Table III.—The Effect of an Increased Period of Incubation in the Presumptive Test Upon the Possible Number of Confirmatory Tests

Total Number of Tubes Incubated and Giving Gas = 495

Gas Formation	Number Tubes Showing Gas Total Additional		Per Cent. Tubes Showing Gas Total Additional		Number Confirmed Total Additional		Per Cent. of Total Tubes Confirmed	Confirmed
at End of	Total		2.6	3.6	17	17	6.2	3.4
24 hours	18	18	3.6		197	180	66.0	36.0
48 hours	263	245	53.1	49.5	-		24.6	13.4
	448	185	90.5	37.4	264	67		
72 hours		The second secon	100.0	9.5	273	9	3.3	1.8
96 hours	495	47	100.0			1 - J Chata	Department	of Health dur

Note—Data obtained from routine analytical determinations in laboratory of Maryland State Department of Health during 1917. Presumptive tests in lactose broth. Confirmatory tests consisted of Endo, secondary lactose broth, and agar slant.

approximately 60 per cent., of the laboratories in question have seen fit to use this exact procedure as a routine measure. The percentage is undoubtedly higher than that which would represent the individual opinions of the analysts in these laboratories, in view of the fact that some of them have adopted the aforementioned methods on account of their official stamp rather than as a result of the conviction that they are superior to others. This conclusion is borne out by the fact that it has been by no means firmly established that the bacterial count, obtained as outlined by the Federal requirements, serves as the best index to the quality of a drinking water. In the light of the data illustrating the wide discrepancy in the method of obtaining the bacterial count, it would appear that effort should be directed towards further study of individual types of bacteria and their relative significance rather than towards an attempt to predicate a standard upon such an elusive and variable factor as the general bacterial count.

## B. Coli

An index to sewage pollution in potable waters is an excellent asset in determining the safety of a supply if it "indicates." Some years ago, perhaps, the presence of B. coli in small quantities of water was considered sufficient evidence upon which to condemn the supply. He certainly would be venturesome who would issue a manifesto to-day as to the allowable frequency of B. coli in a safe water. He would indeed be skilled who can gather sufficiently consistent data out of the present chaotic conception of the significance of B. coli, and of how to obtain it, to be able to establish even a fairly elastic measure of quality.

Table II. illustrates, for instance, that the use of a medium for testing even the elementary phenomenon of gas formation is still open to question, while the significance of gas formation itself is disputed by authorities. Considerable evidence had supported previously the use of lactose bile, but the wind has apparently shifted in recent years and the balance now rests upon the importance of lactose broth as a better medium for an initial B. coli test. Each day brings forth another experimental factor to make the confusion greater as to the significance of lactose fracture.

The data given in Table II. show a close agreement in the laboratories as to the necessary period of incubation in the B. coli presumptive test. In the face of the almost universal choice of a 48-hour period, it is found in the Maryland State Department of Health laboratory that about 25 per cent. of all typical B. coli isolations are obtained from those tubes which show gas only after 72 hours incubation. It is somewhat doubtful, with the evidence shown in Table III., whether even the apparently settled question of period of incubation is not still debatable.

## Uncertainty of Fermentation

In 1907, Phelps discussed a method of estimating the numbers of B. coli from fermentation tube results. His system of numerical interpretation has served, until recently, as a basis of practically all quantitative estimates of B. coli in various waters. A misconception of the method proposed at that time has been responsible in a degree for the eternal cry for standardization. The realization of the fact that "the method is obviously of no value for single tests and finds its most useful application in routine studies in water purification and sewage treatment extending over long periods of time" would tend to emphasize the utility, but uncertainty, of fermentation. Granting the value of establishing a maximum allowable pollution of "x" B. coli per 100 c.c., we are confronted still with the difficulty of estimating such numbers from data afforded by our present bacteriological methods.

## Frequent Sampling Necessary

Past standards for B. coli content have shown a surprisingly patent disregard of the importance of stipulating the necessary frequency of sampling of a source before its quality may be safely postulated. In fact no standard comes to mind at the present time in which the number of samples is apparently considered of sufficient importance to warrant even a cursory establishment of a necessary minimum. In certifying a public water supply to com-

Table IV.—Number of Samples Necessary to Establish a
B. Coli Content to Within the Probable Errors
of 5 and 10 Per Cent.

(From Stein, "Engineering News-Record," May 24, 1917.)

Per Cent of PosiNo. of Samples to Establish Coliper c.c. to Probable Error of

10 15 20 25 30 35 40 45 50	900 760 627 485 365 320 282 235 204	3,600 3,040 2,508 1,940 1,460 1,280 968 940 816 760 684
		1,280
	235	
	190	
60	171	CONTRACTOR OF THE PARTY OF THE
. 65	165	660
70	162	648
75	155	620
80	156	624
85	. 160	640
90	178	712
95	210	840