STUDIES IN THE BACTERIOLOGY OF WATER*

By Denis B. Wood

IT is well known that bacteriological examinations of water supplies are directed to the detection and enumeration of what are known as "indicator organisms"—that is to say, organisms which are always to be found in large numbers in the excreta of man and animals. The presence of such organisms in water indicates potential danger to the health of consumers, for individuals suffering, or having suffered, even at some remote time, from typhoid fever and kindred diseases may void the specific organisms in large numbers in their excreta.

Contamination of water with excretal matter derived from animals is of less potential danger, for the lower animals are not to anything like the same extent subject to these diseases. They are known, however, to be subject to infection with the Gäertner group and paratyphoid bacilli, and with animal parasites, though such infections are rare, and contamination with animal excretal matter must be regarded as undesirable. Moreover, as we have as yet no means of distinguishing between contamination with human and animal excretal matter, so similar are their bacterial flora, we are obliged to take a safe course and regard all waters showing definite evidence of excretal contamination as more or less unsatisfactory.

There are three groups of "indicator organisms": (1) The B. Coli group, (2) the Streptococcus group, and (3) the Enteritidis Sporogenes group. The average numbers in which they are present in one gramme of human fæces are:—

B. coli.	Streptococci.	Spores of B. Enteritidis Sporogenes.
100-1,000 million.	100-1,000 million.	1-10 million.

It is evident, therefore, that tests based on the enumeration of B. coli and Streptococci will be the most sensitive. In this paper only these two groups will be considered.

The B. Coli Group

Members of the B. coli group have one characteristic in common—viz., the fermentation of lactose with production of acid and gas. The study of this group reveals the fact that it can be divided and subdivided into a number of types.

From MacConkey's result it appears that 87% of the lactose-fermenting organisms present in human fæces all produce indole, acid and clot in milk, do not liquefy gelatine nor give the Voges and Proskauer reaction. In other words, they possess, practically speaking, the cultural characters of Houston's "typical B. Coli." Houston found that about 85% of the lactose fermenters present in human fæces, and 65 to 85% of those present in sewage, possessed these characteristics.† The results of his investigations on the viability of members of the B. coli group in soil and water show that the atypical varieties persist for longer periods than the typical, and are therefore less characteristic of recent contamination. Houston's definition of typical B. coli is admirably suited to routine bacteriological examination, and has been very generally adopted as a basis for bacteriological reports.

The Streptococcus Group

It sometimes happens that in a water examination only lactose fermenters having the characteristics of Houston's "typical B. coli" are to be found. It is then a great advantage to have recourse to another test, and the most satisfactory corroborative test is based on the enumeration of streptococci. Savage and Read have shown that the majority of waters showing evidence of contamination on the B. coli basis contain streptococci.

In calculating percentages nothing is to be gained by working out the percentage of water-samples which contain streptococci or B. coli respectively in the different amounts. The percentage results are worked out in two ways. In one way each group of B. coli prevalence is taken separately and the percentage prevalence of each group of waters on a streptococcus basis is calculated. In the other way each group of streptococci prevalence is taken separately, and the percentage prevalence of the waters on a B. coli basis is recorded.

An example will make this clear. Take, for instance, 118 samples from deep-water supplies containing B. coli in 10 or 30 cm.; 58 or 49.2% contained no streptococci. Or take the 198 samples from deep-water supplies containing no streptococci; 64 or 32.3% contained B. coli. The mean of these two numbers (49.2 and 32.3) is 40%, and gives a fair average of the number of samples in which B. coli may be present in only moderate numbers and streptococci absent. In other words, the chances are rather more than even that streptococci will be found, and if not found in one sample they probably will be in the next, or at any rate in subsequent examinations. This correlation between B. coli and streptococci is a point the author particularly wishes to emphasise. Apparently the value of this test as carried out by Savage has not been fully appreciated.

The streptococcus group can also be differentiated into a variety of types by fermentation reactions; those producing acid in lactose are particularly typical of excretal contamination. Differentiation of this group, however, is seldom practised.

Index of Recent Contamination

The streptococcus enumeration is particularly valuable as an index of recent contamination. This was shown by the results of an investigation carried out by the present author in conjunction with Dr. W. G. Savage on the relative viability of B. coli and streptococci in water under conditions closely resembling those of a well. In this investigation an earthenware pipe cemented at the bottom and holding about 40 litres of water was used as the model well. The water was contaminated with excretal matter emulsified with water, or with sewage, and examinations were made at regular intervals to ascertain how rapidly the organisms diminished in numbers and finally disappeared.

These experiments with excreta or sewage added to a large bulk of water yield minor differences in the individual experiments, but in general they show a rapid diminution and elimination of the streptococci and a continuous, yet not quite so rapid, diminution in the number of B. coli.. With the latter it was more common to find persistence in small numbers for a period extending to many weeks. The elimination of the streptococci was particularly uniform and rapid. At the end of three weeks in only two experiments were they present in more than insignificant numbers.

The decline curves of both organisms agree very closely, as could be seen most readily when the figures were plotted out as graphs.

Comparison with Typhoid Bacillus

Comparison with the viability of the typhoid bacillus is of special interest. Sir A. C. Houston's investigations in the viability of uncultivated typhoid bacilli in river water showed that they died out within three weeks, even when as many as 38,000 were present per c.c. of the infected water.

Several lactose-fermenting organisms isolated from various sources gave the Voges and Proskauer reaction, very few of which—only 6.3%—were represented in human fæces. These types have recently attracted the attention of American bacteriologists, and have been shown by them to be rare in the excreta of man and animals, more common in sewage and surface water, and the predominant types in soil and grains. They can be distinguished in the laboratory by two tests—(1) the methyl-red test, and (2) the Voges-Proskauer reaction, alluded to above. These two tests are very closely correlated, and by them the lactose-fermenting organisms can be divided into two main types, which are known as—

*From a paper read at the annual summer meeting of the Institution of Water Engineers, England.

†Since the large majority of organisms fermenting lactose and producing indole fulfil the other tests also, according to Houston's later definition, the other tests are dispensed with in routine examinations.