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COLIPHAGES AND BACTERIOPHAGES IN CANADIAN DRINKING WATERS

by

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ABSTRACT

Eighteen samples of treated drinking water and their source waters in three Canadian cities were tested for coliphage, bacteriophage and bacterial content. In the source waters, bacterial concentrations varied from nondetectable to over 5,500 TC per 100 mL, but they were all found to contain coliphage and bacteriophage. After treatment all the drinking waters were found to have residual total and free chlorine levels, to be negative for bacterial indicators, however, all contained coliphage and bacteriophage. Implications of these findings are discussed.

RÉSUMÉ

La teneur en bactéries, en bactériophages et en coliphages a été déterminée dans 18 échantillons d'eau brute et d'eau potable traitée provenant de trois villes canadiennes. Dans les eaux brutes, qui contenaient toutes des coliphages et des bactériophages, les teneurs en bactéries variaient de non décelables à plus de 5 500 CT par 100 mL. Après traitement, toutes les eaux potables renfermaient du chlore libre et total résiduel, ne renfermaient pas d'indicateurs bactériens mais contenaient tous des coliphages et des bactériophages. Les implications de ces résultats sont traitées.

MANAGEMENT PERSPECTIVE

Recent reports on the incidence of coliphage in coliform free drinking water (tap and bottled) in developing countries provides strong support for the inadequacy of the present microbiological indicator system to fully protect the consumers. This was generally believed to only be a problem of developing and third world countries who were not able to provide the technology or funding necessary to ensure adequate water treatment and quality monitoring. However, recent studies in Canada, a developed, scientifically-aware and health caring nation, have shown that the presence of coliphage and bacteriophage in coliform free, treated drinking water is not uncommon. This finding implies that human enteric viruses can also survive the treatment processes given to Ontario drinking waters, and thus a previously unsuspected health hazard may be present in these drinking water supplies. The data and their implications are supportive of the need to install the coliphage test as one of the essential tests for potable water quality testing. Also, research should be initiated to investigate treatment processes which allow for coliphage/bacteriophage survival.

PERSPECTIVE - GESTION

Les rapports récents portant sur la présence de coliphages dans de l'eau potable ne renfermant pas de coliformes (eau du robinet et eau embouteillée) dans les pays en voie de développement illustrent combien les indicateurs microbiologiques utilisés à l'heure actuelle ne protègent pas totalement le consommateur. L'on croyait généralement qu'il s'agissait d'un problème limité aux pays en voie de développement, qui n'avaient ni la technologie ni les fonds nécessaires au traitement adéquat des eaux et à la surveillance de la qualité. Cependant, des études récentes réalisées au Canada, pays industrialisé, soucieux de sa santé, et dont les moyens scientifiques sont nombreux, ont montré qu'il n'était pas rare de trouver des coliphages et des bactériophages dans l'eau potable traitée libre de coliformes. Cela veut donc dire que les entérovirus peuvent aussi survivre aux traitements auxquels sont soumises les eaux potables de l'Ontario; l'eau potable peut donc présenter un risque pour la santé, insoupçonné jusqu'à maintenant. Il est donc nécessaire d'ajouter aux tests essentiels de mesure de la qualité de l'eau potable le test de détection des coliphages. De plus, des études devraient être entreprises en vue d'étudier les traitements qui n'empêchent pas la survie des coliphages et des bactériophages.

There is evidence that viral contamination of water systems throughout the world is increasing, therefore, more attention is being focussed on the evaluation of the associated health hazards and their effective controls (Simkova and Cervenka, 1981). Concomitantly, it has been shown, rather convincingly, that bacteriological indicators of water viral quality are completely inadequate in protecting the public (Berg et al., 1978). This is especially important as the health and well-being of the majority of the world's population is dependent to a large extent on the quality of water they drink.

Several recent reports from developing countries (Sim and Dutka, 1987; El-Abagy et al., 1988; Ratto et al., 1989) on the incidence of coliphage in coliform free drinking water (tap and bottled) provide strong support for the inadequacy of the present bacteriological indicator system to fully protect the users. This was generally believed to only be a problem of developing and third world countries who were not able to provide the technology or funding necessary to ensure adequate water treatment and quality monitoring.

Recent studies in Canada, a developed, scientifically-aware and health-caring nation, have shown the presence of coliphage and bacteriophage in coliform free, treated drinking water. Thus, the finding of coliphage in these drinking water samples without coliform or indicator organism presence, suggests that viruses can also survive the normal treatment and disinfection process accorded these potable water samples (Grabow, 1968; Chamber, 1971; Havelaar, 1986). In this paper we will present examples of results obtained from water distribution lines of the three cities' whose raw water came from 3 different sources.

METHODS

Water Samples

Eighteen water samples were collected from taps in public buildings in three different cities in the province of Ontario, over a two month period. For two of the cities, the source waters were two different lakes in the Canadian Great Lakes System and for the third city, the water source was a small river. Samples were dechlorinated with sodium thiosulphate (APHA, 1985) and maintained at cool temperatures until processing was completed, within a maximum of 4 hr of collection.

Source water samples (18) were collected from pipes immediately prior to their entry into the drinking water treatment plant. Samples were cooled and tested within 4 hours.

Microbiological Tests

All water samples were subjected to the following microbiological population estimation techniques: total coliforms by membrane filtration procedures using Gelman GN6 membranes, LES Endo agar with 24 hour incubation at 35°C; fecal coliforms by membrane filtration procedures using m-Tec agar with 24 hour incubation at 44.5°C; fecal streptococci by membrane filtration procedures using m-Enterococcus agar with incubation at 35°C for 48 hours; and Standard plate count using membrane filtration procedures and modified SPC agar with incubation at 35°C for 48 hours. Details of the microbiological procedures can be found in the Ministry of Environment Handbook of Analytical Methods of Environmental Samples (HAMES, 1984).

Coliphage and Bacteriophage Tests

The procedures detailed by Havelaar (1986) for enumerating bacteriophage from natural water samples, were used in this study. Basically, 2 litres of raw water and 10 L of treated water, for each host used, was filtered through an electropositive filter at pH 6.0 in the presence of 0.005 M imidozole buffer. Elution was conducted using a solution of beef extract at 10 g/L, pH 7.0 with 0.1 M Mg^{2+}. This method was found to underestimate bacteriophage populations by at least 30-50%.

Four bacterial hosts were used in this study, two for coliphage and two for F specific (RNA) bacteriophage detection. The E. coli hosts were E. coli C and E. coli HrfH. The S. typhimurium hosts were WG49 specific for F+ specific (RNA) bacteriophage and WG45 for F-somatic bacteriophage (Havelaar, 1986).

Chemical Tests

The Ontario Ministry of Environment procedures were used to test for total and free Cl_2 (C.P.W.S, 1987).

RESULTS AND DISCUSSION

Examples of typical results found in the distribution systems of the three cities are shown in Table 1. Here it can be seen that in all instances, as well as the other data not shown, total coliform, fecal coliform, fecal streptococcus counts were not detected in 100 mL water samples. The only bacteria detected were those observed on the Standard Plate Count Tests. Also, it can be seen that in all the samples presented as well as those not shown, coliphage and bacteriophage were present before treatment and after treatment. It should also be noted that the coliphage and bacteriophage levels after treatment were equal to, less than and greater than intake levels. However, it must be recognized that the intake and treated water samples were usually collected within a 20-30 minute period, except for sample site A which was approximately 50 kilometres from the treatment plant, thus the water masses being tested were not the same, but are representative of the system.

A striking feature of these data was the 100% recovery of four different types of bacteriophage even when total and free chlorine levels were greater than 2.70 ppm, a level rarely, if ever, encountered in other North American water treatment distribution lines.

It has now been well documented that coliphages are more resistant to inactivation by chlorination than are E. coli. Scott et al. (1979) demonstrated that E. coli were nonviable after a 5 minute contact with 6 mg of chlorine per litre while coliphages survive an exposure to 25 mg of chlorine per litre for 80 minutes.

Earlier Grabow (1968) reported the results of many studies that indicated that most common pathogenic viruses are more resistant to chlorination than are E. coli. Kott et al. (1978) in a later study reported that coliphages are similarly resistant to chlorination as polio viruses. Thus the presence of coliphage and bacteriophage in these drinking water samples, without coliform presence, strongly suggests that virsuses can also survive the normal treatment and disinfection processes accorded these potable water samples. Another major implication of these data is that coliform-free potable waters are not necessarily pathogen-free.

Studies by Petrovicova et al. (1988) have produced similar results in Czechoslovakian waters and they state "presence of coliphages in drinking water is an indication for aimed virological examination". Furthermore, since 1987, the coliphage enumeration procedure as a means of water quality control has been adopted by the Hygiene Services and Water Supply Authorities in Slovakia.

On a speculative note, based on our finding of coliphage and bacteriophage in all of the 18 water samples tested, perhaps the presence of bacteriophage/coliphage in water systems are indicators of the mode of transport of many human viral infections requiring the oral route for infections to occur.

In conclusion, we believe, on the basis of these Canadian data and the other referenced data (El-Abagy et al., 1988; Ratto et al., 1989; Sim and Dutka, 1987; Simkova and Cervenka, 1981) that the coliphage test merits further investigation as an indicator of inadequate potable water treatment.

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Table 1. Coliphage and bacteriophage levels (as plaque-forming units per litre) and bacterial levels in raw drinking water and treated drinking waters in three Ontario cities.

Tests	Raw Water Samples				
	City A[5] June 24	City A July 16	City B[6] May 20	City B June 11	City C[7] June 4
ECC/L[1]	17.5	32.5	10.0	18.4	1695.0
CCHFR/L[2]	32.5	67.5	-	8.4	180.0
WG49/L[3]	50.0	23.8	8.0	40.0	32.5
WG45/L[4]	40.0	11.3	10.0	26.7	20.0
TC/100 mL	52	40	<1	<1	5900
FC/100 mL	30	16	<2	<2	420
FS/100 mL	neg	16	<4	<4	460
SPC/100 mL	-[8]	-	2300	-	-

	City Water Samples				
	City A June 25	City A July 16	City B May 20	City B June 11	City C June 4
ECC/L	15.0	15.8	10.0	35.0	0.0
CCHFR/L	14.0	33.6	-	-	7.0
WG49/L	17.0	16.5	10.0	60.0	15.0
WG45/L	8.5	8.5	10.0	45.0	0.0
TC/100 mL	ND[9]	ND	ND	ND	ND
FC/100 mL	ND	ND	ND	ND	ND
FS/100 mL	ND	ND	ND	ND	ND
SPC/100 mL	3000	3000	142	254	700
Free Cl_2 (ppm)	0.4	0.2	0.7	0.4	2.7
Total Cl_2 (ppm)	0.5	0.3	1.0	0.7	3.2

[1]E. coli C coliphage
[2]E. coli HfrH coliphage
[3]F+ bacteriophage (RNA)
[4]F- somatic bacteriophage
[5]Lake Huron source water
[6]Lake Erie source water
[7]River source water
[8]Not tested
[9]Not detected