

Ratio. In these columns are given the ratios between the bacterial counts on the same plates at the different periods of incubation, as indicated. These ratios indicate the relative increases under the different conditions.

Original sample, untreated, shows the bacterial count in the control sample, before dilution with sterile water, and corresponds also to the count before treatment in the Toronto city water or in the water used in the laboratory experiment.

Experiment No. 1.
Toronto City Water.

Treatment with available chlorine equals 0.25 part per million. City plant.

Plated.		Count, after					Ratio.	
Time.	Day.	48 hours.	72 hours.	96 hours.	120 hours.	144 hours.	2:6 days.	2:5 days.
10	1	17,000	21,440	liquid	11,880	—	—	—
12	1	1,570	5,820	12,000	—	11,960	1:6.9	—
2	1	752	630	1,250	3,480	3,680	1:4.9	—
4	1	692	1,440	2,910	4,520	5,240	1:7.5	—
10	2	165	410	620	930	—	—	1:5.6

Control, lagoon.

Plated.		Count, after					Ratio.	
Time.	Day.	48 hours.	72 hours.	96 hours.	120 hours.	144 hours.	2:6 days.	2:5 days.
10	1	260	296	338	380	404	1:1.5	—
12	1	254	304	310	384	412	1:1.6	—
2	1	246	244	236	368	432	1:1.7	—
4	1	318	474	528	510	540	1:1.7	—
10	2	12,880	12,440	13,960	14,000	—	—	1:1.08
10	3	288,000	508,000	630,000	—	—	—	—

Original sample, untreated, 23,700.

Experiment No. 2.
Toronto City Water.

Treatment with available chlorine equals 0.25 part per million. City plant.

Plated.		Count, after					Ratio.	
Time.	Day.	48 hours.	72 hours.	96 hours.	120 hours.	144 hours.	2:6 days.	2:5 days.
10	1	14,000	18,800	28,000	22,800	26,000	1:7.6	—
12	1	3,400	5,700	16,200	5,000	7,100	1:13.6	—
2	1	520	2,100	3,440	3,020	5,200	1:7.0	—
4	1	740	1,900	2,860	3,720	—	—	1:7.4
10	2	500	2,300	2,480	—	—	—	—
4	3	1,120	1,910	—	—	—	—	—
10	3	2,400	4,150	9,300	—	—	—	—

Control, lagoon.

Plated.		Count, after					Ratio.	
Time.	Day.	48 hours.	72 hours.	96 hours.	120 hours.	144 hours.	2:6 days.	2:5 days.
10	1	180	180	268	liquid	—	—	—
12	1	124	138	256	230	238	1:1.9	—
2	1	136	118	140	176	188	1:1.4	—
4	1	210	158	240	318	340	1:1.6	—
10	2	6,800	7,900	10,800	12,300	—	—	1:1.8
4	3	25,200	26,400	28,400	—	—	—	—
10	3	640,000	866,000	960,000	—	—	—	—

Original sample, untreated, 22,800.

Experiment No. 3.

Control, laboratory tap water.

Plated.		Count, after					Ratio.		
Time.	Day.	48 hours.	72 hours.	96 hours.	120 hours.	144 hours.	2:6 days.	2:5 days.	2:4 days.
11	1	121	134	285	liquid	—	—	—	—
12	1	115	171	223	380	—	1:3.4	—	—
2	1	109	122	221	362	375	1:3.4	—	—
4	1	121	172	251	410	415	1:3.4	—	—
10	2	6,200	8,500	8,800	8,900	—	—	1:1.4	—
10	3	425,000	650,000	670,000	—	—	—	—	1:1.5

Available chlorine equals 0.1 part per million. Treatment in laboratory.

Plated.		Count, after					Ratio.		
Time.	Day.	48 hours.	72 hours.	96 hours.	120 hours.	144 hours.	2:6 days.	2:5 days.	2:4 days.
11	1	520	940	1,350	2,380	2,780	1:5.3	—	—
12	1	390	770	1,080	2,040	2,320	1:5.8	—	—
2	1	187	260	690	1,540	2,080	1:10.4	—	—
4	1	91	130	280	760	840	1:9.2	1:22	—
10	2	42	120	670	920	—	—	—	1:10.9
10	3	320	1,210	3,500	—	—	—	—	1:2.9
10	4	8,700	14,200	26,000	—	—	—	—	—

Available chlorine equals 1.0 part per million. Treatment in laboratory.

Plated		Count, after				
Time	Day	48 hours	72 hours	96 hours	120 hours	144 hours
11	1	2	5	7	8	10
12	1	1	1	2	2	4
2	1	0	0	0	2	2
4	1	1	2	2	6	6
10	2	0	0	0	1	—
10	3	0	0	—	—	—
10	4	5	13	16	—	—
10	5	79	166	—	—	—

It is evident that chlorine, in quantities smaller than 0.25 part per million, under the conditions of the experiments, mainly retards the growth of bacteria and kills very few.

One part of available chlorine per million acts as a germicide, and there is no evidence of revivication. Since small quantities retard the growth when placed in such a suitable media as nutrient gelatin, it is probable that reproduction in water with a low organic content would be still further diminished. This is also indicated by the results.

From the results of these experiments and many others of a similar nature, it was decided to abandon the two-day incubation period recommended by the American Public Health Association and to count all plates on the third day. Three days is the period during which the formation of visible colonies proceeds most rapidly, and very little advantage can be gained by increasing it to four days. A three-day incubation period raises the standard of quality if numbers are given any consideration, and eliminates the anomalous results so often found when counts are made of chlorinated waters.

In this connection it is interesting to note the experience of Clark and De Gage, at Lawrence, Mass., during 1909. In the report of the Board of Health, page 319, 1910, they state: "It has frequently been observed that the numbers of bacteria determined at body temperature in the disinfected samples were much higher than the numbers determined at the usual room temperature. This phenomenon of reversed ratios between counts at the two temperatures has been occasionally observed with natural waters but a study of the records of many thousands of samples shows that the percentage of such samples is very small, not over 3 to 5 per cent. On the other hand 20 to 25 per cent. of samples

Available chlorine equals 4.0 parts per million. Treatment in laboratory.

Plated.		Count, after					Ratio.	
Time.	Day.	48 hours.	72 hours.	96 hours.	120 hours.	144 hours.	2:6 days.	2:5 days.
11	1	1	1	1	4	5	—	—
12	1	0	0	1	3	—	—	—
2	1	0	0	1	0	—	—	—
4	1	0	0	1	0	—	—	—
10	2	0	0	0	—	—	—	—
10	3	0	0	0	—	—	—	—
10	4	0	0	0	—	—	—	—
10	5	1,620	2,230	2,800	5,620	3,840	1:2.3	—

treated with calcium hypochlorite show higher counts at body temperature than at room temperature. Similar counts have been noted elsewhere where waters are being treated with hypochlorites, but in many cases such results have been considered abnormal and have been omitted from the records. A phenomenon which has a frequency of 25 per cent., however, cannot under any circumstances be considered abnormal, and the omission of such counts from records is entirely unjustifiable. A careful study has been made of the conditions under which such reversed counts occur, and it appears that these counts are found in a considerable percentage of samples of water, sewage, etc., in which the room temperature counts have been reduced to less than 100 to 200 per c.c. by the use of hypochlorites, permanganates, and other oxidizing agents. A study of the types of bacteria remaining after disinfection shows that the proportion of spore-forming bacteria is practically the same after disinfection as it was in the untreated water, and the appearance of the reversed ratios apparently cannot be attributed entirely to the non-destruction of spores. The true significance of this phenomenon cannot be stated at the present time. It is evident, however, that, if the body temperature counts are omitted and reliance placed upon those at room temperature, a wrong and possibly a dangerous interpretation may be made as to the quality of the water which has been treated with hypochlorites."

This is possibly due to the cause previously mentioned, and the phenomenon would probably disappear by increasing the period of incubation.