## THE COLON BACILLUS IN GROUND WATERS'.

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DURING the past four years it has been part of our routine work to examine samples of ground waters from wells and springs in various portions of Ohio, and, in so doing, organisms have been obtained from time to time conforming to the tests usually given as differentiating faecal bacteria.

The mere finding of *B. coli* in ground waters is not new, as its presence in several cases has been mentioned by other workers<sup>2,3</sup>, but we desire especially to call attention to the significance of the presence of *B. coli* in ground waters, *i.e.* ground waters derived from springs or drilled wells.

Naturally there have been many samples from dug wells in the waters of which the colon bacillus was present, but in the present article we omit all reference to such samples, and include only those derived from springs and drilled or bored wells that at the same time contained *B. coli*. The list includes 25 samples from wells varying in depth from 16 to 243 feet, although most of the wells were from 25 to 80 feet deep. Almost without exception these wells were cased, and with the casing extending above the level of the ground, thus preventing the direct entrance of surface water. The list also shows 12 samples of water from springs in which the colon bacillus was found.

<sup>1</sup> Read at the meeting of the Laboratory Section of the American Public Health Association, New Orleans, La., Dec. 8, 1902.

<sup>2</sup> Weissenfeld, Zeitschr. f. Hygiene, Vol. xxxv. p. 78.

<sup>3</sup> Savage, Journ. of Hygiene, Vol. 11. p. 320.

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Part of the springs were open to surface pollution, while others appear to have been well protected.

With a few of the samples the analysis was bacteriological only, but generally there was also a more or less complete sanitary chemical examination. The number of bacteria per cubic centimetre varied from 16 to 900,000 in the well waters, and from 90 to 400,000 in the samples from the springs. Some of the chemical and bacterial analyses gave marked evidence of sewage pollution, indicating a somewhat open course from some sewage source to the water-producing strata in question. In other cases the chemical evidence of pollution was small, while in still others there was no chemical evidence of sewage pollution, and the waters would have been passed without question had there been only a chemical analysis.

Since some of the waters under examination were those proposed as public supplies for villages or cities, and some were from localities where typhoid fever was or had been present, we did not feel warranted, at the present time, in accepting as final any of the gross, or preliminary, or presumptive tests, although such were made use of in connection with the more laborious isolation of species and identification by cultivation on various media, etc. The final conclusion depended almost entirely on the results obtained by the latter process.

During the period of this study different methods have been resorted to for the isolation, and much of the time two or more methods were employed simultaneously. The methods have been as follows:

(a) Preliminary cultivation in Dunham's peptone, using 40 or 90 c.c. of the water in question, and followed by plating to lactose-litmus-agar (or in a few cases to gelatin) from which colonies were fished and subcultivations made.

(b) Cultivation in glucose broth, using 40 or 90 c.c. of water with subsequent plating, etc.

(c) Same as (b) except that 1 c.c. of the water was used.

(d) Cultivation in carbolated sugar broth, using 40 or 90 c.c. of water with subsequent plating, etc.

(e) Same as (d) except that 1 c.c. of water was used.

After plating, it was customary to fish from four to six colonies and test as follows: morphology, motility, litmus milk, nitrate solution, peptone for indol, neutral-red, gelatin stab, gas production in sugar broth, gas formula of 2:1. A portion of the above individual tests were omitted with some of the samples, but not with the more particular ones.

In view of the work by Prescott<sup>1</sup> on B. coli and allied forms, the question arises as to whether the organisms isolated were colon bacilli, and, accordingly, other than bacteriological evidence was sought in the history of these waters in relation to typhoid fever among the users. Of the 37 samples, 27 gave a history of typhoid fever, the number of cases per well ranging from 1 to 16, and from 1 to 17 for the springs. Of the remaining samples two were from springs that could hardly escape animal pollution. With the evidence at hand we feel warranted in saying that the organisms isolated were colon bacilli, and that the presence of this organism is at present to be looked upon in spring waters with grave suspicion, unless there is known opportunity for surface pollution carrying waste material from the lower animals. The results obtained lead us to believe that the presence of B. coli in waters from drilled wells ought in most cases to condemn the use of that water for domestic purposes.

An exceptional case may be stated here. A drilled well 35 feet in depth was put down for the proposed supply of a village near the central portion of the State. On examining the water the more important chemical findings were as follows in parts per million: oxygen consumed, 46; N as albuminoid ammonia, 039; N as free ammonia, 014; nitrites, none; nitrates, trace; chlorine, 33.1. (It should be stated that the chlorine is not a close indicator in some parts of Ohio owing to the presence of salt or salt waters.) The number of bacteria per c.c. was 18. In the qualitative bacterial examination, each of the above individual tests was applied to the organisms isolated with positive results, *i.e. B. coli* was present. A second sample a month later gave the same organism, but in view of the excellent chemical analysis, the low number of bacteria, and the unqualified recommendation of the Civil Engineer of the State Board of Health, who had made a personal inspection of the locality, the sample was allowed to pass. Subsequently an examination was desired, but unfortunately the portion for bacterial examination was not taken by the collector, and only a chemical analysis was made. This analysis showed even better results than the former one.

<sup>1</sup> Prescott, *Abstract in Science*, N.S. xv. p. 363 (1902). (Also a paper read at New Orleans, La., American Public Health Association Meeting, Dec. 8, 1902.)

## CONCLUSIONS.

1. We believe the organism found in these waters from springs and drilled wells was *B. coli*.

2. The presence of *B. coli* in spring water should be looked upon with suspicion, unless there is apparent opportunity for pollution by the lower animals.

3. The presence of B. coli in water from a drilled well should generally condemn that water for domestic use.

4. The fact that a water is derived from a drilled well should not be taken as an absolute guarantee that it is a potable water.

5. Both chemical and bacterial examinations of a water are desirable, but the bacterial will sometimes prove the more delicate indicator of direct pollution, which is the more dangerous pollution.