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Project Summary

Interrelationship of Bacterial Counts With Other Finished Water Quality Parameters Within Distribution Systems

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This study's objective was to obtain realistic information concerning the interrelationships among temperature, chlorine, turbidity, coliforms, and Standard Plate Count (SPC) densities present in finished water after treatment and distribution. Bacterial identifications were performed to determine types and densities of isolates from the SPC and coliform tests.

The frequency of coliform isolation was independent of free chlorine, turbidity, and temperature. SPC's were not contingent on low level turbidity and varied with respect to free chlorine residual and temperature. SPC's exhibited no interrelationship with coliform counts when the SPC was less than 50 organisms/mL. A slight inverse relationship was noted between free chlorine residual and turbidity. Of the physical and chemical parameters measured, free chlorine residual had the greatest influence on the microbial population.

Encapsulated *Klebsiella pneumoniae, Enterobacter agglomerans, Enterobacter aerogenes* and *Enterobacter cloacae,* which gave typical coliform results, exhibited the ability to survive a free chlorine residual of 0.2 mg/L or more. The diversity of organisms identified by the SPC method strongly suggests the phenomenon of an established microbial ecosystem within the distribution networks.

This Project Summary was developed by EPA's Municipal Environmental Research Laboratory, Cincinnati, Ohio to announce key findings of the research project that is fully documented in a separate report of the same title (see Project Report ordering information at back).

Introduction

Recently, increased attention has been placed on water quality and the monitoring procedures that ultimately determine the quality of water delivered to the consumer. Questions have been raised about bacterial, chemical and physical standards used in water quality monitoring and their interrelationships in the final product reaching the tap. These standards and some of their relationships have been studied and documented in the laboratory and in distribution systems. Previous studies, however, have tended to focus on those systems that do not provide full treatment to their water supply and that have not generally monitored seasonal fluctuations of water quality parameters. Water supplies providing full treatment (chlorination, rapid mixing, flocculation, sedimentation, filtration) have tended to be ignored in previous studies, presumably since they were assumed to be supplying good and safe water to their consumers. This study involved Salem and Beverly (Massachusetts)—each

Table 1. *Effect of Free Chlorine on Coliform Densities in Two Distribution Systems*

System	Coliform	Free chlorine (mg/L) ≥0.0 % samples	≥0.2 % samples	≥0.5 % samples	≥1.0 % samples
	100 mL				
Salem	<1	78	80	80	86
		(900)*	(336)	(176)	(80)
	≥1	22	20	20	14
		(257)	(84)	(44)	(13)
	≥5	9	6	7	7
		(107)	(27)	(16)	(5)
Beverly	<1	82	78	78	81
		(863)	(291)	(106)	(13)
	≥1	18	22	22	19
		(193)	(82)	(30)	(3)
	≥5	8	10	11	14
		(60)	(37)	(15)	(3)

*The numbers in parentheses are the number of samples meeting the imposed test statement criteria.

with its own distribution system but sharing a common water source and treatment plant.

Good quality water leaving the treatment facility has long been known to undergo deterioration within the distribution system, but the extent of chemical, physical, and biological degradation before the water reaches the consumer has not yet been fully studied. The objective of this research was to determine if monitoring the fundamental parameters of temperature, chlorine residual, turbidity, pH, coliforms, and Standard Plate Count (SPC) adequately characterized the microbial quality of water as it traveled through a distribution system.

Results

During the study of the Salem and Beverly distribution systems, the frequency of coliform isolation was found to be independent of the amount of free chlorine present in the sample at the time of collection. Despite the fewer number of samples taken at each increasing free chlorine level (read across, Table 1), the frequency of coliform isolation did not significantly decrease. This occurred not only at the ≥1 coliform level, but the maximum contaminant level (MCL, ≥5) as well.

The effect of free chlorine residuals on SPC was also analyzed. The results demonstrate a very definite reduction of the SPC with increased free chlorine residuals (read across, Table 2), in contrast to the coliform results presented in Table 1. Increased chlorine residual levels from ≥0.0 mg/L to ≥0.1 mg/L effectively dropped the SPC percentage more dramatically in Salem than in Beverly. It may be speculated that the older, more encrusted and slower flowing (higher retention time) Salem distribution system had a greater proportion of its pipe network harboring SPC organisms than the cleaner Beverly system. Beverly's results deviated from Salem's at the ≥1.0 mg/L percent level, again on the lower side, probably because of Beverly's newer distribution system not allowing a suitable environment for the establishment of the microorganisms.

If this is correct, then why were the ≥0.1 mg/L and ≥0.5 mg/L results from Beverly similar to those of Salem? Possibly the threshold of "effective" disinfection in the "clean" Beverly distribution system was lower than it would have been in the older, more encrusted Salem system. The "effective" level for Beverly was somewhere between 0.5 mg/L and 1.0 mg/L, where a sharp drop in the percentage was noted. Salem's "effective" level must have been higher than 1.0 mg/L because no such percentage reduction occurred between the 0.5 mg/L and 1.0 mg/L levels.

Another analysis (Table 3) revealed the coliform frequency was approximately the same throughout the SPC ranges of 0, 3, 10, and 50. The 500 level may be statistically invalid since only 24 samples in Salem and 8 in Beverly exceeded the 500 level. The table reveals three conclusions. First, the SPC levels do not affect the frequency of coliform recovery (read across). Second, the fact that coliforms appear with the same frequency, regardless of SPC levels, strongly suggests that the coliforms are a part of the distribution system's microbiological flora. The third conclusion is that high or low SPC densities do not indicate either the presence or absence of coliform organisms.

The diversity of organisms isolated from the distribution systems (Table 4) strongly suggests these organisms have established an ecosystem within those pipe networks. The ability of organisms, including coliforms, to establish and maintain this ecosystem is not surprising when it is realized how the environment and the microorganisms genetic potential combine to form an ideal habitat for bacteria. If the distribution systems have a diversified microbial flora consisting of the organisms in Table 4 with their varied characteristics, then that flora cannot and should not be expected to react in an absolute manner to one parameter(e g., free chlorine residual or turbidity) with any sort of consistency. These organisms, including coliforms, are able to survive a variety of physical, chemical, and biological phenomena by being encapsulated. This dense polysaccharide capsular coat, that is generally absent from the pure laboratory

Table 2. *Effect of Free Chlorine on SPC Densities in Two Distribution Systems*

System	SPC/mL	Free chlorine (mg/L) ≥0.0 % samples	≥0.1 % samples	≥0.5 % samples	≥1.0 % samples
Salem	<3	46	57	60	65
	≥3	54	43	40	35
	≥10	27	17	11	9
	≥50	8	4	2	1
	≥500	1	0	0	0
Beverly	<3	55	59	62	73
	≥3	45	41	38	27
	≥10	18	15	11	0
	≥50	5	4	2	0
	≥500	0	0	0	0

cultures but not from environmental strains, enables the various environmental organisms to protect themselves from the "hostile" conditions of a distribution system.

Encapsulation is essential to the success of bacteria in natural environments because the capsule coat collects useful materials and also binds harmful ions and molecules in the environment. The implication is clear, the dense polysaccharide coat has not only the physical barrier capacity to protect itself from free chlorine molecules and ions but the chemical capability as well. With the "neutralization" of free chlorine, the water utility's primary defense mechanism, the distribution system, becomes an ideal environment for the survival and growth of microorganisms.

Coliform organisms are no different from other groups of microorganisms listed in Table 4 with regard to their sustaining and replicating capability within the distribution system. They are able to replicate in water with trace organics present as evidenced by the fact that 22.4% of the microorganisms randomly selected from the SPC during this study were coliforms. The coliform group makes up a rather remarkably large percentage of the SPC population identified in Table 4. Although taken in the context of the large number of coliforms isolated throughout the study period by the membrane filtration method and the hypothesis that these organisms comprise part of the ecosystem in the distribution system, it is not a remarkably large percentage and it is in fact a normal phenomenon that might be predicted. With a microbiological flora established throughout the distribution system, the generally independent nature of the results concluded for coliform and SPC populations, when compared with temperature, turbidity, and chlorine, may be understood.

Conclusions

The similarity of the ecosystems' bacterial isolates from the two separate and distinct distribution systems of Salem and Beverly was probably related to sharing the same source of water and treatment—the Salem and Beverly Water Supply Board's filtration plant. Coliform bacteria were found to be a part of the ecosystem established within the distribution systems, and the occurrence of coliforms in the distribution networks was independent of free

Table 3. *Effect of SPC Densities on Coliform Densities in Two Distribution Systems*

		SPC/mL				
		≥0	≥3	≥10	≥50	≥500
System	Coliform	% samples	% samples	% samples	% samples	% samples
	100mL					
Salem	<1	78	74	72	71	75
		(900)*	(521)	(242)	(64)	(18)
	≥1	22	26	28	29	25
		(257)	(183)	(94)	(26)	(6)
	≥5	9	11	14	10	0
		(107)	(73)	(42)	(7)	(0)
Beverly	<1	82	81	79	75	75
		(863)	(431)	(147)	(45)	(6)
	≥1	18	19	21	25	25
		(193)	(101)	(39)	(15)	(2)
	≥5	6	10	10	12	25
		(60)	(39)	(17)	(4)	(1)

* *The number of samples meeting the imposed test statement criteria.*

Table 4. *Organisms Identified from Salem and Beverly Distribution Systems*

m-Endo agar LES	Plate Count Agar
Klebsiella pneumoniae	*Klebsiella pneumoniae*
Klebsiella rhinoscleromatis	*Enterobacter agglomerans*
Klebsiella ozaenae	*Enterobacter cloacae*
Enterobacter cloacae	*Enterobacter hafnia*
Enterobacter aerogenes	*Serratia marcescens*
Enterobacter agglomerans	*Proteus*
Escherichia coli	*Pseudomonas cepacia*
Citrobacter freundii	*Pseudomonas fluorescens*
Serratia liquifasciens	*Pseudomonas maltophilia*
Acinetobacter calcoaceticus	*Pseudomonas putida*
CDC Group 11K	*Pseudomonas vesicularis*
Aeromonas hydrophila	*Bacillus*
	Bacillus subtilis
	Streptomyces
	Streptococcus
	Lactobacillus
	Arthrobacter
	Achromobacter
	Achromobacter xylosoxidans
	Corynebacterium
	Flavobacterium
	Moraxella
	Rhizobium
	Nitrococcus
	Micrococcus
	Acinetobacter antratum
	Actinomyces
	Clostridium
	Vibrio alginolyticus
	Aeromonas hydrophila
	CDC Group 11F
	CDC Group UE1
	Alcaligenes
	Yeast

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chlorine residuals and turbidity fluctuations of less than 2.0 Turbidity Units. Also, SPC of 50 colonies or less did not reflect the presence or absence of coliforms.

The best quality water may be produced at a treatment facility and a high chlorine residual may be employed throughout the delivery system, but if the distribution system has a microbial ecosystem throughout its network, then regardless of that high quality chlorinated water, microorganisms from that flora, including coliforms, may be isolated from that system.

Analysis of all the parameters in the above conclusions proved difficult because of the variation of physical and chemical processes at the filtration plant and the inherent complex nature of the dynamic ecosystems within the distribution networks. It is recommended other distribution systems should be studied with respect to the ecosystems established within them. Also studies should be instituted to evaluate the ability of environmental organisms, specifically coliforms, to withstand the effects of free chlorine residuals. Critical considerations of these studies would be confirmation of encapsulated bacteria. The authors consider a study of pH and its effect on the environmental organisms, in conjunction with chlorine, to be very critical. Finally, additional research should be undertaken to study the SPC enumeration procedures along with improved media and new recovery methods and to define the health significance and impact on coliform populations by the SPC population

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The complete report, entitled "Interrelationship of Bacterial Counts with Other Finished Water Quality Parameters Within Distribution Systems," (Order No. PB 81-168 726; Cost: $8 00, subject to change) will be available only from:

National Technical Information Service
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