PHILLIPPI CREEK WATER QUALITY REPORT

by

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SUMMARY

Septic tanks can be one of the major sources of water pollution and have been the cause of waterborne disease associated in particular with viruses. Total and fecal coliform bacterial indicators can not consistently indicate the persistence of pathogens, especially viruses in surface waters. F-specific RNA coliphage, *enterococcus* and *Clostridium perfringens* might be better indicators of fecal contamination and public health risks. Five different sites along Phillippi Creek have been sampled and tested for these five indicators in April, July and October 1994. Fecal indicator bacteria and coliphage were detected in every sample during each of the three collections in the Phillippi Creek study and the concentrations of fecal coliforms in 12 of 15 samples were in violation of Florida State Standards for Safe Swimming (200/100mL), averaging from 152 to 2,780 CFU/100mL for each collection. Enterococci, *Clostridium* and coliphage also were found to be in concentrations indicating significant fecal contamination. The site 628 was downstream of a wastewater treatment plant with a deep well injection and was within Standards and showed less impact from fecal indicators than the sites 625 and BR which were influenced by septic tank discharges. The concentrations at site 625 were 10 to 20 times greater than those seen at the canal site 628. The levels decreased as the water became more saline. The overall rankings indicate that site 628 was the least contaminated and sites BR and 625 were found to have the greatest level of contamination. Based on the bacterial analysis and the presumptive

positives in enterovirus detection, a more thorough investigation of viral pathogens present along the Phillippi Creek would be warranted, testing greater volumes, more samples and using molecular techniques to screen for an array of disease causing agents.

INTRODUCTION

Public Health Significance of Enteric Viruses

Water is generally recognized as a vehicle for the transmission of many enteric viruses (Goyal, 1984; Gerba *et al.*, 1985). There are about 120 enteric viruses which have been identified in human feces including hepatitis, enteroviruses and Norwalk virus. Enteric viruses are associated with a wide spectrum of clinical illnesses, including fever, rash, meningitis, paralysis, myocarditis, hepatitis, eye infection and most often, gastroenteritis (Melnick, 1980; 1984). Enteric viruses may originate from untreated and treated wastewater but septic tanks were the most frequently reported sources of contamination of groundwater (USEPA, 1977). Septic tanks are the source of approximately 1 trillion gallons of waste disposed to the subsurface each year (U.S. Congress, 1984). The overflow or seepage of sewage, primarily from septic tanks and cesspools, was responsible for 43% of the reported outbreaks and 63% of the reported cases of illness caused by the use of untreated water (Craun, 1985). Yates (1985) reviewed a total of 1,720 cases in six waterborne outbreaks caused by the contamination of groundwater with septic tank effluent. Of these, 115 (6.7%) cases were hepatitis A and 1600 cases (93.0%) were characterized as unknown gastroenteritis, a significant portion of which can be caused by viruses.

Due to their special structure, the enteric viruses are able to survive water treatment, and survive in fresh and marine waters much longer than coliform bacteria (Bitton *et al*, 1983; Craun,1984; Payment 1985; Haavelar *et al*. 1993), which are generally used to monitor the water quality. When released into coastal waters, the viruses may either remain suspended in seawater and be transported to recreational areas or they may be accumulated in bottom sediments and/or filter-feeding shellfish. Viruses in marine waters have been detected far from the original source of pollution and in the absence of bacterial indicators (Metcalf *et al*. 1967; Vasl *et al*. 1981).

Through septic tanks, sewage effluent discharges and landfills, these viral pathogens contaminate the aquatic environment, which may subsequently transmit the viruses to human beings. Public health risks are associated with the accumulation of these viruses in sediments, in shellfish and resuspension into the water. Exposure can occur through the food chain or through recreational use of the water body.

The Suggested Water Quality Indicators

The presence of human enteric viruses in water used for drinking, recreation, or growing shellfish pose a risk to human health. Treatment processes and watershed management strategies designed on the basis of bacteriological criteria (coliforms) do not necessarily protect against virus infection because viruses are

generally more persistent in the water environment and are not removed as well by treatment processes (Havelaar *et al.*,1993). Several types of alternative microorganisms have been suggested as indicators of water quality, fecal pollution and public health risks. These include *enterococci*, *Clostridium perfringens* bacteria and F-specific coliphage (a bacterial virus) (Table 1).

F-specific coliphage

F-specific RNA coliphage are viruses which infect the F$^+$ *E. coli* bacteria and can be found in fecally contaminated water. They have been suggested as adequate model organisms for enteric viruses in water. Havelaar *et al* (1993) detected culturable enteroviruses and three groups of indicator microorganisms in a wide variety of environments. They found that bacteria were relatively low in disinfected effluents and relatively high in surface waters open to nonhuman fecal pollution whereas coliphages were found to be higher in effluents than in waters which were impacted by nonhuman wastes. The concentrations of F-specific RNA coliphage were highly correlated with enterovirus concentrations in all environments.

F-specific RNA coliphage may also be a possible indicator of faecal pollution in water because they were found consistently and abundantly in wastewaters of human domestic origins (Dhillon, 1976). The concentrations of F-specific coliphage in mixed wastewater were found between 10^3-10^4 pfu/ml in dry periods but were lower during periods of heavy rainfall (Havelaar *et al.*,1993).

Table 1. Comparison of Microbial Water Quality Indicators

Name	Type	Application
Total Coliform	Bacteria group associated with human and/or animal fecal contamination.	National standards For drinking water(<1/100ml). For recreational water (<1000/100ml).
Fecal Coliform	Bacteria subgroup of total coliform, more restrictive than total coliform to indicate human and/or animal fecal contamination.	State standard for recreational water (<200/100ml). Also associated with standard for wastewater disposal.
F-specific RNA coliphage	Virus specifically infects F+ E.coli More resistant than coliform in water treatment and in the environment.	Suggested to be an human enterovirus indicator in water.
Enterococcus	Subgroup of fecal streptococci (bacteria cocci of fecal origin). More resistant than bacterial pathogen and coliform in water treatment and in environment.	Suggested to be better fecal pollution indicator of human health risks.
C. perfringens	Anaerobic spore-forming bacteria. Spores are very resistant in water.	Suggested virus or remote fecal pollution indicator, particularly applicable to marine water.

In addition, F-specific RNA coliphage are detectable by simple, rapid, and inexpensive methods. Thus, F-specific RNA coliphage can serve an indicator function with regard to human pathogenic viruses in the water environment.

Enterococcus

Enterococcus is a subgroup of the fecal streptococci (bacterial cocci of fecal origin) which possess the group D antigen and conform to the Sherman criteria (Clausen *et al.*, 1977). The *enterococcus* group includes *S. faecium*, *S. faecalis*, *S. durans*, and related biotypes (Facklam, 1972; Clause. *et al.*,1977).

In surface waters, fecal streptococci are generally less numerous than fecal coliforms and are present in lower numbers than total coliforms (Litsky *et al.*,1955; Leninger and McClesdey, 1953). However, fecal streptococci have been consistently recovered from waters known to receive fecal contamination, especially from polluted wells and springs in which fecal coliforms were absent (Cohen and Shuval, 1973). There were no enterococci detected in non-contaminated water in which total coliforms were recovered(Leninger and McCleskey, 1953). Furthermore, there was no indication that they multiply in natural or fecally polluted waters or soils.

Enterococcus generally appears to be more persistent than either bacterial pathogens or fecal coliforms (Coheb and Shuval,1973; Davies-Colley *et al*, 1994; Sinton *et al*, 1994). Cohen

and Shuval (1973) have compared survival of total coliforms, fecal coliforms, fecal streptococci, and viruses before and after sewage treatment. Removal of fecal streptococci was considerably less than that of coliforms and, thus, more closely paralleled virus survival.

In summary, *enterococci* do not multiply and are more persistent than fecal coliform in polluted waters. Therefore they may be a safer indicator of pollution. Whereas fecal coliforms may better parallel die-off of enteric bacterial pathogens such as *salmonellae*, fecal streptococci better indicate the possibility of viral contamination.

Clostridium perfringens

C. perfringens is an enteric gram positive, anaerobic spore-forming pathogenic bacterium found in feces. But there are no data suggesting that waterborne transmission is significant in the epidemiology of diseases caused by this organism which is associated with foodborne diseases. Klein and Houston (1899) first suggested that this organism could be used to detect fecal pollution. Although there are considerable controversies about using *C. clostridium* as a water quality indicator(Cabelli, 1977), a number of scientists (Fujioka and Shizumura, 1985; Payment, 1993) continue to recommend *C. perfringens* due to its spore-forming property as a valuable supplement to other water quality tests, particularly in situations where the detection of viruses or remote

faecal pollution is desirable. This organism is consistently present in municipal wastewater at concentrations of 10^3 to 10^4 colony-forming units/100ml, and its resistance to chlorination and other environmental factors resembles that of enteric viruses (Fujioka and Shizumura, 1985).

Total coliforms

Breed and Norton (1937) first suggested that total coliform be used to describe the lactose fermenting bacteria found in polluted water and that the organism be used as a measure of pollution. The broad general characteristics which define this group have allowed it to be one of the most useful bacterial indicators and at the same time have been responsible for its displacement as an indicator of fecal contamination.

The total coliform are used to test for drinking water quality, and their presence in any drinking water should initiate an immediate search for a contaminating source. The use of the total coliform as a water quality indicator has successfully helped decrease the incidence of several major bacterial water-borne diseases, such as typhoid fever and cholera. But presence of the total coliform in wastewaters or untreated surface waters is to be expected and does not have the significance of the presence of fecal contamination (Mack, 1977).

Fecal coliforms

Fecal coliforms are a subgroup of the total coliform bacteria which can ferment lactose with the production of gas in 24 hr with the incubation temperature increased from 35°C to 45°C.

Fecal coliform bacteria in aquatic environments suggest that the water in question has been contaminated with fecal material from warm-blooded animals. Although the fecal coliform test was a great improvement in methodology for detecting fecal contamination in aquatic environments, it still has its shortcomings ---- (i) pathogens such as human enteric viruses have been recovered from natural waters that were determined to be safe based on low densities of fecal coliforms (Berg and Metcalf, 1978), (ii) fecal coliforms have reported to be capable of multiplying in environmental water under some conditions (Dutka, 1973), (iii) some fecal coliforms such as *Klebsiella pneumoniae* do not have a fecal source (Knittek, 1975), positive tests for fecal coliforms have been observed in the absence of a source for human or warm-blooded animal fecal inputs, particularly for tropical waters (Fujioka *et al*, 1988), (iv) and laboratory results show that fecal coliforms are less resistant than some pathogens (such as human enteric viruses) to chlorination or less stable in natural waters (Gerg and Metcalf, 1978).

Florida Standards for Class III Waters

Classifications of water and surface water quality are determined by the State. The Florida State standards are set by the Department of Health and Rehabilitative Services (HRS) or the Department of Environmental Protection (DEP). Table 2 shows the Florida State Standards which are currently in place for Class III waters used for recreational purposes. Water quality standards had been last evaluated in 1987 in Florida, but were not changed from the 1979 guidance.

The standards promulgated by DEP are expressed in 3 tiers in recognition of the fact that sampling will produce a range in results. The DEP standards for fecal coliform state that the average level in the samples taken cannot exceed 200 colony forming units per 100mL (CFU/100mL) (This average is calculated as a geometric mean in which each value is expressed as a logarithmic value, an average of the logged values is taken, and this average is then converted back to the original units of measure). In addition, the fecal coliform level in 10 percent or more of the samples cannot exceed 400 CFU/100mL and none of the samples can exhibit a fecal coliform level above 800 CFU/100mL.

Table 2. Ambient Water Quality Standards in Florida

Indicator Bacteria	Concentrations Allowed	Agency
Total coliforms	<1,000 CFU/100ml	HRS*
Total coliform	<1,000 CFU/100ml(geometric mean) <2,400 CFU/100ml (maximum level) <1,000 CFU/100ml (no more than 20% exceed this level).	DEP**
Fecal coliform	200 CFU/100ml(geometric mean) 800 CFU/100ml (maximum level) 400 CFU/100ml (no more than 10% exceed this level).	DEP**

* Three samples per day, three days per week, for three consecutive weeks. Health and rehabilitative Services.

** Ten samples per 30 days. Department of Environmental Protection.

OBJECTIVES

Sarasota Bay is a very important area used for recreation and shellfish harvesting in the mid western part of Florida on the Gulf of Mexico. Due to the large amount of nutrient and pathogenic microbial input from septic tanks along Phillippi Creek and other rivers, the water quality of Sarasota Bay has become questionable. The National Estuary Program had identified septic tanks near Sarasota Bay tributaries as a major source of pollution.

Phillippi Creek has one of the largest waterflows in the northern Sarasota County. Along both sides of Phillippi Creek, there are large areas without centralized sewage treatment plants. On-site septic tanks are the major form of waste disposal. It is estimated about 32,000 septic tanks exist in that area. Septic tank effluent may be the source of a ninth of the creek's water during times when flow levels are lowest with approximately 4.8 million gallons a day of effluent discharging from septic tanks. Therefore, Phillippi Creek may be generally considered unfit for drinking, recreation and shellfish harvesting.

The objective of this study was to evaluate the water quality along Phillippi Creek using various fecal indicators for assessing the effects of septic tank effluents. The focus of this study was on microorganisms which may indicate public health risks particularly associated with viruses.

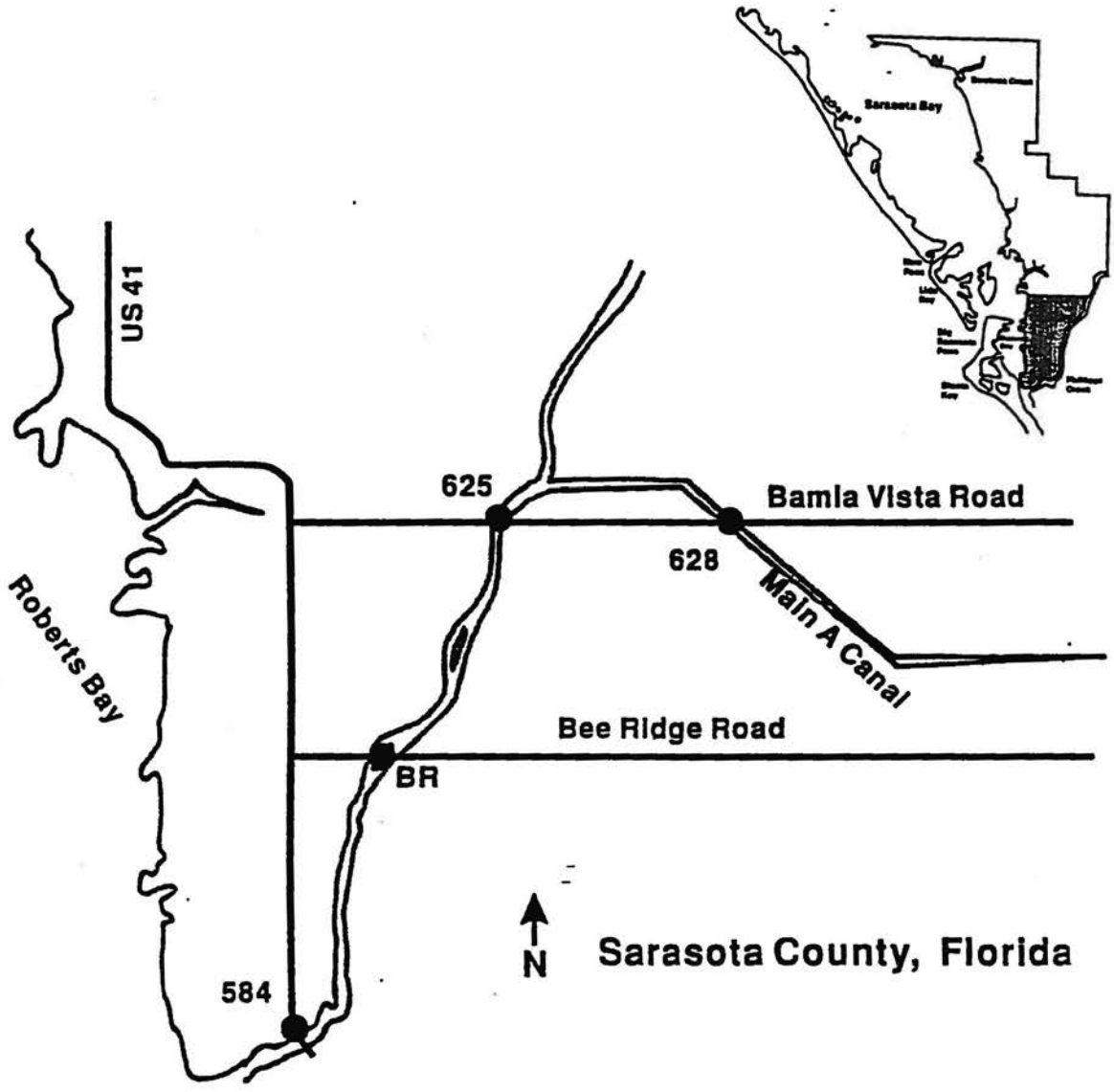
MATERIALS AND METHOD

Sampling Sites

Five different sampling sites were chosen along the Phillippi Creek (Figure 1). Site 628 was located on a canal labeled as main "A" canal. There was a secondary sewage treatment plant located upstream of site 628 with deep well injection of its effluent. Site 625 was located where Phillippi Creek crossed Bahia Vista street. Along the bank upstream of this site, there were large numbers of septic tanks discharging to the creek. Site BR was downstream of site 625 and site 584 was downstream of site BR. Site BC was on the Bowless Creek, which was located in a area with a centralized sewer.

Sampling Methods

Twenty liters of water from each site were collected into a 20L bleached carboy. This was to be used for the membrex concentration analysis. In addition, two liters of water from each site was collected in autoclaved bottles for all the bacterial analysis. The water was collected by bucketing or with a gasoline driven water pump. The water samples were placed on ice immediately and brought back to the University of South Florida laboratory. Salinity and temperature were measured in the field. Collection buckets were bleached between sample collections or different buckets and hoses were used for each site.


US 41
625
Bamla Vista Road
628
Main A Canal
Roberts Bay
Bee Ridge Road
BR
584
N
Sarasota County, Florida

Sample Concentration

Water samples for coliphage detection were concentrated to approximately 50 ml by a Benchmark Gx Flow Filtration System which concentrated water samples by vortex flow (Paul *et al*, 1991). The other samples for the bacterial indicators were assayed directly on non-concentrated water samples.

Sample Analysis

Total Coliforms

Volumes of 0.1ml, 1ml and 10ml of each water sample were filtered through membrane filters (0.45um, 47mm, Gelman Sciences). The filters were placed on M-Endo medium (Difco Laboratories, Detroit, MI) and incubated for 24 hours at 35+-0.5°C. All bacteria that produced a red colony with a metallic sheen were counted as total coliform bacteria (Standard Methods for Examination of Water and Wastewater, APHA, 1989).

Fecal Coliforms

The same volumes of water from each sample was filtered as described for the total coliform analysis. The filters were placed on M-FC medium (Difco Laboratories, Detroit, MI) and sealed in plastic bags within 30 min after filtration. The plates were

incubated for 24 hours in a water bath at 44.5+-0.2°C . The bacterial colonies with various shades of blue were counted as fecal coliform bacteria (Standard Methods for Examination of Water and Wastewater, APHA, 1989).

Enterococci

Water samples were filtered as described above. The filters were placed on M-EC media (Difco Laboratories, Detroit, MI) and incubated at 41+-0.5°C. After 48 hours incubation, enterococci showed pink or red colonies on the membrane filters. The filters were transferred to EIA medium (Difco Laboratories, Detroit, MI)and incubated at 41+-0.5°C for 20 min, the colonies which developed a black or reddish-brown precipitate on the underside of filter were counted as enterococci (Standard Methods for Examination of Water and Wastewater, APHA, 1989).

Clostridium perfringens

Water samples were filtered as described above. The filters were placed on the M-CP (Acumedia Manufactures, Inc. Baltimore, Maryland) plates and sealed with anaerobic gas paks (BBL GasPak, Becton Dickinson). After 24 hours incubation at 45°C, the yellow colonies were exposed to ammonium hydroxide fumes and the colonies which turned red or dark pink were enumerated as *C. perfringens* (Bisson and Cabelli, 1979).

F-specific Coliphage

The concentrated phage sample was analyzed by plaque assay using a soft agar overlay technique (Adams, 1959) with *Escherichia coli* C-3000 (courteously offered by Dr. Farrah from University of Florida) as a host. Serial dilutions were made from samples in PBS buffer, added to test tubes containing 3 ml of melted 1% TSB top agar (48°C) and 1 ml of a 3 hour culture of *E. coli* and poured onto solid TSA (1.5% agar) plates. The plates were incubated for 24 hours at 37°C, after which the plaques where the viruses had grown and lysed the bacterial lawn were enumerated. Plaque forming units (PFU) per liter were calculated (Standard Methods for Examination of Water and Wastewater, APHA,. 1989).

Culturable Enteroviruses

Concentrated water samples from Membrex filtration were prefiltered through 0.2um filter (25mm,Corning) then stored at -70°C. The samples were quickly melted in a 37°C water bath before inoculation onto cells and were kept on ice during the processing. One milliliter of sample was inoculated onto each of a total of twenty 25mm^2 bottles with a Buffalo green monkey (BGM) kidney cell monolayer without cell culture media. After the bottles with the sample were incubated with the cell side down at 37°C for two hours, the maintenance medium (E-MEN with 5% fetal calf serum) was added to each bottle. The bottles were incubated at 37°C for two

weeks and were evaluated daily for cell destruction caused by viruses known as cytopathic effects (CPE). Both positive and negative samples were frozen at -70°C and thawed at 37°C before being transferred (1ml of each) to a 13 X 100mm tube with a new BGM monolayer. The tubes were incubate at 37°C for two more weeks and were checked for CPE everyday (Standard Methods for Examination of Water and Wastewater, 1989).

RESULTS AND DISCUSSIONS

Three collections were made from five sites on May 4, July 24 and October 10,1994 (Table 3). The water temperatures ranged from 27°C to 33.5°C. The average water temperature in May was 30.7°C, in July was 29.2°C and in October was the lowest (28°C). The turbidity ranged from 2.22 in site 628 in May to 10.29 in site 584 in July which was collected right after a heavy rain. The salinity ranged from 0.5 to 18 in May with a sequence of 628, 625, BR, 584 and BC. The pH of all the water samples was between 7.55 to 8.16.

Fecal indicator bacteria and coliphage were detected in every sample during each of the three collections in the Phillippi Creek study. The data are shown as colony forming units (CFU) per 100 mL for the bacteria and as plaque-forming units (PFU) per 100 mL for the coliphage in Table 4. The standard in Florida for recreational waters is an average of 200 fecal coliform CFU/100mL and the average for the April collection was 2,780 CFU/100mL with 3 of the 5 sites exceeding the maximum allowable level of 800 CFU/100mL; in July the average was 152.4 CFU/100mL, with none of the samples exceeding the 800 CFU/100mL maximum and one sample exceeding the 400 CFU/100mL (10% standard) and in the October collection, the average was 1,147 CFU/100mL with 2 of 5 sites exceeding the 800 CFU/100mL maximum allowable standard. The guidance from the Environmental Protection Agency for safe water for recreational purposes is based on enterococci at 35 CFU/100mL. The averages for

Table 3. Physical and Chemical Analyses of Water Along the Phillippi Creek

Date	Site	Temperature oC	Turbidity (NTU)	Salinity (ppm)	pH
5/4/94	628	33.5	2.22	0.5	8.16
	625	29	4.36	0.5	7.83
	BR	30	7.58	2.5	7.55
	584	31	3.16	12	7.71
	BC	30	5.22	18	7.88
7/24/94	628	29	2.75	-	-
	625	29	4.29	-	-
	BR	29	7.25	-	-
	584	30	10.29	-	-
	BC	29	6.16	-	-
10/10/94	628	28	-	-	-
	625	27	-	-	-
	BR	28	-	-	-
	584	29	-	-	-
	BC	28	-	-	-

Table 4. The Concentrations of Bacteria and F-specific Coliphage

Date	Sample ID	TC(1) CFU(5)/100mL	FC(2) CFU/100mL	CP(3) CFU/100mL	EC(4) CFU/100mL	Coliphage PFU(6)/100ml
4/4/94	628	-	250	200	50	1
	625	-	8500	350	40	118
	BR	-	3000	200	250	136
	584	-	2000	14	200	1
	BC	-	150	300	335	1
	Average	-	2780	213	193	52
7/24/94	628	1000	12	9	32	61
	625	775	5	26	3200	22
	BR	3600	10	26	950	64
	584	13250	505	47	5000	209
	BC	1500	230	9	165	26
	Average	4025	152	23	1869	77
10/10/94	628	400	285	20	80	4
	625	6500	1500	40	575	3
	BR	8000	3000	350	690	7
	584	3000	250	20	146	<6
	BC	5500	700	20	154	<6
	Average	4680	1147	90	329	3

Notes:
(1) TC: Total coliform
(2) FC: Fecal coliform
(3) CP: C. perfringens
(4) EC: Enterococci
(5) CFU: Colony-forming units
(7) PFU: Plaque-forming units

enterococci were 193, 1,869.4 and 328.8 CFU/100mL in the April, July and October collections respectively. There are no standard levels suggested for coliphage or for *Clostridium* although the presence of either are linked to fecally contaminated water and potential disease risks. The *Clostridium* averaged 212.8, 23.2, and 90 CFU/100mL and the coliphage averaged 51.5, 76.5, and 2.9 PFU/100mL for the April, July and October collections, respectively.

These data demonstrate that the Phillippi Creek system is heavily contaminated with fecal material and can not be considered safe to swim in. In addition, the standards for harvesting shellfish are set at 14 CFU/100mL fecal coliform and therefore this water body may be a significant source of disease risk to any estuaries in the Sarasota Bay.

The averages by sampling site along the Phillippi Creek for all three collections are shown in Table 5. Site 628 was not influenced by any major wastewater discharges and was within Standards and showed less impact from fecal indicators than the sites 625 and BR which were influenced by septic tank discharges. The concentrations at site 625 were 10 to 20 times greater than those seen at the canal site 628. The levels decreased as the water became more saline. This is expected due to the limited survival of the bacteria in marine waters (Davies-colley *et al.*, 1994; Sinton *et al.*, 1994). The exception was the *Clostridium* which produces spores with high resistance and can survive for extended periods of time (years) (Bisson and Cabelli, 1980).

Table 5. The Average Concentrations of Bacteria and Coliphage in Phillippi Creek

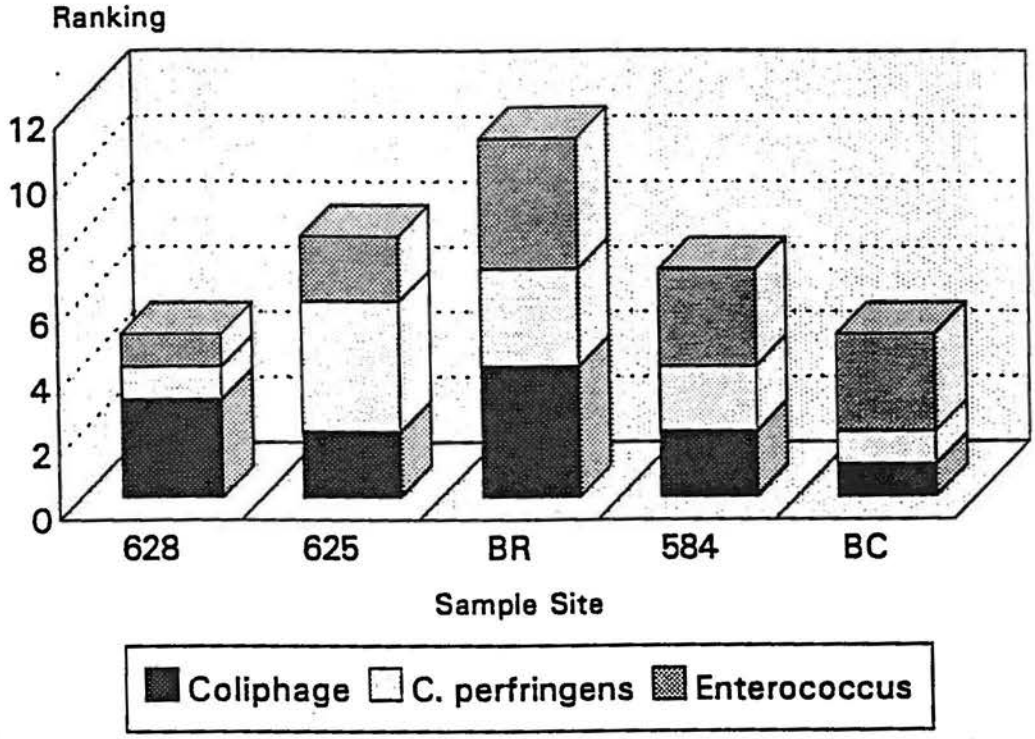
Sample ID	Ave.TC(1) CFU(7)/100m	Ave.FC(2) CFU/100ml	Ave.CP(3) CFU/100ml	Ave.EC(4) CFU/100ml	Ave.Coliphage CFU/100ml
628	700	182.3	76.3	53.8	22.2
625	3637.5	3335	138.7	1271.7	47.5
BR	5800	2003.3	191.8	630	69
584	8125	918.3	27	1780.5	70.3
BC	3500	360	109.5	384.5	9.3

Notes:
(1) The average concentrations of the total Coliform bacteria for each site
(2) The average concentrations of the fecal Coliform bacteria for each site
(3) The average concentrations of Clostridium perfringens for each site
(4) The average concentrations of the Enterococci for each site
(5) CFU/L: Colony-forming unit
(6) PFU/L: Plaque-forming unit

Each site was ranked for each of the bacterial test with number 1 indicating the lowest concentration of bacteria or coliphage up to number 5, indicating the greatest level of contamination relative to the other sites (Table 6). These were combined to give an overall ranking for each site. The overall rankings are shown in Table 7 and Figure 2 and indicate that site 628 was the least contaminated and sites BR and 625 were found to have the greatest level of contamination. The fecal indicator bacteria (fecal coliforms, enterococci, and *Clostridium*) all showed similar trends, although the enterococci were lower at the 625 site than those down stream, perhaps indicating greater loading as the water moved down stream. The salinity at site BC may have reduced the bacteria levels as previously mentioned. The coliphage showed a higher ranking at the 628 site. Still higher numbers were detected at the BR site, which had high septic tank discharges.

Fecal coliforms may not always be adequate for predicting health risks associated with fecal contamination particularly in tropical waters (Fujioka *et al.*, 1988). This is due to the fact that some fecal coliforms may become native to the natural vegetative environment over time due to high temperature and high organic content of the water. *Enterococci*, coliphage and *Clostridium* have been suggested as better indicators of fecal contamination in tropical waters. Figure 2 demonstrates the ranking of the sites using only these three indicators and the greatest level of contamination was found in BR, followed by 625, 584, 628 and BC.

Figure 2. Bacteria and Coliphage Rankings by Sites Along Philippi Creek for Fecal Contamination



* The lower the number, the lower the contamination level.

Table 6. The Rankings of Levels of Contamination for Each Sampling(7)

Date	Sample ID	TC(1)	FC(2)	CP(3)	EC(4)	FSC(5)	Total(6)
4/4/94	628	-	2	2	2	3	2
	625	-	5	4	1	4	4
	BR	-	4	2	4	5	5
	584	-	3	1	3	1	1
	BC	-	1	3	5	2	3
7/24/94	628	2	3	2	1	3	1
	625	1	1	4	4	1	1
	BR	4	2	3	3	4	3
	584	5	5	5	5	5	4
	BC	3	4	1	2	2	2
10/10/94	628	1	2	1	1	3	2
	625	4	4	2	4	2	4
	BR	5	5	3	5	4	5
	584	2	1	1	2	1	1
	BC	3	3	1	3	1	3

Notes:
(1) TC: Total coliform
(2) FC: Fecal coliform
(3) CP: C. perfringens
(4) EC: Enterococci
(5) FSC: F-specific coliphage
(6) Total: The total rankings for all the bacteria and F-specific coliphage
(7) Ranking: The higher the number, the greater the level of contamination.

Table 7. The Combined Rankings of Levels of Contamination for Each Site(7)

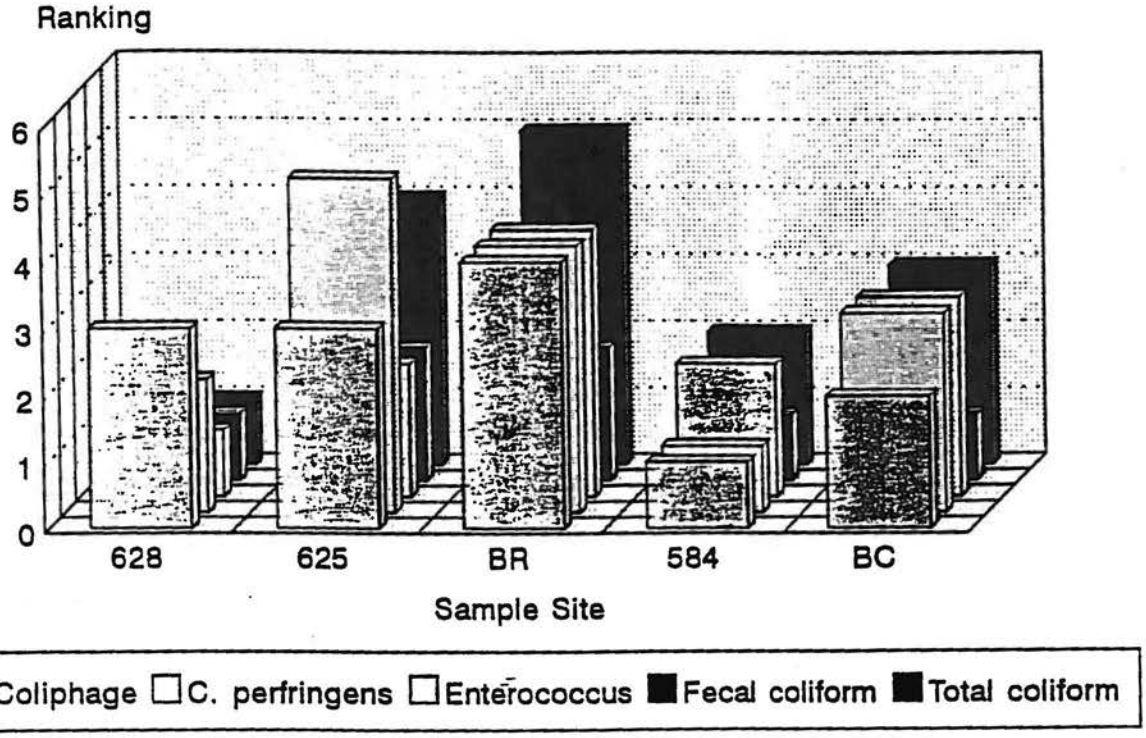
Sample ID	TC(1)	FC(2)	CP(3)	EC(4)	FSC(5)	Total(6)
628	1	1	1	1	3	1
625	2	5	4	2	2	4
BR	5	4	3	4	4	5
584	4	3	2	3	2	3
BC	3	2	1	3	1	2

Notes:
(1) TC: Total coliform
(2) FC: Fecal coliform
(3) CP: C. perfringens
(4) EC: Enterococci
(5) FSC: F-specific coliphage
(6) Total: The total rankings for all the bacteria and F-specific coliphage.
(7) Ranking: The higher the number, the greater the level of contamination.

Figures 3 and 4 show the rankings of the level of contamination by site for the April and October sampling (Figure 3, dry season) when there was little precipitation and the July collection (Figure 4, wet season), when there was significant rainfall. Although site 625 still was found to contain the highest levels in some cases, there appeared to be a shift downstream during the rainy season with greater numbers appearing at site 584 perhaps indicating a flushing of the streams during high flow events.

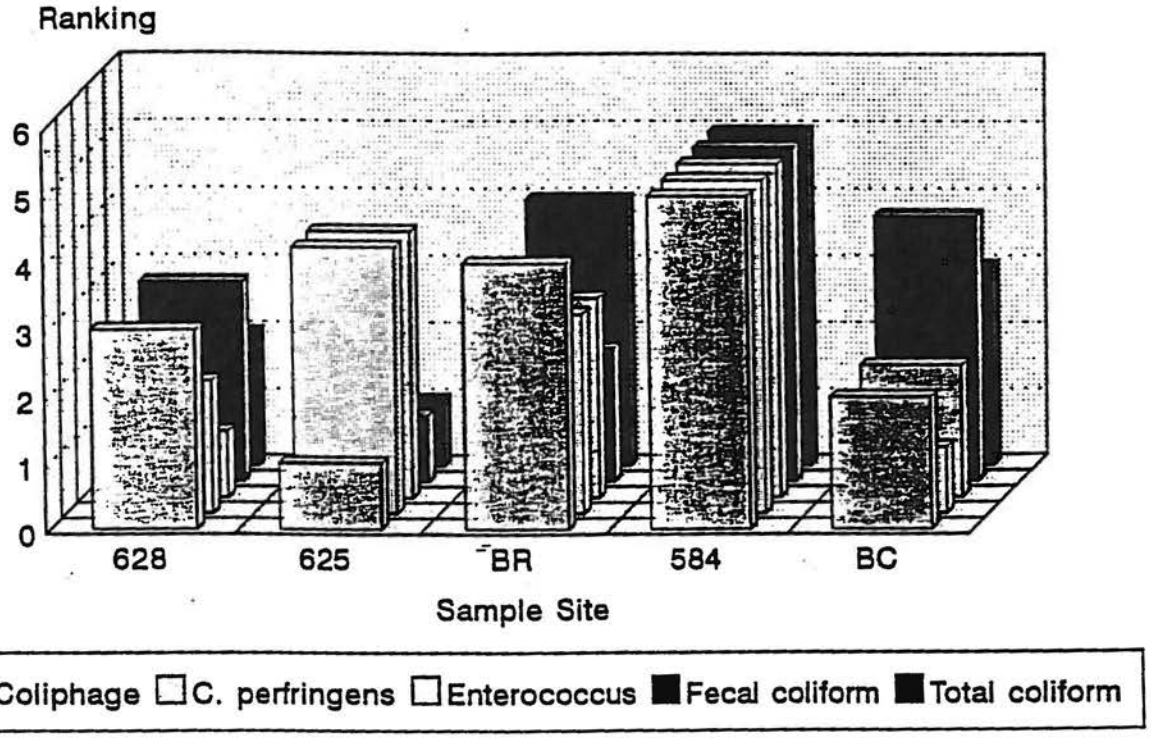
No enteroviruses were found in any of the ten samples during the April and July collections. However, only 10 L of water was examined and the contamination may have been below the sensitivity of the method used. Two of the samples (April collection site BR and July collection site 625) were found to be positive upon initial screening on BGM cells for enteroviruses however these were not confirmed positive after a second passage. Some viruses will die out after initial culturing. Also, it is well known that cell culture only detects a small percentage of the possible 100 human enteric viruses. Norwalk virus, hepatitis A virus those most commonly associated with waterborne disease would not be detected using cell culture and molecular techniques would be needed. Based on the bacterial analysis and the presumptive virus positives, a more thorough investigation of viral pathogens present along the Phillippi Creek would be warranted, testing greater volumes, more samples and using molecular techniques to screen for an array of disease causing agents.

Figure 3. Ranking of the Various Indicators for Fecal Pollution(Dry Seasons)



* The lower the number, the lower the contamination level.

Figure 4. Ranking of the Various Indicators for Fecal Pollution(Wet Seasons)



* The lower the number, the lower the contamination level.

CONCLUSIONS

Fecal contamination all along the Phillippi Creek is evident based on the use of four indicators of pollution. The water quality can not meet Florida State standards or Federal guidelines for safe swimming and should not be open for recreational purposes in order to protect the public from acquiring waterborne disease. The sites impacted by septic tanks (625 and BR) were found to have the greatest levels of contamination. Further studies may be warranted to examine approaches needed for remediating the pollution level and to better determine public health risks for those individuals using the Phillippi Creek for recreational purposes.

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