nitrogen-free water is not always available in large quantities in a sewage laboratory. In this laboratory two gallons and a half of nitrogen-free water can be made in about four and one-half hours. Starting with the water in the flasks cold, eight Kjeldahls can be distilled in about fifteen minutes. Figuring on this basis the cost of distilled water necessary for the direct method by Kimberly and Roberts' procedure is about one-fifth greater than by the distillation process, and by Whipple's procedure, about three times as great. If many determinations are to be made by the latter method the saving in the cost of water used would in a short time pay the cost of a still.

In the direct method the digestate has to be made up to volume at least twice and a definite amount measured twice, while in the distillation method but one measurement is necessary. The chance for error in manipulation is therefore four times as great by the direct as by the distillation method.

Conclusions.

The direct Kjeldahl method undoubtedly has its own place in sewage work, but it does not seem as if it should take the place of the distillation method in a permanent sewage laboratory handling many samples because of:

1. The greater amount of bothersome and bulky apparatus necessary.
2. The large amount of nitrogen-free water required.
3. The greater chance for error in manipulation.
4. The necessity of having the excess of caustic within narrow limits to avoid turbidity, this practically requiring a rough titration of each determination.
5. The greater length of time required for the determination.

The method, however, is without doubt an excellent substitute for the distillation method in a temporary laboratory where it is necessary to incur the least possible expense for apparatus or in a small laboratory where but a very few determinations are to be made daily.

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STUDIES OF INCUBATION TESTS.

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For the past seven years incubation tests have been made in the laboratories at the Lawrence Experiment Station to determine the stability of the effluents of contact and trickling filters. These studies have shown that the development of odor is, perhaps, the surest proof of putrescibility. Oxygen consumed and oxygen dissolved tests before and after incubation are of value but are sometimes contradictory. The so-called methylene-

blue test has been used to a considerable extent, but studies have shown that according to this test, some samples are putrescible, which, judging from the odor test only, are stable. In following out the work with this methylene-blue test, quite a number of dyes have been experimented with to find, if possible, one which would decolorize only when putrefaction as determined by odor, had also taken place. The dyes tested in this way were indigo carmine, methylene green, congo red, alizarin blue, acid brown, alkali blue, acid violet, aniline yellow, curcumein, ponceau red, martius yellow, methyl orange, tropaeolin, coccinine, toluidine blue and azo blue. Of these dyes, indigo carmine and methylene green are more readily decolorized than methylene blue, as will be shown later. Congo red, methyl orange and tropaeolin were the only others affected, but were too stable to use as an indicator of putrescibility in an incubation test. The results with indigo carmine and methylene green, however, looked promising enough to study.

During these studies, a test for hydrogen sulphide in the sample incubated, was made in connection with the other putrescibility tests. The test used is based on the formation of methylene blue from dimethyl-*p*-diamino-benzene sulphate in the presence of hydrochloric acid, hydrogen sulphide and ferric chloride, and by this method approximately 0.01 part of hydrogen sulphide per hundred thousand can readily be determined.

Samples of the effluents tested were incubated at 80° F. with methylene blue, methylene green and indigo carmine in bottles with tightly fitting stoppers, a blank being incubated at the same time. In each case enough dye was added to the sample tested to give it a decided color. The amount of dye used, however, within reasonable limits is not important, since samples with twice the amount of dye usually used were decolorized in the same period of time. Hydrogen sulphide was determined before and after incubation and the time required to decolorize the three dyes by incubation of the samples noted.

The procedure followed for the determination of hydrogen sulphide was as follows: Standards were made up in 100-cc. Nessler tubes from hydrogen sulphide water, the strength of which was determined by titration with iodine, and 2 cc. of strong hydrochloric acid containing $^1/_2$ per cent. ferric chloride and 1 cc. of a 1 per cent. solution of dimethyl-*p*-diamino-benzene sulphate. With small amounts of sulphide about thirty minutes are required for a good color to develop. Reagents were added to the proper amount of the samples to be tested, these samples being, also, in 100-cc. Nessler tubes and the colors developed in the samples compared with the standard colors. It was found that the same set of standards might be used for several weeks without change, and it was also found that equal amounts of reagents must be added to standards and to samples in order that the same shade of blue be obtained.

The following table shows the results of incubation of nineteen samples which did not putrefy. In these samples hydrogen sulphide did not form or odor develop, and with two exceptions, none of the three dyes was decolorized. Following this is a table showing the results of incubation of samples, all of which did putrefy.

TABLE SHOWING RESULTS OF INCUBATION OF NON-PUTRESCIBLE SAMPLES.

Effluent of Filter No.	Number of days incubated.	Hydrogen sulphide. Parts per 100,000. At start.	After incubation.	Methylene blue. Methylene green. Indigo carmine.	Appearance, odor, etc.
233	5	0.01	0.01	Not decolorized	No change
248	5	0.01	0.00	" "	" "
175	5	0.03	0.00	" "	" "
235	5	0.00	0.00	" "	" "
233	6	0.01	0.01	" "	" "
235	6	0.01	0.00	" "	" "
251[1]	6	0.01	0.01	" "	" "
136[1]	6	0.01	0.00	" "	" "
248	5	0.01	0.01	Decolorized in four days	Very slight increase in odor
251	6	0.00	0.00	Decolorized in four days	Very slight increase in odor
175	5	0.04	0.01	Not decolorized	No change
135	5	0.00	0.00	" "	" "
233	6	0.01	0.01	" "	" "
235	6	0.00	0.00	" "	" "
248	6	0.01	0.00	" "	" "
235	5	0.00	0.00	" "	" "
248	5	0.01	0.01	" "	" "
176	6	0.01	0.01	" "	" "
175	5	0.01	0.00	" "	" "

The results of the incubation of the 26 samples which may be said to have been putrescible are shown in the table on the next page.

Examining this second table, it will be noticed that in every case if one dye was decolorized the other two were also, and that the average time required for indigo carmine to be decolorized was 2 days; for methylene green, $2^1/_2$ days and for the methylene blue, nearly 4 days. In every sample tested after incubation there was an increase in the amount of hydrogen sulphide present, but no close relation could be noted between the amount of hydrogen sulphide formed and the appearance and odor after incubation.

It is probable that the hydrogen sulphide is largely formed from the decomposition of albuminous compounds in the effluents tested. Several samples shown in the above table were, as designated in the table, incubated with the addition of one part sulphur, as potassium sulphate,

[1] One part sulphur as potassium sulphate added before incubation, had no effect.

TABLE SHOWING RESULTS OF INCUBATION OF PUTRESCIBLE SAMPLES.

Effluent of filter No.	Days required to decolorize.			Hydrogen sulphide. Start. Parts per 100,000.	After incubating.			
	Indigo carmine.	Methylene. Green.	Methylene. Blue.		Days.	Hydrogen sulphide. Parts per 100,000.	Appearance.	Odor.
176	1½	2	5	0.03	5	0.40	sl. black	str.
176	1	1	2	0.01	2	0.05	....	d.
					5	0.10	....	d.
221	2	4	4		4		black	str.
221	2	1	3		3		black	str.
221	1	1	3		3		black	str.
221	1½	3	5	0.03	5	0.20	black	str.
221	¾	5	5	0.02	5	0.28	black	str.
221	2	4	4	0.05	4	0.25	black	str.
233	4	5	6		6		....	d.
233	5	4	6		6		....	d.
233	2	3	3	0.01	3	0.05	....	d.
247	2	2	4		4		black	str.
247	2	1	4		4		black	str.
247	1	¾	2		2		black	str.
247	1	¾	3		3		black	str.
247	1½	1½	5	0.02	5	0.80	sl. black	str.
247	¾	¾	¾	0.01	¾	0.06	....	str.
					5	0.30	black	str.
247	¾	3	3	0.01	3	0 25	black	str.
					5	0.25	black	str.
247	2	3	5	0.01	5	1 50	black	str.
247	3	4	4	0.02	4	0.35	black	str.
247	1	¾	3		3		black	str.
248	3	2	3		3		....	d.
251	3	¾	5	0.01	5	0.15	....	str.
251	3	4	6	0.01	6	0.20	....	d.
251	4	4	4	0.01	4	0.08	....	d.
					5	0.22	....	d.
251		Not decolorized		0.01	5	0.40	....	d.

and these showed no increase in the hydrogen sulphide formed above that formed in duplicate samples without the addition of the sulphate; on the other hand, a 1 per cent. solution of peptone in distilled water, seeded with one drop of sewage, developed 2.5 parts of hydrogen sulphide by five days' incubation. However, if putrefaction occurring is great, inorganic sulphates may be reduced as happened in the following experiment: Twelve bottles were filled with *sewage*. To six of them 1 part of sulphur as potassium sulphate, was added and incubated for one, two, three, four, seven and eight days, respectively. The hydrogen sulphide formed is shown in the following table, and it will be noticed that after three days' incubation there was a rapid development of hydrogen sulphide in the samples to which potassium sulphate had been added.

HYDROGEN SULPHIDE.

(Parts per 100,000).

Days incubated.	Sewage only.	Sewage + sulphate.
Start	0.00	0.00
1	0.04	0.04
2	0.10	0.10
3	0.30	0.35
4	0.35	1.20
7	0.10	2.50
8	0.10	2.50

One or two samples of filter effluents decolorized all of the dyes in four days without the production of hydrogen sulphide or odor, and, on the other hand, one sample showed a considerable development of hydrogen sulphide and odor without decolorizing in 7 days. These two or three results were, of course, abnormal and simply show that absolute reliance cannot be placed on incubation results obtained by the methylene-blue test; in fact, these studies have shown (1) that the degree of putrescibility of such effluents as experimented with can probably be better estimated by odor and appearance after incubation than by the time required to decolorize dyes; (2) the hydrogen sulphide formed comes very largely from albuminous compounds in the effluents and the amount formed is, to some degree, a measure of the putrescibility of the sample tested; (3) on the whole, it would seem that if a putrescibility test of the methylene blue kind is to be adopted, equally good results can be obtained in a shorter time by the use of indigo carmine or methylene green.

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NOTE.

The Quantitative Determination of Arsenic by the Gutzeit Method.—In the issue of *Chemical Abstracts* for April 10, 1908, p. 976, is an abstract of a note by T. F. Harvey on the estimation of arsenic by the Gutzeit test. As this immediately follows the abstract of an article by Sanger and Black on the quantitative determination of arsenic by the Gutzeit method, the casual reader may be led to infer that Sanger and Black were anticipated by Harvey in the method published by them.

I have already called the attention of the editor of the *Journal of the Society of Chemical Industry* to the misleading nature of Harvey's article, and Mr. Harvey himself has assured me that it is quite clear to him that his work had not come to our notice. The Harvey method, however, is merely a quantitative treatment of the ordinary Gutzeit test, while the paper of Sanger and Black not only introduces a different