	= 10.324 grammes
"	"	+ ash	.	.	= 10.358 "
<hr/>					
Ash	= 0.034 gramme
But this is the ash from 5 grammes of milk ;					
∴ the percentage of ash = 0.034×20					
= 0.68%					

THE FAT

Apparatus required

1. A Schmidt-Werner tube.
2. A stout test-tube, carrying a wash-bottle arrangement.
3. A platinum dish.
4. A 20 c.c. pipette.

The Process

1. 10 grammes of milk are quickly and accurately weighed into the platinum dish.
2. The Schmidt-Werner tube is clamped in a vertical position, after having been thoroughly cleaned and drained ; and a small funnel (cut short as to its tube) is placed in the mouth of the tube.
3. The milk is poured down the funnel, the remains of the milk are then washed on to the funnel from the dish with the strong HCl (which must be pure) as contained in the extemporized wash-bottle. The milk is then washed out of the funnel with the acid, and finally the sides of the tube are washed down with the acid, and sufficient acid added so that the mixture of milk and acid reaches the 20 c.c mark.

4. The tube is shaken so as to mix the milk and acid well, and the milk is then boiled, the tube being constantly shaken. When the liquid is of a fairly deep brown colour, the heating is stopped.
5. The whole is then allowed to cool, by immersing the tube in water if desired.
6. When the whole is cool, ether is poured in to the 50 c.c. mark. A cork is now inserted into the mouth, and the tube inverted gradually so that the whole of the brown liquid collects in the upper end of the tube.
This must be repeated fifteen or twenty times, so that the ether may come into contact with, and take up all the fat. The tube is then held vertically and rotated quickly between the hands in order to get the débris to settle well, so that the level of the ether can be easily read.
7. A 20 c.c. pipette having indiarubber tubing at the upper end is now inserted into the tube, and exactly 20 c.c. of the ether are sucked out of the tube. The ether can be easily held in the pipette by pinching the indiarubber tubing.
8. The 20 c.c. of ether are then allowed to run into a clean platinum dish, which is then placed in a water oven, at 60° C., in order to drive off the ether.
9. When the ether is completely evaporated, the dish now containing the fat is weighed.
10. The ether left in the tube is read off and noted for the subsequent calculation.

Explanation

The fat in the milk being in the form of a perfect emulsion cannot be taken up by ether without previous treatment. Boiling with HCl converts the albuminous envelopes round the fat into soluble acid-albumin, the fat is set free and rises to the top. The ether is now able to dissolve it.

EXAMPLE

Weight of milk taken	.	.	.	= 10.000 grammes
„ dish	.	.	.	= 42.312 „
„ dish + fat	.	.	.	= 42.638 „
„ fat	.	.	.	= 0.326 gramme
Ether left in tube	.	.	.	= 3.5 c.c.

Total fat = $\frac{23.5}{20} \times 0.326$ gramme (since

0.326 gramme of fat was present in
20 c.c. of ether) . . . = 0.383 gramme

But this quantity is present in 10 grammes of milk ;

∴ 100 grammes contain 3.83 grammes ; or the milk contains 3.83% of fat.

Notes

The Werner-Schmidt tube is fixed vertically to prevent the milk from getting into the upper bulb, and a funnel is used to the same end.

If several estimations are to be made, the 20 c.c. pipette should not be cleaned out, but the ether must be drained each time into the platinum dish.

After having done this, it will be noticed that there is a modicum of fat lining the pipette and this amount is constant. From this it is evident that the first estimation done with a new or clean pipette will be slightly under the proper figure.

In mixing the ether with the fat, HCl, etc., care must be taken not to shake the tube whilst the brown sediment and ether are mixed together. If it is done, a froth will be formed which is difficult to get rid of.

When all the ether is at one end, that end should be well shaken, in order to get the ether well in contact with the walls of the tube.

ADULTERATION OF MILK

When a milk vendor finds that the consumption of milk exceeds the production, there are certain courses open to him. Firstly, he may refuse to supply some of his customers ; secondly, he may increase the production ; thirdly, he may make the production fit the consumption. The last course is not altogether unknown ; and, in order to follow it, the dishonest vendor adds water to the milk. Sometimes he wishes to sell cream, and then he may deplete some of his milk of fat, and sell the resulting skimmed milk as genuine.

If he abstracts cream he will raise the specific gravity of the milk ; if he adds water he will lower it. It may possibly occur to him that some of his customers have lactometers ; and so, judiciously, he abstracts cream *and* adds water. If these operations are carefully performed, the resulting product will have a normal specific gravity ; and the dishonest milkman may trade unsuspected, until an inclement fortune sends some of his milk to an analyst. Such frauds in connection with milk are often referred to by the kindly name of sophistication.

Standards for Milk

In order to detect a fraudulent milk vendor, it is necessary to have a standard to which milk should conform; if the milk fails to come up to the standard, then it can be said not to be genuine.

It has been ascertained that milk, if genuine, should contain :—

1. At least 3% of fat.
2. At least 8.5% of solids not fat.

And that skimmed milk, if unwatered and genuine, should contain at least 9.0% of solids not fat.

These are legal standards; and if a milk sample fails to attain to these it is considered to be adulterated and not genuine.

Addition of Water

When water is added to milk the specific gravity of the sample is lowered, and the solids both fatty and non-fatty are diminished. The amount of water added is determined from the amount of solids not fat. Thus a sample of milk was found to contain 8% of solids not fat; but 8.5% of solids not fat denotes 100% of genuine milk. Therefore 8% of solids not fat denotes $\frac{8 \times 100}{8.5} = 94.1\%$ of genuine milk; or, in other words, about 6% of water has been added to the milk.

Again, the amount of water added may be calculated from the ash. The ash in milk is fairly constant at 0.7%, although there is no legal standard for it. A milk sample was found to give 0.6% of ash: now 0.7% of ash shows 100% of pure milk; therefore 0.6% of ash shows $\frac{0.6 \times 100}{0.7} = 85.7\%$ of pure milk. So about 15% of water has been added to this sample.

In legal work and ordinary routine work, however, it is more convenient to calculate the added water from the figure for solids not fat, as has been shown above.

Abstraction of Cream

Removal of cream raises the specific gravity of milk, and lowers the amount of fat present. The amount of cream abstracted is measured by the estimation of the fat.

Thus a sample of milk was found to have only 2.5% of fat.

Now 3% of fat show that the fat in the milk is all present; or a figure of 3 denotes 100% of fat. Therefore 2.5 denotes $\frac{2.5 \times 100}{3} = 83.3\%$ of fat originally present. So more than 16% of the original fat has been abstracted.

Where the fat is low and the solids not fat are average, it may be inferred that the diminished fat figure is due to the abstraction of cream and not to the addition of water. Where the fat is much reduced, and the solids not fat slightly reduced, it may be inferred that cream has been abstracted and water added.

Rarely condensed milk is added to skimmed or watered milk in order to supply the deficiency in fat. The fraud is detected by the abnormally high figure for total solids, and by the increase in sugar. Condensed milk contains much sugar and solids—sometimes as much as 40%.

Note

It has been found that the specific gravity, total solids, and fat bear a somewhat constant relation to one another. Advantage has been taken of this fact, and a formula has been calculated for deducing the fat from the specific gravity and total solids. This is a great advantage when many samples of milk have to be estimated; because the estimation of the fat requires more attention than that of the total solids.

The formula is:—

$$\text{T.S.} = \frac{\text{Spec. Grav.} - 1000}{4} + 0.14 + \frac{6F}{5}$$

Where T.S. = Total solids, and F = fat.

EXAMPLE

A milk sample had a specific gravity of 1.032, and had 12.34% of total solids. Then:—

$$12.34 = \frac{1.032 - 1.000}{4} + 0.14 + \frac{6F}{5}$$

$$\therefore F = 3.5.$$

Preservatives

The most common additions to milk are sodium carbonate or bicarbonate, boric acid or borax, formalin, and occasionally salicylic acid.

Sodium Carbonate or Bicarbonate

is added in order to neutralize the acidity generated by the growth of micro-organisms, and to delay the curdling.

These may be detected

1. By boiling the suspected milk for an hour. After prolonged boiling, normal milk has only a faint tinge, but milk to which either of these salts has been added assumes a fairly deep brown colour.
2. By the reaction on rosolic acid. To 10 c.c. of the milk add 10 c.c. of alcohol and a few drops of a 1% alcoholic solution of rosolic acid.

Normal milk will show a brownish colour, whereas milk with NaHCO_3 added will turn the rosolic acid a rose colour.

Boric Acid or Borax

The presence of these preservatives may be detected as follows :—

1. Evaporate 50 c.c. of the milk, which has been rendered slightly alkaline, to dryness, and incinerate.
2. Dissolve in the minimum quantity of HCl and again evaporate to dryness.
3. Dissolve the residue in a small quantity of hot water and moisten a piece of turmeric paper with the solution. Dry the turmeric paper.

If boric acid or borax was present in the milk, the dry turmeric paper will assume a rose or cherry-red colour.

The quantitative estimation is a very lengthy and delicate process, and beyond the scope of this work.

Salicylic Acid

This acid may be detected as follows :—

1. Acidify about 25 c.c. of the milk with HCl and filter.
2. Shake up the filtrate well with ether and decant it.
3. Evaporate the ether and moisten the residue with ferric chloride.

The presence of salicylic acid is indicated by the development of a violet colour.

Formalin

The presence of small quantities of formalin in milk may be detected by the method suggested by Hehner.

If a few c.c. of milk are poured into a test-tube, and a like quantity of strong commercial sulphuric acid is poured down the side of the tube so that the two do not mix freely; at the junction of the H_2SO_4 with the milk, a purple ring will be formed which becomes more extensive upon gradually agitating the test-tube.

Notes

This test is only available for formalin in milk, since no such reaction can be obtained with pure formalin. Pure H_2SO_4 also gives no such reaction, so that the commercial acid must be employed.

A still better test is as follows:—

10 c.c. are placed in a test-tube and 2 c.c. of 10% KOH solution and 1 c.c. of a watery solution of phloroglucinol are added. If formalin is present a pink colour will be at once produced.

ANALYSIS OF BUTTER

THE term “butter” has been defined in the Margarine Act, 1887, to “mean the substances usually known as butter, and made exclusively from milk or cream or both, with or without salt or other preservatives, and with or without the addition of colouring matter.”

As in the case of milk, the composition of butter varies within certain limits. An average pure butter has the following percentage composition:—

Fat	85.45
Curd	2.75
Salt	3.25
Water	8.55

but even in genuine butters the fat may vary from 82% to 87% and the water a corresponding amount, and no butter should be condemned as being adulterated with water unless it contains less than 80% of fat.

The proximate analysis of a sample of butter may be undertaken as follows :—

ESTIMATION OF WATER

Apparatus required

1. A platinum dish and a piece of glass rod.
2. A water bath and a water oven.
3. A good balance.

The Process

1. Weigh about 2 or 3 grammes of butter into the clean dry platinum dish.
2. Place the dish on the water bath and stir from time to time with the glass rod, leaving the latter in the dish the whole time.
3. When the visible water has evaporated, wash the fat off the rod with a little ether, and place the dish in the water oven until all the water and ether have disappeared.
4. Remove to a desiccator until the dish is cool, and weigh.
5. Replace in the oven for half an hour, again place in the desiccator and re-weigh.

If the two weighings are alike or very approximate the last weighing may be taken as the correct weight of the dry butter.

The loss in weight of the butter represents the water present.

EXAMPLE

Platinum dish	.	.	.	= 10.324 grammes
Weight of dish + butter	.	.	.	= 13.524 „
				<hr/>
Weight of butter	.	.	.	= 3.200 „
Weight of dish + dried butter	.	.	.	= 12.960 „
Loss in weight	.	.	.	= 0.564 gramme

$$\therefore \text{percentage of water} = \frac{0.564}{3.2} \times 100 = 17.60$$

Note

Unless the butter is stirred from time to time during the process of evaporation, the melted butter floats on the surface of the water, and so prevents its evaporation.

Care must of course be taken to wash all the fat off the glass stirring rod.

ESTIMATION OF SALT

Apparatus required

1. A separating funnel.
2. The apparatus for estimating chlorine in water.

The Process

1. Weigh out 5 grammes of the butter to be analysed, and carefully transfer it to a clean filter funnel, washing the remnants off with hot water.
2. Pour about 200 c.c. of hot distilled water on to the butter and shake up well.
3. Pour off the water into a measuring glass, pipette off 20 c.c. into a porcelain evaporating basin, and estimate the chlorine with standard silver nitrate as in the case of water.

EXAMPLE

Butter taken 5.2 grammes
 Measure of water after shaking with butter 220 c.c.
 Standard AgNO_3 required for 20 c.c. = 4.4 c.c.
 \therefore 0.004 gramme Cl are present in 20 c.c. of the water
 \therefore butter contains

$$\frac{0.004}{1} \times \frac{58.5}{35.4} \times \frac{220}{20} \times \frac{100}{5.2} \% \text{ of NaCl}$$

$$= 1.42\%$$

Detection of Adulteration with Foreign Fats

The most important chemical examination of butter is the determination of the presence of fats which are not those of milk.

Margarine, which now is a good and cheap substitute for butter, is made chiefly of beef fat. According to the definition of butter given above, the addition of beef or other fat to butter is a legal offence, unless the mixture is sold as margarine, and it is illegal to mix more than 10% of butter fat with margarine.

In order to test the properties of butter fat and margarine fat it is necessary to obtain these free from water, curd, salt, etc.

Perhaps the simplest method of doing this is to fill a beaker of about 50 c.c. capacity with the butter and place it in the water oven at 100°C . until the butter has melted, and the water, curd, etc., have sunk to the bottom.

The supernatant fat is then carefully poured on to a dry filter-paper—care being taken that no water gets on to the paper—and allowed to filter into a clean dry beaker in the oven.

The same method is adopted for margarine. In the process to be described, although butter only is used, it will be understood that exactly the same procedure is to be adopted for margarine.

The general compositions of butter and margarine fats are—

	BUTTER		MARGARINE
Olein	42·21	...	30·4
Palmitin and stearin .	50·00	...	69·2
Butyrin	4·67	}	0·4
Caproin	3·02		
Caprylin	0·10		
	<hr/> 100·00		<hr/> 100·0

These fats are the salts of the respective fatty acids with glycerol. Oleic, palmitic and stearic acids are termed non-volatile or insoluble fatty acids, and butyric, etc., the volatile or soluble acids.

The differences in the composition of the fats are responsible for the chemical and physical properties which are next to be determined.

Estimation of the Specific Gravity

The procedure is similar to that adopted for determining the sp. gr. of milk.

Instead of weighing at the room temperature, however, the melted butter fat and the clean sp. gr. bottle are placed in an incubator whose temperature is 37° C., for an hour. The bottle is quickly filled, restoppered, wiped, and replaced in the incubator for a few minutes. It is then weighed as quickly as possible. The sp. gr. is then calculated from the figure thus obtained and that obtained on weighing the same bottle full of water at 37° C.

EXAMPLE

Weight of bottle + water	= 18·143 grammes
" " "	= 8·401 "
	<hr/>
Weight of water	= 9·742 "

Weight of bottle + butter	= 17.279 grammes
" " 	= 8.401 ,,
Weight of butter	= 8.878 ,,

$$\therefore \text{sp. gr. of butter} = \frac{8.878}{9.742} \text{ of 1}$$

$$= 0.9113$$

The sp. gr. of true butter fat is never below 0.911, whereas that of margarine never rises above 0.906.

If we are dealing with an adulterated butter, the estimation of the percentage of butter present is merely a proportion sum.

EXAMPLE

The sample had a sp. gr. of 908.5. Find the percentage of adulteration.

We must accept the lowest sp. gr. of butter, i.e. 911, and the highest of margarine, 906.

$$x \times 906 + (100 - x) 911 = 100 \times 908.5$$

$$5x = 250$$

$$x = 50\%$$

The Valenta¹ Test Modified by Jean

This test depends upon the different amounts of glacial acetic acid taken up by butter and margarine respectively.

Apparatus required

1. A graduated test-tube 1 cm. in diameter.
2. A graduated pipette.
3. Water bath.

The Process

1. Pour 3 c.c. of the melted fat heated to 50° C. into the test-tube and place in the water bath at 50° C.
2. By means of the pipette add 3 c.c. of glacial acetic acid to the fat. Leave the tube in the water bath until the temperature of the whole of the contents is 50° C.

¹ The original Valenta test was as follows: Equal parts of the fat and acetic acid were mixed together and heated to 100° C. They were subsequently cooled and the temperature observed at which a cloudiness appeared. This was found to occur with margarine at 96.5° C. and with butter at 61.5° C.

3. Shake well two or three times and return to the bath. Allow the acetic acid to settle to the bottom and read off the height of the acid, i.e. the junction between the acid and the oil.

The loss in volume of the acetic acid represents the amount dissolved by the fat.

EXAMPLES

Samples of butter and margarine were tested. The level of the acetic acid after the experiment was 1.2 c.c. in the butter tube, and 2.2 in the margarine tube.

$$\begin{aligned} \therefore \text{acetic acid absorbed by butter} &= \frac{1.8}{3} \times 100 = 60\% \\ \text{margarine} &= \frac{0.8}{3} \times 100 = 26.6\% \end{aligned}$$

The average figures are

Butter	63.3
Margarine	26.6

If the sample submitted to analysis comes up to the standard of the above tests it can be certified as free from admixture with foreign fats. If, on the other hand, the sample is below the standards, it is well to apply a further test. This test consists in estimating either the volatile or the non-volatile fatty acids, or both.

ESTIMATION OF THE VOLATILE FATTY ACIDS

REICHERT-WOLLNY PROCESS

Apparatus, etc., required

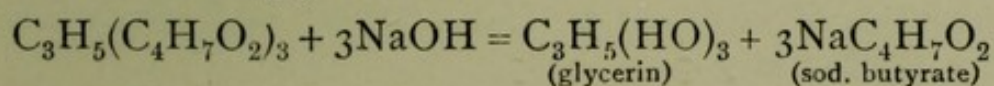
1. Globular flask about 300 c.c. capacity.
2. Small Thorpe's condenser.
3. Graduated measures.
4. Graduated burette.
5. Two small pieces of freshly burnt pumice.
6. Filter funnel and paper.
7. Beakers or small flasks.
8. $\frac{N}{10}$ NaOH.
9. Phenolphthalein.
10. 50% NaOH.
11. 25% sulphuric acid.

The Process

1. 5 grammes of the filtered butter fat are poured into the flask and 2 c.c. of the 50% NaOH and 10 c.c. of absolute alcohol are added to the butter.
2. A reflux condenser is fitted to the flask, and the latter is placed over a water bath at 100° C. The flask is shaken from time to time and the boiling continued for about half an hour.
3. The condenser is disconnected from the flask and the alcohol is allowed to evaporate.
4. 100 c.c. of distilled water are poured in the flask, the whole shaken up well, and placed on the water bath for a quarter of an hour.
5. 4 c.c. of 25% H₂SO₄ are mixed with 36 c.c. of distilled water and poured into the flask. At the same time the two small pieces of pumice are dropped in the flask.
6. A cork is inserted into the neck of the flask, carrying a tube bent at an obtuse angle and having a bulb blown on it close to the cork. The whole is then connected to a condenser and within 28–32 minutes exactly 110 c.c. are distilled into a measure through a funnel carrying a filter-paper.
7. 100 c.c. of the distillate are taken, and to this 1 c.c. of phenolphthalein is added.
8. The decinormal caustic soda is then run into the distillate until a permanent rose-pink is obtained. The number of c.c. of soda is noted. To the number used one-tenth is added.
9. This number of c.c. of decinormal soda is known as the "Reichert-Wollny Number" of the butter or fat. Butter gives a Reichert-Wollny number of not less than 24. Margarine gives a Reichert-Wollny number of not less than 3. Mixtures of margarine and butter give Reichert-Wollny numbers between 3 and 24.

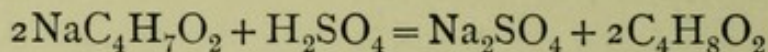
Explanation

Boiling the fat with alcoholic soda converts the glycerol salts into soda salts and glycerin.



The reaction for the butyrate is similar to that for all the others, oleate, stearate, etc.

The addition of the sulphuric acid decomposes the sodium butyrate, etc.; butyric acid, etc., are set free and are distilled over by heat



The object of filtering the distillate is to free it from traces of the non-volatile acids which almost invariably distil over.

Pumice is added to prevent the "bumping" which so often accompanies the distillation of any liquid containing sulphuric acid.

EXAMPLE

Five grammes of butter fat gave a distillate, 100 c.c. of which required 26.8 c.c. of $\frac{\text{N}}{10}\text{NaOH}$ to neutralize it.

Observe that 110 c.c. were distilled and only 100 c.c. neutralized. The acidity of the whole distillate will therefore obviously be $\frac{11}{10} \times 26.8$, or one-tenth more than the observed quantity, namely, 29.48. This is the Reichert-Wollny number of this butter fat, and shows genuine butter.

Suppose the figure obtained had been 18. What percentage of adulteration with margarine would this show?

Let x = the percentage of adulteration (i.e. of margarine).

Then $x + 3 + (100 - x)24 = 100 \times 18$

$$21x = 600$$

$$x = 28.5\%$$

Notes

The Reichert-Wollny number does not give an exact estimation of the volatile fatty acids present in the butter fat. It is an empirical figure only, and in order that it may be obtained accurately, strict care must be taken in performing the various operations; the apparatus must be of standard Reichert-Wollny size; and the distillation must be completed within the specified time. Under such standard conditions butter fats invariably yield figures over 24; other fats (with the exception of cocoanut oil) give numbers of less than 3. Cocoanut oil may give a Reichert-Wollny number at 7-8.

ESTIMATION OF THE INSOLUBLE FATTY ACIDS

Instead of estimating the volatile, the insoluble fatty acids may be estimated. In order to do this, the fat is saponified as before. After saponification it is transferred to a litre flask, and the small flask is washed two or three times with hot water, the washings being added to the contents of the large flask. 4 c.c. of 25% H_2SO_4 are mixed with 30 or 40 c.c. of hot water and poured into the flask, which is then carefully filled to within an inch of the top of the neck with hot water. The fatty acids will now collect on the top of the water.

The mouth of the flask is covered over, and the contents are allowed to cool. When the flask is cold, the mass of fatty acids can with a little care be loosened by means of a glass rod from the sides of the neck, and transferred *en masse* to a clean porcelain evaporating basin. The water remaining in the flask is now filtered through a filter-paper, and when the filtration is complete both paper and flask are allowed to dry.

The mass of fatty acids is dissolved in ether and filtered through the dried filter-paper in a tared platinum dish, and the basin is washed free of fatty acids with a little ether. The large flask is now washed out with ether, and the ether filtered. Finally the filter-paper is washed with ether, so that all the fatty acids are in the platinum dish. The ether is next evaporated in the water oven, and the dish is weighed. The gain in weight represents the amount of the insoluble fatty acids.

Note

In butter fat the insoluble fatty acids form about 88%. In margarine and other fats they are about 96%.

Preservatives

The common preservative added to butter is boric acid or borax. The detection of this is performed in exactly the same manner as in the case of milk.

ANALYSIS OF FLOUR

A GOOD wheaten flour has the following average percentage composition :—

Starch	.	.	59.7	Soluble nitrogen	.	1.8
Dextrin	.	.	7.2	Fat	.	1.2
Cellulose	.	.	1.7	Mineral matter	.	1.6
Gluten	.	.	12.8	Water	.	14.0
Total				.	.	100.0

The chemical analysis of a sample of flour for adulteration is usually confined to the determination of the percentage of water, gluten, ash, and mineral matter.

The best practical and domestic test to apply to flour is to use it for making bread. A good flour makes good bread.

ESTIMATION OF THE PERCENTAGE OF WATER

5 grammes of flour are weighed in a platinum dish, and dried in the water oven until a constant weighing is obtained. The loss in weight represents the water present, and the percentage is calculated therefrom. In a good sample of flour the percentage of water should not exceed 18.

ESTIMATION OF THE ASH

After the dried flour has been weighed it is incinerated in the platinum dish, and the residue weighed.

The percentage of ash varies from 0.5% to 1%, but should not exceed this latter figure unless mineral matter has been added.

ESTIMATION OF THE GLUTEN

1. Weigh 50 grammes of flour, mix it carefully and thoroughly with 50 c.c. of distilled water, and allow it to stand for an hour.
2. Collect the paste thus made in a linen handkerchief, and make the latter into a bag.
3. Allow water from a tap to run on the outside of the bag and knead the paste well until the water running away is clear.
4. Remove the paste from the bag and complete the kneading in the hand until the water which runs away is quite clear.
5. Squeeze the mass and remove all the extraneous water and weigh. The moist gluten should form from 25% to 30% of the weight of the flour.
6. Dry the gluten in a platinum evaporating basin and weigh again. The dry gluten should be 12% to 15% of the flour.

Note

Old or musty flour will not produce an adhesive mass which can be kneaded, but a semi-liquid mass which easily washes away. No such mass of gluten can be obtained with rye flour, however good and new it may be.

ESTIMATION OF MINERAL MATTER

The Chloroform Test

If flour which contains added mineral matter be shaken up with chloroform the flour will float in the chloroform, whilst the mineral matter very quickly sinks to the bottom.

If the majority of the chloroform be poured off, and the sediment shaken up with fresh chloroform two or three times, the mineral matter can be obtained free from flour.

The sediment being washed out of the test-tube and the chloroform evaporated, is now ready for qualitative examination.

If alum has been added to the flour (a rare proceeding, since it is not usual to add it until the flour is in the process of bread-making) it will be detected in the sediment, as it is only very sparingly soluble in chloroform.

ERGOT

Flour, and particularly rye flour, is liable to be ergotized, and it is necessary for the student to know how to detect the presence of this drug. This may be done by warming some of the suspected flour with a solution of KOH. If ergot is present the characteristic odour of propylamine will be detected.

Ergot may also be detected by shaking up 2 grammes of the suspected flour in 10 c.c. of 70% alcohol containing $\frac{1}{10}$ HCl. If ergot is present, after a short time a blood-red colour will develop.

ANALYSIS OF BREAD

AN average sample of bread has the following percentage composition :—

Starch, dextrin, maltose	51.3
Proteins	6.5
Fat	1.0
Ash	1.0
Water	40.0
	<hr/>
	100.0

The chemical analysis of a sample of bread is usually confined to the determination of the acidity, the ash, and the presence of alum.

THE ESTIMATION OF THE ACIDITY

Acetic and lactic acids are both present in bread, but the acidity is usually calculated in terms of acetic acid.

In order to ascertain the acidity, grate a certain quantity of the bread crumb and weigh 20 grammes of the crumbs. Transfer these into a beaker and add 100 c.c. of hot distilled water. Stir well and allow it to stand for two or three hours. Filter 25 c.c. into a flask or evaporating basin and add a drop of phenolphthalein.

Add $\frac{N}{10}$ NaOH from a burette until the fluid is neutral. From the amount of alkali used calculate the amount of acid.

EXAMPLE

20 grammes of bread-crumbs were taken.

25 c.c. of the extract took 9.6 c.c. of $\frac{N}{10}$ NaOH.

The 20 grammes of bread yielded therefore an acidity equal to 38.4 c.c. of $\frac{N}{10}$ NaOH. There was therefore as much acid as in 38.4 c.c. of $\frac{N}{10}$ acetic acid. The molecular weight of acetic acid is 60. Therefore 100 c.c. of $\frac{N}{10}$ acetic acid contain 0.6 gramme of acetic acid.

$$\therefore 38.4 \text{ c.c. contain } \frac{38.4}{100} \times 0.6 \text{ gramme} \\ = 0.2304 \text{ gramme}$$

100 grammes of the crumb would contain 5×0.2304 , i.e. 1.15 grammes of acetic acid.

ESTIMATION OF THE ASH

The estimation of the ash is performed in a manner similar to that which has been previously described.

If the ash exceeds 2% there will be a suspicion of added mineral matter.

DETECTION OF ALUM

In order to detect the presence of alum in bread, a slice should be cut from the middle of the loaf and a few drops of a mixture of a fresh solution of logwood in alcohol and a saturated solution of ammonium carbonate should be poured on to the centre of the slice. When the fluid dries up a distinctly blue colour will develop if alum is present. If no alum is present the colour will be brown.

ESTIMATION OF ALUM

The estimation of alum is a somewhat lengthy process, but may be performed in the following manner:—

100 grammes of bread are carefully incinerated in a platinum dish until the ash does not decrease in weight. After cooling, 3 c.c. of pure strong HCl are added, and the whole diluted with 20 or 30 c.c. of water. The fluid is then boiled and filtered.

The residue should be dried, incinerated, weighed and returned as silica.

The filtrate is now alkalized with ammonia, when the phosphates of calcium, magnesium, iron, and aluminium will be thrown down. The fluid is now made strongly acid with acetic acid, boiled, and filtered. The residue consisting of the phosphates of iron and aluminium is now dried and weighed. After the weight has been ascertained, the residue is dissolved, the iron estimated colorimetrically and deducted from the total residue. The remainder will represent the phosphate of aluminium, and from this the amount of alum can be calculated.

In order to be able to say whether alum has been added to bread it is first necessary to deduct the alumina which is present normally in bread. It has been found that in a normal sample of bread there is as much alum as silica. The weight of silica found must therefore be deducted from the amount of alum found, and any excess will represent added alum.

ANALYSIS OF COFFEE

COFFEE is frequently sold mixed with chicory, a preparation from the root of the wild endive. When it is sold as a mixture no legal objection can be taken to it. Sometimes it happens, however, that such a mixture is sold as pure coffee: this constitutes a fraud, and it becomes necessary to know whether any sample of coffee has been adulterated. The usual adulterant is chicory, and the detection of this substance only will be treated here.

Qualitative Tests for Chicory

- A. Put some of the suspected coffee upon the surface of some water in a tall beaker or cylinder. Roasted chicory sinks at once, making a brown trail in the water through which it passes; coffee will float for several minutes, and takes longer than chicory to colour the water.
- B. The smell of chicory is different from that of coffee.
- C. Examine with the microscope. Chicory shows typical "dotted ducts."
- D. Heat some of the sample in a platinum dish until it is reduced to ash. The ash of coffee is almost white; chicory contains more iron, and the ash has a reddish colour.
- E. Take 5 grammes of the suspected coffee, and pour into it 30 c.c. of boiling water. Filter into a Nessler glass and add 5 c.c. of lead acetate solution. This will precipitate the colouring matter of coffee, leaving the supernatant fluid colourless; if chicory is present the column of fluid will retain its brown colour.

ESTIMATION OF AMOUNT OF CHICORY

FIRST METHOD

Specific Gravity of a 10% Extract

1. Weigh exactly 10 grammes of the sample of coffee, and place the coffee in a beaker.
2. Make a paste of the coffee with a little distilled water, and then dilute so that exactly 100 c.c. of water are present.
3. Cover the beaker and leave it for 10 or 12 hours.
4. Filter 50 c.c., fill into a specific gravity bottle and weigh.
5. Calculate the specific gravity, and from this the percentage of adulteration.

EXAMPLE

The average specific gravities of 10% extracts of coffee and chicory are respectively 1.009 and 1.024. The specific gravity of the 10% extract of a sample of mixed coffee and chicory was found to be 1.0139.

If x is the percentage of coffee in the mixture it is obvious that

$$\begin{aligned}
 x \times 1.009 + (100 - x)1.024 &= 100 \times 1.0139 \\
 15x &= 1010 \\
 x &= 67.3
 \end{aligned}$$

That is, there is 32.7% of chicory.

TABLE GIVING THE APPROXIMATE PERCENTAGES OF COFFEE IN A MIXTURE OF COFFEE AND CHICORY FROM THE SPECIFIC GRAVITY OF A 10% EXTRACT.

Percentage of Coffee	Sp. gr. of 10% Extract	Percentage of Coffee	Sp. gr. of 10% Extract
100	1009.00	45	1017.25
95	1009.75	40	1018.00
90	1010.50	35	1018.75
85	1011.25	30	1019.50
80	1012.00	25	1020.25
75	1012.75	20	1021.00
70	1013.50	15	1021.75
65	1014.25	10	1022.50
60	1015.00	5	1023.25
55	1015.75	0	1024.00
50	1016.50	—	—

SECOND METHOD

The Determination of the Soluble Ash

A weighed quantity of coffee is placed in a platinum dish and incinerated. After cooling, the ash is treated with distilled water until all the soluble matter has been dissolved. The solution is then filtered, the dish and paper washed with distilled water, the filtrate transferred to a tared platinum dish and evaporated to dryness. The dish is then weighed—the increase in weight representing the amount of soluble ash. From the weight the percentage is calculated. In order to determine the adulteration, it is assumed that chicory never yields a soluble ash greater in amount than 1.7%, and that coffee yields a soluble ash never less than 3%.

ANALYSIS OF SPIRITS

THESE, with the various liqueurs, contain a large proportion of alcohol. The Sale of Food and Drugs Amendment Act, 1879, fixes the minimum of alcohol for spirits.

Whisky, brandy, and rum must not be more than 25 under proof.

The term "proof spirit" arose when the test applied was to moisten gunpowder with the spirit in question. On applying a

light, if the gunpowder burnt, the spirit was said to be proof or over proof; if it did not, it was under proof. It has been subsequently defined by Act of Parliament to be a mixture of alcohol and water of such a density that the weight of 13 volumes at 51° F. shall be equal to that of 12 volumes of water. According to this definition, proof spirit contains 49·24% by weight and 57·06% by volume of alcohol.

25 under proof corresponds, therefore, to about 43% by volume of absolute alcohol.

Gin may be as much as 35 under proof, i.e. need only contain about 37% by volume of alcohol.

Alcohol

To determine the amount of alcohol present in a sample of spirit.

Apparatus required

1. Small distilling apparatus with Argand burner.
2. Specific gravity bottle.

The Process

1. 100 c.c. of the spirit are measured into the distilling flask.
2. The spirit is distilled over the Argand burner until it is all but dry.
3. The distillate is made up to 100 c.c. by the addition of distilled water and well mixed.
4. The specific gravity of a portion of this is now taken by the aid of the specific gravity bottle.
5. From the tables¹ the percentage of alcohol is read.

Acidity

Nearly all spirits have a slight acidity, due either to volatile acids (which are returned in terms of acetic acid) or to fixed acids (which are returned as tartaric acid).

The following are approximate figures:—

					TOTAL ACIDITY %
Brandy	0'01 to 0'05
Rum	0'5
Whisky	0'1

To determine the acidity.

1. 50 c.c. of the spirit are measured into a flask, and a few drops of phenolphthalein are added.

¹ These tables are of such a length that they have been relegated to the Appendix.

2. Decinormal soda is added drop by drop until the point of neutrality is arrived at.
3. The percentage acidity is then calculated.

Brandy being made from grapes, the acidity is returned in terms of tartaric acid (1 c.c. $\frac{N}{10}$ NaOH = 0.0075 gramme tartaric acid).

For other spirits the acidity is in terms of acetic acid (1 c.c. $\frac{N}{10}$ NaOH = 0.006 gramme acetic acid).

Sulphuric acid is sometimes present, but the estimation of the free acid is beyond the scope of this work.

The **residue** of the various spirits and the ash resulting from them vary.

	RESIDUE %	ASH %
Brandy . . .	1 to 1.5	0.04 to 0.2
Whisky . . .	0.7	mere trace
Rum . . .	0.7 to 1.5	0.1
Gin (sweetened) .	5 to 6	

The residue is obtained by evaporating a measured quantity in a tared platinum dish, and weighing. The difference in weights is the amount of residue. This is then burnt, and the ash weighed. The percentages are then calculated.

Tannin

Any amount of tannin more than mere traces may be detected by adding a few drops of perchloride of iron solution to the spirit. A darkening in colour is indicative of this substance.

ANALYSIS OF WINES

THE determination of the alcohol and the residue is made as in the analysis of spirits. There is an additional determination necessary, namely, the estimation of the volatile acidity.

Acidity

a. Total acidity.

25 c.c. of the wine are measured into a beaker and titrated with $\frac{N}{10}$ NaOH, the colouring matter in most wines acting as an effective indicator. The acidity is returned in terms of tartaric acid.

b. Volatile acidity.

This may be determined in two ways :—

- (1) 25 c.c. of the wine are diluted with 200 c.c. of distilled water and distilled until only about 20 c.c. are left in the retort.

The distillate is then titrated with $\frac{N}{10}$ NaOH and the acidity returned in terms of acetic acid.

- (2) 25 c.c. of the wine are evaporated over a water bath almost to dryness, the residue is dissolved in distilled water and titrated as before. The difference between the total acidity and that now found will represent the volatile acidity.

TABLE (AFTER DUPRÉ) SHOWING THE PROPORTIONS OF THE ABOVE CONSTITUENTS IN A FEW WINES (grammes per cent).

	Alcohol	Fixed acidity	Volatile acidity	Total acidity	Dry residue
Hock . .	9.56	.348	.057	.420	1.86
Claret . .	8.53	.424	.147	.608	2.14
Sherry . .	17.20	.270	.153	.461	4.20
Madeira . .	17.75	.326	.168	.536	4.34
Port . . .	18.56	.308	.084	.413	7.55
Champagne .	9.22	—	—	.580	11.20

Some wines contain free sugar, whilst others, such as some of the Spanish wines, are “fortified” by the addition of alcohol.

ANALYSIS OF BEER

THE determinations required in the case of beer are alcohol, fixed, volatile and total acidity, and solid residue, and the operations are performed exactly as in the case of wine.

The fixed acidity is returned in terms of lactic acid, so that 1 c.c. $\frac{N}{10}$ NaOH = 0.009 gramme lactic acid.

The best test for the bitter used, whether hop or other, is by means of the taste.

It is well known that in England the beer is brewed with what is termed a top yeast, whilst the well-known Lager beers are brewed with a bottom yeast. In consequence, there is a distinct difference in the amount of alcohol formed. In English beers the alcohol forms 4% to 6%, in German beers from 2% to 5%.

The acidity is fairly constant, about 0.16%, and the residues vary from 2.5 to 15%.

Arsenic in Beer

During the winter of 1900-1901 there was a large outbreak of arsenical poisoning among beer-drinkers, chiefly in the northern parts of England. The beer that gave rise to this poisoning had been manufactured, not from malt, but from invert sugar, which was prepared by the action of dilute sulphuric acid on rice and other starches. Dilute commercial sulphuric acid may contain arsenic, derived from the iron pyrites used in its manufacture, and some invert sugars, made by this process, yield as much as two grains of arsenious acid to the pound. Some of the beers analysed during the outbreak of poisoning showed as much as a grain per gallon of arsenious acid, which is one hundred times the maximum allowed by the Royal Commission on Arsenical Poisoning.

Test for Arsenic

The most convenient test for this metal is that of Reinsch. Some of the beer is placed in a beaker, and acidulated by dilute pure hydrochloric acid. A small piece of bright arsenic-free copper foil is suspended in the liquid, and the whole is then boiled for half an hour and allowed to cool. If the copper foil is unaffected, no arsenic is present: if, on the other hand, there is a grey or black deposit, then arsenic (or antimony) is probably contained in the beer. The copper foil is then washed with water, dried, and placed in a test-tube over the mouth of which is a cover-glass. The tube is heated gently over a Bunsen burner, when the arsenic is oxidized and sublimes and condenses on the cool part of the tube and on the cover-glass. Upon examining the deposit under the microscope the tetrahedral or octahedral crystals of arsenious oxide are seen. If the deposit on the copper is antimony, the microscopic examination shows only an amorphous deposit.

With suitable and obvious modifications this test of Reinsch for arsenic may be applied to the detection of the metal in food, artificial flowers, wall-papers and other substances in which the presence of arsenic is suspected.

VINEGAR

VINEGAR is the well-known condiment, the essential ingredient of which is acetic acid.

Two points are important concerning vinegar. First, that it shall contain at least 3% of acetic acid, and secondly, that it shall not contain more than the merest traces of free mineral acid.

Estimation of the Acetic Acid

The vinegar may be titrated directly with $\frac{N}{10}$ NaOH, using phenolphthalein as indicator. 10 c.c. of the vinegar are diluted with an equal quantity of distilled water before titrating. The acetic acid is calculated directly from the soda : 1 c.c. $\frac{N}{10}$ NaOH = 0.006 gramme acetic acid.

Detection of Free Mineral Acid

Sulphuric acid is the acid most commonly found, although hydrochloric acid has been found. The detection of free acid is easily determined by placing a few drops of vinegar and a drop of a watery solution of methyl violet upon a white slab, the two fluids being separate. With a glass rod, a portion of the methyl violet is brought into contact with the vinegar. If no free mineral acid is present the colour remains. If only a trace of free acid is present the violet changes to blue, and if more than 1% is present a distinctly green colour develops.

A further test consists in testing the reaction of the ash. If a pure sample of vinegar be incinerated the ash will be alkaline, owing to the fact that the organic salts are converted into carbonates upon heating. If free mineral acid is present the ash will be less alkaline, or even neutral if more than traces are present.

LEMON JUICE AND LIME JUICE

These juices are always kept on board ship, and the Board of Trade standards are that the juice shall have a specific gravity of at least 1.030, and shall have an acidity equal to 30 grains per ounce of citric acid (6.8%).

The specific gravity is determined in the manner previously described.

The acidity, which is due to the presence of citric and malic acids, is determined by titrating with $\frac{N}{10}$ NaOH (1 c.c. $\frac{N}{10}$ NaOH = 0.006 gramme citric acid), and returned in terms of citric acid.

Free mineral acids are determined as in the case of vinegar.

ANALYSIS OF AIR

ESTIMATION OF OXYGEN IN AIR

Apparatus, etc.

1. HEMPEL's gas-absorption bulbs.
2. Two graduated cylinders, one furnished with a fine nozzle and glass tap, and bent at right angles at the other end, and the second one bent at right angles at one end and funnel-shaped and open at the other, the two bent ends fitting in wooden supports and connected together with indiarubber tubing, 2 ft. The rubber tubing should not connect the two tubes directly, but should have a piece of glass tubing inserted about the centre.
3. An alkaline solution of pyrogallic acid.

The Process

1. Fill the Hempel bulbs with the pyrogallic acid so that the lower bulb is full and the level of the pyrogallic acid in the U-shaped capillary tube at a certain point, which is recorded by making a pencil mark on the white enamel behind. Attach short lengths of rubber tubing to the open ends, and clamp.
2. Place the two cylinders *A* (the one provided with a stop-cock and nozzle) and *B* (the levelling tube) on the table and pour water into *B* until each tube is about half full. Now raise *B*. The air in *A* will be expelled as the level of the water rises. When all the air is expelled close the tap.
3. The apparatus being in the place whose air is to be examined, the tube *B* is lowered and the stop-cock of *A* opened. The air will enter into *A*. When about 50 c.c. have entered, the stop-cock is closed. *B* is now raised or lowered as required to bring the water in the tubes to the same level, and the quantity of air noted.
4. The capillary tube of the absorption bulb is connected with the nozzle of *A* by the short indiarubber tube and the clamp undone.

5. The stop-cock of *A* is now opened and the tube *B* is raised. The air is by this means driven over into the absorption bulb. The stop-cock is now closed. The bulb may be carefully disconnected after clamping the tube, and shaken gently.

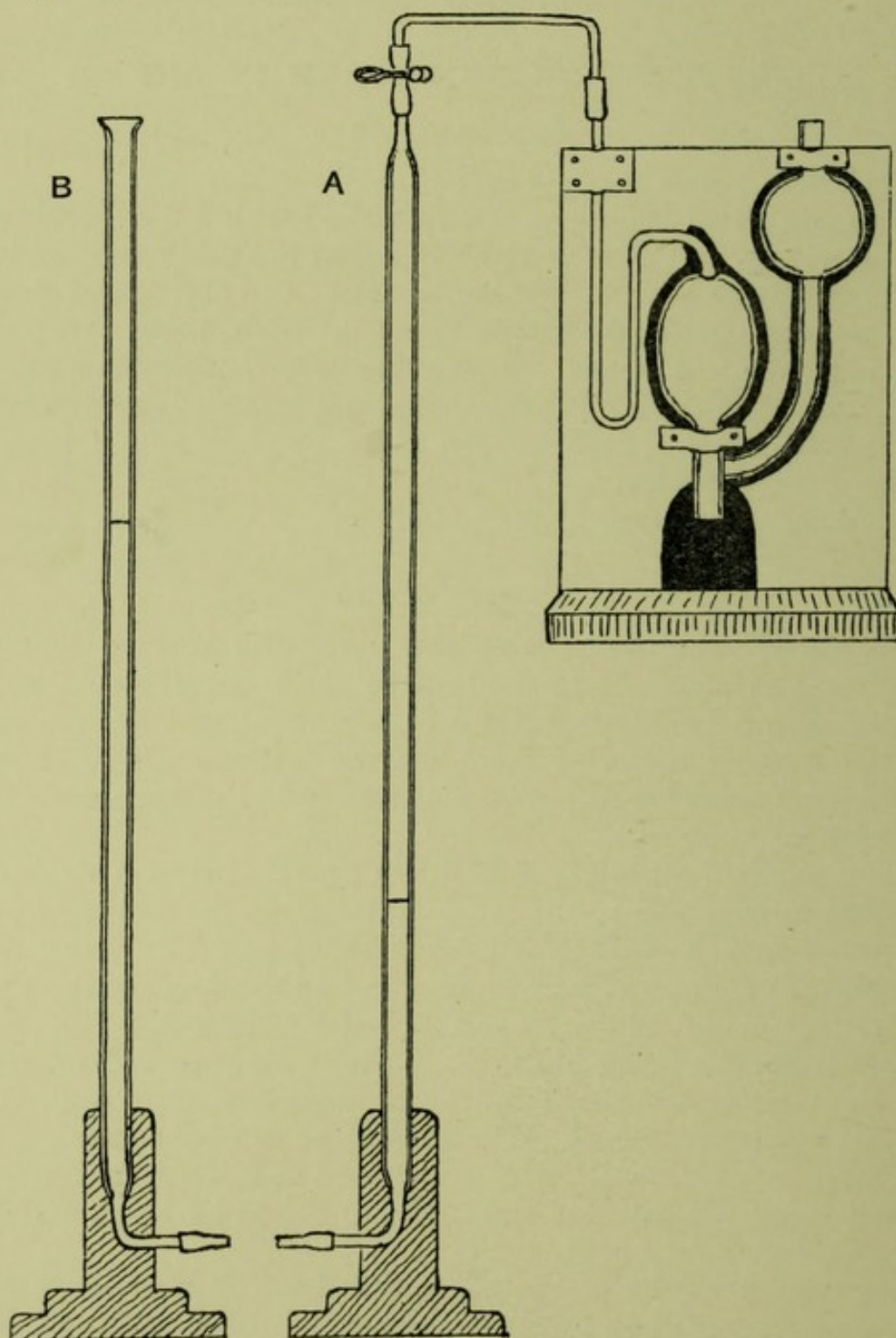


FIG. I. HEMPEL'S BULB

6. After about 15 minutes the tube *A* is again connected, the stop-cock opened, and the tube *B* lowered until the level of the pyrogallic acid in the capillary tube is the same as before the operation.

7. The tube *B* is again adjusted so that the level of the water in *A* and *B* is the same, and the quantity of air in *A* is noted.

The difference between the first and second readings will give the amount of oxygen absorbed, and this difference multiplied by 100 and divided by the original bulk will give the percentage of oxygen in the air.

Expired air may be examined in the same manner. In order to collect the expired air, the tube *A* should be filled with water by raising *A* as before, and the air may be simply blown down the nozzle from the mouth.

This method is not sufficiently delicate for the estimation of CO_2 in ordinary air, and other methods have to be adopted. These are described below.

ESTIMATION OF CARBON DIOXIDE

1. PETTENKOFER'S METHOD

Apparatus, etc.

1. A large bottle.
2. A 50 c.c. and a 25 c.c. pipette.
3. A 50 c.c. burette.
4. A small Erlenmeyer flask.
5. Standard oxalic acid, 1 c.c. = 0.5 c.c. CO_2 .
6. Baryta water.
7. Solution of methyl orange or phenolphthalein.

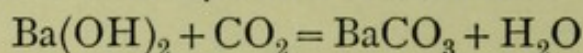
The Process

1. The large jar must be accurately gauged by filling with water to the top and inserting the stopper, and then measuring the amount of water.
2. To fill the jar with the air, first fill it with water and empty the water in the room, the air of which is to be sampled.
3. Add 50 c.c. of baryta water by means of the pipette and replace the stopper. Shake up well and allow the jar to stand for about an hour, shaking from time to time.
4. Meanwhile, measure 25 c.c. of the baryta water into an Erlenmeyer flask, add a drop of methyl orange and titrate with the standard oxalic acid. Note the number of c.c. used.

5. When the baryta water has been in contact with the air for a sufficiently long time, remove 25 c.c. of the baryta water with the pipette and allow it to run into another Erlenmeyer flask.
6. Add a drop of methyl orange and titrate with the oxalic acid, noting the number of c.c. used.

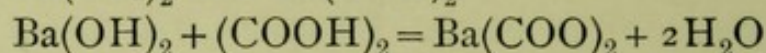
Explanation

The baryta water when in contact with the CO_2 absorbs it, and barium carbonate is formed, which is insoluble.



As BaCO_3 is an insoluble neutral salt, the alkalizing power of the $\text{Ba}(\text{OH})_2$ is diminished in proportion to the amount of the salt formed.

In titrating with the oxalic acid the BaCO_3 is unaffected by the weak acid, and the whole of the acid used is expended in converting the $\text{Ba}(\text{OH})_2$ into $\text{Ba}(\text{COO})_2$.



Therefore the difference between the quantity of oxalic acid used to neutralize the 25 c.c. of $\text{Ba}(\text{OH})_2$ which has not been in contact with the air, and that required to neutralize the $\text{Ba}(\text{OH})_2$ which has been in contact with the air, will represent the amount of $\text{Ba}(\text{OH})_2$ converted into BaCO_3 .

But the oxalic acid was prepared so that 1 c.c. should be the equivalent of 0.5 c.c. of CO_2 ; therefore each c.c. of difference = 0.5 c.c. of CO_2 which has converted the $\text{Ba}(\text{OH})_2$ into BaCO_3 .

EXAMPLE

The jar was found to contain 3950 c.c.

50 c.c. of baryta water were run into the jar, therefore the air experimented upon was $3950 - 50 = 3900$ c.c.

On titrating the $\text{Ba}(\text{OH})_2$ it was found that 25 c.c. of the fresh solution required 22.50 c.c. of standard acid to neutralize.

The $\text{Ba}(\text{OH})_2$ from the jar took 19.35 c.c.

25 c.c. of original $\text{Ba}(\text{OH})_2$.	.	= 22.50 c.c. acid
25 c.c. of used $\text{Ba}(\text{OH})_2$.	.	= 19.35 " "

Difference of acid used . . . = 3.15 " "

But 1 c.c. acid = 0.5 c.c. CO_2 at 0°C . and 760 mm. of mercury,
 $\therefore \text{CO}_2$ taken up by 25 c.c. of $\text{Ba}(\text{OH})_2 = 3.15$ c.c.

As 50 c.c. were used, the CO_2 absorbed by the $\text{Ba}(\text{OH})_2 = 3.15$ c.c.

Now this 3.15 c.c. were present in 3900 c.c.

\therefore there were $3.15 \times \frac{1000}{3900}$ c.c. of $\text{CO}_2\% = 0.80\%$

Notes

In manipulating the jar for emptying, filling, etc., care must be taken not to handle it with the naked hands, as by so doing the sides of the jar will get heated, and the volume will not be correctly obtained, since the air will expand.

The baryta water must be run into the jar and removed with all expedition, and care must be taken not to breathe into the jar during any of the manipulations. The same care must be taken with the flasks during titration, etc.

It is not necessary to wait for the baryta water to clear before pipetting it out for titration, since, as has been said, dilute oxalic acid does not decompose the barium carbonate.

In calculating the CO_2 present per thousand in the jar, in the above example, no notice has been taken of the fact that 1 c.c. of the acid corresponds to 1 c.c. of CO_2 at 0°C . and 760 mm. of mercury, and not to 1 c.c. at the room temperature and pressure.

To correct for this it is only necessary to multiply the 3.15 by $\frac{760}{P}$, and this by $\frac{T}{273}$, when P is the height of the barometer, and T the absolute temperature of the room.

Standard Oxalic Acid

(1 c.c. = 0.5 c.c. CO_2 at N.T.P.)

It is convenient to have the oxalic acid of such a strength that 1 c.c. of it neutralizes as much $\text{Ba}(\text{OH})_2$ or $\text{Ca}(\text{OH})_2$ as 0.5 c.c. of CO_2 . Now 1 c.c. of CO_2 weighs 22×0.0000895 grammes = 0.001969 gramme. Oxalic acid crystallizes with 2 molecules of H_2O , its molecular weight is therefore 126, for the formula will be $(\text{COOH})_2 + 2\text{H}_2\text{O}$. Therefore in 1 c.c. of the standard solution there must be

$$\frac{1}{2} \text{ of } \frac{126}{44} \text{ of } \frac{0.001969}{1} \text{ grammes} = 0.00281925 \text{ gramme.}$$

For convenience a stronger solution than this is made by dissolving 28.19 grammes of crystallized oxalic acid in a litre of freshly boiled distilled water. Each c.c. of this will be equivalent to

5 c.c. of CO_2 ; when the acid is required for use, 10 c.c. of the strong acid are diluted with 90 c.c. of distilled water; 1 c.c. of the weak acid then equals 0.5 c.c. CO_2 .

2. HALDANE'S METHOD

Haldane has devised a small and portable gas analysis apparatus for the estimation of carbon dioxide in air. The advantages claimed by it are that the estimation can be completed in a few minutes, only a small volume of air is required, and the apparatus is easily portable: the disadvantages are the relatively large cost, and the difficulty, to the beginner, of manipulation. The results obtained by it are sufficiently accurate for practical purposes; the principle on which the analyses are made being the absorption of the CO_2 by means of potash. Full details are given in text books larger than this, and are supplied also with the apparatus itself.

Detection of Carbon Monoxide in Air

If CO is present in the air or any mixture of gases in any considerable quantity it may be absorbed by a solution of cuprous chloride; if it is only present in minute percentage this method is quite useless.

If a sample of the suspected air be shaken up with a few c.c. of a 1% solution of blood, the latter acquires a pink colour which is quite different from the colour of the normal blood.

Blood thus treated with carbon monoxide gives, in weak solution, a characteristic spectrum, showing two well-marked bands with sharp edges in the yellow and green parts of the spectrum: when the blood is treated with dilute ammonium sulphide these bands persist, in contradistinction to oxy-hæmoglobin, which loses the bands after such reduction. The persistence of these two bands after reduction with $(\text{NH}_4)_2\text{S}$ means that carbon-monoxide-hæmoglobin is present, and that CO was present in the air that was tested.

Estimation of Carbon Monoxide in Air

The following account is that of Haldane, who elaborated the method:—

Estimation of the Degree of Saturation of Blood with CO

“When only a rough quantitative estimate of the percentage

saturation is required, as in ordinary post-mortem examinations or in examining the blood of a patient suffering from gas poisoning, all that is necessary is to prepare in three test-tubes of even size (1) a solution of normal blood well diluted; (2) some of the same solution saturated with coal gas; and (3) a solution of the suspected blood diluted to the same *depth* of colour as the other two solutions. One can then tell roughly by the relative pinkness of the suspected blood to what extent it is saturated."

To measure accurately the extent to which blood is saturated with CO he devised the following method:—

"A solution of about 1 of normal blood to 100 of water is made; also a solution of carmine dissolved with the help of a little ammonia, and diluted till its *depth* of tint is about the same as that of the blood solution. Two test-tubes of equal diameter (about half an inch) are then selected. Into one of these 5 c.c. of the blood solution are measured with a pipette; into the other about an equal quantity is poured. Ordinary lighting gas is then allowed to blow into the second test-tube through a piece of rubber tubing for a few seconds. The test-tube is then quickly closed with the thumb before the gas has had time to escape, and the blood solution thoroughly shaken up with the gas for a few seconds. The hæmoglobin is thus completely saturated with carbonic oxide, and the solution has now the characteristic pink tint. The carmine solution, which has a still pinker tint, is now added from a burette to the 5 c.c. of normal blood solution in the other test-tube until the tints are the same in the two test-tubes. Not only, however, must the tints be equal in *quality*, but they must also be sensibly equal in *depth*. If the carmine solution is too strong or too weak, the latter will not be the case, and the solution must be diluted or made stronger accordingly. It is usually easiest to make the carmine a little too strong at first, so that on adding both carmine solution and water equality can be established. From the amount of water which required to be added it is easy to calculate the extent to which the original carmine solution needs to be diluted. The solutions are now ready for use, and the actual analysis is made as follows: 5 c.c. of the solution of normal blood are measured into one of the test-tubes, and a drop of the suspected blood placed in the other test-tube and cautiously diluted with water till its *depth* of tint is about equal to that of the normal solution. If carbonic oxide be present in the hæmoglobin, a difference of quality in the tints of the two solutions will now be clearly perceptible. Carmine

solution is then added from the burette to the normal blood, and water, if necessary, to the abnormal blood, till the tints are equal in both quality and depth. The carmine is added by about 0.2 c.c. at a time, the points being noted at which there is just too little and just too much carmine, and the mean being taken. The solution of abnormal blood is then saturated with coal gas, and the addition of carmine to the other test-tube continued until equality is again established, and the amount of carmine noted. The percentage saturation with carbonic oxide of the abnormal blood can now be easily calculated, since we know how much carmine solution its saturation represented as compared with what complete saturation represented.

"The method of calculation is illustrated by the following example: To 5 c.c. of normal blood solution, 2.2 c.c. of carmine is required to be added to produce the tint of the blood under examination, and 6.2 c.c. to produce the tint of the same blood fully saturated. In the former case the carmine was in the proportion of 2.2 in 7.2 and in the latter of 6.2 in 11.2. The percentage saturation (x) of the hæmoglobin with carbonic oxide is thus given by the following proportion sum:—

$$\frac{6.2}{11.2} : \frac{2.2}{7.2} :: 100 : x$$

x is therefore = 55.2. As the compound of carbonic oxide and hæmoglobin is to a slight extent dissociated when the blood is diluted with water, the value found is a little too low. The corrections needed are as follows: Add 0.5 if 30% saturation be found, 1.1 if 50%, 1.6 if 60%, 2.6 if 70%, 4.4 if 80%, 10.0 if 90%. Thus, in the above example, we must add 1.3, so that the true saturation is 56.5%. In comparing the tints the test-tubes should be held up against the light from a window, but bright light should be avoided as much as possible, as it increases the dissociation. Failing daylight, an incandescent burner with a chimney of blue glass and an opal globe may be used as the source of light.

"Hæmoglobin brought into intimate contact with air containing 0.07% of CO will finally reach a state of equilibrium in which it is saturated to an equal extent with CO and oxygen. If the percentage of CO or oxygen in the air be increased or diminished, there will be an exactly corresponding increase or diminution of the *relative* share of the hæmoglobin which either gas obtains. Air containing $2 \times 0.07 = 0.14\%$ of CO will, for instance, produce

two-thirds saturation with CO, and one-third saturation with oxygen, and so on. In the living body the proportion of CO taken by the hæmoglobin from respired air containing a given percentage of CO is not so large as outside the body, about 0.1% of CO in the air breathed being necessary to produce half saturation of the hæmoglobin. The general law of absorption is, however, much the same, and it follows that there is a certain maximum of saturation for each percentage. With less than 0.05% of CO in the air this maximum does not exceed 33% saturation, and the corresponding symptoms are scarcely appreciable, except on muscular exertion. With more than about 0.2% the maximum exceeds 60% saturation.

"The detection and determination of small percentages of CO in air was formerly a matter of great, and often almost insuperable, difficulty. I have recently, however, introduced a simple, and I think very satisfactory, method, depending on the already described action of CO on blood solution in presence of air. The sample of air is collected in a clean and dry bottle of about 4 oz. capacity. The cork of the bottle is removed in the laboratory under a 0.5% solution of blood, and about 5 c.c. of the air allowed to bubble out, a corresponding volume of the blood solution entering. The cork is then replaced, covered with a cloth to keep off the light, and shaken continuously for about ten minutes, when the hæmoglobin will have reached the point of saturation corresponding to the percentage of CO present. The solution is then poured out into a test-tube, and the saturation determined with carmine solution in the manner described above. It is evident that as in each case the saturation found corresponds to a definite percentage of CO in the air, it is easy to calculate this percentage. If p be the percentage required, and s the percentage saturation found, p is calculated from the following formula:—

$$p = \frac{s \times 0.07}{100 - s}$$

Thus, if $s = 60$, p is 0.105. This method may also be used for the direct determination of carbonic oxide in lighting gas. The latter must, however, be first diluted to $\frac{1}{100}$ (or with carburetted water-gas to $\frac{1}{400}$) with air. As it is quite easy to make this dilution with perfect accuracy, the method is an exact one, and is not only rapid, but avoids the difficulties and sources of error connected with the ordinary method of determination by cuprous chloride, or by explosion."

Ozone

Ozone (O_3), an allotropic modification of oxygen, is found in the air in the neighbourhood of the sea and after electric discharges.

In order to detect its presence in the atmosphere, a piece of blotting-paper is saturated with a solution of KI and starch, and exposed to a current of air for from six to twenty-four hours, shaded meanwhile from the sun. If ozone is present the paper will have acquired a blue tinge from the liberation of iodine from the potassium iodide, and subsequent combination of the iodine with the starch.

In the neighbourhood of chemical works the above test is not available, since other gases, such as chlorine, will cause the appearance of the blue colour. Instead, two strips of neutral litmus paper are taken, one of which has been steeped in KI solution, and exposed to the air. If ozone is present the litmus paper soaked in KI will be turned blue from the conversion of the KI into K_2O by the ozone. The control litmus paper is in order to ensure the absence of ammonia.

Noxious Gases in Air

The air in the neighbourhood of chemical and other works frequently contains traces of chlorine, hydrochloric acid, sulphur dioxide, and various other gases. These gases when concentrated are certainly harmful; but when diluted with air, as they are usually found, their danger to life is doubtful. The student for D.P.H. examinations, however, is expected to be able to identify various gases, which are generally supplied to him in the undiluted condition. The gases that may be set at such examinations are included in the following list:—

Acid gases.

HCl.
HNO₃.
N₂O₃, etc.
Cl₂.
SO₂.
CO₂.

Alkaline gases.

NH₃.
(NH₄)₂S.

Neutral gases.

H₂S.
CS₂.
CO.

Method of Procedure

1. Take the reaction with litmus paper which has been made slightly moist. This will give an indication whether the gas is acid, alkaline, or neutral.
2. Smell the gas. Chlorine and hydrochloric acid gas have a characteristic odour. So has sulphur dioxide. Ammonia and ammonium sulphide are easily distinguished, the latter giving, besides the smell of ammonia, the unpleasant odour of rotten eggs. Sulphuretted hydrogen also smells like these; and carbon disulphide has the odour of concentrated bad cabbages. The oxides of nitrogen have their own particular smell, reminiscent of strong nitric acid. Carbon monoxide and carbon dioxide have no odour.

With these aids the student will be enabled to diagnose that the gas he is examining is, at the most, one of two or three. He should now apply confirmatory tests, as follows:—

Dissolve the gas by shaking in 10 c.c. of water, and test the solution.

HCl. AgNO_3 gives a white precipitate insoluble in HNO_3 , but soluble in NH_4OH .

HNO_3 . Brucine test.

N_2O_3 (now HNO_2). Metaphenylene-diamine test.

Cl_2 . Bleaches litmus paper. Moist KI paper is blackened by the liberation of free iodine.

SO_2 . Characteristic smell. AgNO_3 gives a white precipitate soluble in HNO_3 .

CO_2 . Lime water or $\text{Ba}(\text{OH})_2$ gives turbidity.

NH_3 . Nessler's reagent gives a yellow-brown colour.

$(\text{NH}_4)_2\text{S}$. Odour characteristic. Sodium nitro-prusside gives a violet colour.

H_2S . Lead acetate paper or solution is darkened.

CS_2 . On burning, sulphur is deposited.

CO. Characteristic colour and spectrum when shaken with a dilute blood solution.

ANALYSIS OF SOIL

THE chemical and physical examinations of the soil are attended by many and great difficulties, and the training necessary to become an expert in the subject is both long and laborious. Fortunately the examinations which develop upon the hygienist are comparatively simple.

DETERMINATION OF THE SIZE OF THE PARTICLES

A series of sieves is taken, having meshes of 2 mm., 1 mm., and 0.5 mm. respectively.

100 grammes of air-dried soil is taken and broken as finely as possible between the finger and thumb. The large pebbles, sticks, roots, etc., are removed by hand and weighed. The remainder is next transferred to the 2 mm. sieve. After as much of the soil is through as will pass, the remainder is again rubbed between the finger and thumb in order to break up any cohering masses. The amount left on this sieve is then weighed. In a similar manner the amount left upon the other sieves, and the amount which passes the 0.5 mm. sieve is weighed. The result is then tabulated as follows :—

1. Coarse pebbles, etc., removed by hand.
2. Pebbles and coarse sand not passing a 2 mm. sieve.
3. Sand not passing a 1 mm. sieve.
4. Fine sand not passing a 0.5 mm. sieve.
5. Fine earth passing a 0.5 mm. sieve.

DETERMINATION OF THE MOISTURE

Since the moisture-containing property of the soil is chiefly possessed by that portion of the soil which passes a 2 mm. sieve, 5 grammes of such air-dried soil are carefully weighed in a tared dish. The dish is then placed in a water oven and heated for five hours. It is then transferred to a desiccator, allowed to cool, and weighed. The heating, cooling, and weighing are repeated at intervals of two hours, until the weight is found to be constant. The loss in weight then represents the moisture in 5 grammes.

DETERMINATION OF THE POROSITY OF A SOIL

The porosity of a soil depends upon the volume of the solid particles as compared with the volume of the interstitial spaces. Three factors affect the porosity: (1) the state of divisibility or the number of particles per unit volume; (2) the nature and arrangement of these particles; and (3) the interstitial space.

The porosity is most easily determined by finding the real and apparent specific gravity of the soil in question, and dividing the latter by the former.

The real specific gravity is determined by means of a pyknometer having a capacity of 25 or 50 c.c. 10 grammes of soil dried at 100° C. to a constant weight are boiled for a time with a few c.c. of distilled water in order to remove any air, and poured into the pyknometer. The vessel is rinsed with distilled water, so that all the soil is transferred to the pyknometer. After cooling to the requisite temperature, 15° C., distilled water is added to the mark, and the whole weighed. The weight of the pyknometer and the pyknometer filled to the mark with water being known, the weight of the water displaced by the 10 grammes of soil is easily obtained.

Suppose this to be 4.791.

The sp. gr. of the soil is then $\frac{10}{5.106} = 2.08$.

The apparent specific gravity is obtained in the following manner:—

An open cylinder holding 1 litre is taken and filled—small quantities at a time—with the soil. As each portion is placed in the cylinder, the bottom is struck fairly hard with the palm of the hand. When the cylinder is full it is covered with a glass plate and weighed. The weight of the cylinder and plate is deducted, and the apparent specific gravity thus obtained.

The real sp. gr. of a sample of soil was found to be 2.64, and the apparent sp. gr. 1.28. The porosity is therefore $\frac{1.28}{2.64} \times 100 = 48.4\%$.

Schübler gives the weights of different kinds of soil:—

	lbs. per cu. ft.		lbs. per cu. ft.
Sand	110	Heavy clay	75
Sand and clay	96	Vegetable mould	78
Common arable soil	80-90	Peat	30-50

The specific gravity thus decreases as the amount of humus increases.

ESTIMATION OF CLAY AND SAND

The constituents of soil are spoken of as sand and clay, the sand being the coarser particles which sink rapidly in water, the clay being the very fine particles, consisting chiefly of silicate of alumina, which remain suspended in still water for a considerable time. This is only a rough division, because in any sample of soil, every grade can, by appropriate methods, be found between particles 3 mm. in diameter and particles 0.001 mm. in diameter.

In order to estimate the clay and sand, 10 grammes of air-dried soil are taken and placed in a beaker which holds about 200 c.c. The soil is first moistened with distilled water containing 0.01% of NH_4Cl , and about 50 c.c. or 100 c.c. of the distilled water are added and the soil well stirred. The soil is allowed to settle for five minutes and the supernatant fluid poured into a large clean cylinder. 50 c.c. or 100 c.c. more of the water are added, the soil well mixed, and again allowed to settle for five minutes, when the supernatant fluid is poured off. This is repeated until the supernatant fluid is quite clear.

The sand remaining in the beaker is transferred on to a filter and well washed with distilled water, dried, weighed and returned as sand.

The fluid in the cylinder is allowed to stand for from 12 to 24 hours, when the clay will have settled at the bottom. The whole of the upper part of the fluid is filtered through filter-paper without disturbing the sediment. When only a thin layer of water is left, the clay is stirred up and transferred to the filter-paper. The cylinder is well rinsed with distilled water and the washings are poured on to the filter-paper. The clay is then well washed (at this stage filtration is so slow that frequently two days are required to complete the washing), dried, weighed, and returned as clay.

Good loamy soil often contains from 10–15% of clay. Stiff soils contain from 20–30%. Sandy soil contains only 1 or 2%, and brick clay or kaolin contains 80–95%.

DETERMINATION OF THE SPECIFIC HEAT OF SOILS

The specific heat of any substance is the relation between the amount of heat required to raise a given mass of the substance through a given number of degrees, and the amount of heat required to raise the same mass of water through the same number of degrees.

The principle involved in the determination is that a certain mass of the soil heated, say, to boiling point when added to water at a certain temperature will raise the temperature of the water a certain number of degrees, whereas the same mass of boiling water will raise the temperature a different number of degrees. As the operation is a delicate one and necessitates the use of a sensitive calorimeter it hardly lies within the province of this book. Suffice it to say that there is a very considerable difference in the specific heats of various soils.

The specific heat varies from 0.19 to 0.51, the latter being a peaty soil. Speaking generally, the specific heat increases with an increase of humus in the soil.

ESTIMATION OF THE ORGANIC MATTER

An approximate estimation of the organic matter in soil may be obtained by taking 10 grammes of air-dried soil and heating it in a platinum dish at 110° C. until a constant weight is obtained. The dish is then transferred to an Argand burner and the soil oxidized at a low red heat. When the oxidation is complete, the dish is transferred to a desiccator, allowed to cool and weighed. The loss in weight gives approximately the amount of organic matter.

ESTIMATION OF NITRATES IN SOIL

In order to estimate the nitrates and nitrites in soil, the sample should be spread in a thin layer in an oven having a temperature of 50° – 60° C. in order to prevent any further nitrification.

After the soil is dry, 1000 grammes are finely powdered, weighed, and placed in a large flask. 2000 c.c. of distilled water are then added, well mixed, and allowed to stand (with frequent shaking) for 48 hours. 1000 c.c. are then filtered. A small quantity of Na_2CO_3 is added to the filtrate, which is then evaporated to about 100 c.c.

The nitrates are then estimated by the phenol-sulphonic method.

ANALYSIS OF GROUND AIR

IN order to collect ground air for analysis a convenient method is to have a hollow tube furnished with a steel cone. The tube is provided at its lower end with perforations, and when it has been driven into the ground to the required depth, the upper end is connected with an aspirator full of water; the water is allowed to flow slowly out. When the necessary amount of gas is collected, the apparatus is transferred to the laboratory and the gas analysed.

Ground air contains about the normal amount of nitrogen. The CO_2 varies from 1% to 8%, and the O_2 is correspondingly decreased. From time to time NH_3 , H_2S , CH_4 , etc., are found. The methods for the detection of the first two will suggest themselves to the student. CH_4 can only be estimated in a proper gas apparatus, which is somewhat expensive, and which requires special practice to use.

DISINFECTANTS

CERTAIN disinfectants and antiseptics (boric acid, formalin, and salicylic acid) have already been discussed in the chapter on milk analysis: it remains now to consider other disinfectants that are not usually added to food as preservatives, but which are employed in public health work in connection with the control of infection. A bacteriological standard is here obviously of more value than a chemical analysis; and for the consideration of the Rideal-Walker method of standardizing disinfectants the student is referred to text-books on bacteriology.

Occasionally, however, the D.P.H. candidate is asked to identify some disinfectant; and possibly to determine whether the sample submitted to him has been adulterated by some inert substance: in other words, he is required to estimate the quantity of the disinfectant present in the sample.

BLEACHING POWDER

ESTIMATION OF AVAILABLE CHLORINE

Apparatus, etc., required

1. A flask to hold 1 litre.
2. A burette graduated in 0.1 c.c.
3. A solution of KI in water.
4. Freshly prepared starch solution.
5. $\frac{N}{10}$ $\text{Na}_2\text{S}_2\text{O}_3 \cdot 5\text{H}_2\text{O}$ (24.8 grammes to the litre).
6. A porcelain dish.

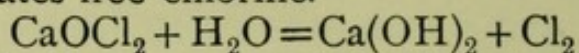
The Process

1. Weigh out 10 grammes of the bleaching powder and transfer to the porcelain dish.
2. Add small quantities of distilled water and mix with the bleaching powder until it is thoroughly suspended in the liquid.

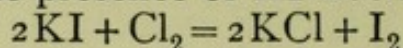
3. Transfer the liquid to the litre flask and wash out the dish with more distilled water. Transfer this to the litre flask and make up to 1 litre with distilled water.
4. Shake the flask thoroughly.
5. Measure 20 c.c. of the solution into a dish and dilute with about 50 c.c. of distilled water.
6. Add a drop of acetic acid, and excess of KI to the bleaching powder solution in the dish. Free iodine will be liberated in proportion to the amount of available chlorine.
7. Estimate the iodine by means of the decinormal thiosulphate solution, judging the end point more accurately by the addition of some of the starch solution.

Explanation

Bleaching powder consists of a number of compounds; the one, however, which gives rise to the available chlorine probably has the formula CaOCl_2 . This in contact with water and a dilute acid liberates free chlorine.



The chlorine in the presence of KI liberates free iodine.



This iodine, and so the chlorine, is estimated directly by the decinormal thiosulphate solution.

Notes

The thiosulphate solution should be freshly prepared.

A good bleaching powder will give as much as 33% of available chlorine.

CARBOLIC ACID (PHENOL)

Qualitative Tests

1. Ferric chloride gives a deep violet colour with a solution of phenol.
2. Bromine water gives with phenol a white crystalline precipitate of tri-bromo-phenol.
3. KNO_2 and strong H_2SO_4 gives a brown colour, changing to green and blue.

ESTIMATION OF PHENOL

TRI-BROMO-PHENOL METHOD

Apparatus, etc., required

1. Two stoppered flasks, each of about 150 c.c. capacity.
2. 500 c.c. flask.

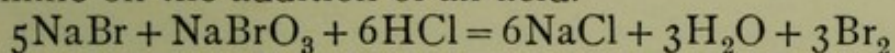
3. Graduated pipettes—25 c.c. and 5 c.c.
4. Solution of KI in water.
5. Solution of sodium thiosulphate (10 grammes to the litre).
6. Standard solution of NaBr and NaBrO₃ (1 c.c. = 0.0012638 grammes of phenol).
7. Starch solution.
8. Graduated burette.

The Process

1. Weigh out 1 gramme of the sample phenol and dissolve in 500 c.c. of distilled water, to which a trace of NaOH has been added to facilitate the solution.
2. Take 25 c.c. of this solution and transfer to one of the stoppered flasks. To the same flask add 25 c.c. of the standard bromine solution and 5 c.c. of pure HCl. Bromine will be liberated.
3. Stopper the flask.
4. To the control flask add 25 c.c. of the standard bromine solution and 5 c.c. of pure HCl. Bromine will be liberated. Stopper the flask.
5. To each flask add excess of the KI solution. The free bromine will combine with the KI and iodine will be liberated.
6. From the burette run the thiosulphate into the control flask until the colour has disappeared. Add a little starch solution as in the estimation of available chlorine. A few c.c. of chloroform added to the contents of the flask sharpens the reaction. Note the number of c.c. of thiosulphate used.
7. Add thiosulphate solution similarly to the flask containing the sample phenol, until all the colour is discharged. Note the amount of thiosulphate solution used.

Explanation

The standard solution containing NaBr and NaBrO₃ liberates free bromine on the addition of an acid.



The free bromine thus liberated combines with the phenol present to form tri-bromo-phenol.

The amount of bromine used up by the phenol is represented by the difference in the amounts of thiosulphate solution required respectively by the control and by the sample solutions. Knowing the amount of bromine used up in converting the phenol

into tri-bromo-phenol, the percentage of phenol actually present in the sample can be easily determined.

EXAMPLE

The control flask required 20.3 c.c. of thiosulphate solution to decolourize its contents.

The flask containing the sample required 8.6 c.c. of thiosulphate solution.

∴ the phenol in the sample absorbed bromine corresponding to 20.3 – 8.6 c.c. of thiosulphate solution, or 11.9 c.c.

Now 20.3 c.c. of thiosulphate solution = 25 c.c. of the standard bromine solution

$$\therefore 11.9 \text{ c.c. of thiosulphate solution} = \frac{25 \times 11.9}{20.3}$$

$$= 14.65 \text{ c.c. of standard solution.}$$

But 1 c.c. of the standard solution = 0.0012638 grammes of phenol

∴ 14.65 c.c. of the standard solution = 0.01851 grammes of phenol.

∴ in 25 c.c. of the sample there were 0.01851 grammes of phenol.

∴ in 500 c.c. of the sample there were 0.37 grammes of phenol.

Therefore in the gramme of the sample of phenol there was present but little more than a third of pure phenol.

Notes

The flasks should be kept stoppered as much as possible, in order to prevent the escape of any bromine vapour.

The standard bromine solution has the following composition: Sodium bromide 8.0 grammes, sodium bromate 2.04 grammes. Distilled water to 1 litre. 1 c.c. of this solution is equivalent to 0.0012638 grammes of phenol.

Potassium or sodium permanganate is used sometimes as a disinfectant. It is easily recognized from its colour. When an acid solution of oxalic acid is added the colour disappears. The strength of a permanganate solution can be estimated by means of decinormal oxalic acid in the presence of H_2SO_4 .

Copper sulphate and zinc chloride and salts of iron are used occasionally for their disinfecting properties. The detection of

these metals has already been considered in the section devoted to water analysis.

Sulphites and sulphurous acid are not used on a large scale for disinfection except when SO_2 is used for fumigating rooms. The smell of SO_2 and of sulphurous acid is characteristic.

Salicylic acid, boric acid, and formalin have been referred to in the chapter on milk analysis.

Benzoic acid is too expensive to use on a large scale. It is occasionally found in foods. Fe_2Cl_6 gives a red precipitate. When heated with lime benzene is evolved.

Mercuric chloride and the other salts of mercury have all disinfectant properties. H_2S gives a black precipitate with mercuric solutions, and KOH gives a yellow precipitate of HgO . On placing copper foil in a solution of a mercury salt the mercury is deposited on the copper.

MICROSCOPY

THIS part of the book deals with the microscopical examination of food, clothing, parasites, water sediment, etc., and includes descriptions of all the microscopical material about which a D.P.H. candidate is likely to be questioned in his examination.

No pretence is made that this section is a complete treatise on such a vast subject as parasitology, and if the student wishes for more minute descriptions he is recommended to consult one of the many text-books on that science. For the ordinary D.P.H. candidate, however, the subject matter in this part of the book will be found amply sufficient.

It is almost impossible to follow a routine plan in the arrangement of this section ; but, as far as possible, the subjects have been kept together : for instance, the parasites of wheat are dealt with in connection with wheat, rather than with the other parasites.

FOOD

EXAMINATION OF STARCHES

METHODS OF MOUNTING

A. In Water

1. Clean a slide with a handkerchief and see that it is dry.
2. With a clean platinum loop remove a small quantity of the starch to be examined, and place it in the centre of the slide.
3. Take the slide in one hand, holding it by one end, and tap the opposite end against the desk, and remove all the starch which does not readily adhere to the slide.
4. By means of the sterilized loop place three or four loopfuls of water in the centre of the slide and mix the starch well.
5. Breathe on a clean coverslip and whilst the surface is moist with the condensed water, gently drop it—moist surface downwards—on to the mixture of starch and water.
6. If the water runs out round the coverslip remove the excess with a little filter-paper. The preparation is now ready to be examined.

B. In Dilute Iodine Solution

This method is a very useful one, as the concentric rings can very often be seen with great distinctness, even in specimens which only show them faintly when mounted in water.

The process of mounting is identical with that above described, a weak solution of iodine being used instead of water.

The solution of iodine recommended is

Gram's iodine	1 part.
Water	3 or 4 parts.

These specimens prepared in either of the above ways will only last as long as the water remains. In the warm laboratory they very soon dry. For this reason, they should be examined as soon as prepared, and if sketches are to be made, they should be made at once.

C. In Farrant's Solution

Proceed as in *A*, omitting, however, to breathe on the cover-slip.

Mounted in this way, starches will keep for two or three weeks.

The starch granules possess certain characteristic appearances either in their size, shape, concentric rings or hila. These appearances enable us to divide them into five groups.

It is not always easy to differentiate from one another starches in the same group; but not difficult to determine into which group a starch should be placed.

GROUP I

Wheat, Barley, Rye

The granules in this group are circular or oval in appearance, some being large and others small. They have no very apparent hilum and no concentric rings.

1. Wheat (*Triticum vulgare*).
2. Barley (*Hordeum vulgare*).
3. Rye (*Secale cereale*).

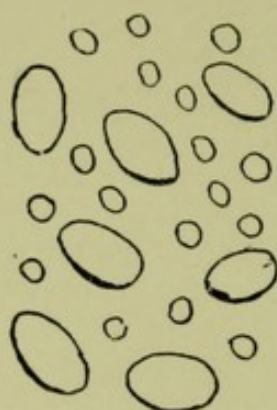


FIG. 2
WHEAT STARCH
X 200

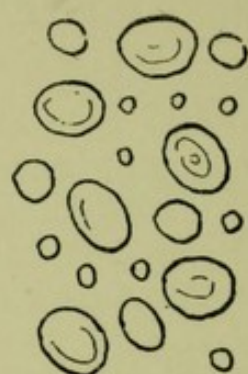


FIG. 3
BARLEY STARCH
X 200

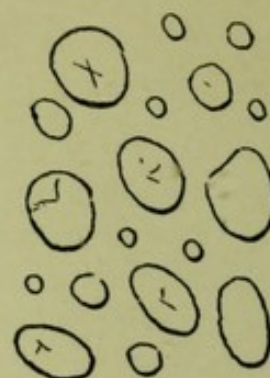


FIG. 4
RYE STARCH
X 200

1. The wheat granules are generally very perfect. They are chiefly of two sizes, large ones varying in shape from circular to oval, and very small ones. Sizes intermediate between these are rare.
2. The granules of barley resemble those of wheat, but (1) the large granules are more circular, (2) the number of intermediate sizes is proportionally much greater, and (3) slight indications of concentric rings may be seen.
3. The granules of rye resemble the two preceding ones, but (1) the large granules are larger than either wheat or barley, (2) the large, intermediate, and small granules are more nearly equal in number, (3) the granules are more frequently cracked and occasionally show a stellate hilum but no concentric rings.

GROUP II

Potato, Arrowroot

The granules of this group are large and oval, and show a distinct hilum and well-marked concentric rings.

1. Potato (*Solanum tuberosum*).
2. Arrowroot (*Maranta arundinacea*).

1. The potato granules have the hilum as a point at the narrow end.
2. The arrowroot granules, on the other hand, have a punctate or linear hilum at the broad end.

Note

Although it is not true that every potato granule has the hilum at the narrow end, or is even oviform; or that every



FIG. 5. POTATO STARCH
X 200



FIG. 6. ARROWROOT STARCH
X 200

arrowroot granule has the hilum at the broad end; the majority of the granules in any pure specimen agree with this description.

It would be obviously impossible to differentiate between one oval potato and one oval arrowroot granule.

GROUP III

Pea, Bean, Maize

The granules of this group are round or oval without any evident rings, but with linear or stellate hila.

1. Pea (*Pisum sativum*).
2. Bean (*Faba vulgaris*).
3. Maize (*Zea mays*).

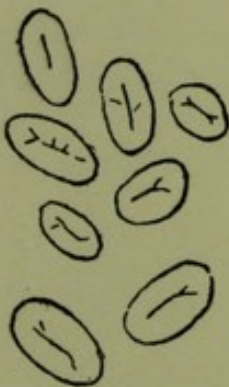


FIG. 7. PEA STARCH
X 200

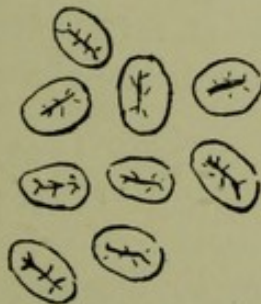


FIG. 8. BEAN STARCH
X 200

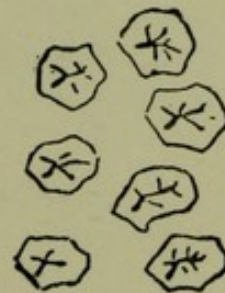


FIG. 9. MAIZE STARCH
X 200

1. The pea granules are generally a long oval in shape, fairly large, but showing different sizes. They generally present a linear hilum, but this is sometimes branched.

2. The bean granules are of a shorter oval than those of the pea, and are more uniform in size. The hilum is linear, but more often branched (sometimes even stellate) than in the case of pea granules.
3. The maize granules are mostly polyhedral in shape—approximating to the oval form. They are often cracked, and present a well-marked stellate hilum.

GROUP IV

Rice, Oats

The granules of this group are much smaller than those of the preceding groups. They are angular, and appear to be faceted.

1. Rice (*Oryza sativa*).
2. Oat (*Avena sativa*).

1. The rice granules are the smallest of all those with which we are dealing. Under a higher power an eccentric hilum may sometimes be made out. The granules are often massed together in angular and irregular shapes.

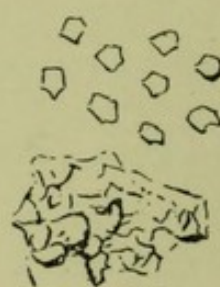


FIG. 10
RICE STARCH
X 200

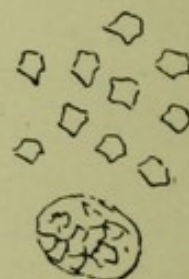


FIG. 11
OAT STARCH
X 200

2. The oat granules are larger than those of rice, and hila are not to be found. These granules are found in masses, but the contour is generally regular and oval, and not irregular and angular.

GROUP V

Sago, Tapioca

The granules of this group are very irregular in shape, and many appear truncated. They have a hilum and badly defined rings.

1. Sago (*Sagus farinifera*).
2. Tapioca (*Jatropha manihot*).

1. The sago granules are large and irregular. They are often rounded at one end and truncated at the other. The hilum is frequently rounded.
2. The tapioca granules are much smaller than those of sago, but are in other respects similar.

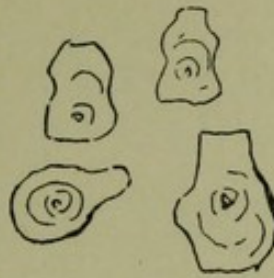


FIG. 12
SAGO STARCH
X 200

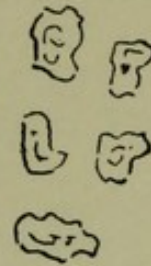


FIG. 13
TAPIOCA STARCH
X 200

The Parasites of Grain, Flour, etc.

Various parasites are found affecting grains, flours, and bread. Some of these parasites are harmless, others are somewhat injurious when consumed.

They may be divided into two classes:—

1. Animal.
2. Vegetable.

I. ANIMAL PARASITES

- a. Corn weevil (*Calandra granaria*).
- b. Meal mite (*Acarus farinæ*).
- c. Pea bruchus (*Bruchus pisi*).
- d. Ear cockle (*Tylenchus tritici*).

a. The *Calandra granaria* is one of many allied species of insect which affect grain.

The corn weevils belong to the order of Beetles (Coleoptera). Two species are well known in England, the *Calandra granaria* (or *Sitophilus granarius*) and the *Sitophilus oryzae*.



FIG. 14
CALANDRA GRANARIA
X 4



FIG. 15. ACARUS FARINÆ
X 100. (ad nat. T.G.S.)

The insect perforates the shell and abstracts the contents of the seed, leaving merely the coverings. All the harm this insect does, therefore, is simply to eat the flour; it does not of course affect any grains which are not attacked, nor is it ground up with the flour as

in the case of some of the parasites. The grains are attacked when the corn is actually standing.

b. Acarus farinæ. This is also an insect which is frequently found in inferior and damp meal, flour, etc.

It bears some resemblance to the *Acarus scabei*, but a close examination will reveal the great difference between the bodies and legs. The body of the *Acarus scabei* is much rounder than that of the *farinæ*, and the legs of the meal mite are fairly thick up to the extremity, whilst those of the *Acarus scabei* are thick at the proximal, but quite thin at the distal end.

c. The Bruchus pisi. The *Bruchus pisi* (the pea bruchus), *Bruchus rufimanus* (bean bruchus), and the *Bruchus granarius* (the grain bruchus) belong to the same order (Coleoptera) as the weevils, which they resemble in their main features. The first two, as their names imply, are connected with peas and beans. The adult female lays her eggs in the young fruit, and the larvæ live in the seeds, eating up all the internal parts and changing to pupæ within the outer shell.



FIG. 16. BRUCHUS PISI
× 4

d. The Tylenchus tritici (Vibrio tritici).

The worms seen when the contents of an infected grain are examined under the microscope are the larval forms of a nematode worm.

In the ears of wheat affected by this worm the grains are misshapen, blackish, and consist of a thick hard scale enclosing a white powdery substance, composed almost entirely of the larval forms of the worm.

In order to examine this powder under the microscope, it is only necessary to place a little on a clean slide and to mount it in water.

Permanent specimens may be prepared by mounting in Farrant's solution, the powder may be thoroughly dehydrated by drying or by treatment with alcohol and xylol, and mounted in Canada balsam. Very pretty speci-

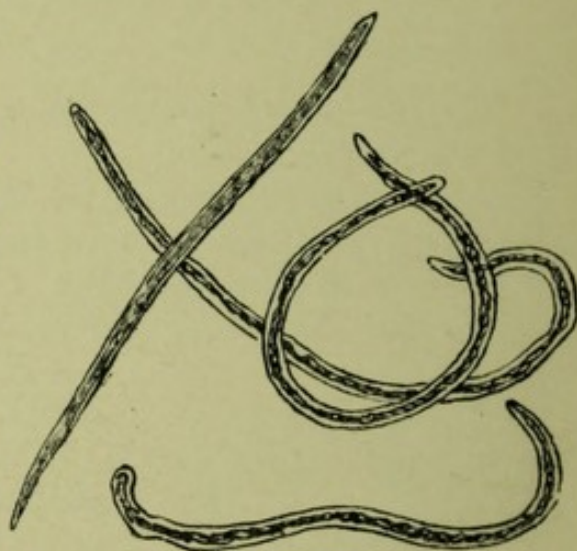


FIG. 17. TYLENCHUS TRITICI
× 60. (ad nat. T.G.S.)

mens may be prepared by staining the worms with eosin and mounting in Canada balsam, in the usual manner.

If the infected grain be sown in ordinary moist ground, the husk simply rots. The larvæ escape and become active. They move along the ground in search of growing blades of corn. When they find one, they slowly creep up and eventually reach the young soft grain, which they penetrate. Here they form gall-like swellings, in the middle of which they are found.

In this position they quickly develop into the adult form. After fertilization by the males, the females lay a large number of eggs, and both males and females die. The eggs subsequently hatch and the larvæ are seen.

Closely allied to this worm is the *Anguillula aceti*, which is sometimes found in vinegar, when this is made from beer or wine, and which is also found in sour paste.

II. VEGETABLE PARASITES

- a. *Penicillium glaucum*.
- b. *Aspergillus glaucus*, *albus*, etc.
- c. *Mucor*.
- d. *Peronosporon*.
- e. *Puccinia graminis* and *Rubigo vera* (rust).
- f. *Ustilago segetum* (smut).
- g. *Tilletia caries* (bunt, *Uredo foetida*, *Ustilago carbo*).
- h. *Claviceps purpurea* (ergot).

a. The *Penicillium glaucum* is a common mould, found very extensively in the air. It is frequently found forming a greenish growth, on damp grain, flour, bread, or cheese.

This mould resembles the others in consisting of many long threads (hyphæ) often interlaced and forming a mycelium. The hyphæ branch and produce special spore-bearing ones. The end of this last hypha then branches into three or more terminal filaments which in their turn divide transversely to the long axis. The protoplasm of these short rods now becomes differentiated and round or oval spores with thick cell walls are found. These soon separate from one another, and free spores are found.

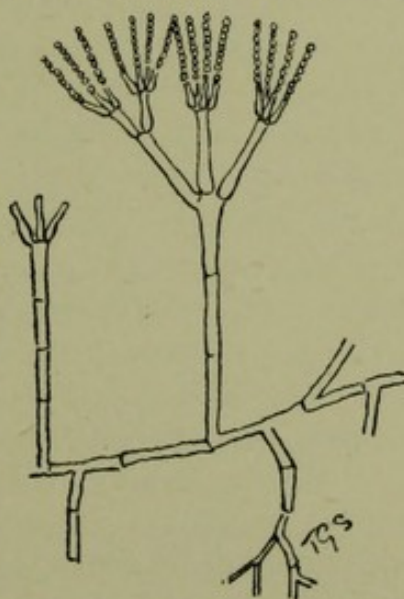


FIG. 18
PENICILLIUM GLAUCUM
X 100

b. Aspergillus glaucus, etc. This species of mould is also found in damp grain, etc. It resembles the penicillium in its mode of growth, but differs from it in the way in which the spores are formed. The end of the spore-bearing hypha becomes enlarged and the spores appear to grow out of the bulbous end, and are at first attached to it by fine pedicles. Generally there are two or three rows of spores on each head, the pedicles of the external row being longer than those of the other rows. When the spores are ripe they simply fall off, and so become free.

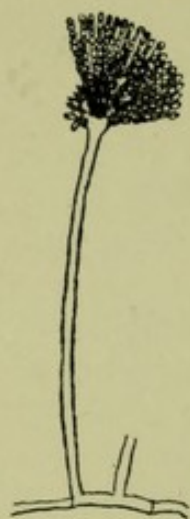


FIG. 19
ASPERGILLUS
X 100
(After Howes)

c. Mucor. This form of mould is found in similar positions to the last two. It resembles these in its mode of growth except that it forms its spores in still another way. The end of the spore-bearing hypha becomes enlarged, and instead of the spores growing out from this enlargement, as in the case of the aspergillus, the end attains a considerable size (quite visible to the naked eye). The protoplasm within the wall of the head

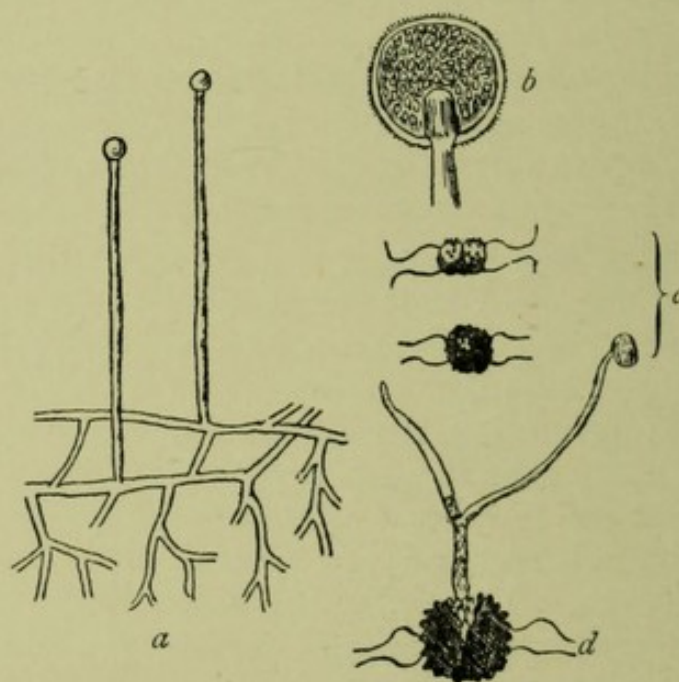


FIG. 20. MUCOR : (a) special spore-bearing hyphæ ; (b) head $\times 100$, showing internal spores ; (c) and (d) conjugation. (After Howes)

undergoes differentiation, and the spores are formed by this means inside the envelope which surrounds the head. When the spores are ripe the envelope is ruptured, and the spores set free.

This mould is occasionally found to multiply by a process of conjugation, as shown in the diagram. The swelling so formed is found to become differentiated in a manner similar to that found in the head produced on a hypha.

d. Peronospora. These moulds do not affect the prepared product so much as the living plant. They were the cause of the great Irish potato famine, and are found affecting many of the "root" crops as onions, parsnips, turnips, etc.

The mould first affects the leaves and gradually travels downwards until finally the tuber or root is affected.

The growth consists of a dense mycelium which produces spore-bearing hyphæ externally.

These hyphæ frequently branch, sometimes many times. At the end of each branch a single spore is produced. This when it is ripe separates and so becomes free.

e. Puccinia graminis. This parasite is found affecting many varieties of corn.

The process of infection is as follows: A spore becomes attached to the, say, wheat grain, and as it grows, it sends free filaments into the interior of the grain. As time goes on minute transparent cellules are developed from the mycelium: these enlarge and become coloured.

As the result of their increase in size, the cuticle first becomes distended, and finally ruptures, and the spores are found at the surface as rust.

The distinctive features of the Puccinia are the uniseptate or double spores, which are attached on a distinct peduncle, and it is in this condition that the parasite is shown at the various examinations.

f. Ustilago segetum (syn. *Uredo segetum*, smut). This—a

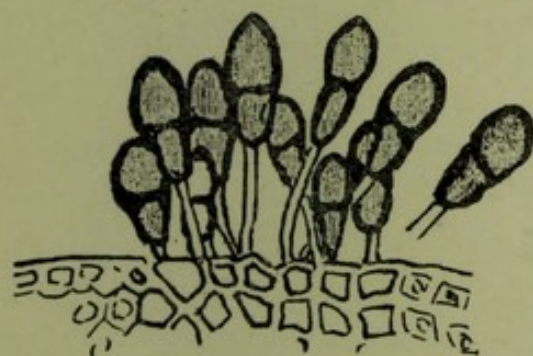


FIG. 22
SPORES OF PUCCINIA GRAMINIS
X 350

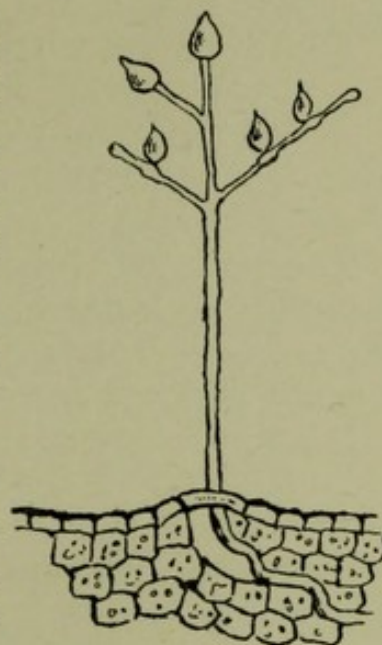


FIG. 21. PERONOSPORON
X 100. (After Vines)

common parasite of corn—is one of a large family of parasites which are found affecting plants.



FIG. 23
USTILAGO
SEGETUM
X 500

Among the standing corn withered heads are often seen, which appear black or brown. If they are rubbed, or even touched, a fine brownish powder falls off. This powder consists of the spores of the parasite.

Under the microscope these are seen to be small spherical spores, which are generally coloured a light brown. They are entirely free, as by the time the spores are ripe the mycelium will have disappeared.

g. Tilletia caries (syn. *Uredo foetida*, *Ustilago carbo*, bunt). This is another member of the ustilaginae, to which order the *Ustilago segetum* belongs.

The parasite is found in the interior of the grain, but does not affect its external appearance, or if it does only by slightly darkening it. In fact it is often stored with the sound grain, and it is not until the corn is ground that its presence is determined.

If an affected grain be cut or broken open, the interior is seen to be filled with a sooty, rather fetid powder. When this powder is rubbed between the fingers it has a greasy feel.

Under the microscope these spores are seen to be brownish spherical bodies with a reticulated surface. They are generally free, but some are found with part of the attached hyphæ, and sometimes two or three are seen to be joined together by hyphæ.

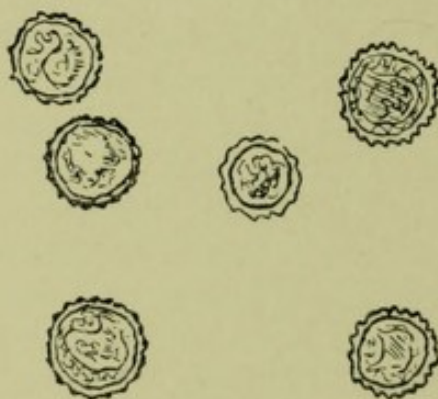


FIG. 24. TILLETIA CARIES
X 200. (*ad nat.* T.G.S.)

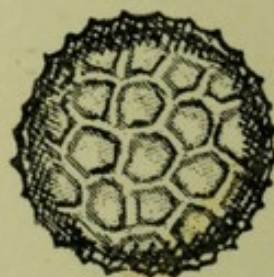


FIG. 25. TILLETIA CARIES
X 500. (*ad nat.* T.G.S.)

h. Claviceps purpurea. This fungus is found chiefly affecting rye, and the mycelial growth—termed the sclerotium—which replaces the actual grain is known as ergot. In the spring time of the year small hair-like growths with a bulb at the end grow out from the mycelium. These are known as stromata, and each contains near its border a row of receptacles (ascocarps), containing oval-shaped bodies known as asci. These ascocarps and the

contained asci are the preparations of ergot usually shown at the examinations.

CLAVICEPS PURPUREA

(ERGOT OF RYE)



FIG. 26. SCLEROTIUM
WITH STROMATA
(Nat. size)

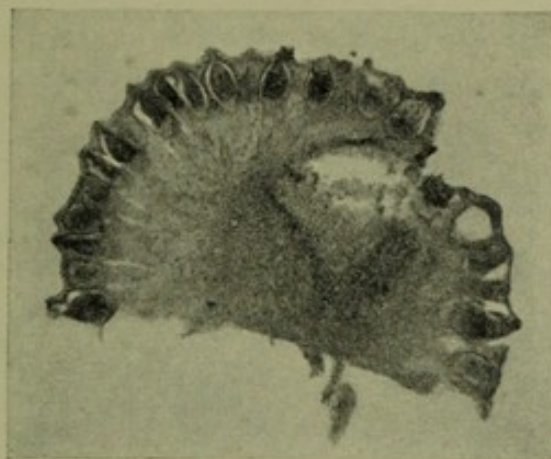


FIG. 27. SECTION OF THE END OF A
STROMA, SHOWING ASCOCARPS AND ASCI
× 80. (Photo)

MILK

The microscopical examination of milk, apart from bacteriological considerations, is not of great value in hygiene. Pure normal milk when viewed with the microscope is seen to consist almost entirely of fat globules, which vary only slightly in size. The globules are all small, the largest being about three times the size of the smallest. In many samples of pure milk cellular elements are absent or rare; but in others obtained from undoubtedly pure sources "leucocytes" may be present, sometimes in large numbers. These leucocytes closely resemble polymorphonuclear leucocytes, but differ from them in slight respects: their presence alone is not sufficient to condemn a milk sample. If, however, typical polymorphonuclear leucocytes are seen, accompanied by pyogenic organisms, the milk may be from a cow suffering from mastitis and then will be unfitted for human consumption.

Gross dirt, such as manure, hairs, shreds of clothing, etc., will also render a milk sample unfit to drink; and shows neglect on the part of the provider to filter the milk before it was distributed.

BUTTER AND MARGARINE

Although the microscopic examination of butter and margarine is not so important as the chemical examination, pure butter differs very markedly from margarine.

In order to examine these substances, a small quantity of the butter or margarine should be spread in a thin layer on a clean slide. A drop of 1% osmic acid in water should then be placed

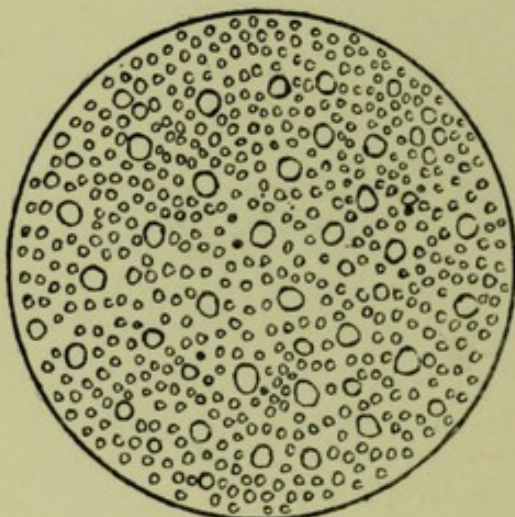


FIG. 28. BUTTER
× 400. (*ad nat.* T.G.S.)

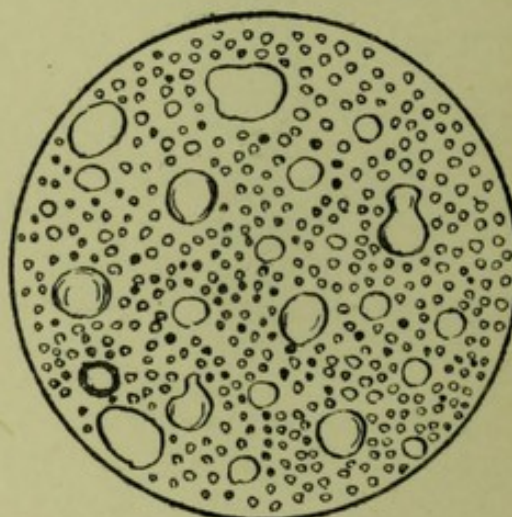


FIG. 29. MARGARINE
× 400. (*ad nat.* T.G.S.)

on the fat and a clean coverslip superimposed. The osmic acid serves to show the contour of the globules very clearly.

In the case of pure butter it will be seen that the globules are small and do not vary much in size. The largest globules are not more than four or five times the size of the smaller ones.

The globules of margarine vary greatly in size, the largest being fifteen to twenty times as large as the smallest ones.

COFFEE

Coffee is obtained from the seeds of the *Coffea Arabica*. For use it is ground and generally mixed with a modicum of chicory, varying from 10 to 90%. So long as such a mixture is sold as a mixture there is no infringement of either law or honesty. It sometimes happens, however, that a mixture of ground coffee and chicory is sold as pure coffee. In order to detect this fraud it is necessary to be able to identify the two substances.

In order to examine them under the microscope the ground substances should be warmed in a watchglass containing 40%

soda solution, and mounted in water or soda. Unless this is done it will be found that the grains are so hard and coarse that it will be impossible to mount them satisfactorily. The soda will

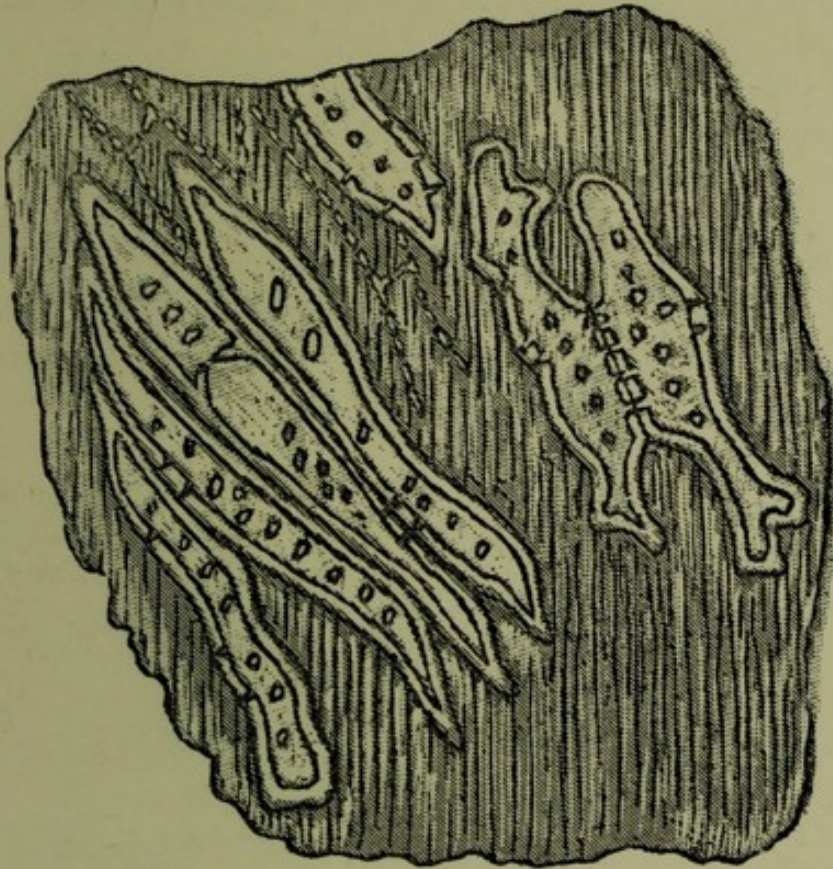


FIG. 30. MEMBRANE OF COFFEE BERRY, SHOWING SPINDLE CELLS
× 100. (*ad nat.* T.G.S.)

take some of the colour out of the grains, but this will be found an advantage rather than a disadvantage.

Under the microscope the endosperm cells form the main bulk



FIG. 31
DOTTED VESSELS OF CHICORY
× 100. (After Möller)

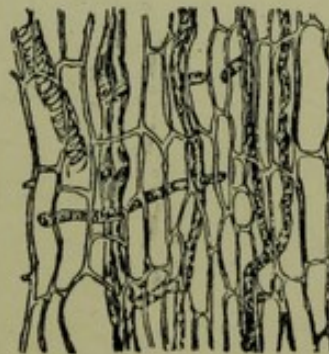


FIG. 32
LACTEAL VESSELS OF CHICORY
× 100. (After Möller)

of the preparation. These are knotted, thickened, thick-edged and polygonal, and may contain remnants of the original contents. Here and there will be found remnants of the membrane lining the berry. This membrane has attached to it a number of very characteristic spindle-shaped cells.

Chicory consists of the ground dried root and contains elements quite foreign to the coffee berry. The parenchyma is much more open than the endosperm cells, and both lacteal vessels and the dotted ducts are numerous.

Numerous other adulterants are from time to time added to coffee, such as various starches or starch-containing tissues, which are obvious upon microscopic examination.

TEA

Tea consists of the dried leaves of the *Camellia thea*, of which there are several varieties.

In order to examine the leaves they should be soaked in water,

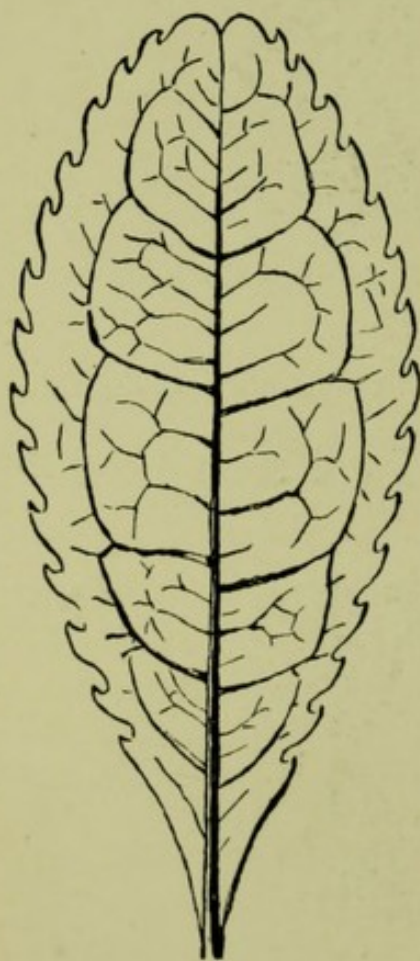


FIG. 33. TEA LEAF
× 4. (*ad nat.* T.G.S.)

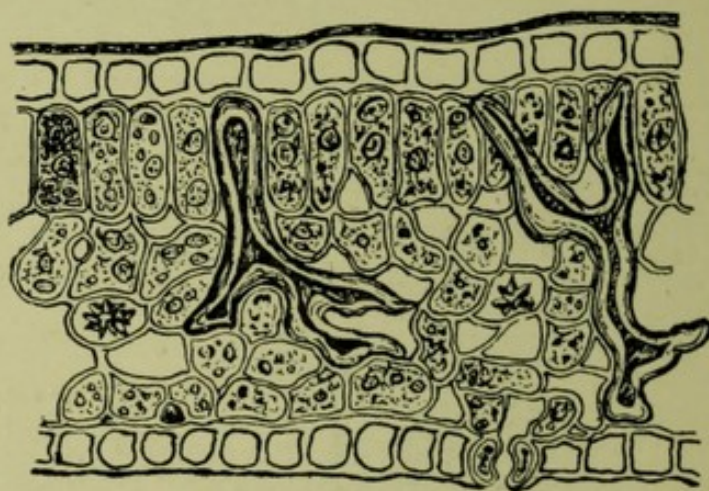


FIG. 34
SECTION OF TEA LEAF, SHOWING IDIOBLASTS
× 160. (After Möller)

and when they have assumed their original shape they may be dried between blotting-paper and mounted on glass in formalin-gelatin or other like substances.

The leaf thus prepared is seen to be elliptical in shape. The margin is serrated and the apex of each serration is surmounted with a minute spine. These serrations do not extend quite to the point of attachment of the stalk. The apex of the leaf is slightly emarginate. The ribs come off from the midrib nearly dichotomously and form a looped network which extends nearly but not quite to the edge of the leaf, leaving a clear margin.

If there is any doubt from the shape of the leaf as to its genuineness, a small portion should be cut off near a rib, warmed in a 20% solution of soda and mounted on a slide, the coverslip pressed down firmly but gently. Upon examining the specimen under the microscope long tough tenacious branched cells are seen. These are termed *idioblasts*, and do not occur in any of the leaves likely to be mistaken for tea leaves.

COCOA

Cocoa is prepared from the roasted seeds of the *Theobroma cacao*. If the cocoa nibs be finely ground in a mortar and mounted in water, it will be found that there is so much fat

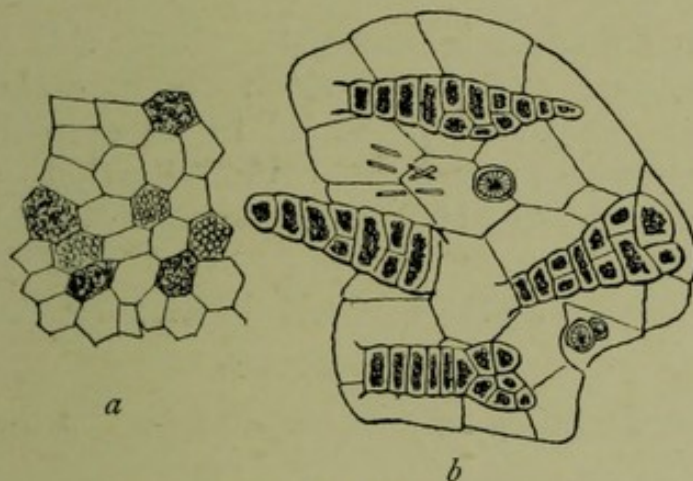


FIG. 35

(a) Parenchymatous cells of the cocoa bean

× 150

(b) Portion of husk, showing characteristic cellular hairs

× 150. (After Rubner)

present that the tissues will be seen with difficulty. In order to avoid this the ground nibs may be treated with ether and subsequently with warm water, and mounted in water.

A second method is to warm some of the ground nibs in a watchglass containing a little 20% soda. Some pieces are then mounted in soda. If, as sometimes happens, the particles are not sufficiently soft to allow the coverslips to be pressed down on to the specimen they may be crushed between two slides before mounting.

Two kinds of tissue will be seen upon microscopic examination, one the external covering and the other the parenchyma. The cells of the external covering are large and have superimposed "hairs" consisting of thick-walled cells arranged as in the illustration. The parenchyma consists of smaller mucilage cells, some of which contain starch granules and others the pigment of the cocoa.

The most of the prepared cocoas consist only of the parenchymatous cells, with or without the addition of other starch, and generally with a portion of the fat removed.

Cocoa starch granules are about the same size as rice, but are rounded in contour.

The addition of foreign starches may be easily ascertained by shaking up some of the cocoa in cold water, allowing the coarser particles to settle, and mounting some of the milky supernatant fluid.

CLOTHING, ETC.

Under this heading are included the fibres used in the manufacture of clothing; and those which are in common use for other purposes. They may be found in water sediments, milk, sewage and elsewhere; and their identification is frequently required of the student at D.P.H. examinations.

A. VEGETABLE FIBRES

EXAMINATION

All the vegetable fibres may be examined under the microscope in water.

The fibres should be macerated in water in order to get rid of the air, and teased out as finely as possible. They may then be placed on a slide with some water, and a coverslip superimposed. The excess of water at the edges of the coverslip should be removed with blotting-paper previous to examination.

Permanent specimens may be made either stained or unstained. The fibres may be coloured with any of the aniline dyes in weak solution, for a few minutes.

After staining, or if they are to be mounted unstained, they should be immersed in absolute alcohol for several minutes, then removed to xylol for 4 or 5 minutes, drained of the excess of xylol and mounted in Canada balsam.

1. Cotton

This is the downy hair of the seeds of plants belonging to the family *Gossypium*. Four species appear to be available for this purpose, the commonest being the *Gossypium barbadense*.

Under the microscope the fibres are seen to be long (from $\frac{1}{2}$ to 1 in.) and thin, the diameter being about 20 or 30 μ . They are flattened and have a very distinct margin which sometimes gives the impression of a double contour. The chief characteristic about the fibres is that they are all twisted, and this, however short the fibres may be.

Cotton fibre is largely used in the manufacture of sheeting, calico, towelling, fustian, velvet, flannelette, paper, etc. Mixed with wool it constitutes merino, which is used for vests, socks, etc. It is used as an adulterant of silk, but not to such a great extent as is jute.

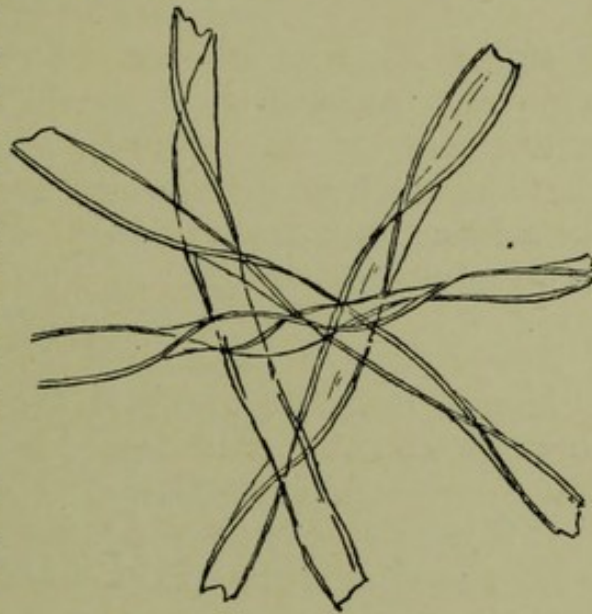


FIG. 36. COTTON FIBRES
× 100. (ad nat. T.G.S.)

2. Linen Fibre—Flax

Flax consists of bast fibres and is obtained from the stalk of *Linum usitatissimum*. The stalks are allowed to rot on the

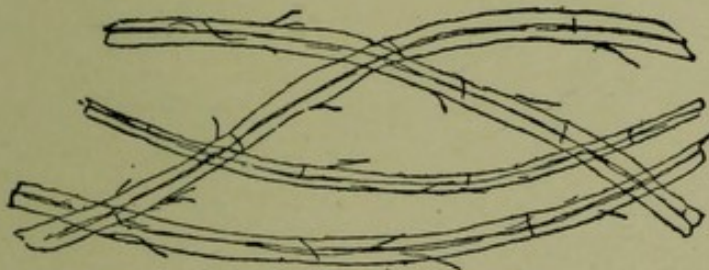


FIG. 37. LINEN FIBRES
× 100. (ad. nat. T.G.S.)

ground and are subsequently beaten and combed, the result of the process being the raw flax of commerce.

Under the microscope the fibres are about the same diameter as cotton, but are cylindrical (not flat). At more or less regular intervals there are distinct nodes or transverse divisions, and from some of these fine hairs (a few μ long) are seen to issue.

Flax is used for shirts, collars, sheeting, and rags made of linen are used to make paper of good quality.

3. Jute

Jute is the bast fibre of the *Corchorus capsularis* or *Corchorus olitorius*, a tropical shrub grown chiefly in Bengal. Microscopically the fibres are seen to be cylindrical and to have a central channel which varies in width and is very distinct. Jute is used for making mats and sacking, and in this country is used largely as an adulterant of silk.

4. Hemp

Hemp consists of the bast fibres of the *Cannabis sativa* and resembles linen very closely. It is coarser than linen, however, and may generally be identified by this character.

It is used chiefly for the manufacture of sacking and ropes, and is little seen as an article of clothing.

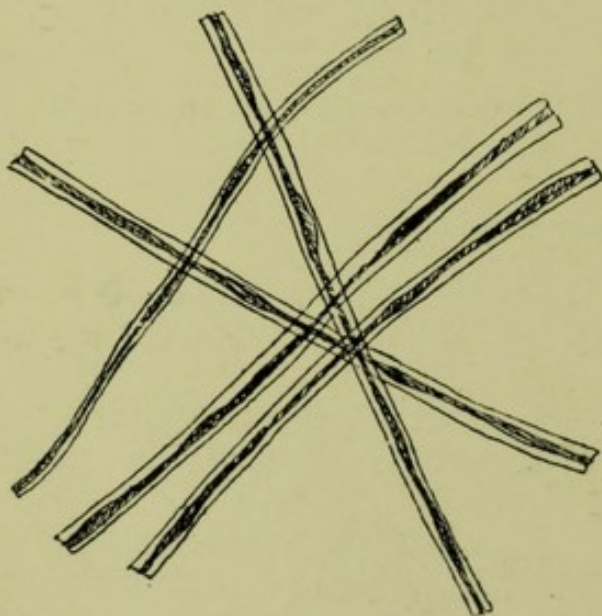


FIG. 38. JUTE
 $\times 100$. (ad nat. T.G.S.)

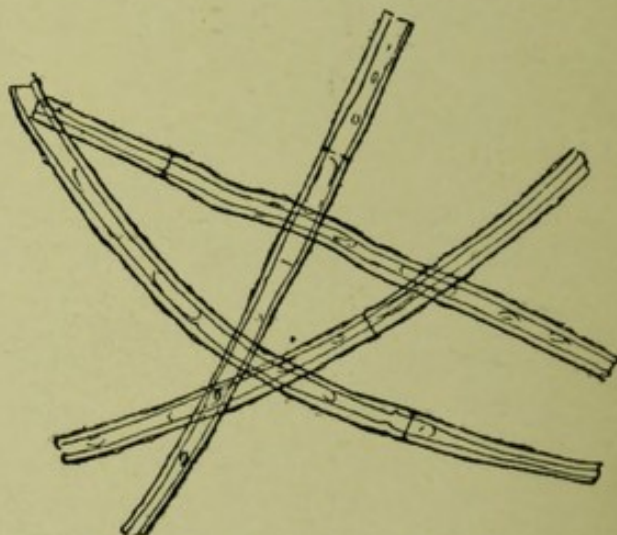


FIG. 39. MANILLA HEMP
 $\times 100$. (ad nat. T.G.S.)

5. Coir

Coir is the coarse fibre obtained from the outer husk of the cocoanut. Under the microscope the fibres are seen to be very coarse and irregular.

It is used chiefly in the manufacture of mats and coarse ropes, and rarely if ever is met with in clothing in this country.

Paper

Paper which is sometimes found in the sediment of water and which has been macerated is seen to consist either of linen or cotton fibres, or of wood fibres. Many toilet papers are made from wood pulp. This paper macerated in water and examined under the microscope is seen to consist of fibres, many of which show "spiral cells" (*A*) and other "pitted ducts" (*B*).

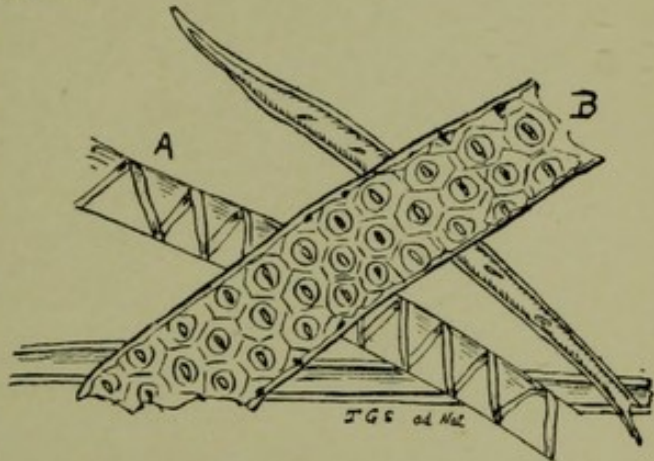


FIG. 40. WOOD FIBRES

× 100

B. ANIMAL FIBRES

The following are best examined by macerating for a short time in dilute soda (2-5%) and mounting in water or dilute soda.

Note

The soda should not be used too strong nor for too long a period, since wool and hair are eventually disintegrated and dissolved by it.

I. Wool

Wool is the prepared fleece from sheep and goats. The wool used in some Jaeger material is derived from the camel.

Under the microscope the fibres are seen to be cylindrical and to be thicker than cotton or linen. The chief characteristic of wool is the

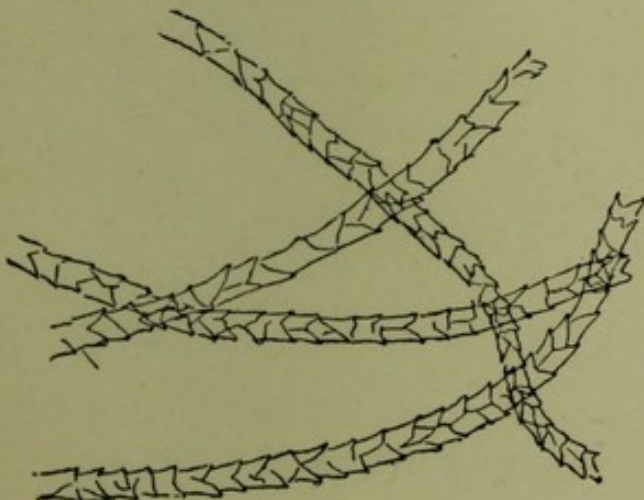


FIG. 41. WOOL FIBRES

× 100. (*ad nat.* T.G.S.)

imbrication of the external scales, which gives the edge a serrated appearance.

Wool is used in making flannel, blankets, worsted stockings, underclothing, etc.

2. Silk

Silk is the fibre produced by the larvæ of several kinds of moth, the *Bombyx mori*, the *Antherea yamamaya*, *Antherea Pernyi*, and *Attacus cynthia*, to serve as a sheath for the chrysalis until it emerges therefrom as the adult moth.

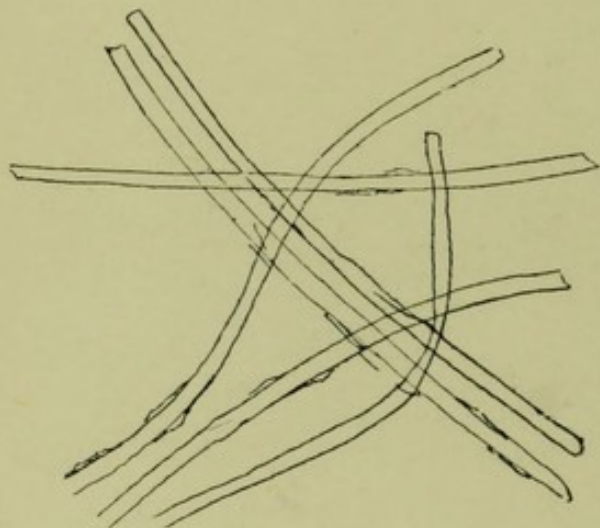


FIG. 42. SILK FIBRES
X 100. (ad nat. T.G.S.)

If silk be mounted in water and examined under the microscope it is seen to be quite structureless and waxy. In reality there is a central core surrounded by albuminous material. The fibres are smaller than any of the preceding, the diameter being half or less than half that of wool. There are no nodes, imbricated scales, or twists.

It is dissolved by strong alkalis and acids, even by acetic acid.

HUMAN PARASITES

Under this heading are considered some of the common parasites that infect the body and clothing of man. Internal parasites such as the worms and bacteria do not, for the most part, come within the scope of this book ; but mention is made, under the section devoted to meat inspection, of certain of the parasitic worms which are introduced to the body through the ingestion of infected meat. At present, however, only those common external parasites, likely to be placed before an examination candidate, are considered.

A. INSECTA

The *Pulex irritans* (the common human flea) does not require much description. There are many species of fleas ; and,

although man is susceptible to the attacks of those which normally affect the lower animals, these insects prefer for the most part their own particular host.

Fleas resemble flies to some extent ; but they have only single eyes and extremely rudimentary wings. A flea is provided with a proboscis for piercing and sucking.

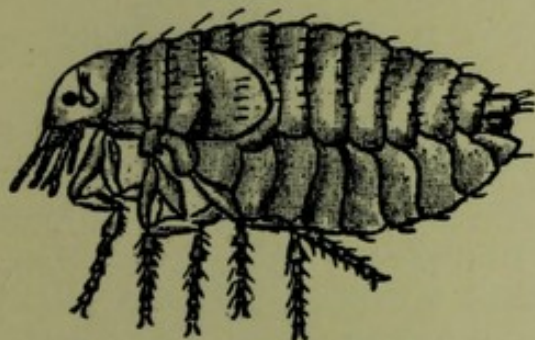


FIG. 43. *PULEX IRRITANS*
× 10. (After Beille)



FIG. 44. *PULEX PENETRANS*
× 10. (After Beille)

The female lays her dozen eggs about the floors of houses, kennels, etc. Six days in the summer suffice for the appearance of the worm-like larvæ, which are provided with a powerful biting mouth. They live on particles of decaying organic matter. They move about by means of the hooks and hairs which are placed on the posterior border of each of the thirteen rings which they possess. At the end of 11 days the larva spins a cocoon and is transformed into a chrysalis ; in another 10 or 11 days the chrysalis emerges as the perfect insect.

The *Sarcopsylla penetrans* (chigoe) is nearly allied to the last described and has a familiar history.

The adult female gets from the ground generally on to the foot. Here she burrows her way beneath the skin, holding on with her powerful mandibles. Soon after she has become parasitic she swells up to twice or three times her normal size with eggs.

The *Cimex lectularius* (bed bug) is a small insect, having a mouth in the shape of a beak or rostrum, adapted both for piercing and sucking.

The adult female lays about 50 white cylindrical eggs in that period of the year between March and September. The eggs have a hinged lid, and after five or six days of incubation, the young cimex opens the lid and walks out. The young do

not come to maturity for 10 or 11 months, and during their adolescence undergo three or four moultings. The young are more slender than the adults and have less colour.



FIG. 45
CIMEX LECTULARIUS
× 10. (ad nat. T.G.S.)

The *cimex lectularius* is wingless: it prefers darkness rather than light, and lives in corners and niches. In the twilight it comes out of its corner and searches for its food until the morning, retreating when the room grows light. The young *cimex* is able to hunt for itself and is independent of the adults.

The odour associated with these insects is due to glands situated in the first segment of the abdomen—the adult bearing two, and the young three.

The **Pediculi** are provided with piercing and suctorial mouth parts. The mouth consists of a soft retractile beak, conical in shape, and furnished below with a row of hooks for attachment. Inside the soft beak are four grooved plates which when juxtaposed form a membranous tube which can be extended beyond the mouth, and which is used for piercing. The thorax is small in comparison with the size of the abdomen, and is not distinctly divided into segments, although as in all insects the three pairs of legs are all attached to it.

The *Pediculus capitis*, as its name implies, is chiefly found on the hairy scalp and obtains its food by piercing the skin and sucking the blood. It is of yellowish brown colour, which is darker at the edges. The legs have a spine at the extremity which can be opposed to the end of the digit. This enables the insect to suspend itself.

The female lays about 50 greyish eggs which are covered at the end with a hinged lid. The eggs are fastened on to the hair with chitinous material and the hair is constricted at this spot. They are nearly always fastened on the hair near the scalp, so that those found an inch or so from the stem are generally empty—the young having been hatched.

The eggs are hatched in about a week and the young *pediculus* opens the lid and crawls out. The young resemble the adults except in size, and undergo no moulting as in the case of the *cimex*. In three



FIG. 46
PEDICULUS
CAPITIS
× 10
(After Beille)

weeks or a month they are full grown and are able to reproduce.

The *Pediculus vestimentorum* or body louse is the same length as the head louse, but is about twice as broad. The colour is the same as that of the head louse, but is not darker at the edges. The head is more triangular in shape than that of the head louse. In other respects the resemblance is almost exact.

The female lays her eggs, to the number of 70 or 80, in the folds of the clothing, where the pediculus lives—only coming on to the body to feed. These eggs hatch in about a fortnight or three weeks and the young pediculus is adult in another fortnight and prepares for egg laying.



FIG. 47. *PEDICULUS VESTIMENTORUM*
× 10. (After Beille)

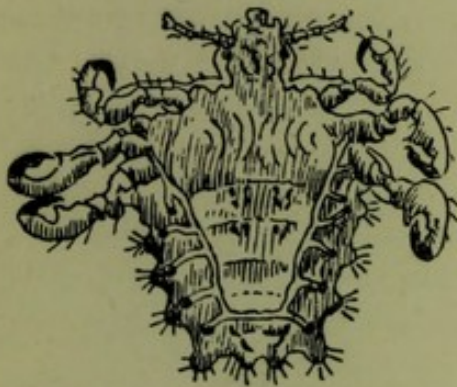


FIG. 48. *PEDICULUS PUBIS*
× 20. (After Beille)

The *Pediculus pubis* differs greatly in shape from the preceding species. It is almost triangular and the abdomen is less broad than the thorax. Between the two there is no constriction. Each leg carries at its free extremity a definite claw, and not merely a spine, but it is with the posterior pairs that the insect hooks itself to the hairs.

The female lays 10 or 12 eggs, which she attaches to the hair quite at the base. The development of the egg resembles that of the preceding exactly.

B. ARACHNIDA

To this class belong several forms of acari which are met with as human parasites. The acari as a class differ from the true insect in several respects. There is no sign of division between the abdomen and thoracic portions, nor is the abdomen segmented; some of the legs are attached to the anterior, and the rest to the posterior portion of the body.

The *Sarcoptes scabiei* (*Acarus scabiei*) has an oval body bearing four pairs of legs, two pairs placed anteriorly and two

postero-laterally on the under surface. The two front pairs terminate in small suckers and the posterior pairs in spines. The animal is greyish in colour and semi-transparent. It carries several pairs of hairs, the longest pairs being on each side of the anus.

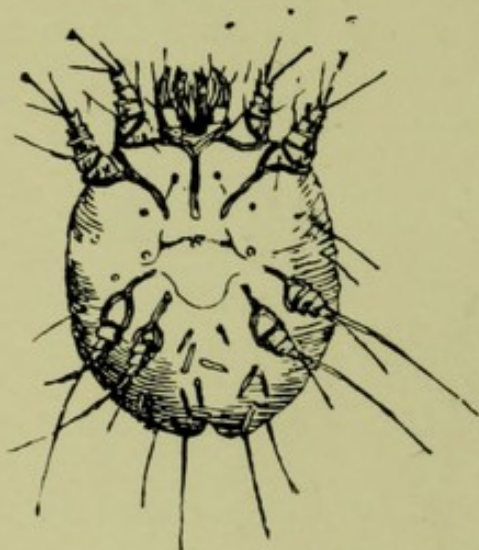


FIG. 49. ACARUS SCABEI
× 60. (Semi-diagrammatic)

The female alone forms the burrows which are characteristic of scabies; she lays about 15 ovoid eggs in the bottom of the tunnel. In about six days these are hatched. They resemble the adult in general shape, but they are completely asexual; they carry only three pairs of legs, two anteriorly and one pair posteriorly. In order to gain their liberty they pierce the vault of the tunnel, and so arrive on the skin. In this stage the larva undergoes two or three separate moults. In the next stage it obtains its fourth pair of legs,

and those which subsequently develop into females are somewhat larger than those that become males. After the next moult the sex of the young is determined and the females are considerably larger than the males.

The male is now adult and undergoes no further change. The adult female again moults before she makes her burrow and lays her eggs. The male is more agile than the female and only excavates the skin sufficiently to find a lodging, where he may be seen as a little brown speck.

The intolerable itching which the presence of the females in the tunnels produces is probably due to the secretion by the acarus of a poisonous fluid. This is only secreted during the night and accounts for the usual phenomenon that the itching is only noticed when the patient is in bed.

The tunnels are found chiefly about the hands, genitals, buttocks and thighs, but may be found everywhere except on the back of the head.

In order to obtain an acarus, the tunnel should be torn up with a sharp needle, and the female picked up on its point.

To examine it, it may simply be mounted in water, or in 5% potash.

Similar acari affect horses, cattle, cats, etc., and these are

occasionally found parasitic on human beings, but they prefer their normal hosts.

The **ticks** or **Ixodes**, of which the common sheep tick (*Ixodes ricinus*) is perhaps the best-known member, are commonly distributed in nature, and are occasionally parasitic on man.

They differ from the sarcopsidæ in having all four limbs at the anterior extremity, and are armed with a powerful beak.

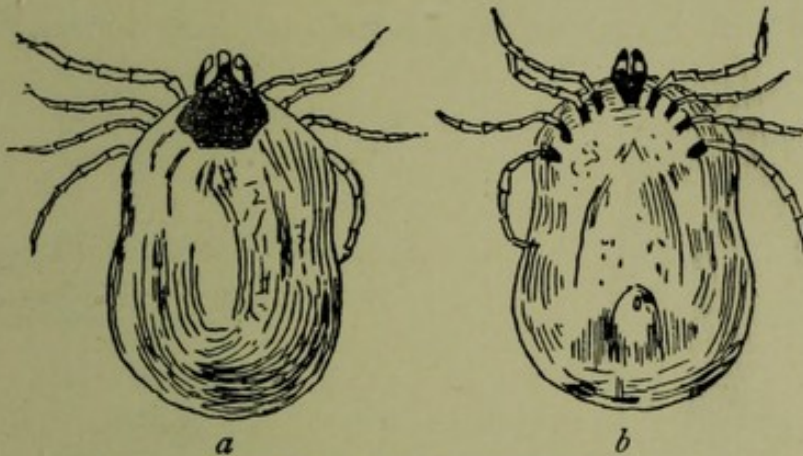


FIG. 50. IXODES RICINUS
× 3 (a) Dorsal view; (b) Ventral view. (After Beille)

The female alone obtains her nourishment from the animal upon which she is parasitic, the larvæ and males being only accidental parasites.

The female grips the hair or skin with the legs and digs her beak through the skin at right angles. There she remains for several days until she is full of blood. She then withdraws her beak and drops off to the ground. If she is brushed off whilst she is sucking, her beak is left *in situ* and may be a source of irritation. The larvæ have a similar life-history to that of the Sarcopsidæ.

WATER SEDIMENT

The following classification comprises the substances that may be found in examining a water sediment :—

- A. *Mineral matter*, sand, clay, etc.
- B. *Vegetable matter*.
 - Diatomaceæ.
 - Schizophyceæ.
 - Schizomycetes.
 - Cyanophyceæ.

Algæ.

Chlorophyllaceæ.

Fungi.

Various more complex plants, or their débris.

C. *Animal matter.*

Protozoa.

Crustacea.

Spongidia.

Various more complex animals, or their débris.

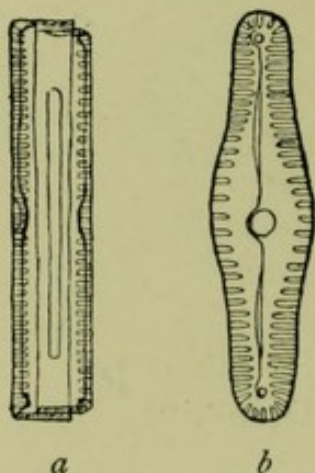


FIG. 51. DIATOMS

X 200

a, surface view ; *b*, side view,
showing two halves of frustule

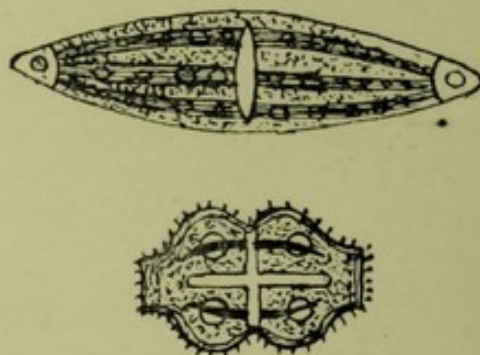


FIG. 52. DESMIDS

X 200

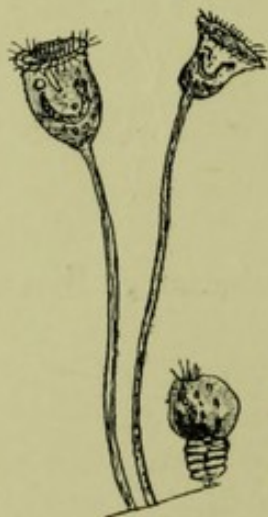


FIG. 53. VORTICELLA

X 150



FIG. 54. EUGLENA VIRIDIS

X 300. (After Ehrenberg)



FIG. 55. SPIROGEIRA

X 60

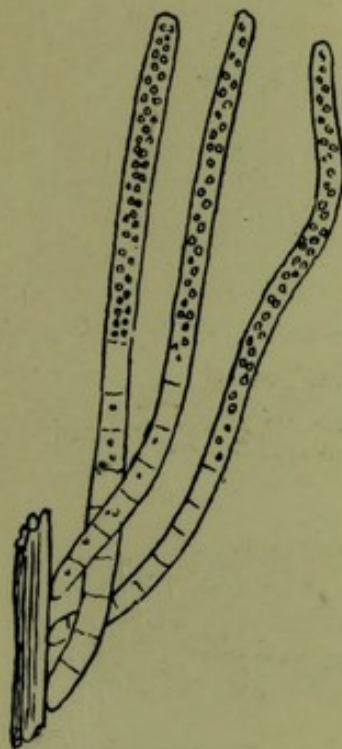


FIG. 56. BEGGIATOA
× 150

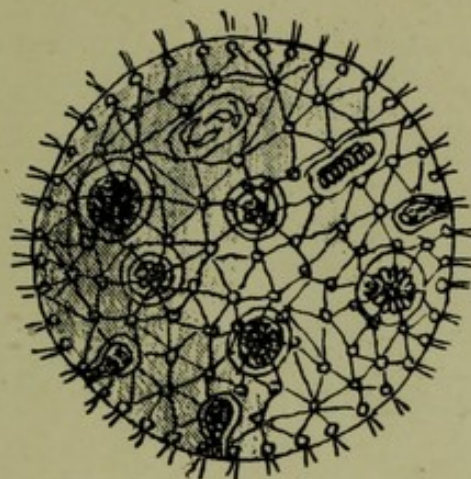


FIG. 57. VOLVOX GLOBATOR
× 60. (Partly after Cohn)

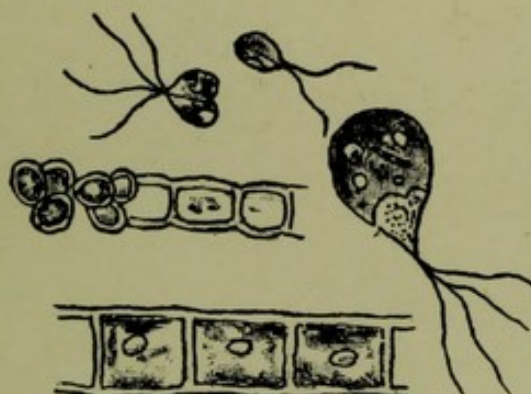


FIG. 58. ULOTHRIX
(After Dodel Port)

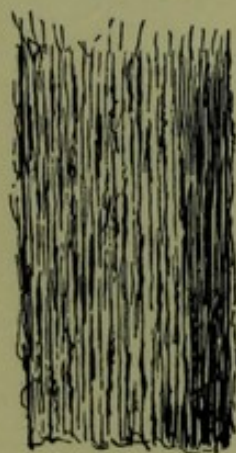


FIG. 59. HUMAN HAIR
× 100. (ad nat. T.G.S.)

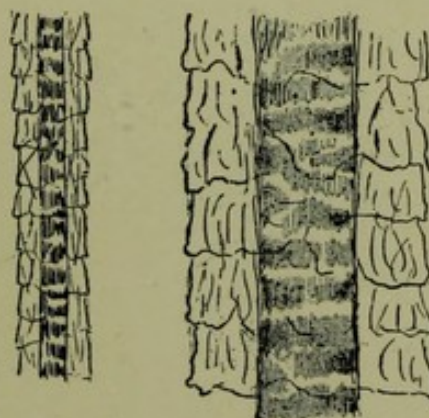


FIG. 60. DOG'S HAIR
× 100. (ad nat. T.G.S.)

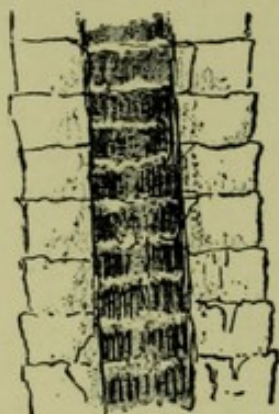
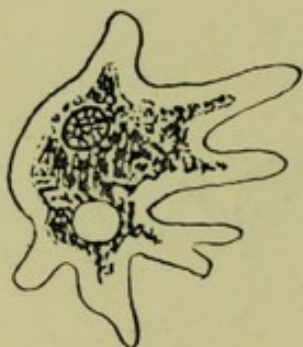


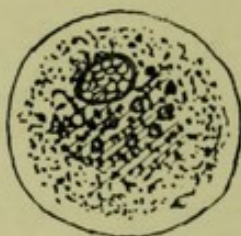
FIG. 61. COW'S HAIR
 $\times 100$. (*ad nat.* T.G.S.)



FIG. 62. RABBIT'S HAIR
 $\times 100$. (*ad nat.* T.G.S.)



a



b

FIG. 63. AMOEBÆA
 $\times 200$
 (*a*) Motile. (*b*) Resting

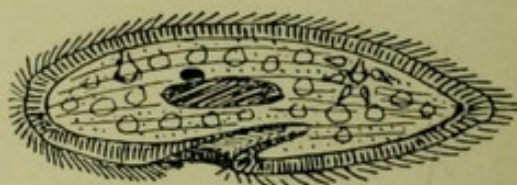


FIG. 64. PARAMOECIUM COLI
 $\times 400$. (T.G.S.)

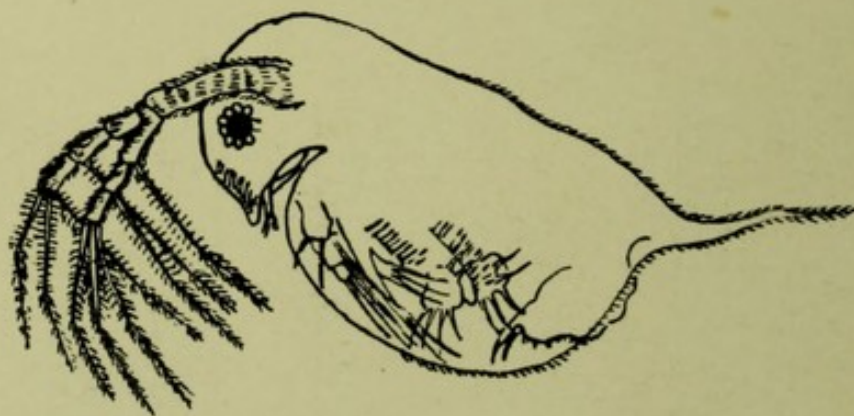


FIG. 65. DAPHNIA



FIG. 66. OVA OF VARIOUS WORMS
 × 400. (After Leuchart)

a. Ascaris lumbricoides
b and c. Oxyuris vermicularis
d. Distoma hepaticum
e. Distoma lanceolatum
f. Trichocephalus dispar

g. Anchylostoma duodenale
h. Bothriocephalus latus
i. Taenia mediocanellata
k. Taenia solium
l. Ascaris mystax

MEAT INSPECTION

THE inspection of meat is often part of the practical examination of the D.P.H. candidate, and he is required to be able to decide if the meat shown to him is fit for human consumption.

Characters of Good Meat

Meat when good and fresh should have a marbled appearance, due to streaks of fat between the bundles of muscle fibres. The colour should be bright and not too dark, and the surface of the meat should be glossy and not dull. Beef is always darker than mutton, veal, or pork—chiefly for the reason that sheep, calves and pigs are bled more freely than oxen at the time of killing. The older the animal, the darker and tougher is the flesh. The connective tissue should be glistening and firm. The diaphragm should be firm, and the abdominal and thoracic parietes should show no evidence of adhesions or staining. The pleura should be intact. The bone-marrow should be set and be light red: the spleen, kidneys, and liver should be regular, of a good red, and without variations in colour.

Good meat is firm and elastic, and does not pit nor crackle on pressure. It is juicy, but not wet; and the juice which adheres to the fingers should be of a bright red colour. The fat is hard and dry, but feels greasy. The kidneys, spleen and liver are firm. All lymphatic glands should be firm.

The smell of sound meat is well known and characteristic. In order to test this more efficiently, the meat should be pierced with a clean knife or skewer in the direction of the bone, and the implement smelt immediately upon its withdrawal.

Good meat gives an acid reaction with litmus paper.

Good meat, when dried upon a water bath, does not lose more than 75% by weight.

Characters of Bad Meat

Meat that is bad is often soft and watery, some parts are harder than others, and there may be emphysematous crackling.

The fat may be liquid or soft, highly coloured or even hæmorrhagic. A deep dark purple colour is seen in meat when the animal has died without being bled, or when some pulmonary congestion or acute septicæmia has affected the animal. The lymphatic glands in bad meat may be enlarged, congested, hæmorrhagic, caseating or calcified. The pleura, peritoneum, or viscera may show evidence of disease such as tuberculosis. Pus may be present between the muscle fibres. The carcass may show signs of emaciation. The odour may be that of putrefaction, and the meat may be alkaline in reaction. In advanced putrefaction the meat may become a greenish tint.

Test for Putrefaction. A mixture is prepared containing 1 part of HCl, 1 part of ether, and 3 parts of alcohol. A few c.c. of this are placed in a cylinder, which is then shaken so as to distribute the reagent over the sides of the glass. A piece of the putrid meat is suspended by a wire inside the cylinder. The white fumes of ammonium chloride will appear if the meat is in a state of putrefaction.

Meat may be unfit for human consumption from one or more of several causes.

1. The animal may have been suffering from a disease which can be communicated to man by ingestion of the meat.

2. The animal may have been suffering from a disease which, though non-communicable to man, may render the meat unwholesome and liable from its contained toxins to produce gastro-enteritis in the consumer.

3. The meat, though derived from a healthy animal, may have undergone decomposition.

4. The meat may not be of the character stated—e.g. horse-flesh may be sold as beef.

Consideration will now be given to certain diseases of animals, which render the whole or parts of the carcass unfit for human consumption.

Anthrax. Anthrax meat rarely comes into the market. The animal afflicted usually dies so quickly that it is impossible to have it slaughtered. The spleen, liver and kidneys are engorged with blood, the intestines are hæmorrhagic, the blood is fluid and the bacillus anthracis is found in vast quantities in the blood and viscera. Of course, all the meat from an anthrax-affected animal must be destroyed—preferably by cremation.

Actinomycosis generally affects the tongue, lower jaw, and lungs. The tongue is wooden in consistence, and may show

flattened white nodules on the dorsum. Occasionally abscesses are formed, and the pus contains grey granules which show typical characters of the "ray fungus" under the microscope. If the disease is not widespread, only the affected parts may be removed, and the remainder of the carcass passed for eating purposes.

Tuberculosis. It should be remembered that tuberculosis in cattle does not frequently lead to pus formation. The tubercles remain firm and typical although the whole body may be filled with the disease. No part of the body is exempt from tuberculosis, although the lungs and lymphatic glands are the sites most commonly infected. The Royal Commission on Tuberculosis in 1898 made the following recommendations with regard to tuberculous meat :—

That the *whole carcass* should be condemned if—

1. There is miliary tuberculosis in both lungs.
2. If there is tuberculosis of both pleura and peritoneum.
3. If there is tuberculosis of the muscles or of the lymphatic glands situated in the muscles.
4. If tuberculosis lesions exist in any part of an emaciated carcass.

And that the *affected parts only* should be condemned if—

1. The lesions are confined to the lungs and the thoracic lymphatic glands.
2. If the lesions are confined to the liver.
3. If the lesions are confined to the pharyngeal lymphatic glands.
4. If the lesions are confined to any combination of the above ; but are collectively small in extent.

The Commission further recommends that the whole carcass of a pig should be condemned if tuberculosis is present even to a slight degree ; and that foreign meat, in which the pleura has been "stripped" should be regarded as tuberculous and should be condemned.

Septicæmia.—In the flesh of an animal dead of septicæmia the blood is fluid and extravasated here and there. The organs will be engorged and the causative micro-organism will be found in the heart blood and generally in the spleen. Local abscesses may be present ; and the flesh will not set, will be moist, and appear purple in colour. Such meat will decompose rapidly. The whole carcass must be condemned.

Glanders affects horses, and not cattle and sheep. Horse-

flesh infected with glanders is entirely unfit for human consumption, as the disease is communicable to man.

Trichinosis. Meat affected with the *trichina spiralis* is seen to be speckled with minute white or grey dots: they are found especially in the diaphragm. The pig is more affected than cattle and sheep. In order to examine the meat, small portions are teased out in dilute potash solution and examined under the low power of the microscope: the small coiled worms are easily distinguished. An account of them is given later under the heading of Parasites of Meat.

As trichinosis is communicable to man, the carcass of the infected pig or other animal must be condemned.

Cysticercus. Pigs, cattle and sheep all suffer from *cysticercus*: the younger animals chiefly are affected, and their flesh is found to be pale and studded with small cysts. These cysts contain the scolex of what, in the second host, would be the tape worm. These are considered later.

All meat that is infected with *cysticercus* must be considered unfit for human consumption.

Sheep Rot is caused by the *distoma hepaticum* invading the portal system. In early stages of the disease it is necessary only to condemn the affected liver: later, however, the animal may become jaundiced and oedematous; and then the whole carcass must be considered as unfit for food.

The *strongylus filaria* is found in the lungs of sheep. If the animal is not emaciated the carcass need not be condemned on this account.

Symptomatic Anthrax (quarter-ill, etc.) and **Malignant Oedema** are septicæmic diseases caused by anærobic bacilli. Animals so affected die quickly. Their flesh is unfit to eat. The symptoms of disease resemble generally anthrax and other septicæmic conditions, and the meat shows the same general characteristics.

Swine Fever. This disease is very severe among pigs and is exceedingly infectious. The flesh in the early stages of the disease shows few lesions; but later a patchy redness of the skin appears which can be traced down through the fat into the flesh. There is much ulceration of the large intestine and patches of congestion or consolidation in the lungs, liver and lymphatic glands. The flesh of a pig dead from swine fever, or killed during the illness, is not fit for human food, and the whole carcass must be destroyed.

Foot and Mouth Disease. The tongue and mucous membrane of the mouth and pharynx show vesicles and ulceration. The feet also show the same condition of ulcers and vesiculation, and the hoofs may be loose, or may even fall off. The disease is very infective, and the carcasses must be condemned.

Horse-flesh is darker in colour than beef, the grain is more coarse, and the fat is more yellow and soft. The bones are stronger in the horse than in the ox, and have better-marked ridges for the insertion of muscles. The tongue of the horse is rounded and smooth: that of the ox is pointed and rough. The liver of the horse has three large lobes and one small one and no gall bladder. The liver of the ox has one large and one small lobe. The kidney of the horse is heart-shaped: that of the ox is long and lobulated. The heart of the ox contains a bone, the os cordis; there is no bone in the heart of the horse.

Fish. Fresh fish should have bright gills and prominent eyes, and their flesh should be firmly adherent to the bones. They should not pit on pressure, nor should their tails hang down unduly. They should give only their characteristic smell.

Inelasticity of the flesh, and an unpleasant odour, are sure signs of decomposition in fish. Fresh fish will sink in water; bad fish floats.

Any fish that is not quite fresh should be considered unfit for food. Fish decomposes rapidly, and even slightly tainted fish may give rise to very severe gastro-enteritis.

Parasites of Meat

Trichina spiralis. This parasite is found affecting man almost throughout the whole world, especially where much pork, and more especially uncooked pork, is eaten.

The natural host would appear to be the rat, and the disease is kept up in them owing to the habit of eating their dead. Pigs become affected by eating portions of dead rats or the refuse of slaughtered pigs, to which they frequently have access. Man acquires it through eating trichinosed pork.

The male measures 1·2 to 1·5 mm. and the unimpregnated female 1·5 to 2·0 mm.

The anterior end is finely pointed and surmounted with the punctiform mouth.

The female has a single ovarian tube which opens into the vagina on the ventral surface about two-fifths of the length of

the worm from the mouth. After copulation the male dies and the female increases in size to 3 to 4 mm.

The young trichinae leave their shell while still in the uterus, and are born free, some five or six days after impregnation. Several thousands are produced before the female dies and is voided.

The young embryo measures about 0.1 mm. and is endowed with activity. Shortly after birth they pierce the intestinal wall, cross the peritoneal cavity by the connective-tissue spaces and so reach the muscles and other tissues. As they proceed they increase in size, and signs of development are

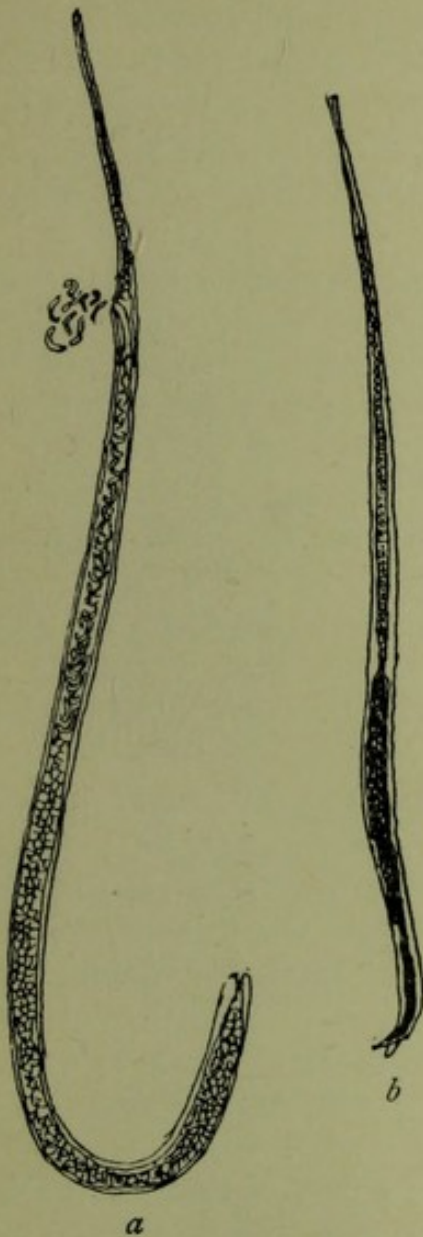


FIG. 67. *TRICHINA SPIRALIS*
a. Female voiding complete embryos. *b.* Male
 × 80. (After Leuchart)



FIG. 68. *TRICHINA SPIRALIS*
 Larval form encysted in muscular fibre
 × 80. (After Leuchart)

found. At the end of 10 days or less they arrive at their destination in the connective-tissue spaces of the muscles, etc.

Their presence gives rise to irritation and subsequent proliferation of the connective-tissue cells in the immediate neighbourhood. The proliferation on the one hand, and the movement of the trichina on the other, lead eventually to the formation of a cyst and wall. At the end of about 18 days the trichina has

increased in size and coiled itself up, and the encystment is complete.

The cysts lie with their long diameter parallel to the muscle fibres and are filled with clear albuminous fluid, whilst the larval trichina lies coiled up but in contact with the wall—especially when alive. It now measures about 1 mm. in length, and is provided with a mouth, alimentary canal and anus, as well as rudimentary sexual organs.

In this condition it remains alive sometimes for years, and is capable, under favourable circumstances, of developing fully. At the end of a certain time, however, if the flesh of the host is not consumed, fatty and calcareous degeneration sets in and the larva dies.

The affected meat can be seen by the naked eye to be "measly," and in order to examine the trichina the muscle fibres containing a cyst should be teased out gently in potash, mounted in potash or water, and a coverslip gently pressed down on the scrapings.

Beautiful specimens can be obtained by hardening the tissue and cutting sections of it.

Tænia mediocanellata (Synonym *T. saginata*, *Tæniarhynchus mediocanellatus*). This is perhaps the commonest and most widely distributed of all human tape worms. It is found in Europe, Asia, Africa, America, and Australia. In this country it is by far the most common, the *Tænia solium* being decidedly rare.

It measures from 4 to 8 metres, and consists of from 1200 to 1300 proglottides. Those near the head are quite small, and gradually enlarge both in length and breadth until near the middle of the worm they measure about 14 mm. square, and are only 2 mm. thick. As the distal end is approached they become narrower, longer, and thicker. As they break away from the main body they are endowed with movement.

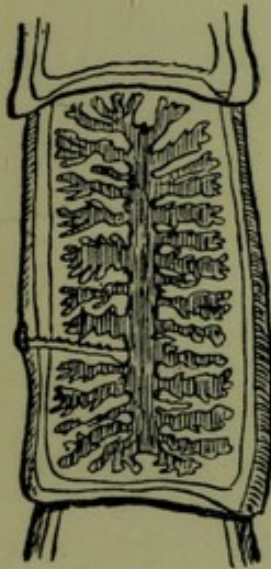
The head is somewhat pear-shaped, and measures from 1.5 to 2 mm. in breadth. It is furnished with four suckers, but has neither hooklets nor rostellum.

The genital pore of the proglottis is marginal, and frequently projects. This leads to the uterus, which is linear, and lies in the long axis of the worm, and which has 20 or 30 lateral branches, which divide dichotomously.

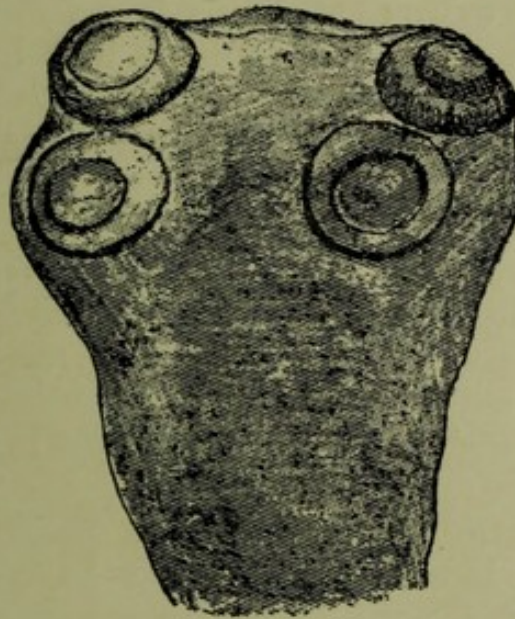
The eggs are contained in the uterus, and when the proglottis is ripe consist of the embryo enclosed in a shell, which is thick,

and is composed of innumerable little rods. The egg is distinctly oval, and is 0.03 mm. in length. The embryo possesses the six hooks.

The ox is generally the intermediate host. This animal swallows the eggs, and the digestive juices dissolve the envelope and set free the embryo, which promptly bores its way into the muscles. It now discards its hooks, and develops the head at



b
FIG. 70



a
FIG. 69

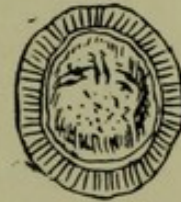


FIG. 71

(a) *TÆNIA MEDIUMCANELLATA*
Head $\times 20$. (*ad nat.* T.G.S.)

(b) PROGLOTTIS, SHOWING NUMEROUS DICHOTOMOUS
BRANCHES OF THE UTERUS
Semi-diagrammatic

(c) EGG, SHOWING EMBRYO
 $\times 400$

the opposite end, whilst the rest of the body becomes a small bladder. The whole cysticercus lies between the muscular fibres and measures from 1 cm. in length. From one end of the short diameter of the cyst the head of the cysticercus may be made to protrude by placing it in warm water. It is seen under the microscope to be an exact reproduction of the head and neck of the mature worm.

The cysticercus is not known in the human subject.

Man is infected undoubtedly from eating underdone beef

affected with cysticerci; quite a low temperature (50° C.) has been found sufficient to render the cysticerci harmless.

The Tænia solium. This tape worm is found wherever swine are badly kept or where the pork is improperly cooked. Hence it is comparatively common in North Germany, and uncommon in this country. It is smaller than the *T. mediocanellata*, rarely measuring 4 m. The proglottides resemble those of the *T. mediocanellata* in shape in the various parts of the body, but are smaller, being not more than 8 mm. broad in the broadest part. The genital pore is marginal, and leads to a linear uterus which has 8 or 10 lateral branches which divide dendritically.

The eggs are more spherical than those of the *T. mediocanellata*, but otherwise resemble them. The head is more or less spherical with a diameter of 1.0 mm. It has four suckers and a rostellum, which can be protruded. The rostellum carries a double row of hooklets numbering 28 in all.

The life-history and cysticercus form is similar to that of the *T. mediocanellata*, except that the pig is the intermediate host.

It differs, however, in that this cysticercus is occasionally found in man.

It is extremely rare to find more than one *T. mediocanellata* in the same patient, but numerous cases have been recorded in which two or more *T. solia* have been found.

Bothriocephalus latus. This parasite—the largest met with in the human subject—is not of very wide distribution. It is

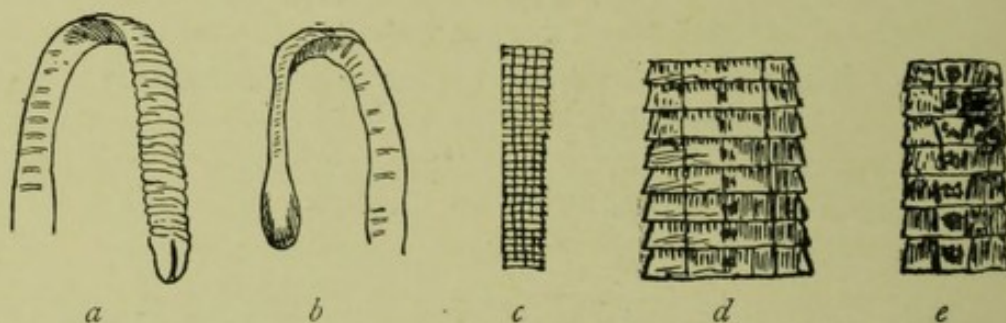


FIG. 72. *BOTHRIOCEPHALUS LATUS*: (a) lateral, (b) front view of head ($\times 10$); (c, d, and e) proglottides in the upper, middle, and lower parts of the worm (nat. size). (After Leuchart)

found in people inhabiting the Franco-Swiss lakes, the Baltic shores, Japan, etc.

It measures from 6 to 12 or even 16 metres in length.

The proglottides are broad and short. The genital pore is in the centre of the proglottis and not marginal as in most others,

and the uterus is rosette-shaped. Each segment is hermaphrodite.

The head is flattened and shaped like an olive or blunt almond. It has two lateral suckorial grooves in place of suckers, and has neither rostellum nor hooklets.

The eggs are oval with a long diameter of about 0.05 mm., the shell is simple, brown, and closed at one end by an operculum.

When the proglottides break away, the ova do not contain the mature embryo enclosed in a shell as in the other tape worm, but

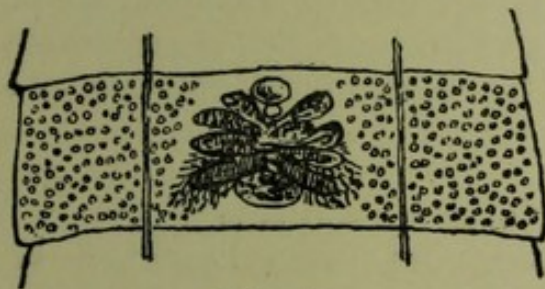


FIG. 73
PROGLOTTIS OF BOTHRIOCEPHALUS
LATUS, SHOWING ROSETTE UTERUS
× 4. (After Leuchart)

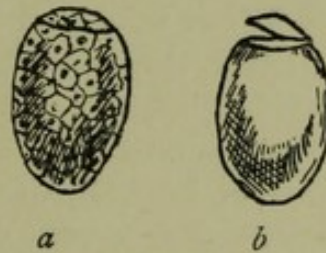


FIG. 74
EGGS OF BOTHRIOCEPHALUS
LATUS: (a) containing em-
bryo; (b) empty
× 200. (After Leuchart)

merely a partially developed egg. These, in order to attain maturity, must remain in water for some time, when the ripe embryo, a ciliated six-hooked one, emerges from the shell by opening the operculum. It then swims about in the water. This is either eaten by fish, or by some animal which is subsequently consumed by fish. From the intestine of the fish the embryo makes its way into the muscles, where it loses its cilia and hooklets, and elongates. After a time evidences of the two suckorial grooves are found and the embryo attains to the size of 1 or 2 cm.; there is no cysticercus form as described in other tape worms, the embryo sometimes lying almost free between the muscular fibres. From the imperfectly cooked or raw fish the embryo is transferred to the intestine of man, dog, cat, etc., where it develops into the mature worm.



FIG. 75. FULLY
DEVELOPED EM-
BRYO OF BOTH-
RIOCEPHALUS
LATUS
(After Leuchart)

The *Distoma hepaticum*. This worm is exceedingly common in the livers of sheep, less common in cattle, sometimes in the horse, rabbit, and even in man.

It is leaf-shaped and flat, being about three or four times as long as it is broad, and varies in length from 16-40 mm.

The worm is enclosed by a skin bearing minute spines attached to the transparent epidermis, and more numerous at the cephalic end. The true skin consists of dense fibrous tissue.

It possesses two suckers, one at the anterior end, that communicates with the mouth and pharynx, and the other, which is placed centrally above the junction of the upper and middle thirds, is blind. These suckers *inter alia* appear to be used for locomotion.

Below the circular pharynx is a band of circular muscular fibres which acts as a sphincter, and prevents the regurgitation of food from the oesophagus, which is a short prolongation of the pharynx, and divides into two canals a little above the ventral sucker. These two canals run parallel to the sides, and give off a series of branches externally, which again divide. A few short branches are given off internally. All these branched tubes terminate abruptly in the parenchymatous tissue, and have no external opening.

The excretory or water vascular system consists of a central canal, which extends from the junction of the upper and middle thirds to the posterior extremity, where it terminates in a small opening, the *foramen caudale*. The anterior end possesses three distinct branches, two lateral and one—a small one—mesial. The main trunk, as well as the three primary branches, all give off smaller branches, which divide dendritically right up to the margin of the worm.

The worm is hermaphrodite. There is a genital pore situated just above the ventral sucker. Into this pore the penis protrudes. The penis is lodged in a pouch, which also encloses the receptaculum seminis. This latter receives the junction of the two vasa differentia, which are formed by the junction of the seminiferous tubules coming from the testes.

The testes are not globular masses, but consist of a large number of vermiform tubes, which are spread out in the middle of the ventral portion of the worm.

The orifice of the vagina is very small, and is situated in the genital pore close to the penis. Just behind the orifice, the vagina widens out into a uterus which is much coiled, and lies between the ventral sucker and the junction of the upper and middle thirds of the body, i.e. above the testes. In the uterus can be seen a large number of ova in various stages of develop-

ment. At the ovarian end the uterus suddenly contracts, and is connected with a tube which suddenly bifurcates, the branches passing laterally across the body and suddenly bending downwards near the lateral margin, to which they run parallel. These tubes give off branches ending in grape-like cæcal extremities, the yolk sacs. These sacs contain minute nucleated cellules.

The ovum when discharged from the uterus is oval and measures 0·14 mm. in length and 0·11 mm. in breadth. It contains a fully developed embryo. This embryo has the form of a cone, the anterior end being flatly convex with a central proboscis-like papilla, and is completely covered with cilia. The ovum is conveyed by the excrement into water or damp soil, and here the embryo leaves the shell, and by means of its cilia moves about in search of an intermediate host, usually a mollusc or crustacean. It bores into this host, loses its cilia and enlarges—forming either a *sporocyst* (a hollow sac without alimentary canal) or a *redia* (a similar structure, but possessing an alimentary canal).

From certain germ cells in the sporocyst or redia, *cercariæ* are developed. These, like the adult distoma, possess suckers and alimentary canal; but, unlike them, they have no genital organs, and have an active and powerful tail. When they are developed they leave the sporocyst or redia and the body of the intermediate host and become free, either in the water or damp soil. The cercaria now either seeks a second intermediate host and becomes encysted, or is taken up by the definite host, where it finds its way into the bile ducts, intestine, etc., and rapidly develops into the adult hermaphrodite worm.



FIG. 76. DISTOMA
HEPATICUM
Nat. size. (After Leuchart)

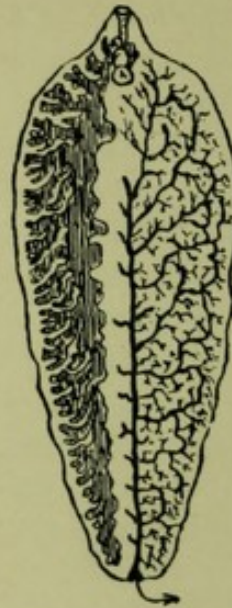


FIG. 77. DISTOMA HEPATICUM
Left half showing the alimentary system,
right half showing the excretory system
(adapted)

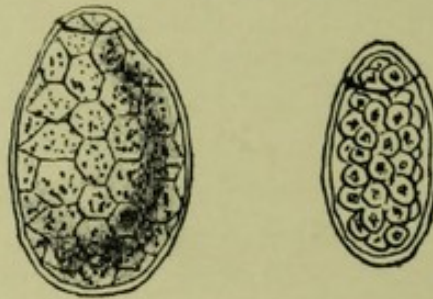


FIG. 78. EGGS OF THE DISTOMA HEPATICUM
X 200



FIG. 79. FREE SWIMMING EMBRYO
OF DISTOMA HEPATICUM

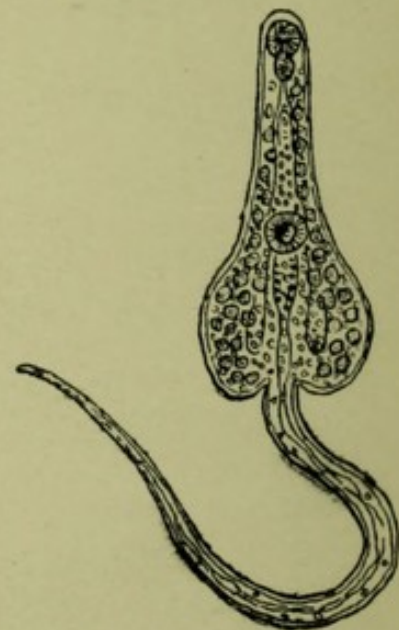


FIG. 80. CERCARIA FORM
OF DISTOMA HEPATICUM

APPENDIX

CHEMICAL SYMBOLS AND APPROXIMATE ATOMIC WEIGHTS OF SOME OF THE ELEMENTARY BODIES

Aluminium .	Al	...	27·5	Manganese .	Mn	...	55·0
Arsenic .	As	...	75·0	Mercury .	Hg	...	200·0
Barium .	Ba	...	137·0	Nitrogen .	N	...	14·0
Calcium .	Ca	...	40·0	Oxygen .	O	..	16·0
Carbon .	C	...	12·0	Phosphorus .	P	...	31·0
Chlorine .	Cl	...	35·5	Potassium .	K	...	39·0
Copper .	Cu	...	63·2	Silver .	Ag	...	108·0
Hydrogen .	H	...	1·0	Sodium .	Na	...	23·0
Iodine .	I	...	126·6	Sulphur .	S	...	32·0
Iron .	Fe	...	56·0	Tin .	Su	...	118·0
Lead .	Pb	...	206·5	Zinc .	Zn	...	65·0
Magnesium .	Mg	...	24·0				

WEIGHTS AND MEASURES

1 gramme = 15·432 grains.	1 square metre = 10·764 square feet.
1 litre = 1000 cubic centimetres.	1 cubic metre = 1000 litres.
35·3 fluid ounces.	= 35·3 cubic feet.
61·027 cubic inches.	1 cubic foot = 6·23 gallons.
1 metre = 39·37 inches.	1 kilogram = 2·204 pounds.

ALCOHOL TABLES. (After Allen)

Specific gravity taken at 15.5° C.

Absolute Alcohol—percentage by weight $\times 1.26$ gives percentage by volume.

Specific gravity.		Alcohol percentage by weight.	Specific gravity.		Alcohol percentage by weight.
.79384	...	100.00	.879	...	67.13
.797	...	98.97	.882	...	65.83
.800	...	98.03	.884	...	65.00
.803	...	97.03	.886	...	64.13
.806	...	96.03	.889	...	62.82
.809	...	94.97	.891	...	61.92
.812	...	93.92	.893	...	61.08
.815	...	92.81	.896	...	59.83
.817	...	92.07	.898	...	58.95
.820	...	91.00	.900	...	58.05
.823	...	89.92	.902	...	57.21
.826	...	88.76	.905	...	55.86
.828	...	87.96	.907	...	54.95
.831	...	86.81	.909	...	54.00
.833	...	86.04	.911	...	53.13
.836	...	84.88	.914	...	51.79
.8382	...	84.00	.916	...	50.96
.841	...	82.92	.918	...	50.09
.843	...	82.15	.919	...	49.24
.846	...	80.96	.920	...	48.96
.848	...	80.13	.922	...	48.05
.851	...	78.92	.925	...	46.91
.853	...	78.12	.927	...	46.00
.856	...	76.88	.929	...	45.09
.858	...	76.04	.931	...	43.95
.860	...	75.14	.933	...	43.00
.863	...	73.79	.935	...	42.05
.865	...	72.96	.937	...	41.05
.867	...	72.09	.939	...	40.05
.870	...	70.84	.941	...	39.05
.872	...	70.04	.943	...	37.94
.874	...	69.21	.945	...	37.11
.877	...	67.96	.947	...	36.00

APPENDIX

161

Specific gravity.		Alcohol percentage by weight.	Specific gravity.		Alcohol percentage by weight.
'949	...	35'00	'977	...	15'00
'951	...	34'05	'978	...	14'00
'953	...	32'87	'980	...	13'00
'954	..	31'94	'981	...	12'00
'956	...	31'00	'982	...	11'00
'958	...	29'87	'983	...	9'99
'959	...	28'87	'985	...	8'89
'961	...	27'93	'986	...	7'99
'962	...	26'87	'988	...	7'02
'964	...	25'86	'989	...	6'02
'965	...	25'00	'991	...	5'01
'966	...	24'00	'992	...	4'51
'967	...	23'00	'993	...	3'49
'969	...	22'00	'994	...	3'02
'970	...	21'00	'995	...	2'51
'971	...	20'00	'996	...	1'99
'972	...	19'00	'997	...	1'51
'974	...	18'00	'998	...	1'02
'975	...	17'00	'999	...	0'53
'976	...	16'00	1'000	...	0'00

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