PHOSPHORUS AND CARBOHYDRATE LIMTATION OF FECAL COLIFORM AND FECAL ENTEROCOCCUS WITHIN TIDAL CREEK SEDIMENTS

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A Thesis Submitted to the
University of North Carolina Wilmington in Partial Fulfillment
Of the Requirements for the Degree of
Master of Science

Center for Marine Science

University of North Carolina Wilmington

2006

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This thesis has been prepared in the style and format

consistent with the journal

Estuarine, Coastal and Shelf Science

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ABSTRACT

Aquatic sediments have been shown to be a significant reservoir for fecal bacteria and at concentations two to three orders of magnitude higher than the waters directly above them. These bacteria represent a potentially serious health threat to humans using these waters. This study was conducted to determine the abundance of fecal bacteria within tidal creek sediments of Bradley Creek and determine if their residence or growth may be limited by concentrations of sediment phosphorus (P), sediment carbon (C), salinity, and water temperature. Sediment fecal coliforms had a mean of 179 CFU/cm^2 (std. dev. = 411, range = 0-3230) for samples collected monthly at 6 stations over the course of this study. Were the bacteria and sediments to be suspended through a 100 cm water column this value would be the equivalent of 179 CFU/100ml which is just below the water quality standard for human contact (200 CFU/100ml). Such disturbances could easily be produced by natural or human activities. Samples for enterococcus had a mean of 285 CFU/cm^2 (std. dev. = 473, range = 0-1730). If these sediments and bacteria were similarly suspended they would equate to 285 CFU/100ml and greatly exceede the standard for human contact in the water column (33 CFU/100ml). Overall, only fecal coliform bacteria were correlated to sediment C, however, the signal from bioavailable C was probably masked by the presence of insoluble C from detrital cellulose. Neither bacteria were correlated to sediment P concentrations which were found to be greater in Bradley Creek sediments than limiting concentrations for coliforms in sediments concluded by previous research. Sediments were a significant reservoir of P as concentrations were recorded as high as 4-5 orders of magnitude greater than in overlying waters. Sediment fecal coliforms were shown to be negatively correlated with salinity and positively correlated with temperature conforming to patterns established by previous research. However, fecal enterococcus was not shown to have a

significant relationship with either salinity or temperature. Fecal coliforms were positively correlated to precipitation over the previous 24 hours.

Experimental addition of bioavailable P (potassium phosphate monobasic) and bioavailable C (dextrose) showed a positive relationship between both fecal bacteria and bioavailable C. Enterococcus was significantly correlated to P in trials with low initial sediment P concentrations. Fecal coliform was significantly correlated to P at $\alpha = 0.1$ where initial P concentrations were low. A higher α was taken into consideration due to the high variability of coliform data and relatively low degrees of freedom for individual experimental trials.

It was concluded that while P and C are important to fecal bacterial residence within sediments, P may no longer be limiting in Bradley Creek due to relatively high background concentrations. Elevated P and bioavailable C concentrations have been correlated to storm water runoff. Limitation of sediment fecal bacteria in Bradley Creek by these nutrients may be alleviated from their introduction via this mechanism.

ACKNOWLEDGMENTS

I must thank the National Science Foundation's GK-12 Teaching Fellowship Program, and the NC Sea Grant # CEHPS-03 for funding a big chunk of my graduate education.

I also must thank my committee chair, Dr. Lawrence Cahoon, whose unequalled brand of guidance, patience, and support has been given far beyond any expectation. Dr. Mallin for insight, advice, and review of my work. Dr. Joan Willey for her exceptional inspiration and compassion for those who most need it. –namely me. I could not have asked for a better support system.

This work would have been incredibly more difficult were it not for the help of Sarah Gillenwater, Sam Romano, Adam Poore, and Michelle Ortwine for all the help in the lab and the field, and Erin Carey for giving me a hand up when I needed it and keeping me from walking in front of traffic.

I owe so much to my friends and family who have supported me long before I began: Shane Baptista, Bob Cutting, Midori Albert, Patricia Turrisi and Ben Snyder, all of whom may never know how much they've done for me, my sister, who's sense of humor and perspective have always added balance, Mother Toothman for reminding me 'you can do it' no matter how much I resisted, and Father Toothman for 5 minute science lessons on the ride home. I'd also like to thank Megan for her love and support through late nights at the library, approaching deadlines, and me. She stuck with me when I needed it most and continues to make it all worthwhile. She clearly doesn't know what she's in for.

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INTRODUCTION

The microbial quality of coastal water bodies in Eastern North Carolina is important from both an economic and public health standpoint and is often the cause of shellfishing and recreational water closures. Aquatic sediments have been shown to be a significant reservoir for fecal coliform bacteria and frequently host concentrations one to three orders of magnitude higher than the overlying waters. (Buckley et al., 1998; Grimes, 1975 and 1980; LaLiberte and Grimes, 1982; Tunnicliff and Brickler, 1984; Doyle et al., 1984; Doyle et al., 1992; Van Donsel and Geldreich 1971). Sediment-bound fecal coliform bacteria commonly exist in high enough concentrations to cause non-attainment of use standards were they to be suspended in the water column by storms, or any other disturbances (Rowland, 2002). Several environmental factors have been identified that may act to enhance the role of sediments as a reservoir for enteric pathogens and can be further altered by anthropogenic influences.

Phosphorous

Carlsson and Caron (2001) and Mallin et al. (2004) found that phosphorus (P) rather than nitrogen could limit bacteria production in freshwater systems. Furthermore, Sundareshwar et al. (2003) and Castillo et al. (2003) both concluded that sediment bacterial production is limited by P in coastal ecosystems and postulated that this is also the case in many other P deficient ecosystems. Sundareshwar et al. (2003) also experimentally found that increases in phosphate concentrations result in bottom-up ecological changes affecting the nitrogen cycle, carbon cycle, and primary production. Among the bacteria that may be enhanced by increasing P concentrations are fecal coliform bacteria (Mallin et. al. 2003). Burkholder et al. (1997) and Mallin et al. (2000) explained that a positive correlation between increased fecal coliforms and increased nutrient concentrations could be due to either a common source or the coincidental

arrival of both to a common location. Sheheta and Marr (1971) found that in areas of low nutrient concentrations *E. coli* growth rates were highly dependent on PO_4^{3-} (<1.0 mM).

Autotrophic organisms are capable of fixing carbon and thereby are not limited by organic carbon as a nutrient. Heterotrophic organisms, enteric bacteria in this case, rely on some external source of carbon. Potential co-limitation of bacterial production by P and carbon (C) has been identified where the organic carbon supporting bacterial production is derived primarily from algal carbon fixation (Castillo et al. 2003). Evidence exists that bacteria are capable of out-competing algal communities for inorganic nutrients and that bacterial production is effectively limited by algal production of fixed carbon (Currie and Kalff, 1984; Kirchman, 1994). By this proposed mechanism, bacteria may be indirectly limited by P or by whatever nutrient is limiting algal production for a given set of circumstances.

Fecal Coliform Bacteria

Fecal coliforms are currently an indicator of coastal water quality (NCDEHNR 1996; APHA, 2001). Fecal coliform bacteria originate as gut flora in warm-blooded animals and are the most commonly used indicators of the presence of fecal material, sewage contamination, and pathogenic enteric bacteria (Dadswell, 1993; Rees et al., 1998; Mallin et al. 2000, 2001; Benson, 2002). Coliform bacteria are used as microbial indicators because they are non-spore forming, facultative anaerobes and are estimated to survive longer than other enteric pathogens. They should therefore provide a more conservative estimate of the presence of potentially harmful bacteria (Benson, 2002). However, some studies have found that fecal coliforms may not be reliable as an indicator of bacterial or viral pathogens in the water column or sediments, underscoring the need for additional testing methods such as the use of fecal enterococcus as a

microbial indicator (Burton et al.1987; Ferguson et. al. 1996; Davis et. al. 1977; LaBelle et. al. 1980; Sayler et. al. 1975).

Fecal Enterococcus

Fecal enterococci are also intestinal flora native only to warm blooded animals and are another microbial indicator of the presence of human or animal sewage. These bacteria are less likely to persist or multiply in the water column than fecal coliforms (APHA, 2001). *Enterococcus faecalis* and *Enterococcus faecium* in particular are associated with human sewage and provide a more reliable distinction between human wastes and that of avian and domesticated/wild megafauna (Farrow et al., 1983; Laukova and Juris, 1997; Pinto et al., 1999).

Survival Of Sediment-Bound Bacteria

Fecal coliform persistence in the water column is relatively short lived due to high susceptibility to solar radiation (Chamberlain and Mitchel, 1978; Solic and Krstulovic, 1992; Gerba and McLeod, 1976), salinity (Cabelli, 1978; Lessard and Sieburth, 1983), and predation (Davies et al., 1995). Fecal coliform bacteria as well as many of the pathogenic enterics with which they are associated have been observed to persist at much higher concentrations in the sediments just below the water/sediment interface than in the overlying waters (Sherer et al. 1992; Burton et al., 1987). Substantially higher concentrations of coliforms and other pathogenic bacteria have been reported persisting for longer periods of time in the sediments than in the water directly above (Van Donsel and Geldreich, 1971). There is also a growing body of evidence that sediment-bound coliforms are capable of growth within the sediments (Chan et al., 1979; Gerba and McLeod, 1976; Hood and Ness, 1982; LaLiberti and Grimes, 1982; Sheheta and Marr; 1979; Rowland, 2002). Effective delivery methods to the sediment-water interface have been identified for both bacteria and nutrients. Precht and Huettel (2003) found that

pressure gradients created by shallow-water wave action and sediment wave ripples increased fluid exchange by a factor of 50 over what could be expected by molecular diffusion alone. This mechanism is capable of transporting nutrient-laden particulate matter as well as dissolved nutrients. Although not directly mentioned, it would follow that bacteria, both free and sediment borne, may also be transported by this method.

Bacteria are thought to recruit to sediments through the sedimentation of the particles to which they are adsorbed (Gannon et al., 1983). Research has shown that sediment-bound bacteria may benefit from the relationship. Sediment particles provide shelter from solar radiation, may ease pressures from predation, and can enhance bacterial persistence (Davies and Bavor, 2000). Populations of sediment-bound bacteria are typically inversely proportional to sediment grain size (Chan et.al., 1979; Erkenbrecher, 1981). Bacteria also adsorb to smaller ($\leq$ 2μm) sediments preferentially, which settle more slowly and provide enhanced mobility and importation of bacteria including coliform bacteria (Dale, 1974). Smaller sediment grain sizes provide higher surface area:volume ratio and are more often associated with organic and inorganic nutrients (Dale 1974, Chan et al. 1974). Others have also concluded that fine grained silts and clays may enhance the survival of fecal coliforms (Burton et al., 1987; Hood and Ness, 1981; Howell et al., 1996).

The Bradley Creek watershed (Fig. 1) is 72% residential and has been identified as having the poorest overall microbial water quality in New Hanover County, NC (Mallin et al. 2000). The New Hanover Tidal Creeks Project and the Wilmington Watersheds Project have sampled 5 of this study's sampling locations for the previous 11 years and past 7 years, respectively. Between 2001-2002 fecal coliform concentrations in Bradley Creek exceeded the North Carolina standard for human contact (200 CFU/100mL) at 5 of 7 stations more than a third

of the times sampled and as often as 83% of the times sampled for one station (Mallin et al., 2003). Residential use of phosphate-containing fertilizers in the Bradley Creek drainage basin is a major source of phosphate and phosphate-adsorbed sediments to the watershed (Cahoon, 2002). Mallin et al. (2000) identified residential pet waste as a major contributor of fecal coliform bacteria to the watershed.

HYPOTHESES

$H_{Ø1}$: There is no correlation between sediment phosphorus concentrations and sediment-bound fecal coliform concentrations.

$H_{Ø2}$: There is no correlation between sediment phosphorus concentrations and sediment-bound fecal enterococcus concentrations.

$H_{Ø3}$: There is no relationship between sediment phosphorus and carbohydrate concentrations and sediment-bound fecal coliform concentrations.

$H_{Ø4}$: There is no relationship between sediment phosphorus and carbohydrate concentrations and sediment-bound fecal enterococcus concentrations.

$H_{Ø5}$: Experimental addition of phosphorus and carbohydrate to sediments will have no effect on sediment-bound fecal coliform growth or persistence within specimen cups anchored to streambeds.

$H_{Ø6}$: Experimental addition of phosphorus and carbohydrate to sediments will have no effect on sediment-bound fecal enterococcus growth or persistence within specimen cups anchored to streambeds.

METHODS

Site Description

Sediments from six sample sites in the Bradley Creek watershed were sampled during this study (Fig. 1). Sample Sites BC-SBU, BC-CR, BC-CA, and BC-NBU are freshwater tributaries to Bradley Creek. BC-CA is a residential storm water detention pond that flows into the watershed. These sites have typically small-grained sands mixed with some organic matter. Sites BC-NB and BC-S are a part of the Bradley Creek estuarine zone.

Sample Collection

Sediment samples were collected from each site once monthly between January, 2003 and March, 2005. The six sample sites were divided into two groups of three. Group 1 (BC-CA, BC-CR, and BC-SBU) was typically sampled during the second week of each month while Group 2 (BC-NBU, BC-NB, and BC-S) was typically sampled during the third week of each month. Tidally influenced sites BC-NB and BC-S were sampled within an hour of low tide.

At each site temperature and salinity of the water were recorded using a YSI 85 multi-parameter water quality meter. Six sediment cores approximately 2 cm deep and 2.36 cm in diameter were collected using sterile, acid washed PVC coring tubes and placed into pre-weighed, acid washed, sterile polypropylene 50 ml centrifuge tubes. Three tubes were designated for membrane filtration and the remaining three tubes were designated for P and total carbohydrate analysis. The tubes were then cooled on ice until returning to the lab for processing. The supernatant was then decanted from each core and discarded. The three cores designated for sediment P analysis were then placed into an ultra cold freezer (-80°C) for 24 hours before lyophilization using a Virtis Benchtop 3.3 Vacu-Freeze lyophilizer.

Laboratory Analysis

The three cores designated for bacterial analysis were placed into 1L flasks and mechanically suspended in 1L of phosphate buffered (0.25M KH_2PO_4, pH adjusted to 7.2 with 0.1N NaOH) rinse water. Magnetic stir bars were used to gently stir the suspension for 2 minutes to homogenize and release the sediment-bound bacteria in the buffer solution. From this suspension coliforms, enterococci, and streptococci were enumerated according to methods outlined by APHA, 2001 (APHA method 9222 D, APHA method 9230 C). Following incubation individual dark blue colonies were assumed to represent colony forming units (CFU). The number of CFU g^{-1} and CFU cm^{-2} were then derived by the following equations respectively:

$CFU \cdot g^{-1} =$ (colonies x 100) / core *weight* in grams

$CFU \cdot cm^{-2} =$ (colonies x 100) / 4.37 cm^2

where 4.37 cm^2 is the area of the sediment / water interface sampled by the core. Fecal bacteria tend to be located in at the top layer of sediments along the sediment/water interface as described by Sherer et al. (1992) and Burton et al. (1987). The results of the three membrane filtrations for each site were then averaged to find a site mean.

The frozen cores were freeze dried and kept dry to be analyzed for total phosphate (TP) and Total Carbohydrate (TC). TP was quantified using a persulfate digestion method outlined by Valderrama (1981). TC quantification was performed according to the methods outlined by Underwood et al. (1995). TP per unit weight of sediment was calculated as follows:

$\mu g\ TP/g = (\mu M\ P \times 0.04L \times 31) / (\text{weight of sediment})$

where 0.04L is the sample volume with oxidation reagent and distilled water, and 31 is the molecular weight of P. The concentration of total carbohydrate was calculated as follows:

$$\mu g\ TC/g = \frac{(Abs - constant) / (coefficient)}{sample\ weight\ (g)}$$

where constant (c) and coefficient (m) were obtained from a linear calibration curve derived from glucose standards, where $y = m x + c$

Experimental Analysis

To experimentally test the effects of varying P and organic C concentrations on sediment bacteria, sediment cores taken in triplicate from the field were incubated in solutions of varying P and C concentrations using a 2x2 design with 2 levels of each (Table 1). Cores for the first two trials were procured from site BC-NBU, which was also sampled for field data collection. Site 210 is beside a bridge on NC Hwy 210 crossing a tributary to the Northeast Cape Fear River located in a relatively pristine and undeveloped area of northern New Hanover County, NC. BWP is a small arboreal pond within the Bluthenthal Wildflower Preserve on the campus of UNCW. Site REC is a storm water detention pond next to the UNCW Recreation Center, also located within the main campus of UNCW. Station BC-CR is an unnamed tributary to Clear Run Branch, a main tributary of Bradley Creek.

Cores were then analyzed by suspension and colony counting as above to determine effects if any on bacterial concentrations. Sodium phosphate monobasic (KH_2PO_4) and dextrose ($C_6H_{12}O_6 \cdot H_20$) were used as the source for each nutrient respectively. Sediment cores were procured during 6 sampling events at 5 separate locations (Fig. 2) by the same methods described for field samples. During each sampling event three cores were collected for initial P analysis,

and three for initial bacterial enumeration of coliforms and enterococci. Another 16 cores were collected for incubation in all combinations of each treatment.

Each treatment consisted of incubating 3 randomly selected cores in a sterile, acid washed 1L beaker containing one of the treatment solutions. Cores were incubated for 24 hours with gentle aeration for circulation. Treatment solutions consisted of a sodium borate/boric acid buffer (pH=8) and one of the four possible treatment combinations of C or P addition/deletion (phosphorus and dextrose, phosphorus only, dextrose only, and deionized water only) as illustrated by Table 1. In order to reflect the relative ratios of nutrient uptake P was supplemented at 100 mg/L and dextrose at 1000 mg/L. Each treatment of 3 replicate sediment cores was incubated at room temperature for 24 hours at ~20^{o}C. Following the incubation 3 cores from each treatment were then processed for coliform and *F. enterococci* enumeration according the modified methods used for the field samples. The remaining core from each incubation was used for a final TC and TP analysis also using the same methods as the field samples.

Statistical Analysis

Statistical analyses were performed using SAS Institute's JMP Version 4.0.2 (Academic) except where indicated. Both fecal coliform and fecal enterococcus data were non-normal as determined by the Shapiro-Wilk test and were normalized through log transformation where possible. Microbial field data were analyzed both as a whole set as well as by site. Correlations for transformed data were determined by calculation of Pearson Product-Moment Correlation Coefficients. Non-normally distributed or bimodal data that could not be reasonably transformed were analyzed for possible correlations using Spearman's Rho Correlation Coefficients. A Simple Logistic Regression was performed to determine if any significant relationships existed between microbial data and TP and TC concentrations. A 2-factor multiple regression analysis

was also employed to determine covariations between microbial and nutrient concentrations. A multiple regression was also used to determine correlation when all measured parameters (temperature, salinity, coliform, sediment P, sediment C, and precipitation for 24 and 72 hours) were taken into account.

Data collected from the experimental manipulation of P and carbohydrate concentrations were non-normally distributed by the Shapiro-Wilk test and log transformed. Data were analyzed using a Two-Way ANOVA to identify differences in treatment effects and possible interactions between treatments. To investigate differences among treatments using pooled data from all trials, net change was analyzed using the Kruskal-Wallis Two-Way ANOVA for non-parametric data (Sokal and Rohlf, 1995) in Microsoft Excel.

RESULTS

Data Description

The field data describe water temperature and salinity, sediment-bound F. coliforms, sediment-bound F. enterococcus, total sediment carbohydrate, total sediment P (Table 2), and precipitation for the past 24, 48, and 72 hours for each sampling event at each site. Bacterial concentrations varied greatly for the entire dataset as well as on a site by site basis. The geometric means for bacterial concentrations ranged from 0 – 3,230 CFU/cm^2 with a mean of 179 CFU/cm^2 and std. dev. = 411 CFU/cm^2 for coliforms, ranged from 0 – 173 CFU/cm^2 with a mean of 285 CFU/cm^2 and std. dev. = 473 CFU/cm^2 for enterococcus and ranged from 10 – 4,920 CFU/cm^2 with a with a mean of 449 CFU/cm^2 and std. dev. = 786 for streptococcus. Sediment total P ranged from 12– 671 μg/g with a mean of 183 and std. dev. = 144. Sediment total carbohydrate ranged from 73 – 45,500 μg/g with a mean of 7,630 and std. dev. = 10,100.

Field data were also analyzed with respect to individual sample sites. High and low means for coliform counts were at Station BC-SBU (340 CFU/cm^2) and Station BC- S (125 CFU/cm^2) respectively. The high and low means for enterococcus counts were at Station BC-NB (528 CFU/cm^2) and station BC-CR (65 CFU/cm^2) respectively. Station BC-S had the highest mean for sediment P (316) while M had the lowest (101 μg/g**).** The high mean for sediment C was at Station BC-S (23,500 μg/g) and the lowest mean was found at Station BC-CR (714 μg/g).

Data for P and coliforms in the water column in Bradley Creek during the same time period were procured (Mallin et al., 2003; Mallin et al., 2004). Coliforms were measured in the water column at BC-CA where NC standards (200 CFU/100ml) were exceeded 82% of times sampled from 2003 -2004 and 86% of times sampled from 2004 – 2005. Bacteria counts ranged from 155 - 13,000 CFU/100mL with a mean of 1093 for 2002-2003 and ranged from and ranged from 60-6,000 CFU/100ml with a mean of 1093 CFU/100mL for 2003-2004. Orthophosphate was measured in the water column at all sites (Table 3). For 2002-2003, orthophosphate ranged from 0.001-0.052 mg/L and means for each site were all ≤0.02 mg/L. For 2003-2004 P ranged from 0.002-0.044 and means for each site were ≤0.019.

Field Data

Combined field data for bacteria from all sites were not normally distributed and were normalized through log transformation to achieve normality. The Pearson Product-Moment Correlation was used to determine correlations between normalized data sets for bacterial counts (Table 3). A strong significant correlation was shown to exist between F. coliform and F. enterococcus (coef. = 0.501, p = 0.0005, df = 45, Fig. 3a). There was also a consistent significant correlation between enterococcus and streptococcus (coef. = 0.388, p = 0.008, df = 45, Fig. 3b).

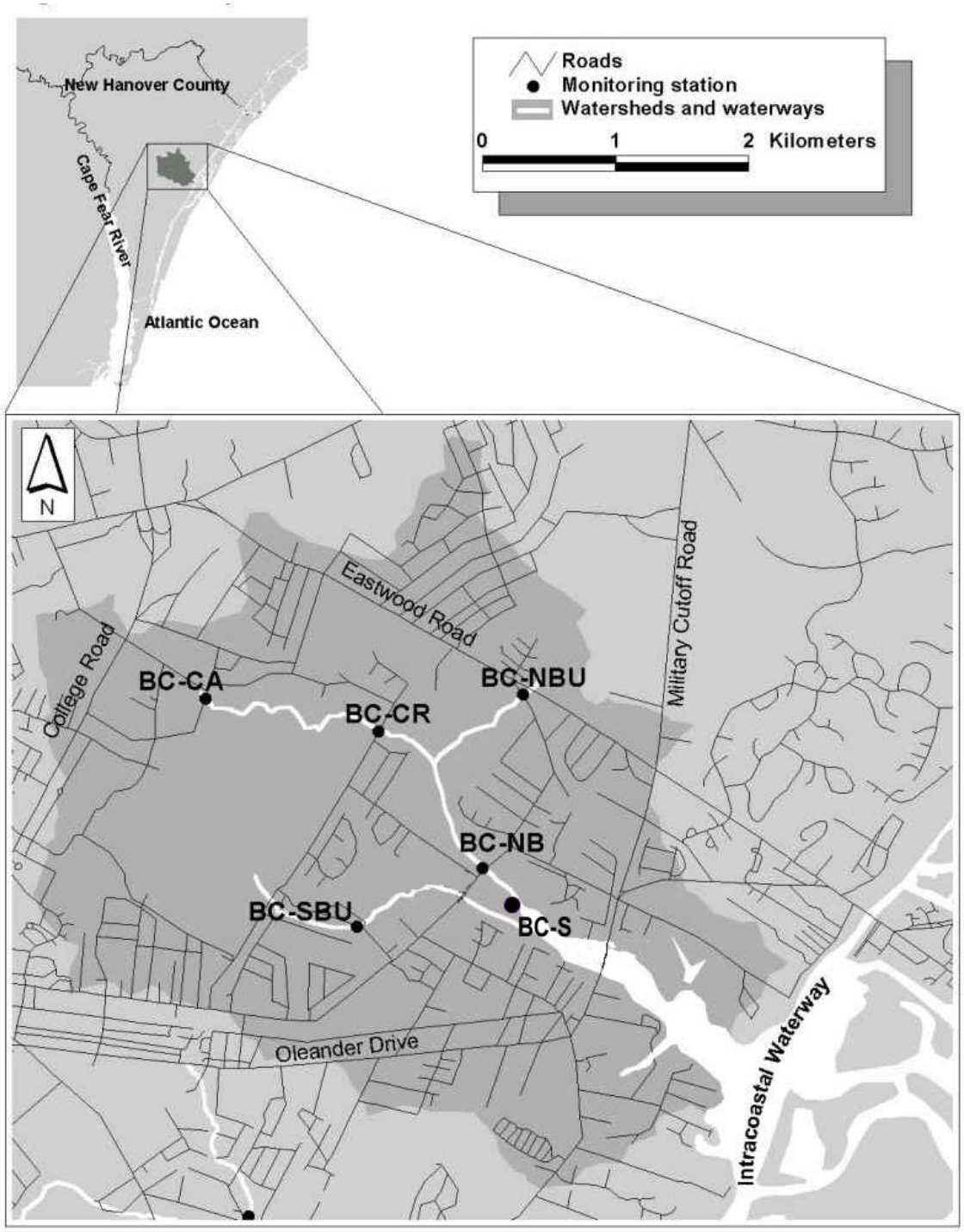


Fig. 1. Bradley Creek Watershed and field sampling locations.

Table 1. 2x2 Experimental Design.

		Phosphorus 100 100ppm	
		P+	P-
Dextrose 1000 ppm	D+	D+ P+	D+ P-
	D-	D- P+	D- P-

Dextrose ($C_6H_{12}0_6$) served as the organic carbon source and is denoted in this Table as 'D', while sodium phosphate ($NaH_2PO_4 \cdot H_2O$) served as the phosphorus source and is denoted as 'P'. Concentrations were measured as elemental phosphorus. The symbols + and – indicate either the addition or no addition of the nutrient in the incubation solution. D- P- served as a control for this experiment.

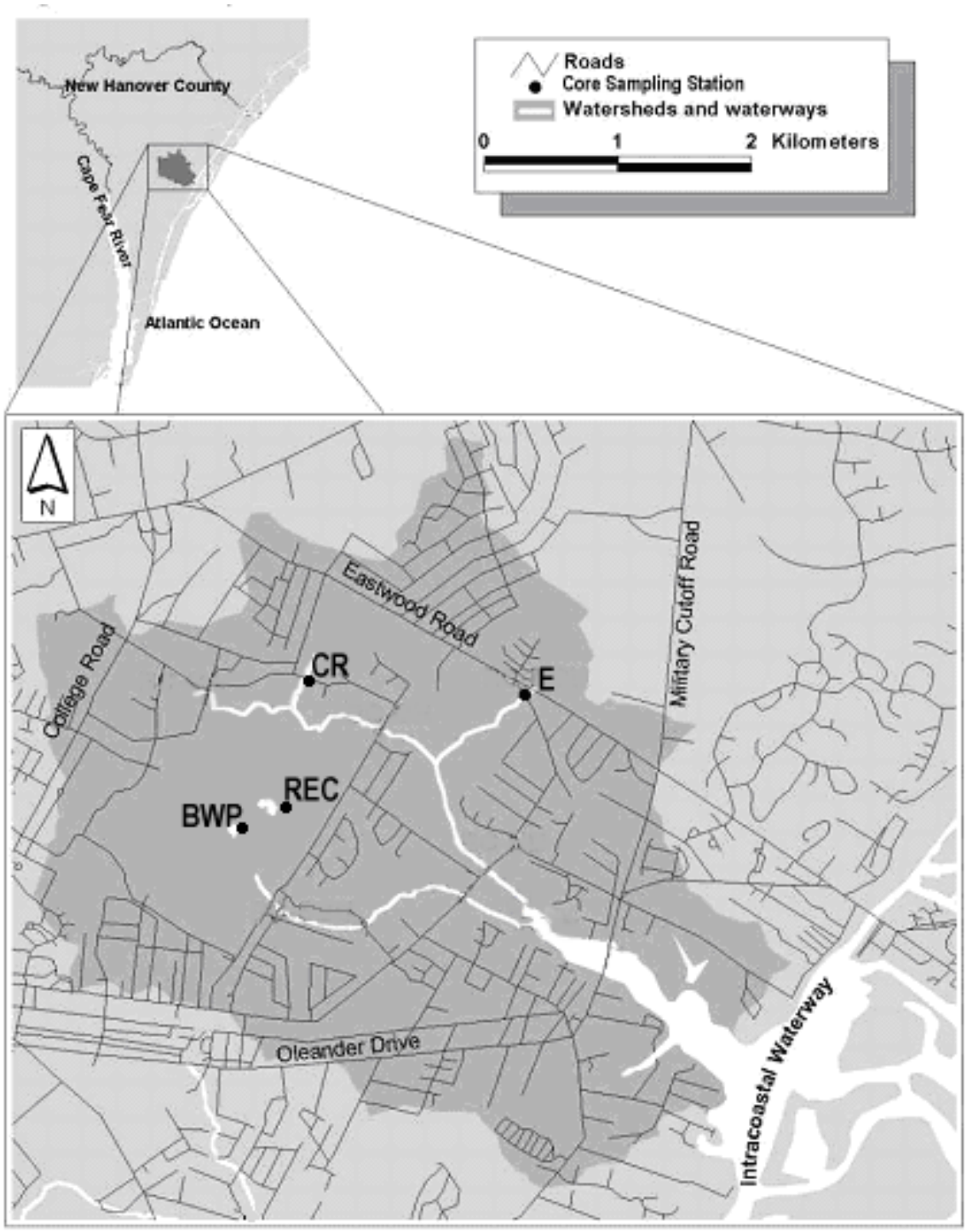


Fig. 2. Locations of sampling sites for cores used in experimental trials. Site 210 is out of the range of this map.

Table 2. Mean, Range, and Standard Deviation for sediment coliform, enterococcus, streptococcus, Total Phosphorus, and Total Carbohydrate for all Field Sites as in Figure 1.

		coliform	enterococcus	Total Phoshorus	Total Carbohydrate
	mean	179	285	182	7630
All	**range**	0-3230	0-1730	12.4-671	73.1-45,500
	σ	411	473	144	10090
	mean	340	332	137	1300
BC-SBU	**range**	2-3230	29-1730	39.8-464	114-2940
	σ	696	587	111	659
	mean	32	202	145	9210
BC-CA	**range**	0-295	0-868	15.8-354	1780-45,500
	σ	68	293	98	9194
	mean	186	365	119	2640
BC-NBU	**range**	0-1430	22-1370	6.1-465	202-14,800
	σ	274	493	136	3520
	mean	257	65	101	714
BC-CR	**range**	3-2770	0-234	26.8-380	73.1-3400.0
	σ	550	90	76	655
	mean	125	251	316	23500
BC-S	**range**	0-1550	4-1230	12.4-671	2070-45,200
	σ	301	448	153	11700
	mean	132	528	261	8910
BC-NB	**range**	0-492	37-1670	65.2-654	2430-18,300
	σ	152	733	141	4440

Table 3. Correlations between indicator bacteria.

Site	Comparison	F-value / correlation coefficient	p-value	r^2	d.f.
	FC v FE	**11.6**	**0.014**	**0.660**	**7**
BC-SBU	FC v FS	1.202	0.315	0.167	7
	FE v FS	3.984	0.093	0.399	7
	FC v FE	2.452	0.168	0.290	7
BC-CA	FC v FS	0.298	0.605	0.047	7
	FE v FS	0.036	0.855	0.060	7
	FC v FE	0.903	0.386	0.153	6
BC-NBU	FC v FS	4.538	0.086	0.476	6
	FE v FS	0.107	0.757	0.021	6
	FC v FE	2.052	0.202	0.255	7
BC-CR	FC v FS	0.796	0.407	0.117	7
	FE v FS	1.228	0.310	0.170	7
	FC v FE	**62.3**	**0.000**	**0.912**	**7**
BC-S	FC v FS	2.509	0.174	0.334	6
	FE v FS	2.361	0.185	0.321	7
	FC v FE	0.011	0.919	0.002	6
BC-NB	FC v FS	0.098	0.766	0.019	6
	FE v FS	0.683	0.446	0.120	6
	FC v FE*	**0.501**	**0.001**	**0.262**	**45**
Field	FC v FS*	0.248	0.101	0.051	44
	FE v FS*	**0.388**	**0.008**	**0.154**	**44**

Pearson's Product-Moment was used to determine correlations between normally distributed data. Spearman's Rho (*) used to determine correlations for data that were not normally distributed. Significant values are indicated by bold type.

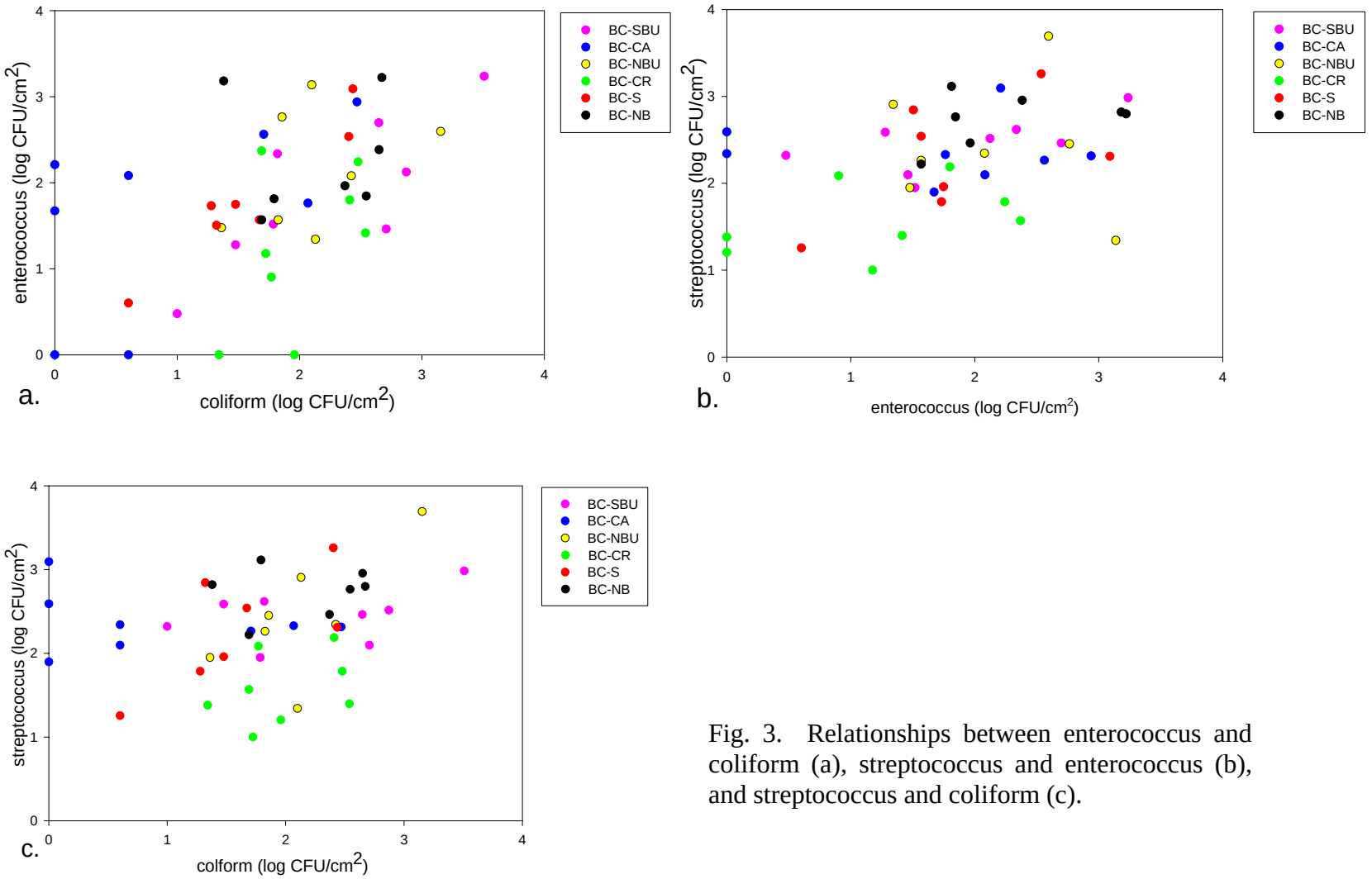


Fig. 3. Relationships between enterococcus and coliform (a), streptococcus and enterococcus (b), and streptococcus and coliform (c).

No correlation was detected between coliforms and streptococcus (Fig. 3c). Significant correlations between coliforms and enterococcus were detected at station BC-SBU ($F = 11.6$, $p = 0.014$, $df = 6$) and Station BC-S ($F = 62.3$, $p = 0.0002$, $df = 6$) when the data were analyzed by site. There were no correlations between enterococcus and streptococcus or between coliform and streptococcus at any of the sites. Sampling efforts for streptococcus were discontinued because there was no correlation with coliform data with relatively high degrees of freedom. It was decided that the streptococcus signal was likely influenced by other sources.

No significant correlation was found between sediment P and either indicator bacteria in combined field data (Table 4). Sediment P did not correlate with either coliform or enterococcus concentrations at any individual site either (Fig. 4). Sediment C concentrations significantly correlated with coliform counts ($F = 4.541$, $p = 0.035$) for combined field data (Fig. 5, Table 4). Sediment P did not correlate with either coliform or enterococcus concentrations at any individual site either (Fig. 4). Sediment C concentrations significantly correlated with coliform counts ($F = 4.541$, $p = 0.035$) for combined field data (Fig. 5, Table 4). There was also a significant correlation between enterococcus and carbohydrate at Station BC-NBU ($F = 9.620$, $p = 0.027$, d.f. = 0.658). Sediment C did not correlate with coliform counts at any individual site. A multiple regression analysis showed that there was no significant combined effect from sediment P and sediment C on coliforms or enterococcus (Table 5).

Only sites S and W were estuarine and subject to increased salinity. Data from both sites were combined to look for significant effects (Table 6). Coliform counts decreased significantly with increasing salinity ($F = 23.678$, $p < 0.0001$, d.f. = 49, Fig. 6 a). Salinity had no significant effect on enterococcus when data from sites were combined (Fig. 6b). When analyzed by individual site salinity had a significant effect on coliform counts at both Station BC-S ($F =$

Table 4. Pair-wise regression between bacteria and phosphorus and carbohydrate.

Site	Comparison	F-value	Prob>F	R2	d.f.
BC-SBU	Fc v TP	3.082	0.090	0.118	23
	Fc v TC	1.320	0.321	0.045	22
	Fe v TP	0.052	0.828	0.010	5
	Fe v TC	<0.000	0.999	<0.000	6
BC-CA	Fc v TP	0.438	0.515	0.186	23
	Fc v TC	0.119	0.734	0.005	22
	Fe v TP	0.001	0.976	0.000	5
	Fe v TC	4.034	0.091	0.040	6
BC-NBU	Fc v TP	3.333	0.080	0.122	24
	Fc v TC	0.041	0.842	0.002	23
	Fe v TP	0.563	0.487	0.101	5
	Fe v TC	**9.620**	**0.027**	**0.658**	**5**
BC-CR	Fc v TP	0.460	0.504	0.018	23
	Fc v TC	0.113	0.470	0.005	21
	Fe v TP	0.180	0.683	0.036	5
	Fe v TC	0.285	0.613	0.045	6
BC-S	Fc v TP	0.463	0.503	0.020	23
	Fc v TC	1.514	0.228	0.068	21
	Fe v TP	1.439	0.284	0.223	5
	Fe v TC	0.882	0.391	0.150	5
BC-NB	Fc v TP	0.215	0.647	0.009	23
	Fc v TC	3.894	0.062	0.456	21
	Fe v TP	0.791	0.414	0.137	5
	Fe v TC	0.067	0.806	0.251	4
Field	Fc v TP	1.432	0.233	0.009	150
	Fc v TC	**4.541**	**0.035**	**0.031**	**139**
	Fe v TP	0.167	0.685	0.004	40
	Fe v TC	0.716	0.790	0.001	43

Pair-wise comparisons are shown for coliform and total sediment phosphorus (Fc v TP), coliform and total sediment carbohydrate (Fc v TC), enterococcus v total sediment phosphorus (Fe v TP), and enterococcus and total sediment carbohydrate (Fe v TC). Prob > F is the probability that the F value would be greater than the critical F – value for significance.

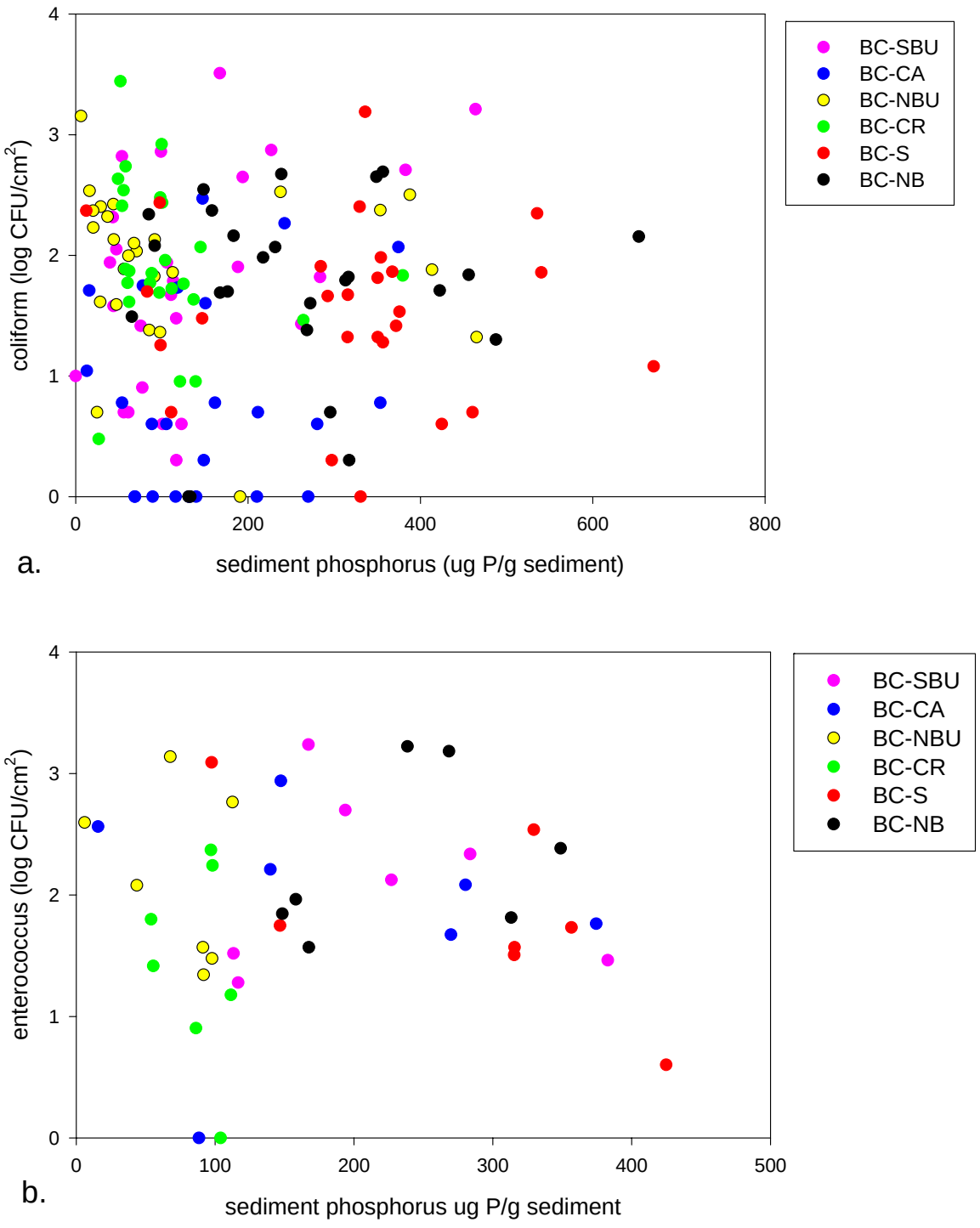


Fig. 4. Effects of phosphorus on coliform (a) and enterococcus (b) in Bradley Creek.

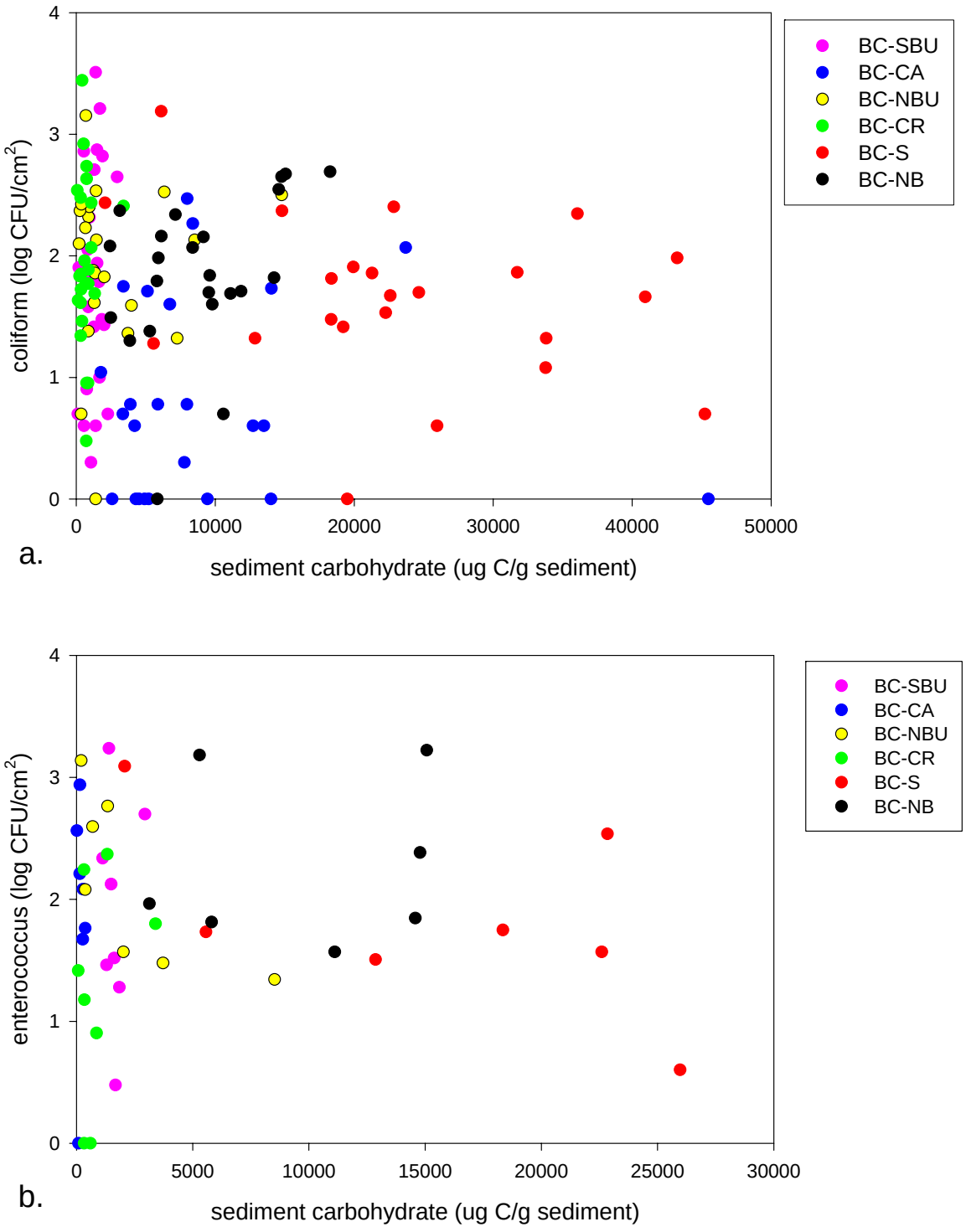


Fig. 5. Effects of carbohydrate measured as total carbohydrate on fecal coliform (a) and enterococcus (b) in Bradley Creek.

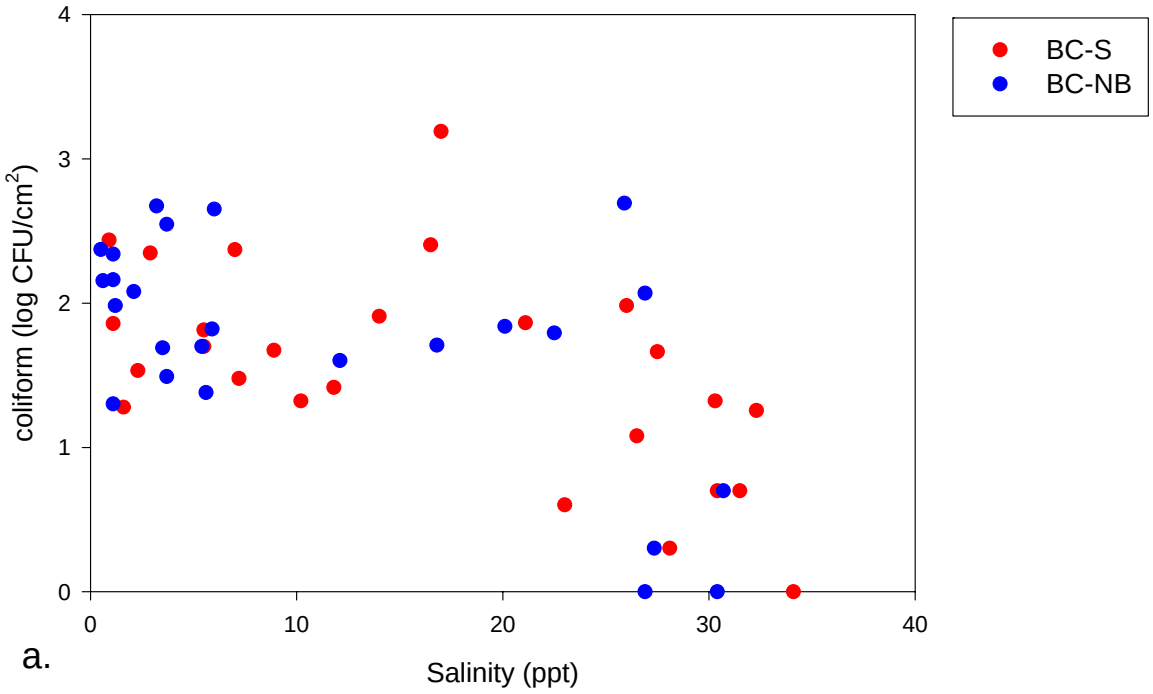
Table 5. Site-wise regression analysis of bacteria and phosphorus and carbohydrate field data.

Site		F-value	p-value	d.f.
BC-SBU	**coliform**	1.729	0.203	20
	enterococcus	0.128	0.884	4
BC-CA	**coliform**	0.633	0.541	20
	enterococcus	0.846	0.494	6
BC-NBU	**coliform**	1.732	0.202	20
	enterococcus	3.890	0.115	4
BC-CR	**coliform**	0.308	0.739	19
	enterococcus	0.120	0.890	0
BC-S	**coliform**	0.734	0.430	20
	enterococcus	0.672	0.560	4
BC-NB	**coliform**	1.925	0.172	20
	enterococcus	0.341	0.730	4
Field	**coliform**	1.877	0.107	135
	enterococcus	0.094	0.910	39

Table 6. Linear regression analysis of bacteria and salinity.

Site		F-value	p-value	r2	d.f.
BC-S and BC-NB	F.C.	**23.678**	**<0.0001**	0.326	49
	F.E.	3.521	0.085	0.227	12
BC-S	F.C.	**10.227**	**0.004**	0.3	24
	F.E.	1.098	0.22	0.282	5
BC-NB	F.C.	**11.577**	**0.002**	0.335	23
	F.E.	0.227	0.654	0.043	5

Data are for estuarine sites S and W only. Significant values are in bold type.



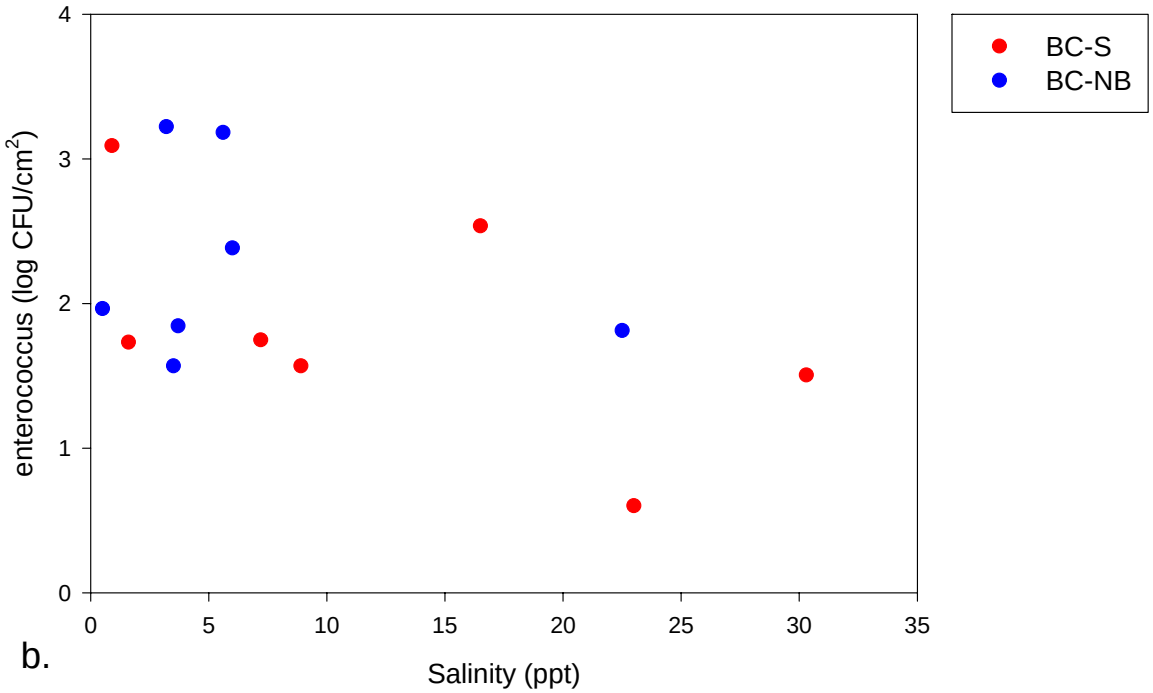


Fig. 6. Effect of salinity on sediment fecal coliform (a) and sediment fecal enterococcus (b).

10.227, p = 0.004) and Station BC-NB (F =11.577, p = 0.002, d.f. = 23). No significant effect from salinity was seen for enterococcus at either site.

Temperature did not have a significant effect on either bacteria for the combined field data (Table 7). When data were examined by site, station BC-SBU (F = 5.385, p = 0.031), CR (F = 8.546, p = 0.008), and E (F = 10.042, p = 0.004) showed that temperature had a significant effect on coliform counts (Table 7, Fig. 7a). Station BC-SBU was the only site at which a significant effect was observed for enterococcus (Fig. 7b). Sites BC-SBU, BC-CS, and BC-NBU are relatively shallow freshwater creeks and are subject to greater changes in temperature than some of the other sampling locations. This may explain the significance effect of temperature at these sites but not for the combined data.

A multi-way analysis of variance was performed to investigate any combined effects on coliforms. Hourly precipitation data was collected from a weather station located at Wilmington International Airport (station KILM 34°16'14" N, 77°54'09" W) for the preceding 24 hours and 72 hours (Figure 8) of each sampling event. Only precipitation for 24 hours preceding sample collection (Table 8) produced a significant effect (F = 17.4894, p = <0.0001). Sediment C, sediment P, salinity, temperature, and rainfall for the previous 72 hours resulted in no significant effects. Coliform data was considered an adequate proxy of enterococcus data because of the strong correlation between the two (Table 3). Coliforms were sampled more times overall than enterococcus and yielded a more powerful test due to greater degrees of freedom.

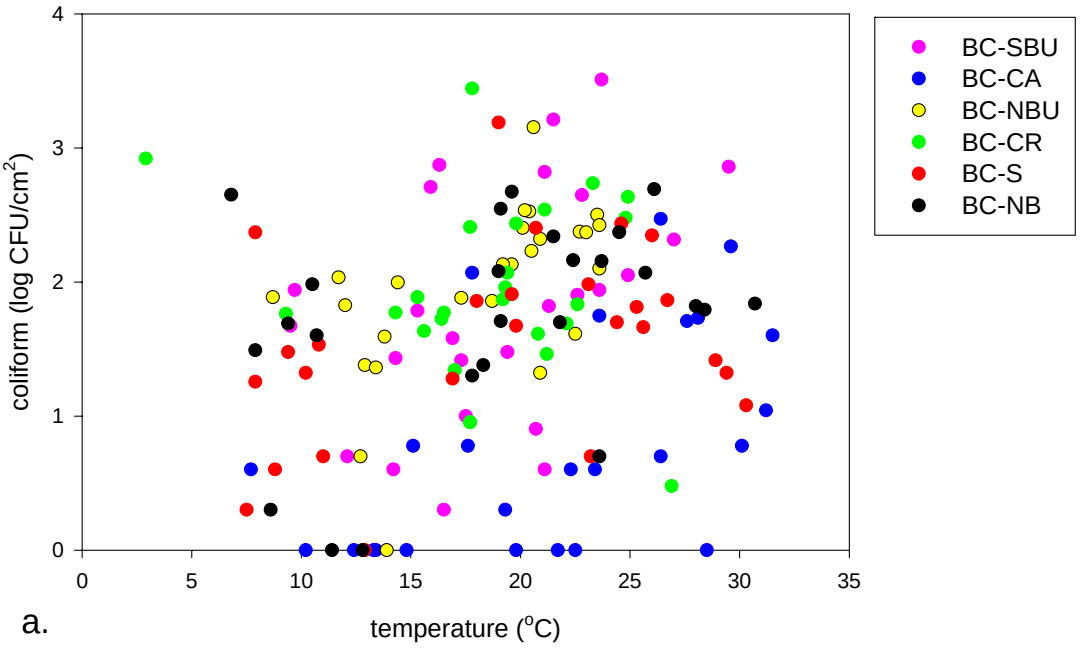
Experimental Data

Experimental data describe initial sediment P and carbohydrate concentrations for each trial as well as initial bacterial counts for coliform and enterococcus for cores before incubation (control) and incubation. Coliform counts following incubation ranged from 25 to 9150

Table 7. Linear regression analysis of bacteria and temperature.

	coliform			enterococcus		
Site	**F-value**	**p-value**	**d.f.**	**F-value**	**p-value**	**d.f.**
Field	1.990	0.166	150	3.096	0.086	43
BC-SBU	**5.385**	**0.031**	23	**6.174**	**0.048**	**6**
BC-CA	**8.546**	**0.008**	23	0.052	0.827	6
BC-NBU	**10.042**	**0.004**	**24**	3.490	0.120	5
BC-CR	0.505	0.484	23	3.771	0.102	6
BC-S	0.094	0.763	22	0.081	0.790	4
BC-NB	3.280	0.083	23	0.003	0.962	5

Significant values are in bold type.



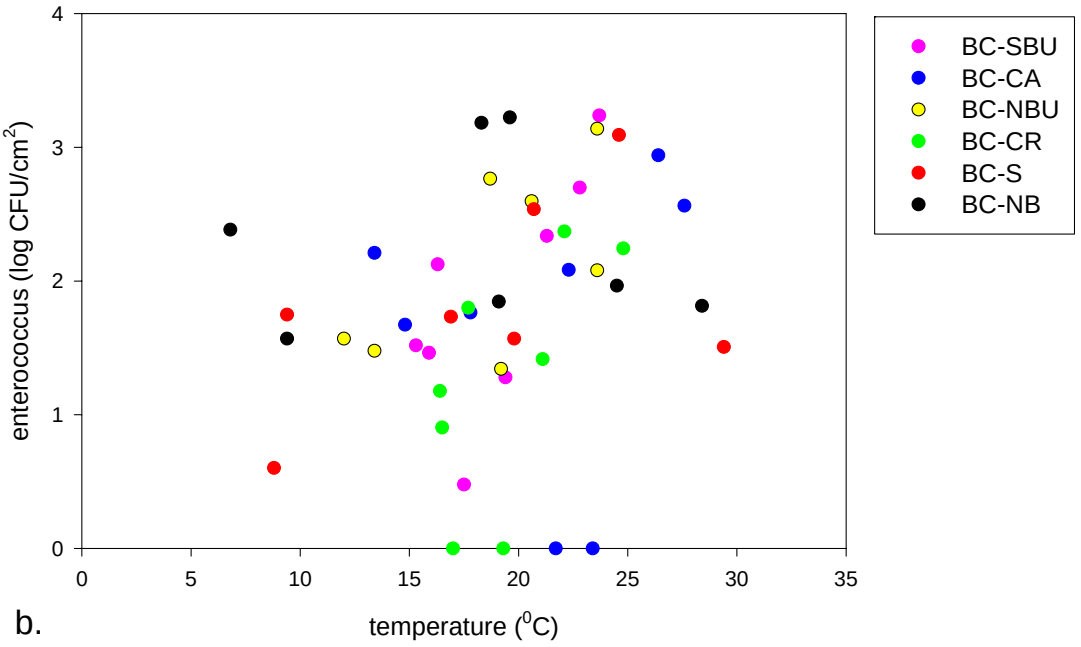
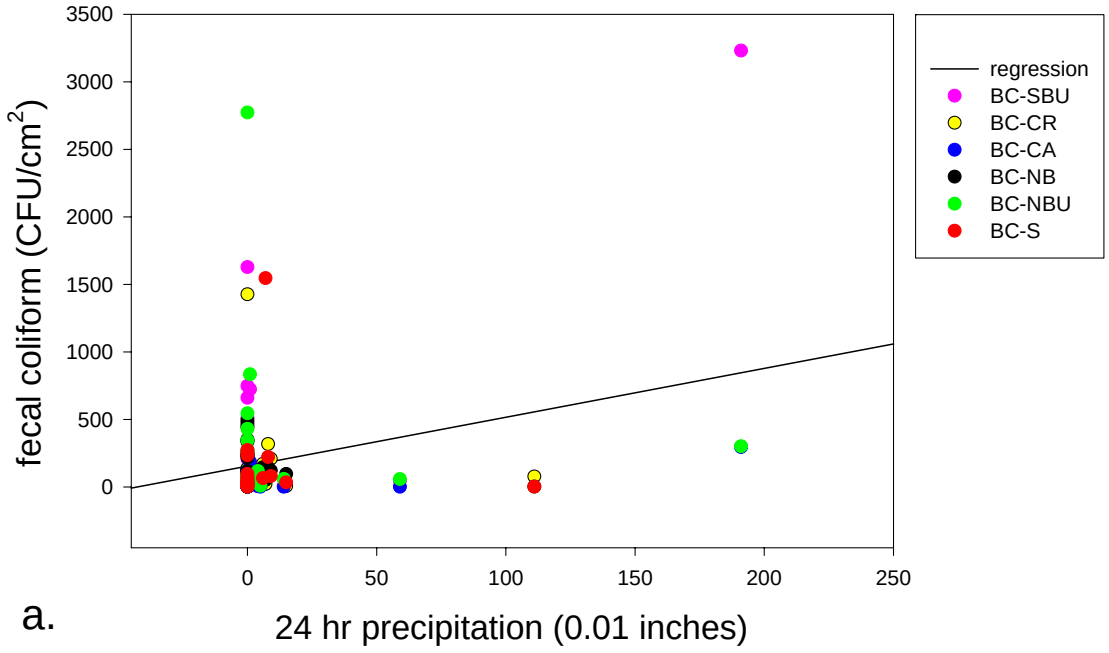


Fig.7 Effects of temperature on fecal coliform (a) and fecal enterococcus (b) in Bradley Creek sediments.



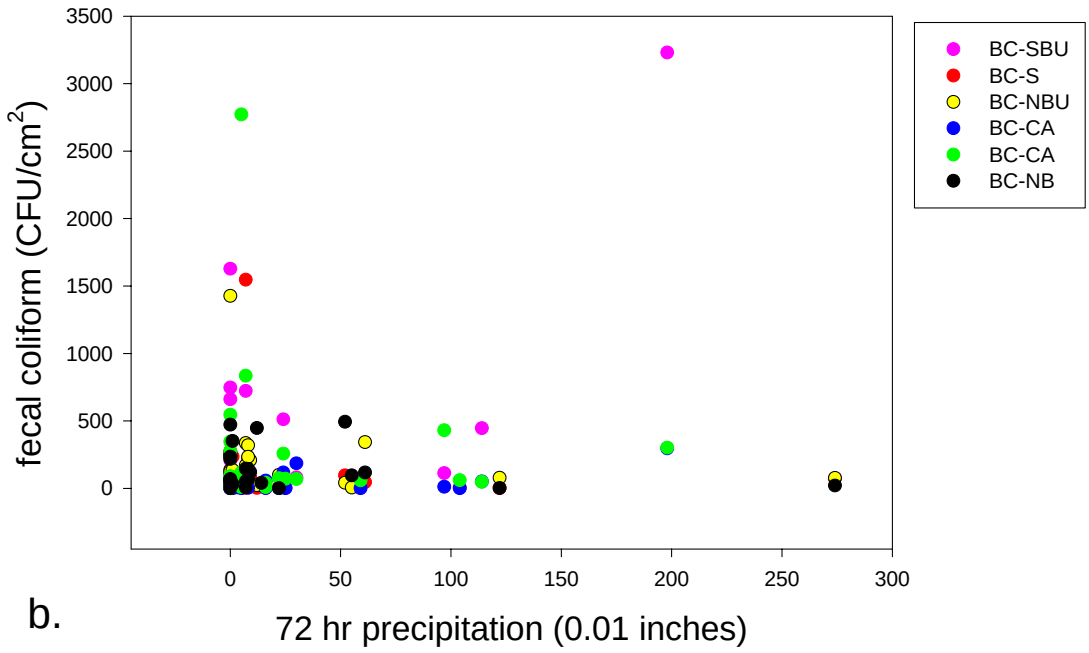


Figure 8. Effects of precipitation over the previous 24 hours (a) and previous 72 hours (b).

Table 8. Multi-way ANOVA of temperature, salinity, carbohydrate, phosphorus, and precipitation for the previous 24 and 72 hrs.

Source	d.f.	S.S.	M.S.	F-value	p-value
Model	6	4022134	670356	**3.9148**	**0.0013**
temp			112	0.0007	0.9796
salinity			4022	0.0235	0.8784
carbohydrate			458146	2.6755	0.1044
phosphorus			135988	0.7941	0.3746
24hr precip			2994796	**17.4894**	**<0.0001**
72hr precip			143975	0.8408	0.3609
Error	125	21404399	171235		
C. Total	131	25426533			

F-values, p-values, and preliminary ANOVA are shown with significant values are in bold type.

CFU/cm^2 with a mean of 1760 CFU/cm^2 (Table 9). Change in coliform counts after incubation ranged from -440 to 8994 CFU/cm^2 with a mean of 1830 CFU/cm^2. Post-incubation counts of enterococcus ranged from 1 to 1650 CFU/cm^2 with a mean of 70 CFU/cm^2. Differences between control and final counts of enterococcus ranged from -5 to 1640 CFU/cm^2 with a mean of 84. Initial sediment P concentrations ranged from 19.6 (E 8/19) to 175 µg P/g sediment (CR 9/6) with a mean of 75.1 µg P/g sediment. Carbohydrate ranged from 290 (BWP 8/29) to 13,100 µg/g sediment (210 9/1) with a mean of 3510 µg C/g sediment.

Experimental trials were grouped together by subtracting initial sediment bacterial counts from final counts for each treatment to find the overall change. The combined data were then analyzed using a Kruskal-Wallis ranked Two-Way ANOVA. Dextrose treatments produced significant effects for both FC ($H = 11.603$, $p < 0.001$, $df = 20$) and FE ($H = 5.590$, $p =$ between 0.025 and 0.01, $df = 20$) (Table 9). Treatments of P produced no significant effects for either coliform or enterococcus.

Data were analyzed for significant effects of treatments during individual trials. Dextrose treatments produced a significant effect on coliform for trials E-8/19., REC-
8/24 and CR-9/6. Significant effects on enterococcus were only seen during trial E-8/19. Significant effects for P were seen at E-7/22 and E-8/19 and were only for enterococcus. There was a significant interaction between P and dextrose treatments for coliform during trial E-7/22. Data on fecal indicator bacteria are often highly variable. Due to the low degrees of freedom of the individual experimental trials it may be worth considering a significance of $\alpha = 0.1$. Under this assumption there were additional significant responses by coliforms to P treatments during trials E-7/22 and 210. Responses by enterococcus to C treatments during trials BWP and REC also became significant.

Table 9. Effects of dextrose (C) and phosphorus (P) treatments and interactions (I) between dextrose and phosphorus treatments in fecal coliform (FC) and fecal enterococcus (FE) in cores for experimental trials.

Site	Date	Initial P µg P (g sed)$^{-1}$	Initial C µg C (g sed)$^{-1}$	Treatment	FC F	FC p	FE F	FE p
E	8/19	20	420	**C**	**29.9**	**0.001**	**11.9**	**0.009**
				P	0.65	0.44	**7.00**	**0.029**
				I	0.89	0.37	0.01	0.92
E	7/22	31	628	C	0.20	0.89	2.38	0.16
				P	3.53	0.09	**5.71**	**0.04**
				I	**57.6**	**0.001**	0.00	1.00
BWP	8/29	47	290	C	1.71	0.23	4.50	0.07
				P	1.05	0.34	0.50	0.45
				I	0.39	0.55	0.50	0.49
210	9/1	74	290	C	2.60	0.15	0.08	0.78
				P	3.36	0.10	0.50	0.49
				I	**8.04**	**0.02**	0.50	0.49
REC	8/24	102	2990	**C**	**7.71**	**0.02**	4.91	0.06
				P	0.22	0.65	0.48	0.51
				I	0.47	0.51	0.78	0.40
CR	9/6	176	3600	**C**	**17.8**	**0.003**	0.08	0.60
				P	0.91	0.37	0.50	0.19
				I	3.06	0.12	0.50	0.19
Combined Data*				**C**	**11.6**	**<0.001**	**5.59**	**<0.025**
				P	0.73	0.40	0.09	0.77
				I	0.73	0.40	1.24	0.28

Parametric data were log transformed and analyzed by Two-Way ANOVA. Non-parametric data were analyzed using Kruskal-Wallis ranked Two-Way ANOVA and are indicated by "*". Significant effects at $\alpha = 0.05$ are in bold type. Significant effects at $\alpha = 0.1$ are in red.

DISCUSSION

Field Data

Throughout this study sediment-bound coliform and enterococcus were repeatedly found at concentrations adequate to be deemed problematic. Sediments containing indicator bacteria may easily be suspended by a number of disturbances. Dredging, changes in changes in stream flow associated with precipitation and/or tidal stage, or physical disturbances may all affect the stability of bacteria within the sediment bed (Grimes, 1980; Brickler et al., 1976; Tunnicliff and Brickler, 1984; Doyle et al., 1992). In the face of a large scale sediment suspension, most of the sites sampled yielded concentrations of bacteria that would have instantly exceeded the NC state standard for coliforms > 20% of times sampled for coliforms and >38% of times sampled for enterococcus (Table 10). Means for coliform at each site exceeded use standards by up to 60% and enterococcus means exceeded standards by as much as nine times the acceptable level for the water column. All bacterial concentrations calculated were expressed as CFU/cm^2. This is a useful unit assessing fecal indicator bacteria considering that if the top layer of stream sediment is suspended into a 100 cm deep water column the resulting volume is then 100ml and can easily be compared to water column measurements. Therefore, assuming the upper 1 cm^3 of sediments containing > 200 CFU were to be mixed into the water column above it by some suspension activity, the overlying waters would then be contaminated beyond the water quality use-standard of 200 CFU/100ml for coliform. Stream depths for sample stations BC-CA, BC-CR, BC-SBU, and BC-NBU were < 100 cm in depth so CFU/cm^2 estimates at those stations are likely to be conservative. According to concurrent studies by the New Hanover County Tidal Creeks Project (Mallin et al., 2003) concentrations of coliforms in Bradley Creek headwaters maintained

Table 10. Means and Theoretical % Non-Compliance for indicator bacteria

Site	**coliform** Mean (CFU/cm^2) %>200 CFU/cm^2	**enterococcus** Mean (CFU/cm^2) %>33 CFU/cm^2
BC-SBU	340 27%	332 63%
BC-CA	32 4%	202 75%
BC-NBU	186 35%	365 71%
BC-CR	257 31%	65 38%
BC-S	125 19%	251 71%
BC-NB	132 23%	257 100%

Means for each site as well as the proportion of times sampled that concentrations of bacteria would have exceeded the NC standard for that indicator bacteria in the event of sediment suspension.

concentrations near or above the use standards for the State of North Carolina (>200 CFU/100ml) with a mean of 807 CFU/100 ml and range of 60 – 6000 CFU/100 ml when sampled during the time period of this study. NCDENR maintains sampling stations on nearby Wrightsville Beach as well as downstream of Bradley Creek in areas in the ocean and estuarine waters surrounding Wrightsville Beach. Several stations were found to be non-compliant with regards to enterococcus in the water column as often as 20% of the times sampled (Table 11). All of these stations are areas with ample tidal flushing and relatively high salinities due to their proximity to the open ocean suggesting that there may be a regular or continuous microbe source. At elevated salinities, coliform and enterococcus are likely to be short lived (Hanes and Fragula, 1967). These values are much less than those measured in nearby Bradley Creek sediments. In the event of sediment suspension existing bacterial concentrations would be increased by the amount of CFU/cm^2 existing in the sediments producing an additive effect and further impairing these economically and recreationally important waters.

Significant correlations between coliform and enterococcus were found at only 2 of 6 sites (site BC-S and site BC-SBU) but when the data from all sites were combined to increase degrees of freedom and statistical power, a correlation between the two bacteria was clear (Table 3). Streptococcus did not correlate with coliform data and was likely influenced by other sources environmental parameters. Collection of data for streptococcus was consequently discontinued. This study has shown that neither coliform nor enterococcus had a meaningful response to changes in sediment P concentrations within sediments in Bradley Creek. Rowland (2002) found a negligible coliform response to sediment P concentrations above 10µg/g of sediment in the field and above 5µg/g of sediment experimentally. Concentrations of sediment ortho-phosphate in Bradley Creek consistently exceeded these by 1-2 orders of magnitude (Table 2) which may

Table 11. Means and % non-compliance of enterococcus in the water column for NCDENR sampling stations near the terminus of the Bradley Creek Watershed.

FIELD DESCRIPTION	LOCATION	CFU/100ml	% non-compliant
BLOCKADE RUNNER	ocean	28	20.8
BANKS CHANNEL - CAROLINA YACHT CLUB	sound	34	20
HANOVER SEASIDE CLUB	ocean	35	13
BASIN	sound	18	16.7
CRYSTAL PIER	ocean	10	0
GREENVILLE SOUND, BANKS CHANNEL	sound	9	0
GREENVILLE SOUND, BANKS CHANNEL - MARKER # 10	sound	9	0
JOHNNY MERCER PIER	ocean	11	0
MASONBORO SOUND, MARKER #135	sound	11	0
MIDDLE SOUND, NIXON'S CHANNEL	sound	9	0
MIDDLE SOUND, S. END OF FIGURE 8	sound	13	3.8
SHELL ISLAND	ocean	9	0
WRIGHTSVILLE SOUND, STOKLEY CUT	sound	10	0

explain the lack of response to varying P concentrations. It is possible that P concentrations within sediments of the Bradley Creek watershed are high enough to have alleviated nutrient limitation from P and shifted to another limiting factor. Residential use of fertilizers on well drained sandy soils has led to an excess P export and accumulation in the drainage basin (Cahoon, 2002). Tidal creeks are exposed to the same dynamic chemistry as estuaries and are capable of acting as a P sink. Particle adsorption, sedimentation, and precipitation due to changes in pH and salinity can strip even low levels of P in the water column and deposit it to the sediment bed below. There it can accumulate to potentially significant concentrations. Mallin et al. (2004) reported that P in the water column has remained relatively low (0.002 – 0.04 ppm) for the same watershed and time period and would not normally present itself as an imperative contributing factor to bacterial growth or persistence (Rowland 2002). However, mean concentrations of P for the same sample stations ranged from 101 – 315 μg/g

(equivalent to ppm by weight) of sediment during the same time period as the New Hanover Tidal Creeks study (Table 12). Measurements of sediment P exceeded concentrations of P in the water column by 4-5 orders of magnitude with the highest averages by site being found at the tidally influenced sites S and W. At concentrations of this magnitude, it is probable that the relevance of P as a limiting factor for growth/persistence is long past. Measurements of P in the water column alone are likely to understate the presence and availability of P as a contributing factor to microbial growth/survival in sediments and possibly the water column as well. Sediment carbohydrate had no significant effect on coliforms or enterococcus for the combined field data and was significant at one site for enterococcus only (Table 5, Fig. 5). No significant effect was observed at any other individual site for either bacteria. Total carbohydrate values were consistent with those found in a previous study of nearby estuaries in New Hanover County

Table 12. Comparisons of phosphorus concentrations in the water column and within the sediments at sampling stations within Bradley Creek.

site	ug P/ g sed.	2002-2003 water column (mg P/L)	2003-2004 water column (mg P/L)
BC-SBU	137	0.009	0.011
BC-CA	145	0.021	0.019
BC-NBU	119	0.004	0.005
BC-CR	101	0.004	0.005
BC-NB	261	0.009	0.011

(Cahoon, 1988). Cahoon (1988) also states that in *Spartina* salt marshes high levels of total carbohydrate are likely due to decaying cellulose (insoluble carbohydrate) and POC and therefore is a poor source of biologically available C. While the total carbohydrate assay may suggest sufficiently abundant levels for bacterial production, the assay does not distinguish between available (soluble carbohydrate) and unavailable C. Sites BC-S and BC-NB were situated within a tidal salt marsh dominated by *Spartina alterniflora* and *Juncus romerianus*. Site BC-CA is a detention pond dominated by *Typha spp*. and various otheraquatic plant species. Elevated levels of sediment total carbohydrate at these sites were probably due to decaying plant matter and were not relevant to bacterial production. C limitation at these sites, if any, might easily have been masked by the signal from refractory carbohydrate.

It is also worth noting that sediment-bound bacteria did not always follow well established patterns for indicator bacteria in the water column. Although coliforms and enterococcus concentrations have been tied to salinity (Hanes and Fragula, 1967; Evison, 1988; and Solic and Krstulovic, 1992) and temperature in the water column (Struck, 1988; Howell, et al., 1996; Solic and Drstulovic, 1992), sediment-bound bacteria seemed to have an attenuated response to these factors (Figs. 6, 7). While coliforms showed a significant response to salinity, enterococcus did not. This may have been due to much lower degrees of freedom for enterococcus (Table 6). A significant effect due to temperature (Table 7) was not seen for either bacteria when the combined field data were analyzed. Significant temperature effects were seen at the uppermost sites in within the watershed. Water depths were typically shallower and subject to more extreme water temperature changes due to changes in air temperature. When analyzed by site a significant effect was seen at 3 of 6 sites for coliforms and only 1 of 6 sites for

enterococcus. This does not conform well with patterns established by previous studies as a response to temperature has been well documented (Davies, 1995; Edmonds, 1976; Van Donsel, 1967). Conditions may be favorable enough within the sediments to overcome the hindrance of growth and persistence associated with cooler temperatures.

Experimental Nutrient Manipulation

Experimental manipulations of P and carbohydrate showed that C is a potential limiting factor for the growth of water quality indicator organisms. When net change in bacteria was taken into account for all trials combined carbohydrate addition resulted in highly significant responses by both coliform and enterococcus ($H = 11.6$, p = between 0.005 and 0.01; and $H = 5.59$, $0.01 < p < 0.05$, respectively) (Table 9). Carbohydrate concentrations were augmented with additions of dextrose. Dextrose provides a highly bioavailable source of soluble organic C. The discrepancy in results from field data and experimental data concerning carbohydrate were probably related to differences in C source. While the total carbohydrate measured in the field probably reflected biologically unavailable cellulose from decaying plant matter, bacteria colonizing the experimentally manipulated cores were provided with a 1000 ppm solution of readily available soluble organic carbohydrate. When experimental data were examined by site, dextrose addition resulted in significantly different bacterial concentrations ($\alpha = 0.05$) for 3 out of 6 trials for F. coliform and only 1 of 6 trials for F. enterococcus. When significance was considered at $\alpha = 0.1$ dextrose addition became significant for 2 during two more trials (BWP and REC) for enterococcus. The significant responses did not correspond to sites with low initial nutrient values with respect to the other experimental trials.

P was not a significant factor in limiting microbial growth at the concentrations measured and is also probably due to high initial P. Bacteria generally have lower C:P and C:N ratios than

other eutrophic organisms and therefore have a greater demand for inorganic nutrient availability (Goldman et al. 1987; Caron 1991). Even in areas where P and nitrogen concentrations are relatively high nutrient limitation may still shift to biologically available DOC (Goldman et al. 1987; Caron 1991; Carlsson and Caron, 2001) or some other limiting factor.

Phosphorus treatments produced a significant response by enterococcus for 2 of 6 trials. The two trials eliciting a bacterial response from P both also had the lowest measured initial P concentrations and greater significance for these two trials corresponded with decreasing initial P values (Table 9). This could be an indicator of the range in which P becomes limiting for sediment-bound enterococcus. There were no significant responses by F. coliform to P treatments at $\alpha = 0.05$. By increasing α to 0.1 coliforms show a significant response during trials E-7/22 and 210. Both of these trials had initial P concentrations < 75 μgP/g sediment and were at the lower end of the range of sediment phosphorus found during this study. Phosphorus should be ruled out as a limiting nutrient for coliforms under these circumstances. As with the field data all initial sediment P concentrations were much higher than growth limiting thresholds established by previous research. There is the possibility that enterococcus is more P limited than coliform and is worth future investigation. There was some significant interaction between treatments for 2 of the 6 trials for coliform and no interaction between treatments for enterococcus, however, neither of these could be explained simply by initial nutrient concentrations.

Discrepancies between the responses of indicator bacteria to carbohydrates in the field data and experimental data support the conclusion that bio-available C sources in Bradley Creek may be masked by sources of detrital plant material. Results from experimental trials in this study have shown that there is potential for limitation by soluble organic C and is supported

elsewhere (Soendergaard et al. 2003). Storm water runoff has been identified as a significant source of bio-available C (Buffam et al., 2001; Seitzinger et al., 2003) and increases in relative concentrations of BDOC being correlated with increased urbanization (Goonetilleke et al., 2003). Future research might benefit from examining the separate fractions of soluble and insoluble carbohydrate.

The existence of large concentrations of fecal indicator microbes within tidal watershed sediments is unquestionable. However, environmental factors controlling persistence and growth are poorly understood. Currently there are no regulations in place to manage or monitor bioavailable carbon, P, or indicator bacteria in aquatic and estuarine sediments. The findings of this study suggest that this leaves a significant population of potentially harmful bacteria and mechanisms regulating its persistence to go largely unchecked. Although sediment-bound bacteria in Bradley Creek do not seem to be limited by P and total carbohydrate, these nutrients play an important role in bacterial persistence. Recent research has suggested that management of microbial water quality may be aided by the management of runoff and nutrient inputs (Sundareshwar et al., 2003: Mallin et al., 2000; Buffam et al., 2001; Seitzinger et al., 2003). Increases in storm water runoff from development and residential areas combined with already elevated sediment associated nutrient concentrations may hinder our ability to easily mitigate microbial pollution by simply managing these individual nutrients.

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