SPECIAL STUDY FINAL REPORT

Influence of Sample Depth on Fecal Indicator Results At Bathing Beaches in North Florida

January 2012

Biology Section
Bureau of Laboratories
Division of Environmental Assessment and Restoration

Quality Manual No. 870346G
NELAC Certification No. E31780

Executive Summary

Under the provisions of the Clean Water Act, the Environmental Protection Agency (EPA) has delegated authority to the State of Florida's Department of Environmental Protection (FDEP) to monitor and remediate waters of the state to the highest attainable quality. As part of this process, the Environmental Regulation Commission (ERC) adopted the Department's Impaired Waters Rule (IWR, Chapter 62-303, F.A.C.) in 2002 wherein a process to evaluate waters of the state was defined. Waters not meeting water quality criteria for its designated use are required to be evaluated for remedial actions through the establishment of a Total Maximum Daily Load (TMDL) for the segment or water body.

Water body segments (WBIDs) around the state include waters that include areas designated by the Florida Department of Health as "Bathing Places" (designated bathing beaches). Although bacteriological criteria different from the FDEP's regulatory criteria are used to assess beach water quality, these data are indirectly used in the assessment of 'Impaired Waters'.

In 2009, the TMDL Section of the FDEP Bureau of Water Resources raised concerns that WBIDs designated at a designated bathing beach were identified as impaired based on FDOH's designation of the number of days under beach advisory. Data underlying the beach advisory designation are frequently a single point used to characterize the entire beach collected on a weekly basis. The concern that data from only one sampling point, even over time, inadequately addresses impairment.

In response to this concern, the FDEP Biology Section, in conjunction with the FDOH's Healthy Beaches Program, initiated this study to answer the following question.

> Do the data from the single sampling location for a beach adequately characterize the water quality at that beach considering both spatial coverage and depth of sampling (ankle, knee, and waist)?

Based on a two-year study of two beaches in North Florida, Carrabelle Beach (Franklin County) and Mashes Sands Beach (Wakulla County), the following conclusions were drawn:

1. Applying the FDOH beach classification criteria categories of Good, Moderate and Poor, the comparison between samples collected concurrently by FDOH and FDEP data at the FDOH designated sampling location for the two years combined showed a high degree of agreement (96%) for both Carrabelle Beach and Mashes Sands Beach. Data for enterococci indicated 65% agreement for Carrabelle Beach and 59% agreement for Mashes Sands Beach. Discrepancies occurred in which the two agencies differed sufficiently to affect the determination of impairment (i.e., Good reclassified to Poor). Spatial data along transects illustrated that depth of sampling (ankle, knee, waist) was statistically related to the log-transformed bacteriological data. In general, the progression of decreasing bacterial counts from the shallow to deeper water suggests that the source of bacteria is land based and that dilution plays an important role in the determination of the bacterial density reported. Sources of bacteria in the shallow waters were not determined during this study. Transect data shows that the characterization of the beaches would potentially shift the FDOH classification of the beach depending on

the depth of sampling and thereby influence the determination of beach water quality and the designation of impairment for the WBIDs in which the beach resides. Human health effects observed during epidemiological studies performed by EPA at designated bathing beaches demonstrated the strongest correlation with fecal indicator concentrations at waist depth (Personnel communication, 2010, John Ravenscroft, EPA).

2. Spatial data for station locations indicated that the beach waters varied depending on the influences to that station. Lateral influences through environmental sources (e.g., river water, bay water, or marsh) were suggested as sources for elevated bacteria. In the absence of more sophisticated source tracking tools, human-associated contributions could not be determined.
3. The differences noted in the bacterial activity profiles between Years 1 and 2 did not suggest a seasonal profile to these data. Additional data, preferably multi-year, are needed to investigate the presence of a seasonal pattern. Although different contributors to the observed elevated densities were proposed, additional work is needed to identify and differentiate among potential influences and resulting sources of bacteria impacting the beach water quality at Carrabelle Beach and Mashes Sands Beach.
4. Based on the data evaluated in this study, the proposed shifting of the FDOH beach monitoring location from knee depth to waist depth will potentially result in fewer waters being deemed Poor and listed as Impaired. This study, however, highlights a potential concern of elevated bacterial densities in shallow waters will remain undetected by routine monitoring at the greater water depths. Additional study is needed to determine both the source and public health impacts of these elevated densities in these shallow areas

Acknowledgements

The FDEP wishes to thank FDOH for their technical assistance in providing valuable discussions on the drafting of the study design and the historical data for Carrabelle Beach and Mashes Sands Beach from the Healthy Beaches Program/ FDOH Bureau of Water Programs:

Patti Anderson, Bureau Chief

Bob Vincent, Environmental Administrator

David Polk, Environmental Specialist I

(Mary)Theresa Howard, Franklin County Health Dept., conducted the FDOH sampling throughout this study.

FDEP acknowledges the following FDEP Biology Staff who conducted sampling and analyses for this study: Chelsea French, Jacqueline Savage, Daisys Tamayo, Leslee Williams, and Jonathan Woods. Additional assistance on IWR data was provided by Barbara Donner, FDEP Watershed Assessment Section.

This report was written by Leslee Williams, Ph.D., reviewed and edited by David Whiting, Biology Program Administrator, Elizabeth Miller, Environmental Administrator, Lori Wolfe, Environmental Administrator, and Daisys Tamayo, Environmental Supervisor.

Table of Contents

List of Tables **Page**

List of Figures **Page**

I. Introduction/Background

Under the provisions of the Clean Water Act, EPA has delegated authority to the State of Florida's Department of Environmental Protection (FDEP) to monitor and remediate waters of the state to the highest attainable quality. As part of this process, the Environmental Regulation Commission (ERC) adopted the Department's Impaired Waters Rule (IWR, Chapter 62-303, F.A.C.) in 2002 wherein a process to evaluate waters of the state was defined. Waters not meeting water quality criteria for their designated use are required to be evaluated for remedial actions through the establishment of a Total Maximum Daily Load (TMDL) for the segment or water body.

Over 200 freshwater bathing beaches are regulated under Florida Department of Health (FDOH) Rule 64E-9.013, F.A.C., where permitting and monitoring criteria are defined. Over 300 marine beaches are tested weekly for two pollution indicator bacteria under the federal Clean Water Act Healthy Beaches grant provided by EPA to the FDOH and implemented by the County Health Departments. Bacterial standards and procedures to protect the beach-going public by communicating beach water quality advisories are defined in this national public health program. Both the water quality monitoring and public health advisory programs for fresh and marine waters are authorized in Chapter 514, FS.

FDEP's IWR (Rule 62-303.360, F.A.C.) defines how data for FDOH's marine Bathing Places (beaches) are to be assessed. The rule requires an assessment of the number of days (within a variety of time frames defined) where closures, advisories, or warnings were set for the beach based on elevated bacterial counts. Raw bacteriological data are not used because FDEP criteria (which use fecal coliform as the water quality criterion) and FDOH criteria (which use both fecal coliform and enterococci as beach criteria) are different. For IWR and TMDL purposes, exceeding any one of the FDOH time-frame criteria (as defined in the FDEP IWR) triggers a potential listing to the Planning List of the entire water-body as impaired, independent of any additional data for the water-body.

Once a water segment is placed on the Planning List, the FDEP must determine whether or not elevated fecal indicators are from an anthropogenic **source**. Waters that have elevated fecal indicators as a result of human-induced pollution are placed on the Verified List, and a Total Maximum Daily Load (TMDL) is implemented to reduce the human sources of fecal indicators in order to restore the bacteriological quality of the water segment. Should it be determined by FDEP that the entire source of elevated fecal indicators is of natural origin, the water segment is lowered in the priority list requiring a TMDL, since the TMDL program is not intended to abate natural wildlife or environmental conditions. This listing process does not affect the FDOH evaluation and advisory process.

A number of waters currently being evaluated as "impaired" include waters listed solely based on the data for FDOH designated Bathing Places (marine beaches). As noted above, FDEP is required to assess whether or not the source of the elevated bacteria is anthropogenic. A number of recent studies (Solo-Gabriel, et.al, 2000; Whitman and Nevers, 2003) have suggested that the elevated densities at bathing beaches may arise from sources other than water-borne fecal pollution, e.g., the shallow beach waters, sands on the beach or possibly the bathers themselves. While it is understandable that FDOH monitors waters in the proximity of the bathers, these in-shore (knee depth) samples may frequently bias the interpretation of impairment of the entire water segment. The two programs (FDEP and FDOH) are not mutually exclusive though, as one influences the other, but the FDOH is only concerned with the public health aspects of the water in the near shore swimmable area usually within 100 feet of shoreline while FDEP must consider public health as well as the natural conditions of the water body as a whole.

Ambient water quality sampling by the FDEP has historically focused on representative samples of the water-body and therefore would be sampled more at a mid-points of the water-body, specifically at greater distances (> 100 feet from shore) and at greater depths (greater than knee deep) than beach samples.

Over the past ten years a number of studies have documented the phenomena of fecal indicator organism persistence and re-growth in environmental media such as soils, sediments, beach sands, pore waters, and surface waters (Hardina et.al., 1991, Cuvelier et.al., 2004, Anderson et.al. 2005; Townsend et.al., 2006, Bonilla et.al., 2007, Hartz et.al., Ishii and Sadowsky, 2008). These phenomena have significant implications for the implementation of fecal indicator-based recreational water quality criteria. The most serious implication being that the relationships between fecal indicator densities to pathogen densities to illness rates no longer meet the assumptions used by the United States Environmental Protection Agency (USEPA) in the development of the 1986 Recreational Water Quality Criteria (Colford et al. 2005), specifically that the fecal indicator organisms are human-associated and are directly linked with pathogen-mediated illnesses. Unlike fecal indicator bacteria, the pathogens responsible for the majority of gastrointestinal illness in human bathers (e.g., viruses, giardia, and cryposporidium) cannot reproduce outside of the human host. This means that fecal indicators from sources other than the gut of warm blooded animals do not share the same relationship to pathogen density or illness rate. That being the case, some waters with naturally elevated levels of fecal indicator bacteria do not pose an increased risk of gastrointestinal illness to bathers and therefore should not be listed as impaired. Unfortunately for risk management decisions, the corollary is true as well; the absence of fecal indicator bacteria does not assure the absence of a human pathogen in sufficient quantity to cause disease. Additionally, the recurring problem of zoonotic transmission of infectious diseases to humans must be considered if public health is the preeminent concern. If only identification of correctable anthropogenic sources is the paramount issue, then other test methods for the offshore water body should be pursued.

Source identification, specifically for human-associated/anthropogenic-induced pollution, most recently referred to as Microbial Source Tracking (MST) is a relatively new but emerging field of study. Few standardized methods are currently available for determining the sources of fecal indicators and none have been “approved” by the US EPA or certified by the national microbial method sanctioning bodies. However, with limits on confidence, a variety of microbiological and chemical analyses are currently being developed that can be used to assist in source identification. In the absence of currently standardized methods for source identification and the need to move toward remediation of impaired waters, the state would benefit from using ancillary techniques to get a better idea of whether the suspected pollution results from an anthropogenic source.

EPA strongly suggests that the use of the Sanitary Survey is one tool that will greatly enhance the characterization of a bacteriological profile of a water body. The USEPA recently published the Great Lakes Beach Sanitary Survey User Manual (EPA-823-B-06-001), which may be helpful in producing a more Florida-specific beach sanitary survey tool. The goal of a beach sanitary survey is to identify potential sources of fecal indicators/pathogens and better understand the conditions that increase the loading of those indicators/pathogens into the bathing beach waters. This in turn will provide critical information on whether the elevated bacterial densities are from anthropogenic sources.

Once these data are available, tools are currently being developed or available to predict when elevated densities may occur and thereby may provide FDOH with better predictive capabilities in its mission to protect the public health of beach goers. The USEPA is developing a beach modeling tool known as Virtual Beach. The Virtual Beach software is used to model up to a dozen variables such as those listed in Table 1 from Frick, Ge, and Zepp, 2008.

Table 1: List of Nowcast and Forecast Explanatory Variables Considered in This Study

nowcast variable			
variable	**unit**	**source**	**transformation**
turbidity	NTU	USGS[a]	natural log
water temperature	F	USGS[a]	none
wave height	category 1-4	USGS[a]	square root
24-/48-h rainfall	in	USGS[a]	square root
solar radiation (8am, 9am)	watts/m^2	OARDC[e]	none or square
cloud cover	category 0-5	NWS KCLE[d]	none or square
dewpoint temperature (8am)	F	NWS KCLE[d]	none
wind direction	deg	NWS KCLE[d]	none
wind speed	mph	NWS KCLE[d]	none
along shore wind component	mph	derived from wind direction and speed	none or asymmetrical
cross-shore wind component	mph	derived from wind direction and speed	none or asymmetrical
24 h forecast variable			
air temperature	F	KCLE[b]	none
dewpoint temperature	F	KCLE[b]	none
cloud cover	%	KCLE[b]	none or square
precipitation potential	%	KCLE[b]	square root
wave height	ft	NOAA[c]	square root
wind direction	deg	KCLE[b]	none
wind speed	mph	KCLE[b]	none
along shore wind component	mph	derived from wind direction and speed	none or asymmetric[f]
cross-shore wind component	mph	derived from wind direction and speed	none or asymmetric[f]

[a] www.ohionowcast.info.
[b] National Weather Service, Cleveland-Hopkins International Airport (KCLE): www.erh.noaa.gov/ifps/MapClick.php?FcstType)digital&textField1)41.492627&textField2)-81.827063&site)cle.
[c] www.weather.gov/forecasts/graphical/sectors/eastgrtlakes.php.
[d] National Weather Service, Cleveland-Hopkins International Airport (KCLE): www.erh.noaa.gov/data/obhistory/KCLE.html.
[e] www.oardc.ohio-state.edu/newweather.
[f] An asymmetrically transformed real number has the same sign but an absolute value square-root of its original absolute value.

The Virtual Beach models are useful for predicting beach water quality 24 hours in advance. Variables that are strongly correlated with accurate predictions of actual indicator organism densities may indicate potential source(s) of the indicators/pathogens or the conditions that cause them to occur in higher densities.

However, while the scientific communities move forward in the development of EPA approved tools and methods to assess the anthropogenic sources of fecal pollution, the FDEP is mandated to remediate its waters wherever impaired. This project was initiated to evaluate the concern that waters of the state are being unduly classified as impaired based on data sets which may not be reflective of the water body as a whole.

II. Study Design

In early 2009, FDEP proposed a Beach Study to investigate whether the current FDOH practice of sampling a minimum of once weekly at a beach at a single depth was sufficient to characterize the bacteriological profile of a beach. Of particular interest was whether a beach which demonstrated exceedance of bacterial concentrations (predominately exceedances of enterococci bacteria) was sufficiently characterized by a single sample at a single sampling depth to be deemed impaired.

Working closely with FDOH, FDEP identified six beaches within close proximity of the FDEP Biology Laboratory in Tallahassee. Two beaches (shown in **Table 2**) were chosen from a list of candidate beaches based on the following criteria:

1- Proximity of the beach to FDEP Biology Laboratory in Tallahassee so that samples could be collected and processed within the regulatory required 6 hours holding time.
2- The number of days of Beach Advisories/Closures for the years 2004-2007.
3- Whether the Beach has been placed on the IWR Planning List.
4- Whether research data or additional monitoring data were available for the beach, in addition to the routine FDOH Healthy Beaches Monitoring Data.
5- Review of the available data indicated that enterococci and fecal coliform monitoring data, while not consistently above recommended guidelines or WQ criteria, were sufficiently high to warrant further evaluation of sources of these organisms.

Table 2: Study Sites chosen showing location and the number of days with Advisory or Closure by FDOH based on Bacteriological exceedances for years 2004-2007.

FDOH Sampling Location # Days with Advisory/Closure

ID	COUNTY	BEACH	LAT	LONG	IWR*	2004	2005	2006	2007
110	Franklin	CARRABELLE BEACH	N 29° 64.759718′	W 84° 89.313678′	YES	112	252	180	267
296	Wakulla	MASHES SANDS BEACH	N 29° 97.31033′	W 84° 34.049353′	YES	91	300	92	160

* IWR (Impaired Waters Rule)-FAILURE

After an initial reconnaissance of the selected beaches, the study was conducted as follows:

- Sampling was conducted bi-weekly (every other week).
- Three stations were established at each beach along the shoreline with the central station being concurrent with the FDOH Beach Monitoring location.
- A remote location, outside of the influence of the beach or beach activity, was established at each beach as a control.
- Three different depths of sampling were established for each station location: ankle (A), knee (K), and waist (W) deep for the person collecting the sample. Based on the shallow nature of some of the sampling sites, it was predetermined that if waist depth was beyond any reasonable expectation of swimmer activity a sample would not be taken and this information would be duly noted in the sampling log.
- Environmental parameters were taken at the knee depth for all stations sampled and included: Salinity (S‰), Temperature (° C), Dissolved Oxygen (mg/L DO), and pH.

- Sampling was conducted concurrently with the routine sampling of the Healthy Beaches Monitoring Program on the FDEP designated sampling dates.
- Bacteriological analyses included fecal coliform and enterococci enumeration using EPA approved membrane filter analyses (SM9222D and EPA Method 1600, respectively).
- Evaluation of the bacteriological data included an assessment based on the FDOH Healthy Beaches criteria. (see **Table 3** below

Table 3: Department of Health Bathing Beach Criteria

FDOH Bathing Beach Criteria

***Enterococcus* Results Description**

GOOD	MODERATE	POOR*
0-35 *Enterococcus sp* per 100 ml of marine water 0-35 CFU/100 mL *Enterococcus sp Geometric Mean*	36-104 *Enterococcus sp* per 100 ml of marine water	105 or greater *Enterococcus sp* per 100 ml of marine water 36 and over CFU/100 mL *Enterococcus sp Geometric Mean*

Fecal Coliform Results Description

GOOD	MODERATE	POOR*
0-199 fecal coliform organisms per 100 ml of marine water	200-399 fecal coliform organisms per 100 ml of marine water	400 or greater fecal coliform organisms per 100 ml of marine water

*A Poor rating may result in a resampling event to confirm poor conditions, otherwise a Health Advisory or Warning will be issued immediately. These indicate that contact with the water at this site may pose increased risk of infectious disease, particularly for susceptible individuals. A reading of **NR** means "No Result." This could indicate that no sample was taken at this point because of weather or other factors, or that an analysis result was not obtained from the laboratory.

- (http://esetappsdoh.doh.state.fl.us/irm00beachwater/default.aspx)

III. Sampling Locations

A. Carrabelle Beach (Franklin County, Florida)

Carrabelle Beach is located directly adjacent to US Highway 98 in Franklin County, Florida. The beach is seaward of a series of covered pavilions and restrooms (see **Figure 1**) fronting on US 98. The beach is unobstructed and offers an opportunity to investigate fate and transport issues for historically elevated fecal coliform and enterococci bacteria. This beach's appearance is considered to be representative of established small to moderately sized bathing beaches in Florida. Carrabelle Beach is one of the many dog-friendly beaches in Florida, meaning that tourists and local residents with pets are attracted to this beach. Carrabelle Beach RV Resort is located on US Highway 98 directly across from Carrabelle Beach.

The extensive history of fecal bacteria exceedances and concerns that septic tank leachates were a major pathogen contributor served as the basis for a FDEP State Revolving Fund (SRF) construction grant to the City of Carrabelle to build a centralized wastewater treatment facility. This facility was completed and permitted on 10/26/2006 (Kenneth B Cope AWT (FLA 100641)).

However, the persistence of bacterial exceedances in 2007 and subsequent years have prevented this beach from being removed from the FDEP Impaired Waters List.

Figure 1 illustrates the sampling stations used during Year 1 of the study. These original stations were established to evaluate the designated bathing beach area near the pavilions (Stations CB-1, CB-2 and CB-3). Station CB-4, the control station was chosen to be outside of the influences at the beach and reflects an unimpacted or natural background site. The closest access point not constrained as waterfront personal property was approximately 3 miles north of the beach adjacent to US 98. During the second year of the study additional stations were established to assess the potential influence of fecal bacteria associated with the Carrabelle River on the beach waters via eddies or longitudinal transport westward to the beach area. Therefore for the second year's study additional stations were added (Stations CB-5, CB-6 and CB-7, **Figure 2**) replacing stations CB-1 and CB-3. Station CB-5 was dropped after an initial sampling.

Figure 1: Station locations CB-1, CB-1, CB-3 for the Carrabelle Beach Stations for Year 1 of the Study. Remote Station CB-4 can be seen in Figure 2.

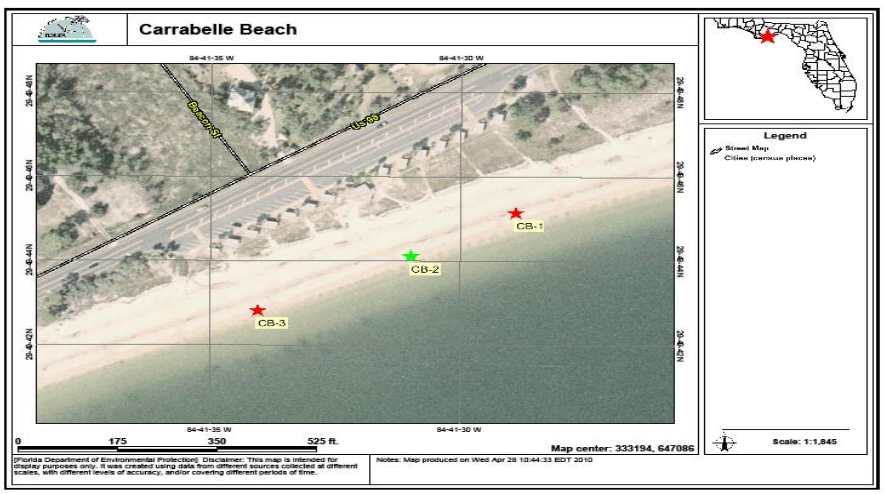
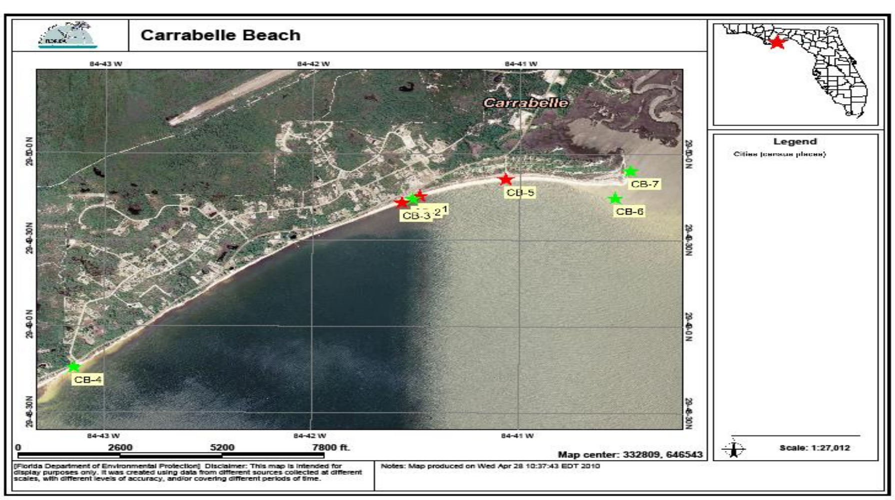


Figure 2: All Station locations CB-1, through CB-7 for the Carrabelle Beach Stations for this study. Year 2 of the Study focused on Stations CB-1, CB-4, CB-6 and CB-7.



B. Mashes Sands Island Beach (Wakulla County, Florida)

Mashes Sands Island is located in Wakulla County, Florida where US Highway 98 intersects the Ochlocknee Bay Bridge on Mashes Sands Road. Mashes Sands Park, south of Mashes Sands Road, is located on one of four finger canals which empty directly to Ochlocknee Bay. Mashes Sands Beach is located at the terminus of Mashes Sands Road approximately ¼ mile east of the Park and fronts on Apalachee Bay. This Beach is surrounded by both extensive marshes and small residential communities along the Mashes Sands Road and is characterized by coarse sand bottom and extremely shallow waters that infrequently rise above knee depth during low and rising tides. While these recreational waters attract fishermen, canoe and kayak enthusiasts, it is only north of the central portion of the beach which offers unobstructed wading or swimming (MS-1). This area is relatively free of beach debris and numerous sand bars create tide pools and extended shallows where young children can safely play. While a number of restrooms, covered grills and a public boat ramp are located at the designated Park, restroom facilities at the beachfront, originally limited to three portables located near the end of the road at the beachfront (landward of MS-2) during Year 1, were replaced by a permanent two-facility restroom structure prior to the Year 2 Study. One permanent structure (formerly a small pavilion) is present near MS-3 but has not been maintained. The numerous sand bars noted at low and rising tides attract numerous aquatic birds. Mashes Sands is a dog-friendly beach. This beach would be better characterized as a recreational waterfront access point similar to many coastal areas in Florida rather than the commonly thought of idea of white sand Florida beaches.

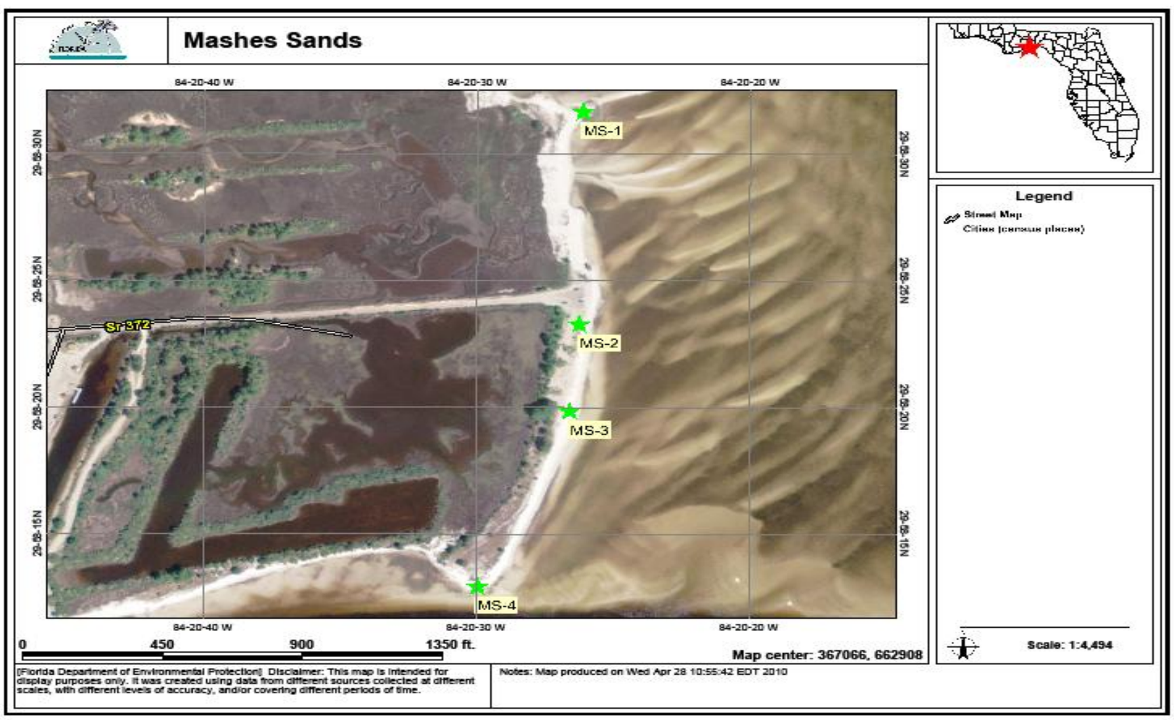
Figure 3 illustrates the sampling locations used for both Years 1 and 2 of the study. The stations encompassed the area of the beach from the northernmost contiguous part of the beach up to the first cut-through from the marsh (MS-1) to the junction of the beach waters with the outflow of the Ochlocknee River (MS-4).

The area between MS-1 and MS-2 was predominately medium grained sand and free of debris. Closer to MS-1 marsh grass was apparent both at the water's edge and appeared to be an extension of the extended marsh at the cut-through. This portion of the beach from the road northward toward the marshes was generally free of debris with only emergent patches of marsh grass and shallow sand bars. The out flow of waters from the marsh appears to be connected with cut-throughs from Apalachee Bay. The extensive biological community associated with marsh areas may be one of the factors in the elevated densities of bacteria noted in this area. Historically, septic tank leachates have been proposed as the source of elevated bacteria at the beach. However, FDOH indicates that centralized wastewater treatment (Wakulla County WWTP, FLA010225) serves the Mashes Sands communities, but it is not know whether all residents connected or the number of residents that still are on septic tanks (Padraic Juarez, Acting Administrator, Wakulla CHU, personal communication). Anecdotal information of undocumented lift station overflows within the local community has been suggested as an alternate cause for the elevated bacteria at this beach. Additionally, the numerous migratory birds attracted to the sand bars as well as the surrounding marshes themselves have been suggested as contributing causes of the fecal indicator bacteria exceedance.

The area between Mashes Sands Road (MS-2) and the tip of the land at the junction of Ochlocknee Bay (MS-4) has a rough, coarse sand bottom scattered with aquatic vegetation debris, roots, and submerged unknowns. MS-3 has tree debris and roots in the shallow waters beyond which a smoother sandy bottom was encountered. Care needed to be taken when collecting the MS-4 sample as the shallows rapidly dropped off to the deep water of Ochlocknee Bay. Reports of sharks in this area also deterred going into the area closest to the bay waters and resulted in samples being taken closer to MS-3 than originally designated.

While the Mashes Sands Beach was not among the locations with the greatest number of Advisories, this beach has been subject to a number of state-funded studies and offered an ability to build on previous studies [Harwood et.al., 2005, Harwood et.al., 2007] and provide additional data for this area.

Figure 3: Mashes Sands Beach sampling locations for Year 1 and 2 identified as MS-1, MS-2, MS-3 and MS-4.



IV. Sampling and analyses

A. Bacteriological sampling

The duration of the FDEP study was intended to focus on the time frame of historical bacterial exceedances in order to investigate potential sources of the elevated bacterial densities. Therefore when routine monitoring demonstrated that sampling events did not show persistence in the exceedances, the study for that year was suspended pending the recurrence of elevated bacteria. The Year 1 study period covered a six month time-frame from March 10 to August 23, 2009. The Year 2 study period covered an eight month time frame from April 21 to November 23, 2010.

Water samples were collected by FDEP Biology Staff bi-weekly for the duration of the sampling periods designated as Year 1 (Y1) and Year 2 (Y2) at each of the designated sampling locations. Sterile Whirl-pak bags were used from 4-21-09 to 5-29-09 to collect two 100 mL samples at each location to ensure against data loss in the event that one Whirl-pak was damaged in transit. Beginning in June 2009 and for the remainder of the two year study, sterile 500 ml polypropylene bottles were used to collect a single sample per sampling station for all bacteriological analyses. Samples were placed in a cooler and maintained on ice at $4^{o}C$ after collection and during transit to the FDEP Microbiology Laboratory in Tallahassee, FL. A field duplicate was collected at each beach at one randomly chosen sample location during each sampling event.

FDEP bacteriological analyses were initiated upon arrival in the laboratory and processed within 2 hours of receipt. All samples met the regulatory requirement of analysis within 6 hours. Membrane filter analyses for both fecal coliform (SM9222D) and enterococci (EPA Method 1600) were conducted on a minimum of three dilutions per sample per method per sampling location. Sample aliquot dilutions routinely run were 50 mL, 10 mL, and 1.0 mL. Method and quality assurance requirements on processing, incubation, enumeration of colony counts, and reporting requirements followed FDEP SOPs MB-01.1 for fecal coliform and MB-01.3 enterococci respectively.

With the exception of the reconnaissance trips for each beach, FDEP sampling occurred concurrently with the FDOH sampling. Sampling was conducted for FDOH by Theresa Howard, Franklin County Health Department, as part of the on-going Healthy Beaches weekly monitoring program, and for FDEP by FDEP Biology Section Staff. FDOH samples were analyzed for both fecal coliform and enterococci bacteria by a NELAC certified contract laboratory in Tallahassee. FDOH protocols for analyses were consistent with those used by the FDEP laboratory. FDEP samples were analyzed in the FDEP Microbiology Central Laboratory in Tallahassee. Both laboratories maintain current NELAC certification for each analytical method.

No attempts to evaluate tidal influences were made in this study. All sampling events were conducted within a 2 hour window (between approximately 10 AM to 12 noon) in order to coordinate sampling with FDOH's routine beach sampling schedule for the FDEP day of sampling.

Raw data and additional photos of the sampling areas for this study are located at http:\\tlhlab5\biology\beach monitoring study. Selected photos are included in the Appendices.

B. Environmental Parameters

Environmental parameters (water temperature (Temp, °C), dissolved oxygen (DO, mg/L), salinity (S‰), and pH) were taken concurrently with bacteriological samples at each of the designated knee (K) depth locations at each beach using YSI Model 650 MDS equipped with a 600 XL Sonde (YSI, Inc., Yellow Springs, Ohio) capable of reading all four environmental parameters. Equipment was calibrated and measurements documented in the Biology Laboratory Equipment QA logbook before field deployment and verified and documented upon return to the lab.

C. Data Evaluation

Raw data for fecal coliform and enterococci were $\log_{10}$ transformed prior to graphing and data evaluation. Illustrations noted as **Figures** display the $\log_{10}$ transformed data to visually investigate data profiles and comparisons between FDEP and FDOH data. For **Figures** in which FDEP data are displayed, the FDOH category thresholds of water quality are shown as horizontal bars within each **Figure**. The green line denotes the upper threshold for the Good category and the red line denotes the upper threshold for the Moderate category. Data above red line threshold indicates waters categorized as Poor. **Figures** which lack these thresholds indicate that all data presented are below the maximum criteria acceptable for Good beach water quality. The threshold values used were fecal coliform are Green threshold (Good) at 2.3= $\log_{10}$ 199 and Red threshold (Moderate) at 2.6= $\log_{10}$ 399; for enterococci are Green threshold (Good) at 1.5 = $\log_{10}$ 35 and Red threshold (Moderate) at 2.0= $\log_{10}$ 104. All data above the Red threshold are categorized as Poor water quality. The Blue line appearing at 0.3= $\log_{10}$ 2 indicates the limit of detection (LOD) for this study. Geometric mean data were not evaluated in this study. Comparisons related to thresholds were used only for comparisons between individual data points. FDOH Beach Advisories are triggered for data identified as Poor. The FDEP IWR evaluation uses both the number of samples deemed Poor and the number of days of Advisories for that sampling location to identify impaired waters.

Spearman Rank Correlation Coefficient (SRCC) analyses were used for all statistical evaluations and comparisons related to spatial depth and transect relationships. Spearman's rho (ρ) is a non-parametric analysis of data based on the rank rather than numerical density of the data and is generally less affected by outliers than other non-parametric analyses. Rho (ρ) can best be described as the linear correlation coefficient computed on the ranks of the data. Where no correlation exists $\rho=0$; and, a perfect correlation would be either -1 or +1 depending on the direction of the correlation. Statistical significance for values determined by Spearman Rank Correlation for this study was set at $p=0.05$ and are shown in the SRCC tables in red.

V. Results and Discussion

A. Microbiological Analyses

1. Carrabelle Beach

Over the course of the two year study 272 samples were processed for both fecal coliform and enterococci bacteria from Carrabelle Beach waters. The data are summarized in Table 4 and in Figures 4 through 7.

Table 4: Descriptive Statistics by year for Carrabelle Beach identifying the number of samples analyzed (N samples), the percent (%) of non-detect results, and the maximum sample result reported.

	N samples	***Fecal Coliform***	***Enterococci***
Year 1	129		
% non-detects		46%	46%
Maximum sample result		70	370
Year 2	143		
% non-detects		31%	34%
Maximum sample result		1200	5800
Total Samples	**272**		

Figures 4 and **5** illustrate fecal coliform data for Years 1 and 2, respectively, and **Figures 6 and 7** illustrate enterococci data for Years 1 and 2 respectively. Data are plotted as are Log_{10} transformed bacterial density by depth of sample (ankle, knee, and waist) by sampling date. The two color-coded horizontal lines for these and all data graphs shown and discussed in this report are the FDOH category of beach water quality as discussed in Table 3 (see Data Evaluation Section).

Figure 4 illustrates that all fecal coliform data for Year 1 were below the threshold for the Good beach water quality category. **Figure 5** illustrates that Year 2 was a more active year with nine data points in the Poor category. Six were from ankle depth and three from knee depth.

Figure 6 illustrates enterococci data were generally within the Moderate category; however, four data points were in the Poor category where two were at ankle depth, one at knee depth and one at waist depth. Year 2 enterococci data shown in **Figure 7** illustrates substantially more enterococci activity resulting in 33 samples categorized as Poor with 17 at ankle depth, 14 at knee depth and 2 at waist depth.

Since the sampling locations were changed between the first and second year and the bacterial profiles for the two years were so dissimilar, conclusions regarding annual or seasonal trends could not be made.

Table 9 shows a further characterization of the data that fell into the Poor water quality classification by date, sampling depth, and station. This analysis was done to determine if any station(s) were more likely to occur in the Poor category. These data further clarify the data displayed in **Figures 4-7**. As previously described, Stations CB-6 and CB-7 were chosen to investigate the potential influence of the Carrabelle River via eddies and lateral flow patterns on elevated bacterial densities at the beach. What is interesting to note is that Poor beach water quality at Stations CB-6 and CB-7 were persistent, supporting the concern that given the right environmental conditions Carrabelle Beach waters could be influenced by the river's outflow.

Transects established at each station were established to investigate whether the depth at which a sample was taken influenced bacterial densities. The three depths evaluated were ankle, knee and waist. As shown in the SRCC analyses in **Tables 5-8**, Data were reviewed by year merging all data by depth of sample independent of station locations. In this analysis, data for all comparisons illustrated a significant correlation between ankle, knee, and waist depth samples. This was not an unexpected result since depth of sample was relatively close in proximity to each other for this beach. In other words, the gradient of the beach was such that swimmers could be fully immersed within 75 feet of the beach water's edge. This presents a generalized picture of the data set which must be taken in context to the data profiles. **Figures 4-7** profiles appear to clarify that, although SRCC shows correlations within the sampling depths, bacterial densities generally decrease as depths of water increases.

The control station (CB-4) was chosen to be outside of the source influences at the beach and reflect an unimpacted or natural background site. The closest access point not constrained as waterfront personal property was approximately 3 miles north of the beach adjacent to US 98. A number of emergent tree trunks and biological debris were noted at this site. This section of US 98 was under road construction for all of Year 1 and part of Year 2 study. Unconsolidated fill used as a road underlayment made access to the sampling site difficult. Birds were frequently noted at this station perched on the emergent tree trunks and sand bars near the two adjacent residential properties. It is difficult to state that Station CB-4 was appropriate as a control station for this beach given differences in waterfront characteristics. Interpreting data for this location is difficult particularly when the bacterial densities exceeded those of the beach and may reflect site-specific environmental conditions. CB-4 was frequently noted as exceeding the Poor category for enterococci in Year 2 (**Table 9**) in contrast to the beach samples. Additional work is needed to clarify the sources and interpretation of the data observed for this station.

During the Year 2 sampling events, Stations CB-6 and CB-7 were consistently shallow in nature yielding only ankle and knee depth data and while sand bars in the area appeared to restrict major flow or influence to the beach data, these stations show elevated densites for both fecal coliform and enterococci data. A review of the exceedances (**Table 9**) for Year 2 data indicates fecal coliform densities resulted in a water category of Poor on dates 6.22.10 (A) and 8.16.10 (A and K) for Station CB-6 and 4.27.10 (A and K) for CB-7. The enterococci profile shows that CB-6 had a Poor classification eight of the seventeen Year 2 sampling dates (47%) and that CB-7 was classified as Poor in seven of the seventeen sampling dates (41%) for Ankle, Knee or both depths. Since all of the Carrabelle Beach samples are collected within a one hour window, the elevated densities at CB-6 and CB-7 did not appear to influence the densities at CB-2 for the comparable sampling dates. The only way to determine a specific influence would have been to appropriately delay the sampling at CB-2 to account for the transit time of the waters of CB-6 and CB-7 to specifically reach CB-2. Although not corroborated by a delayed sampling scenario, these data, particularly the enterococci data, suggest that waters at CB-6 and CB-7 may, under certain flow conditions, affect the beach bacterial densities. As a result of the very shallow nature of these stations, the source of the bacteria, however, is questionable. Whether the densities identified at CB-6 and CB-7 were a result of sand-associated bacterial populations or ambient river waters remains undetermined. Additional work is needed to investigate and identify potential sources and the conditions under which the observed elevated densities at CB-6 and CB-7 influence Carrabelle Beach waters.

SRCC analyses were additionally done to investigate the relationship between individual stations. Results from these analyses are shown in **Figures 10-13**. Year 1 fecal coliform data show that Station CB-1 was significantly correlated with Station CB-2. For Year 1 enterococci data Station CB-2, CB-4 and CB-6 were mutually correlated. Year 2 fecal coliform CB-1 correlated with CB-3. Year 2 enterococci data show correlations mutual correlations for CB-2, CB-6, and CB-7. One important observation is that the speculation that waters from the river outflow as denoted by CB-6 and CB-7 may influence the beach waters were supported by this Year 2 evaluation.

CB-4 was not correlated to any other station during the 2 years of study, further suggesting that the site-specific characteristics of this station were different than the Carrabelle Beach stations.

Figure 4: Fecal coliform data for Carrabelle Beach Year 1 for Stations CB-1, CB-2, CB-3 and CB-4 by depth by sampling date. Note: All data fall below the threshold for Good beach water quality

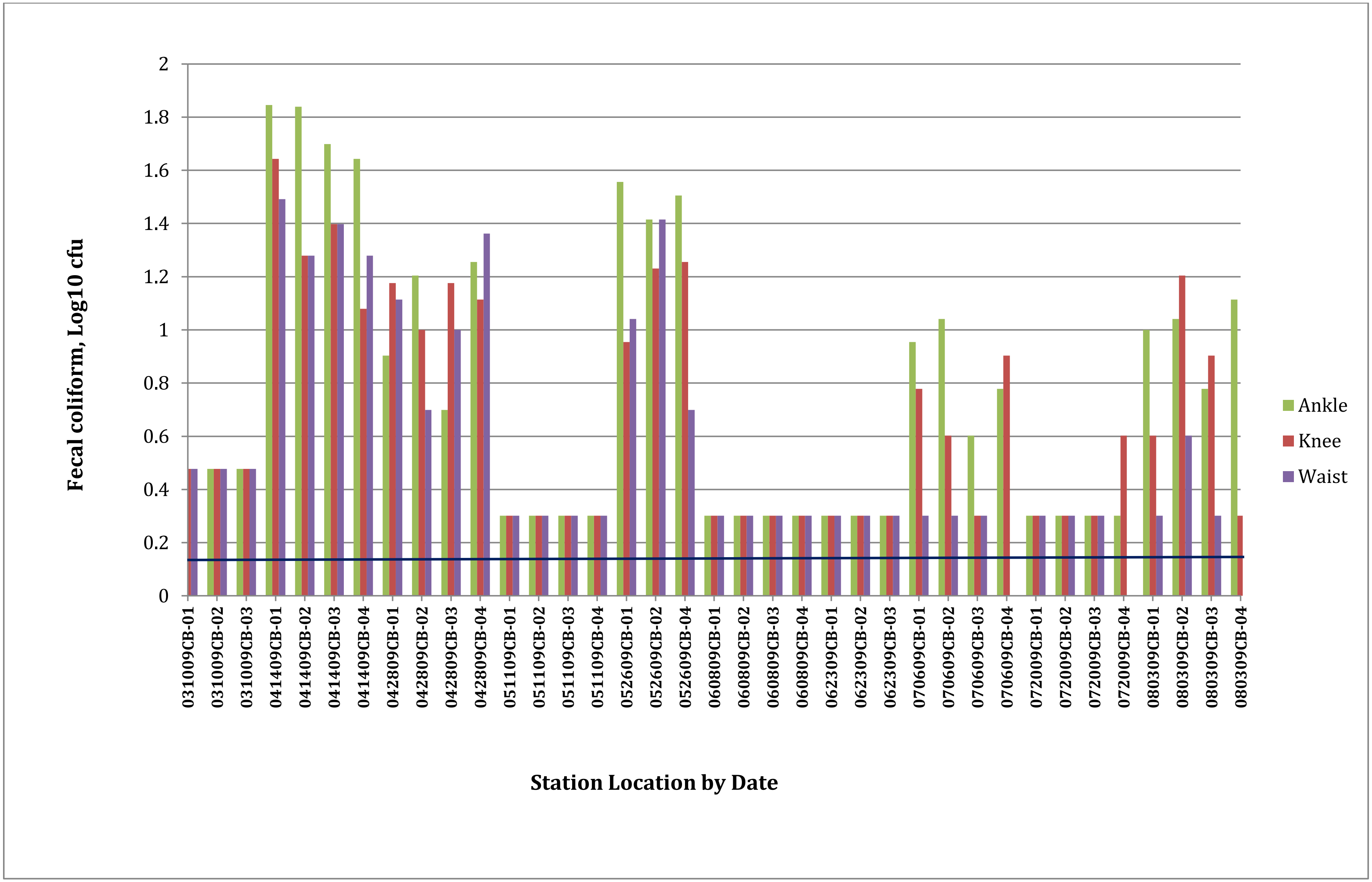


Figure 5: Fecal coliform data for Carrabelle Beach Year 2 for Stations CB-2, CB-4, CB-6 and CB-7 by depth by sampling date.

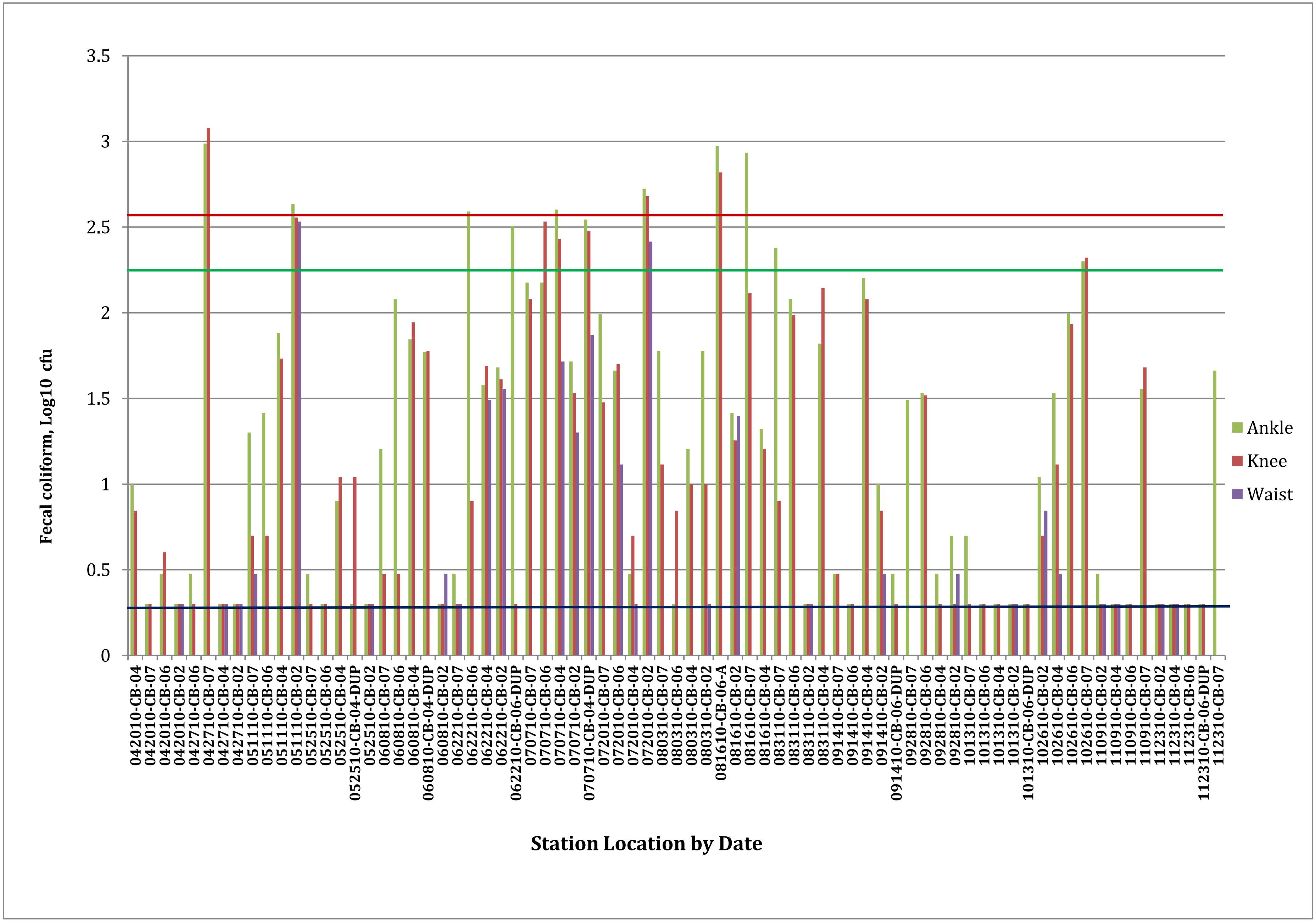


Figure 6: Enterococci data for Carrabelle Beach Year1 for Stations CB-1, CB-2, CB-3 and CB-4 by depth by sampling date.

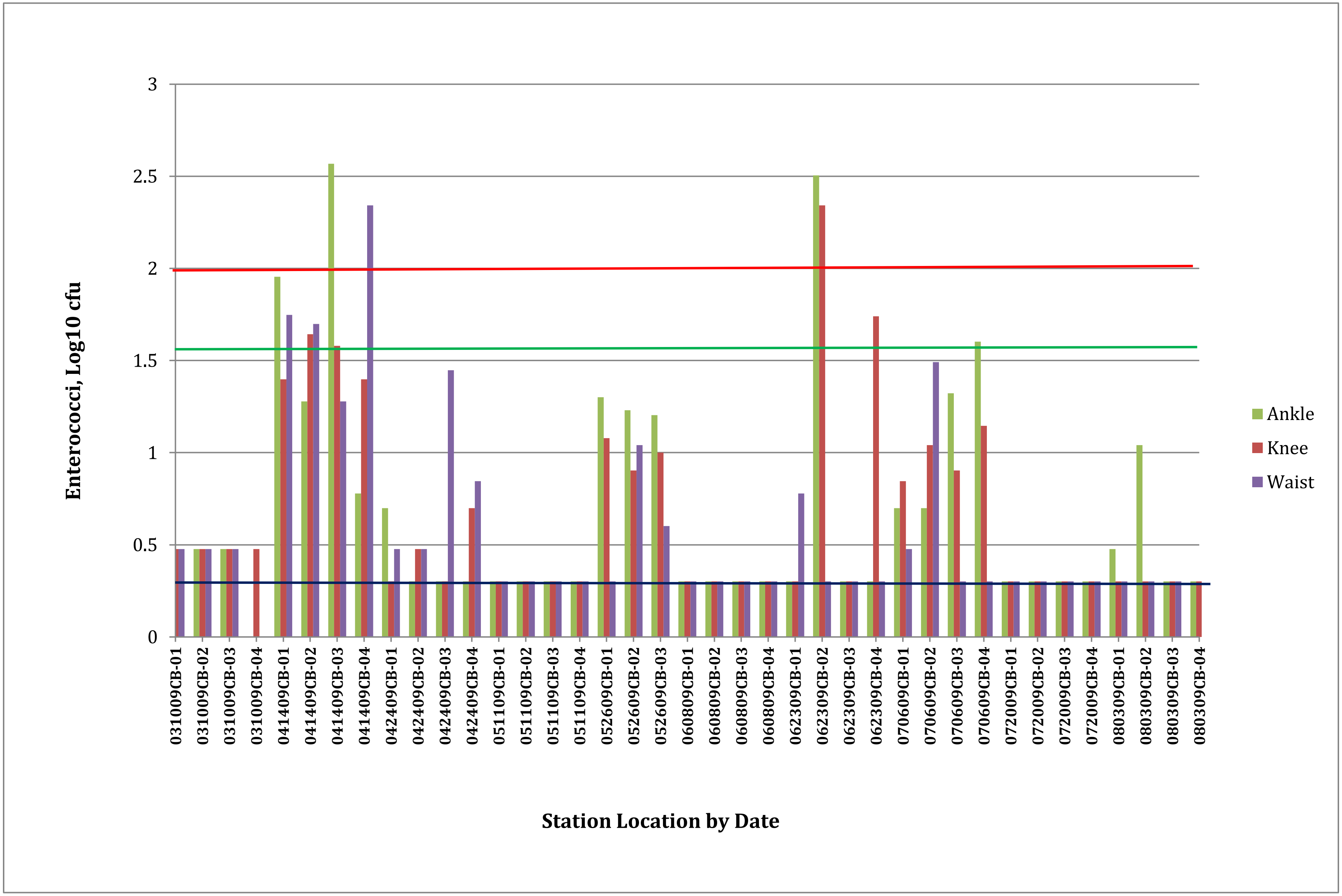
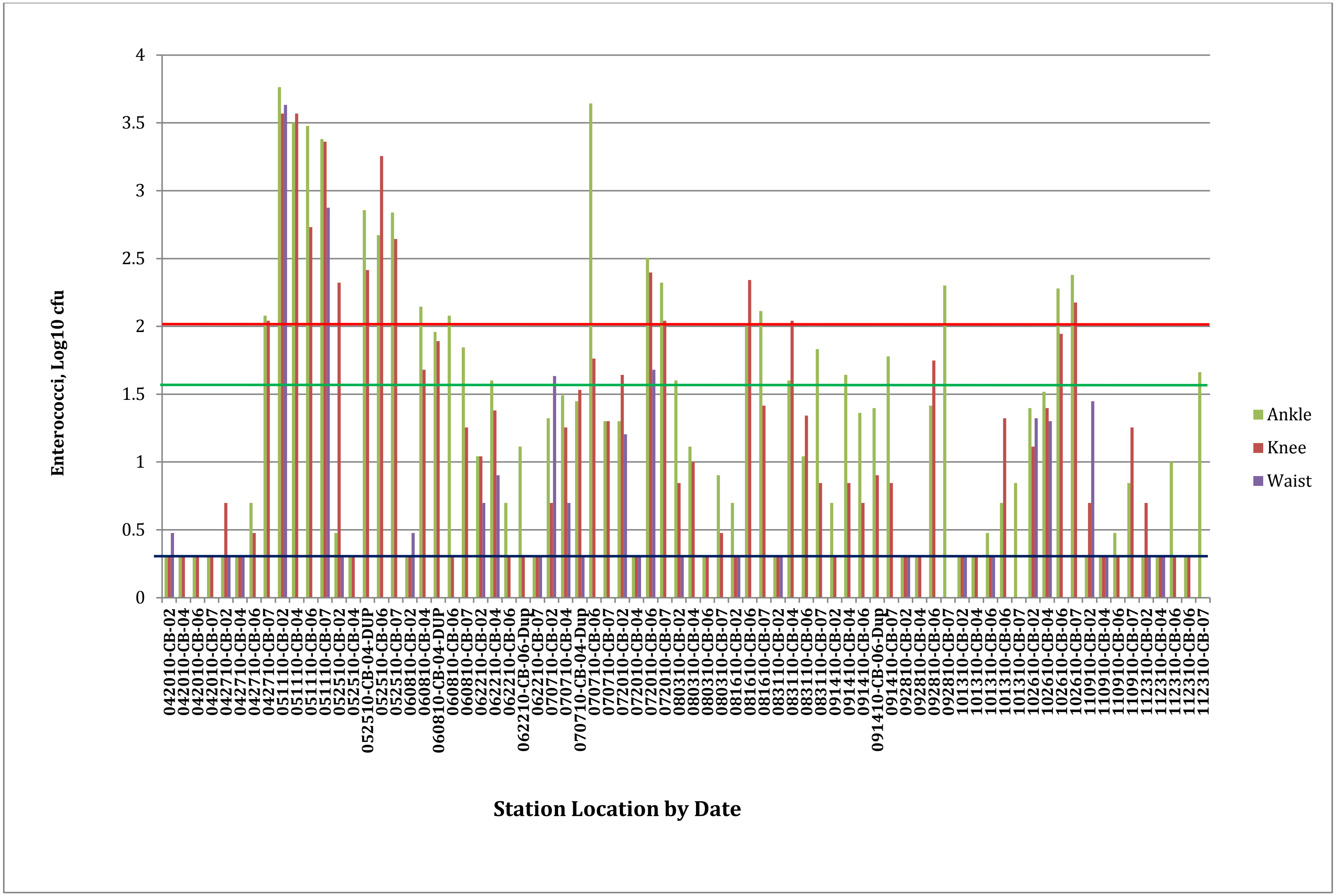


Figure 7: Enterococci data for Carrabelle Beach Year2 for Stations CB-2, CB-4, CB-6 and CB-7 by depth by sampling date.



Tables related to the Spearman Rank Correlation Coefficient (SRCC) analyses for Carrabelle Beach Data

Table 5: Spearman Rank Correlations comparing Depth locations and Log_{10} fecal coliform densities at Carrabelle Beach Year 1 (N = 129)

Variable	Spearman Rank Correlations MD pairwise deleted Marked correlations are significant at p< .05000		
	Ankle	Knee	Waist
Ankle	1.000000		
Knee	0.868368	1.000000	
Waist	0.820867	0.875985	1.000000

Table 6: Spearman Rank Correlations comparing Depth locations and Log_{10} fecal coliform densities at Carrabelle Beach Year 2 (N=143)

Variable	Spearman Rank Correlations MD pairwise deleted Marked correlations are significant at p< .05000		
	Ankle	Knee	Waist
Ankle	1.000000		
Knee	0.835013	1.000000	
Waist	0.833985	0.858836	1.000000

Table 7: Spearman Rank Correlations comparing Depth locations and Log_{10} enterococci densities at Carrabelle Beach Year 1 (N=129)

Variable	Spearman Rank Correlations MD pairwise deleted Marked correlations are significant at p< .05000		
	Ankle	Knee	Waist
Ankle	1.000000		
Knee	0.795981	1.000000	
Waist	0.473028	0.593440	1.000000

Table 8: Spearman Rank Correlations comparing Depth locations and Log_{10} enterococci densities at Carrabelle Beach Year 2 (N=143)

Variable	Spearman Rank Correlations MD pairwise deleted Marked correlations are significant at p< .05000		
	Ankle	Knee	Waist
Ankle	1.000000		
Knee	0.853607	1.000000	
Waist	0.662955	0.661314	1.000000

Table 9: Dates of the exceedance of bacterial densities for Carrabelle Beach above the Poor category (log_{10} FC > 2.6 and Log_{10} enterococci > 2.0) by study year, organism, depth of sample and station location.

Location	Year	Organism	Date	Depth	Station	Comment
Carrabelle Beach	1	FC				No data above the criterion
	2	FC	4.27.10	A,K	CB-7	
			5.11.10	A	CB-2	
			6.22.10	A	CB-6	
			7.07.10	A	CB-4	
			7.20.10	A,K	CB-2	
			8.16010	A,K	CB-6	
			8.16.10	A	CB-4	
Carrabelle Beach	1	ENTERO	4.14.09	A	CB-3	
			4.14.09	W	CB-4	
			6.23.09	A,K	CB-2	
	2	ENTERO	4.27.10	A,K	CB-7	
			5.11.10	ALL	ALL	
			5.25.10	W	CB-2	
			5.25.10	A,K	CB-4	DUP
			5.25.10	A,K	CB-6	
			5.25.10	A,K	CB-7	
			6.08.10	A	CB-4	
			6.08.10	A	CB-6	
			7.07.10	A	CB-6	
			7.20.10	A,K	CB-6	
			7.20.10	A,K	CB-7	
			8.16.10	K	CB-6	
			8.16.10	A	CB-7	
			8.31.10	K	CB-4	
			9.28.10	A	CB-7	
			10.26.10	A	CB-6	
			10.26.10	K	CB-7	

Tables related to the Spearman Rank Correlation Coefficient (SRCC) analyses for Carrabelle Beach by Year, Station location, Log_{10} bacterial density

Table 10: Spearman Rank Correlations comparing station locations and Log_{10} fecal coliform densities at Carrabelle Beach Year 1 (N=129)

Variable	Spearman Rank Correlations MD pairwise deleted Marked correlations are significant at p< .05000			
	CB-1	CB-2	CB-3	CB-4
CB-1	1.000000			
CB-2	0.941828	1.000000		
CB-3	0.284771	0.228720	1.000000	
CB-4	0.357801	0.315000	0.314181	1.000000

Table 11: Spearman Rank Correlations comparing station locations and Log_{10} fecal coliform densities at Carrabelle Beach Year 2 (N = 143)

Variable	Spearman Rank Correlations MD pairwise deleted Marked correlations are significant at p< .05000			
	CB-2	CB-4	CB-6	CB-7
CB-2	1.000000			
CB-4	0.2875409	1.000000		
CB-6	0.325019	0.175943	1.000000	
CB-7	0.162838	0.276330	0.241610	1.000000

Table 12: Spearman Rank Correlations comparing station locations and Log_{10} enterococci densities at Carrabelle Beach Year 1 (N = 129)

Variable	Spearman Rank Correlations MD pairwise deleted Marked correlations are significant at p< .05000			
	CB-1	CB-2	CB-3	CB-4
CB-1	1.000000			
CB-2	0.572223	1.000000		
CB-3	0.538992	0.470275	1.000000	
CB-4	0.222473	0.236582	0.344748	1.000000

Table 13: Spearman Rank Correlations comparing station locations and Log_{10} enterococci densities at Carrabelle Beach Year 2 (N=143)

Variable	Spearman Rank Correlations MD pairwise deleted Marked correlations are significant at p< .05000			
	CB-2	CB-4	CB-6	CB-7
CB-2	1.000000			
CB-4	0.155947	1.000000		
CB-6	0.390975	0.110827	1.000000	
CB-7	0.394005	0.013185	0.654202	1.000000

2. Mashes Sands Beach

Over the course of the two year study 241 samples were processed for both fecal coliform and enterococci bacteria from Mashes Sands Beach

Table 14: Descriptive Statistics by year for Mashes Sands Beach identifying the number of samples analyzed (N samples), the percent (%) of non-detect results, and the maximum sample result reported.

	N samples	***Fecal Coliform***	***Enterococci***
Year 1	98		
% non-detects		14%	19%
Maximum sample result		800	440
Year 2	143		
% non-detects		20%	38%
Maximum sample result		2600	22,000
Total Samples	**241**		

The shallow nature of this area resulted in a lack of data for waist depth samples. Early in Year 1 the decision was made that if the depth of water was less than approximately one meter in depth (designated as the waist depth and deemed the reasonable adult swimming depth), the sample would not be taken. Of the potential number of waist samples that were designated to be taken only 18% of the Year 1 and 27% of year 2 samples were taken. Although subsequent statistical analyses were conducted including the waist depth category, these data must remain suspect as the numbers of samples were so low.

Fecal coliform data for Year 1 and 2 for all stations and all depths are shown in **Figures 8 and 9.** Year 1 fecal coliform densities show only a single occurrence of a Poor quality occurring at the ankle depth on 7.20.09. Year 2 data show substantially more bacterial activity and striking pattern of occurrence with a marked increase in densities between 4.27.10 to 5.25.10 with Poor water quality shown in five of the 17 sampling dates with three at ankle depth and two at knee depth. A secondary peak in the profile was noted for the month of June.

Enterococci data for Year 1 and 2 are shown in **Figures 10 and 11**. Densities for Year 1 show three exceedances of Poor (two for ankle depths and one for knee depth). Year 2 data show a significant trend of exceedances similar to that seen for the fecal coliform within the time frame of 4.27.10 and 5.25.10. In particular, on 5.11.10 all stations all depths were elevated to the Poor category. The pervasive elevated densities for the 5.11.10 date remain undetermined.

Data were evaluated to see if there were correlations among the different depths for the bacterial densities (**Tables 15-18**). **Table 15** shows significant SRCC for all data except that for the Year 1-fecal coliform-waist depth. It is unclear whether this result detects a true difference or if the data set was too small compared to the other depths analyzed.

Table 19 shows the evaluation of the Poor quality responses for dates and depth locations for the two years of the study. A review of this table shows that periodic excursions into Poor water quality occurred at each of the stations. Evaluating both years the numbers of exceedances by station were MS-4 (7), MS-3 (4), MS-2 (3) and MS-1 (3). Although MS-4 was intended to be the control station for this beach, data indicate that the Ochlocknee River waters may be the over-riding influence for this station.

SRCC analyses investigated the individual associations between stations. The results of these analyses are shown in **Tables 18-23**. Year 1 fecal coliform data show that Stations MS-1 and MS-2 were significantly correlated and Stations MS-3 and MS-4 were significantly correlated. Year 2 fecal coliform data show Station MS-1 indicated a correlation with Stations MS-2 and MS-4, and MS-3 was correlated with MS-4. Year 2 fecal coliform data show correlations for MS-1 with stations MS-2, and MS-4 and Station MS-3 was correlated with MS-4. Year 2 enterococci data show MS-1 was significantly correlated with Stations MS-2, MS-3, and MS-4 and Station MS-3 was correlated with MS-4. From these evaluations, it was interesting to note that Station MS-2 was only correlated with Station MS-1 for fecal coliform and enterococci for Year 1 and only for enterococci for Year 2. Although the limitation in the size of the data set may affect these evaluations, it appears that Station 2 (MS-2) alone may not adequately characterize the quality of the Mashes Sands beach water.

Although not confirmed, it appears that the stronger influences of the out flow of the cut through at MS-1 may affect the waters at MS-2 and that the stronger out flow of the Ochlocknee River at MS-4 may have an effect on MS-3. The correlations noted between MS-1 and MS-4 for fecal coliform in Year 1 remains unclear.

Although this study was unable to identify specific sources for the elevated densities noted, it appears that a number of complex contributing elements may be influencing this area. Among the potential factors are the three portable restrooms, the unknown number of residents who may still use septic tanks, and the extensive marsh flats that maintain a significant biological population which may contribute directly or offer nutritional support to bacterial populations of fecal coliform and enterococci from non-anthropogenic sources. Additionally the two water sources, the cut-through from the portion of Apalachee Bay surrounding the inland portion of the marshes at the north end of the beach and the Ochlocknee River at the southern point of the beach can impact this beach depending on flow patterns and eddies that may spin off of these waters.

Additional work is needed to investigate and segregate these various component influences to better determine the complex sources for elevated bacteria noted at this beach.

Figure 8: Fecal coliform data for Mashes Sands Beach Year1 for MS-1, MS-2, MS-3, and MS-4 by depth by sampling date.

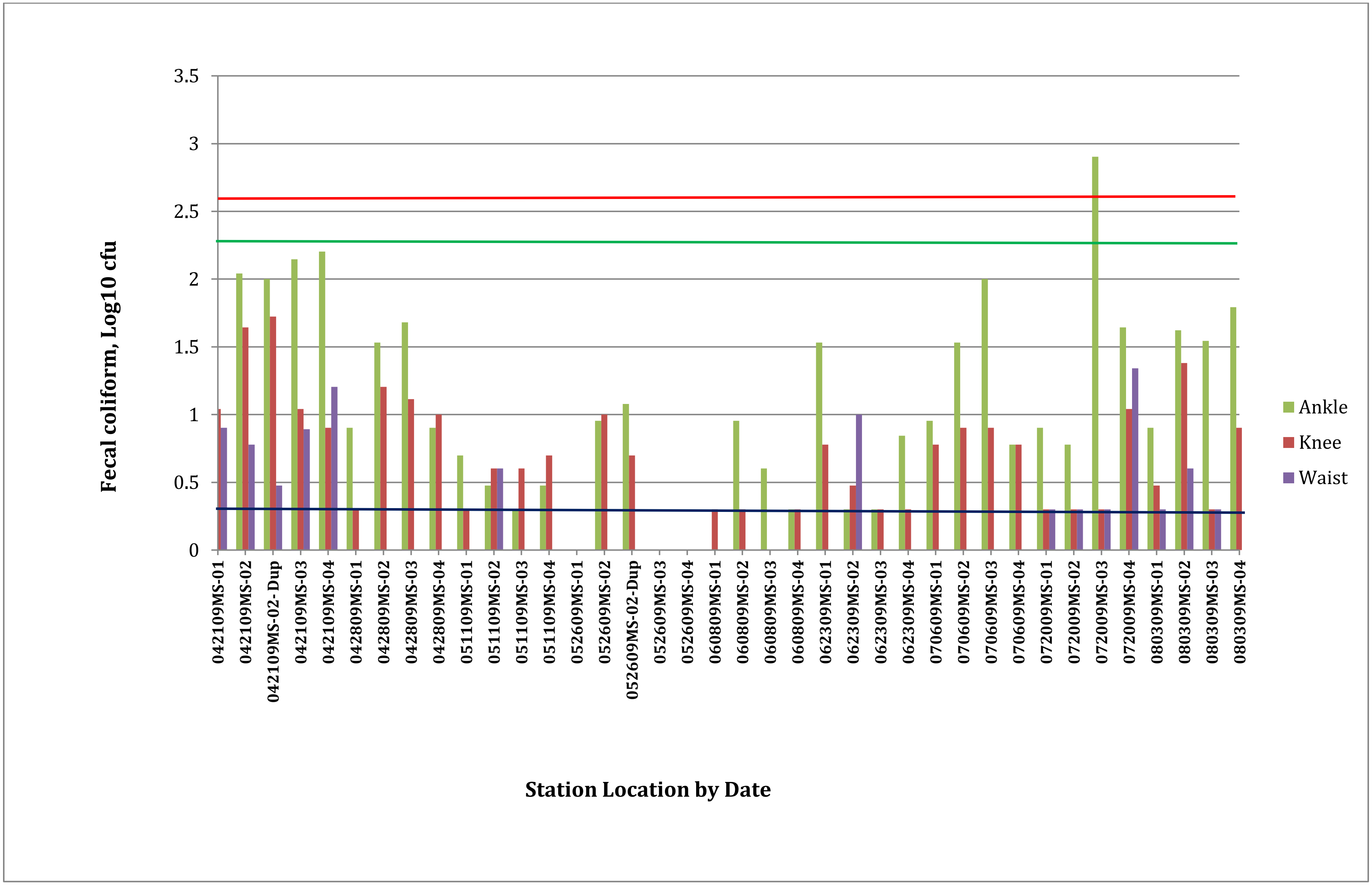


Figure 9: Fecal coliform data for Mashes Sands Beach Year2 for MS-1, MS-2, MS-3, and MS-4 by depth by sampling date.

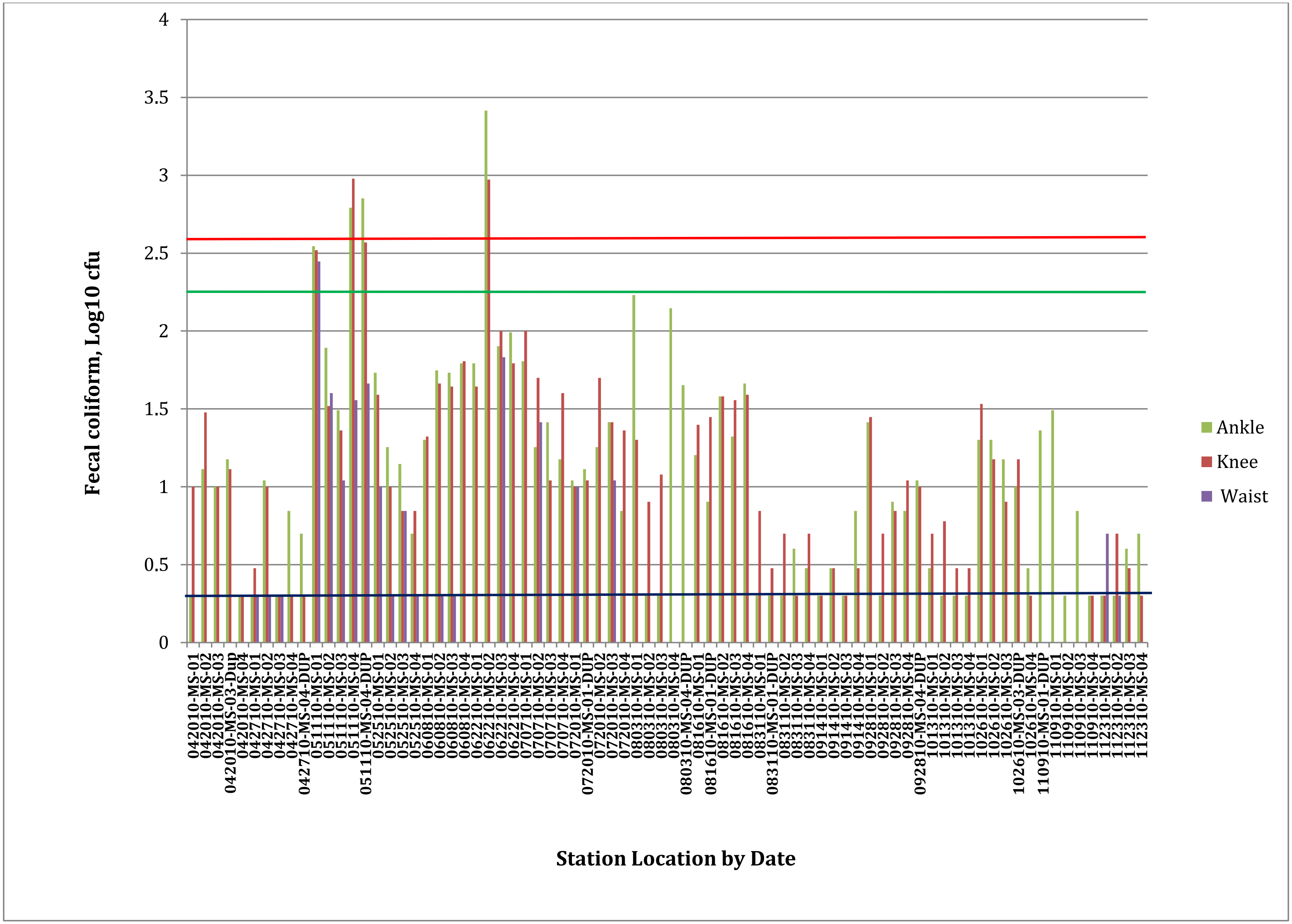


Figure 10: Enterococci data for Mashes Sands Beach Year 1 for MS-1, MS-2, MS-3, and MS-4 by depth by sampling date.

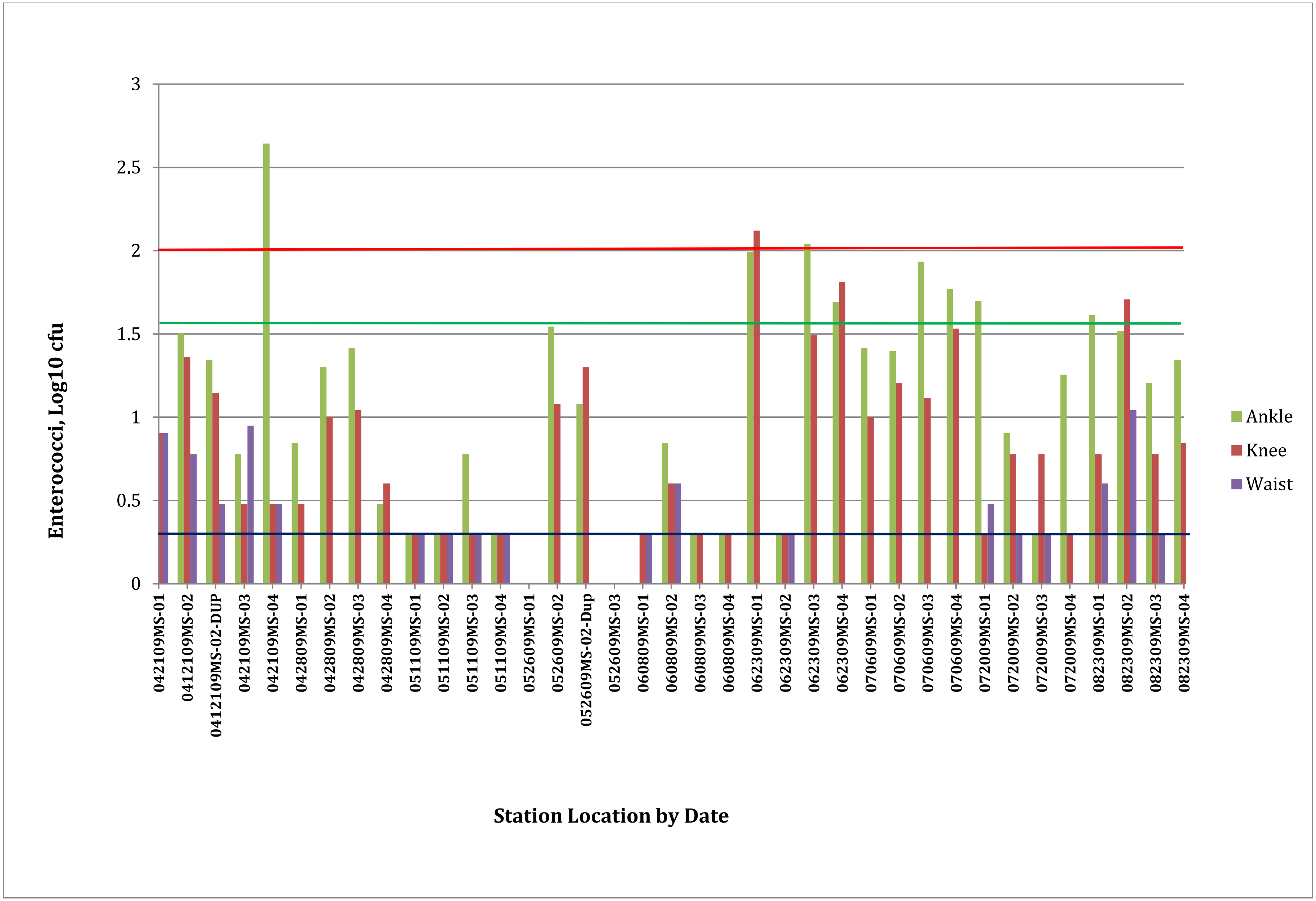
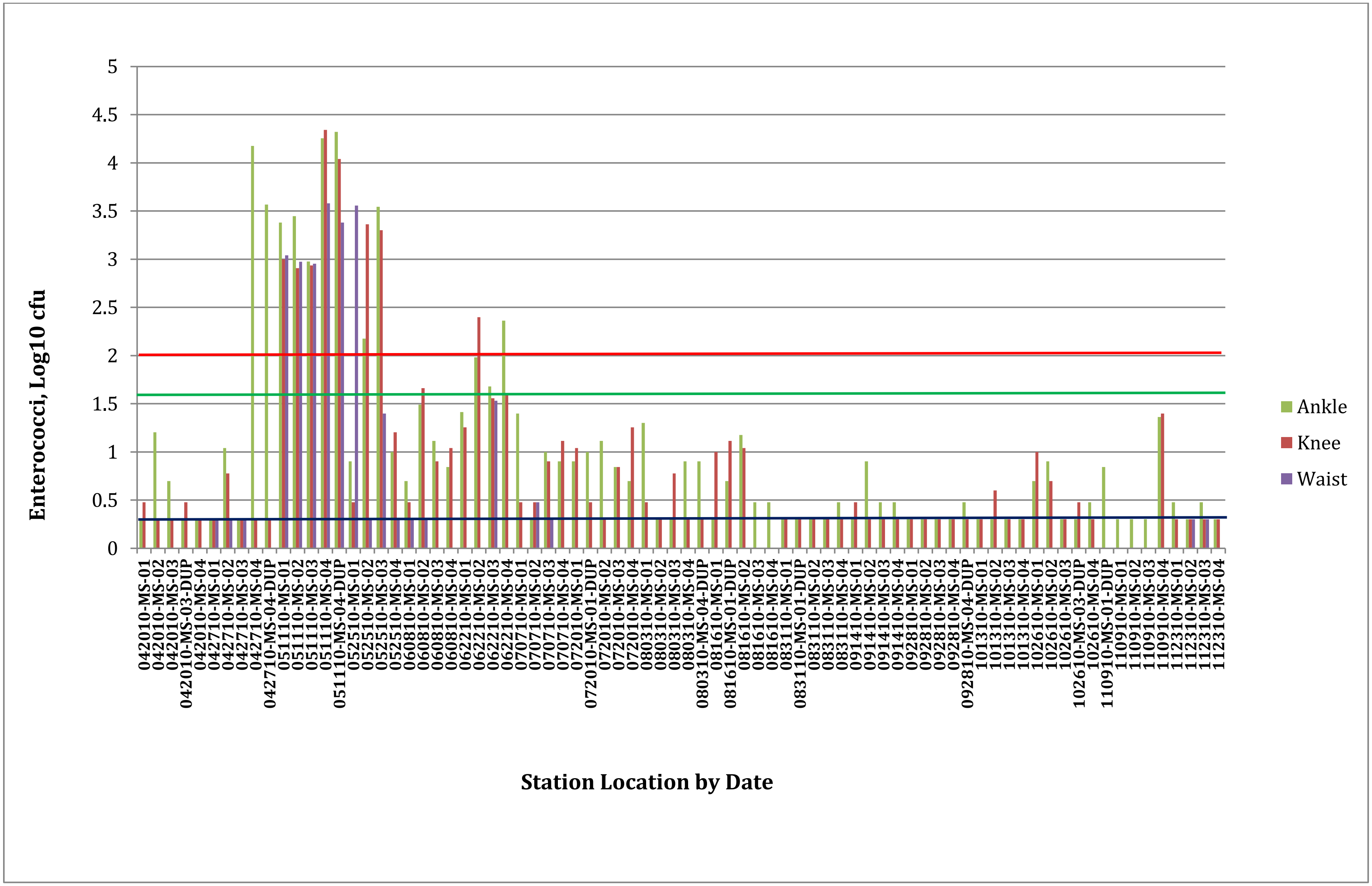


Figure 11: Enterococci data for Mashes Sands Beach Year2 for MS-1, MS-2, MS-3, and MS-4 by depth by sampling date.



Tables related to the Spearman Rank Correlation Coefficient (SRCC) analyses for Mashes Sands Beach by year by depth

Table 15: Spearman Rank Correlations comparing Depth locations and Log_{10} fecal coliform densities at Mashes Sands Beach Year 1 (N=98)

Variable	Spearman Rank Correlations MD pairwise deleted Marked correlations are significant at p< .05000		
	Ankle	Knee	Waist
Ankle	1.000000		
Knee	0.630258	1.000000	
Waist	0.470444	0.617540	1.000000

Table 16: Spearman Rank Correlations comparing Depth locations and Log_{10} fecal coliform densities at Mashes Sands Beach Year 2 (N=143)

Variable	Spearman Rank Correlations MD pairwise deleted Marked correlations are significant at p< .05000		
	Ankle	Knee	Waist
Ankle	1.000000		
Knee	0.856681	1.000000	
Waist	0.731303	0.712075	1.000000

Table 17: Spearman Rank Correlations comparing Depth locations and Log_{10} enterococci densities at Mashes Sands Beach Year 1 (N=98)

Variable	Spearman Rank Correlations MD pairwise deleted Marked correlations are significant at p< .05000		
	Ankle	Knee	Waist
Ankle	1.000000		
Knee	0.671642	1.000000	
Waist	0.558921	0.638417	1.000000

Table 18: Spearman Rank Correlations comparing Depth locations and Log_{10} enterococci densities at Mashes Sands Beach Year 2 (N=143)

Variable	Spearman Rank Correlations MD pairwise deleted Marked correlations are significant at p< .05000		
	Ankle	Knee	Waist
Ankle	1.000000		
Knee	0.623465	1.000000	
Waist	0.629099	0.562352	1.000000

Table 19: Dates of the exceedance of bacterial densities for Mashes Sands Beach above the Poor category ($\log_{10}$ FC > 2.6 and Log_{10} enterococci > 2.0) by study year, organism, depth of sample and station location.						
Location	**Year**	**Organism**	**Date**	**Depth**	**Station**	**Comment**
Mashes Sands	1	FC	7.20.09	A	MS-3	
Beach	2	FC	5.11.10	A,K	MS-4	
			5.11.10	A	MS-4	DUP
			6.22.10	A,K	MS-2	
Mashes Sands	1	ENTERO	4.21.09	A	MS-4	
Beach			6.23.09	K	MS-1	
			6.23.09	A	MS-3	
	2	ENTERO	4.27.10	A	MS-4	
			4.27.10	A	MS-4	DUP
			5.11.10	ALL	ALL	
			5.25.10	W	MS-1	
			5.25.10	A,K	MS-2	
			5.25.10	A,K	MS-3	
			6.22.10	K	MS-2	
			6.22.10	A	MS-4	

Tables related to the Spearman Rank Correlation Coefficient (SRCC) analyses for Mashes Sands Beach by year, station location, Log_{10} bacterial density

Table 20: Spearman Rank Correlations comparing station locations and Log_{10} fecal coliform densities at Mashes Sands Beach for Year 1 (N=98)

Variable	Spearman Rank Correlations MD pairwise deleted Marked correlations are significant at p< .05000			
	MS-1	MS-2	MS-3	MS-4
MS-1	1.000000			
MS-2	0.549909	1.000000		
MS-3	0.186957	0.407486	1.000000	
MS-4	0.242388	0.357517	0.256452	1.000000

Table 21: Spearman Rank Correlations comparing station locations and Log_{10} fecal coliform densities at Mashes Sands Beach for Year 2 (N=143)

Variable	Spearman Rank Correlations MD pairwise deleted Marked correlations are significant at p< .05000			
	MS-1	MS-2	MS-3	MS-4
MS-1	1.000000			
MS-2	0.387959	1.000000		
MS-3	0.251189	0.198014	1.000000	
MS-4	0.346539	0.252717	0.319420	1.000000

Table 22: Spearman Rank Correlations comparing station locations and Log_{10} enterococci densities at Mashes Sands Beach for Year 1 (N=98)

Variable	Spearman Rank Correlations MD pairwise deleted Marked correlations are significant at p< .05000			
	MS-1	MS-2	MS-3	MS-4
MS-1	1.000000			
MS-2	0.327666	1.000000		
MS-3	0.095204	0.158093	1.000000	
MS-4	0.299519	0.315883	0.679732	1.000000

Table 23: Spearman Rank Correlations comparing station locations and Log_{10} enterococci densities at Mashes Sands Beach for Year 2 (N=143)

Variable	Spearman Rank Correlations MD pairwise deleted Marked correlations are significant at p< .05000			
	MS-1	MS-2	MS-3	MS-4
MS-1	1.000000			
MS-2	0.378419	1.000000		
MS-3	0.422968	0.143041	1.000000	
MS-4	0.374971	0.29918	0.638664	1.000000

B. Environmental parameter analyses

Environmental parameters were taken at each station designated Knee for each of the two beaches for the duration of the study. **Table 24** illustrates the maximum, minimum, and average by the study year for the four environmental parameters monitored. Again, it is noted that the stations for the Carrabelle Beach were changed during Year 2 (2010).

Table 24: Comparison of the environmental parameters for Year 1: March 10, 2009 – August 3, 2009 (6 months); Year 2: March 31, 2010 – November 23, 2010 (8 months); and the composite (ALL) for each site.

Carrabelle Beach																
2009	**Sal ‰**	**Temp °C**	**D.O.**	**pH**		**2010**	**Sal ‰**	**Temp °C**	**D.O.**	**pH**		**ALL**	**Sal ‰**	**Temp °C**	**D.O.**	**pH**
Max	33.5	32.9	9.6	9.9		Max	32.8	32.8	14.2	9.6		Max	33.5	32.9	14.2	9.9
Min	18.4	20.5	3.6	6.1		Min	18.7	14.5	3.5	4.5		Min	18.4	14.5	3.5	4.5
Avg	28.8	27.7	6.5	8.1		Avg	28.0	26.2	6.8	7.8		Avg	28.3	26.6	6.7	7.9
Mashes Sands Beach																
2009	**Sal ‰**	**Temp °C**	**D.O.**	**pH**		**2010**	**Sal ‰**	**Temp °C**	**D.O.**	**pH**		**ALL**	**Sal ‰**	**Temp °C**	**D.O.**	**pH**
Max	35.2	33.7	8.7	8.7		Max	29.9	36.5	12.1	8.9		Max	35.2	36.5	12.1	8.9
Min	19.3	20.7	4.5	7.5		Min	17.8	15.9	5.5	4.7		Min	17.8	15.9	4.5	4.7
Avg	29.6	28.4	7.0	8.0		Avg	25.0	28.2	7.8	7.7		Avg	26.5	28.3	7.6	7.8

Spearman Rank Correlation analyses were done to determine whether significance was observed both between individual environmental parameters as well as the parameter associations with the Log10 fecal coliform or enterococci densities. Analyses for Carrabelle Beach and for Mashes Sands Beach are shown by study year in **Tables 25-28** and **Tables 29-32**, respectively, below.

One striking observation was that the data for both Year 1 and Year 2 Carrabelle Beach for both fecal coliform and enterococci data show significant inverse correlations with salinity whereas data for Year 1 and Year 2 Mashes Sands for both fecal coliform and enterococci show no significant correlation with salinity. The cause of this unexpected result was not determined; however, due to the small data sets and the large number of non-detects among the data this disparity may be an analytical artifact. There was nothing in the range of the data for either year or beach location that suggested substantial differences in the environmental data sets.

The significance of associations between parameters, both environmental and bacterial, varied between years for both beach locations. Therefore, discernable trends were could not be identified either within the 2 year time frame at a single beach or in general terms between beaches. See **Tables** on the following page.

Data for Carrabelle Beach shows that significance between environmental parameters and bacterial densities varied. Since station locations were changed during the Year 2 portion of the study, comparisons between the different years could not be made for Carrabelle Beach data.

Year 2 data for fecal coliform was striking as these densities were correlated to all four parameters. This was not seen elsewhere in any other yearly data set.

Data for Mashes Sands Beach shows similar difficulties in demonstrating significance among the environmental parameters or in association with the Log_{10} bacterial densities. The singular remarkable observation was the lack of correlation with salinity for both fecal coliform and enterococci for both years of the study.

Carrabelle Beach Analyses

Table 25: Spearman Rank Order Correlations for Carrabelle Beach Fecal coliform (FC) Data Year 1 (2009)

	Spearman Rank Order Correlations MD pairwise deleted Marked correlations are significant at p<.05000				
Variable	FC(log10)	DO (mg/L)	pH	Temp,° C	S ‰
FC(log10)	1.000000				
DO (mg/L)	0.150598	1.000000			
pH	-0.351989	0.631161	1.000000		
Temp,° C	-0.193844	-0.643544	-0.193223	1.000000	
S ‰	-0.594392	-0.331237	0.117866	0.364197	1.000000

Table 26: Spearman Rank Order Correlations for Carrabelle Beach Fecal coliform (FC) Data Year 2 (2010)

	Spearman Rank Order Correlations MD pairwise deleted Marked correlations are significant at p<.05000				
Variable	FC(log10)	DO (mg/L)	pH	Temp,° C	S ‰
FC(log10)	1.000000				
DO (mg/L)	-0.199124	1.000000			
pH	-0.492165	0.052644	1.000000		
Temp,° C	0.238067	-0.366357	-0.310780	1.000000	
S ‰	-0.224233	-0.051909	-0.308405	-0.326172	1.000000

Table 27: Spearman Rank Order Correlations for Carrabelle Beach Enterococci Data Year 1 (2009)

	Spearman Rank Order Correlations MD pairwise deleted Marked correlations are significant at p<.05000				
Variable	Entero (log10)	DO (mg/L)	pH	Temp,° C	S ‰
Entero (log10)	1.000000				
DO (mg/L)	0.153990	1.000000			
pH	-0.147114	0.631161	1.000000		
Temp,° C	-0.163762	-0.643544	-0.193223	1.000000	
S ‰	-0.706670	-0.331327	0.117866	0.364197	1.000000

Table 28: Spearman Rank Order Correlations for Carrabelle Beach Enterococci Data Year 2 (2010)

	Spearman Rank Order Correlations MD pairwise deleted Marked correlations are significant at p<.05000				
Variable	Entero(log10)	DO (mg/L)	pH	Temp,° C	S ‰
Entero(log10)	1.000000				
DO (mg/L)	-0.084886	1.000000			
pH	-0.522397	0.052974	1.000000		
Temp,° C	0.112410	-0.367081	-0.316621	1.000000	
S ‰	-0.285536	-0.050088	0.308066	-0.328520	1.000000

Mashes Sands Beach Analyses

Table 29: Spearman Rank Order Correlations for Mashes Sands Fecal coliform (FC) Data Year 1 (2009)

	Spearman Rank Order Correlations MD pairwise deleted Marked correlations are significant at p<.05000				
Variable	FC(log10)	DO (mg/L)	pH	Temp,° C	S ‰
FC(log10)	1.000000				
DO (mg/L)	0.051222	1.000000			
pH	-0.184347	-0.212987	1.000000		
Temp,° C	-0.293543	-0.178829	0.412779	1.000000	
S ‰	-0.157583	-0.564337	0.327040	0.798923	1.000000

Table 30: Spearman Rank Order Correlations for Mashes Sands Fecal coliform (FC) Data Year 2 (2010)

	Spearman Rank Order Correlations MD pairwise deleted Marked correlations are significant at p<.05000				
Variable	FC(log10)	DO (mg/L)	pH	Temp,° C	S ‰
FC(log10)	1.000000				
DO (mg/L)	-0.294295	1.000000			
pH	-0.402121	0.534007	1.000000		
Temp,° C	0.220179	-0.261069	-0.296096	1.000000	
S ‰	-0.107609	-0.221021	-0.164576	0.134167	1.000000

Table 31: Spearman Rank Order Correlations for Mashes Sands Enterococci Data Year 1 (2009)

	Spearman Rank Order Correlations MD pairwise deleted Marked correlations are significant at p<.05000				
Variable	Entero (log10)	DO (mg/L)	pH	Temp,° C	S ‰
Entero (log10)	1.000000				
DO (mg/L)	-0.044598	1.000000			
pH	-0.243149	-0.235120	1.000000		
Temp,° C	0.024144	-0.185005	0.397460	1.000000	
S ‰	0.150532	-0.545123	0.276648	0.795461	1.000000

Table 32: Spearman Rank Order Correlations for Mashes Sands Enterococci Data Year 2 (2010)

	Spearman Rank Order Correlations MD pairwise deleted Marked correlations are significant at p<.05000				
Variable	Entero (log10)	DO (mg/L)	pH	Temp,° C	S ‰
Entero (log10)	1.000000				
DO (mg/L)	-0.167840	1.000000			
pH	-0.417999	0.528454	1.000000		
Temp,° C	0.034536	-0.259237	-0.304083	1.000000	
S ‰	-0.086775	-0.218391	-0.168400	0.136006	1.000000

C. FDOH data Analyses

With the exception of the initial reconnaissance event on 3-31-09, FDEP sampling occurred concurrently with the FDOH beach sampling for each beach. The single FDOH designated sampling station at each beach (Lat-Long previously identified in the Sampling Design section) was concurrent with FDEP's Stations CB-2-K and MS-2-K throughout the entire study. On a single occasion (11-09-10, Mashes Sands Beach) the sampling log indicates that the water on this date was very shallow resulting in both FDEP and FDOH sampling at ankle depth rather than at knee depth. The time of sampling was contingent upon the FDOH sampling schedule and occurred within a two hour window on the morning of the sampling date. No attempt to assess tidal influences was made during this study.

FDOH's Bathing Beach Criteria is established based on a range of values (cfu/100 mL of sample) to determine categories of beach water quality. **Table 3** (Section II. Study Design) identified categories as Good, Moderate, and Poor based on a range of numerical bacterial densities. In this Table, the category definitions apply to either a single sample max (SSM) or the rolling geometric mean (GM) of data collected over a 30-day period of time. A Poor category in either fecal coliform or enterococci bacteria resulting from a SSM or GM exceedance results in a Beach Advisory and this designation remains in effect until waters for that beach return to acceptable bacteriological quality of Good or Moderate. The FDEP TMDL Program uses the category of beach waters and the number of days of the Beach Advisory in its assessment of water body impairment. For the purposes of this study FDEP used only the SSM criteria in FDOH/FDEP comparative evaluations.

One of the primary goals of this study was to evaluate whether the FDOH single sample adequately characterized the designated beach area. A preliminary assessment of the FDEP/FDOH data compared the category of the waters for the sampling dates. Using the FDOH Criteria above, FDEP data were assigned the appropriate category and then compared by category to that of the category given to the result of the FDOH sample. Comparisons are presented below in **Tables 33-36**. These evaluations were based on the comparison of SSM only since FDEP data were not evaluated for the rolling GM. Classification Agreement is self explanatory. Classification Non-agreement occurred when either agency differed in the category designation and included both the single (e.g., Good vs. Moderate or Moderate vs. Poor designations) and double (e.g., Good vs. Poor designations) division separations. Classification Major non-agreement is the subset of the Classification non-agreement data including only the double division separation.

Data for fecal coliform analyses for the Year 1 demonstrated 100% concurrence between the FDEP and FDOH classifications for both Carrabelle Beach and Mashes Sands Beach. Year 2 data (**Table 33**) however show that one sample for Carrabelle Beach (7-20-10) and one sample for Mashes Sands Beach (6-22-10) resulted in a difference of one category with FDEP a Moderate classification and FDOH a Good classification.

Data for enterococci were evaluated comparing Year 1 and Year 2 separately, **Table 33 and 34** respectively, and combined, **Table 36**. The combined analysis was appropriate since Carrabelle Beach Station 2 was monitored for both years of the study. Year I data show that Carrabelle Beach enterococci data were concurrent with FDOH data 56% of the time and Mashes Sands Beach Year 1 enterococci data shows a concurrence of 33% Classification agreement. Year 2 data show that

Carrabelle Beach shows concurrence 82% of the time and Mashes sands Beach data were concurrent 72% of the time. Combined data sets for Year 1 and Year 2 show concurrence in Classification agreement at Carrabelle Beach 65% of the time and Mashes Beach 59% of the time. Discrepancies that were considered Major non-agreement occurred in less than 20% of the comparisons.

Table 33: Comparison of fecal coliform data between the FDEP Station 2 and the FDOH Beach Monitoring Station by beach for Year 1 and Year 2 data combined.

Fecal coliform	Classification Agreement*	Classification Non-agreement (FDOH > FDEP)	Classification Non-agreement (FDEP>FDOH)	Classification Major non-agreement
Carrabelle Beach	96% (25/26)	0%	4%(1/26)	0%
Mashes Sands Beach	96%(26/27)	0%	4%(1/27)	0%

Table 34: Comparison of enterococci data between the FDEP Station 2 and the FDOH Beach Monitoring Station for the two beaches studied for Year 1 Data only.

Enterococci	Classification Agreement	Classification Non-agreement (FDOH > FDEP)	Classification Non-agreement (FDEP>FDOH)	Classification Major non-agreement**
Carrabelle Beach	56% (5/9)	33% (3/9)	11%(1/9)	33%(3/9)
Mashes Sands Beach	33% (3/9)	56% (5/9)	11%(1/9)	33% (3/9)

Note: **Major Non-agreement: 2 FDOH >FDEP and 1 FDEP>FDOH at CB; 3 FDOH>FDEP and 1 FDEP>FDOH at MS

Table 35: Comparison of enterococci data between the FDEP Station 2 and the FDOH Beach Monitoring Station for the two beaches studied for Year 2 Data only.

Enterococci	Classification Agreement	Classification Non-agreement (FDOH > FDEP)	Classification Non-agreement (FDEP>FDOH)	Classification Major non-agreement
Carrabelle Beach	82% (14/17)	18% (3/17)	0%	0%
Mashes Sands Beach	72% (13/18)	17% (3/18)	11%(2/18)	2/18 (11%)*

Note: *2 FDEP samples were Poor when FDOH identified Good.

Table 36: Comparison of enterococci data between the FDEP Station 2 and the FFDOH Beach Monitoring Station for the two beaches studied for combined Year 1 and Year 2 data.

Enterococci	Classification Agreement	Classification Non-agreement (FDOH > FDEP)	Classification Non-agreement (FDEP>FDOH)	Classification Major non-agreement
Carrabelle Beach	65% (17/26)	23% (6/26)	12%(3/26)	12%(3/26)
Mashes Sands Beach	59% (16/27)	30% (8/27)	11%(3/27)	19%(5/27)

In addition to the evaluation of the category determinations, data were reviewed as numerical densities for both FDEP and FDOH data. FDOH data were graphed against the FDEP Station 02-K data and additionally against all water depths at Station 02.

Figures 12-27 graph FDOH data against the FDEP Station 02 data for the companion FDOH sampling station as well as all depths for that station. Threshold bars for the FDOH category classification are illustrated as the colored horizontal bars: Green is the upper threshold for Good (FC: 2.3=Log_{10} 199; Enterococci: 1.5=Log_{10}35), Red is the upper threshold for Moderate (FC: 2.6=log_{10} 399; Enterococci: 2.0=log_{10}103) and data above the Red bar are categorized as Poor (comparable to greater than 400 cfu/100mL for FC and 105 cfu/100mL for Enterococci). Data graphed or otherwise analyzed were log_{10} transformed (log_{10} cfu/100 mL sample) of the final value reported for each sample. The horizontal Blue line denotes the MDL for the analyses.

Figure 12: Comparison of fecal coliform for FDOH data with Carrabelle Beach Station CB-2 for Year 1 by date for the Knee Depth by date

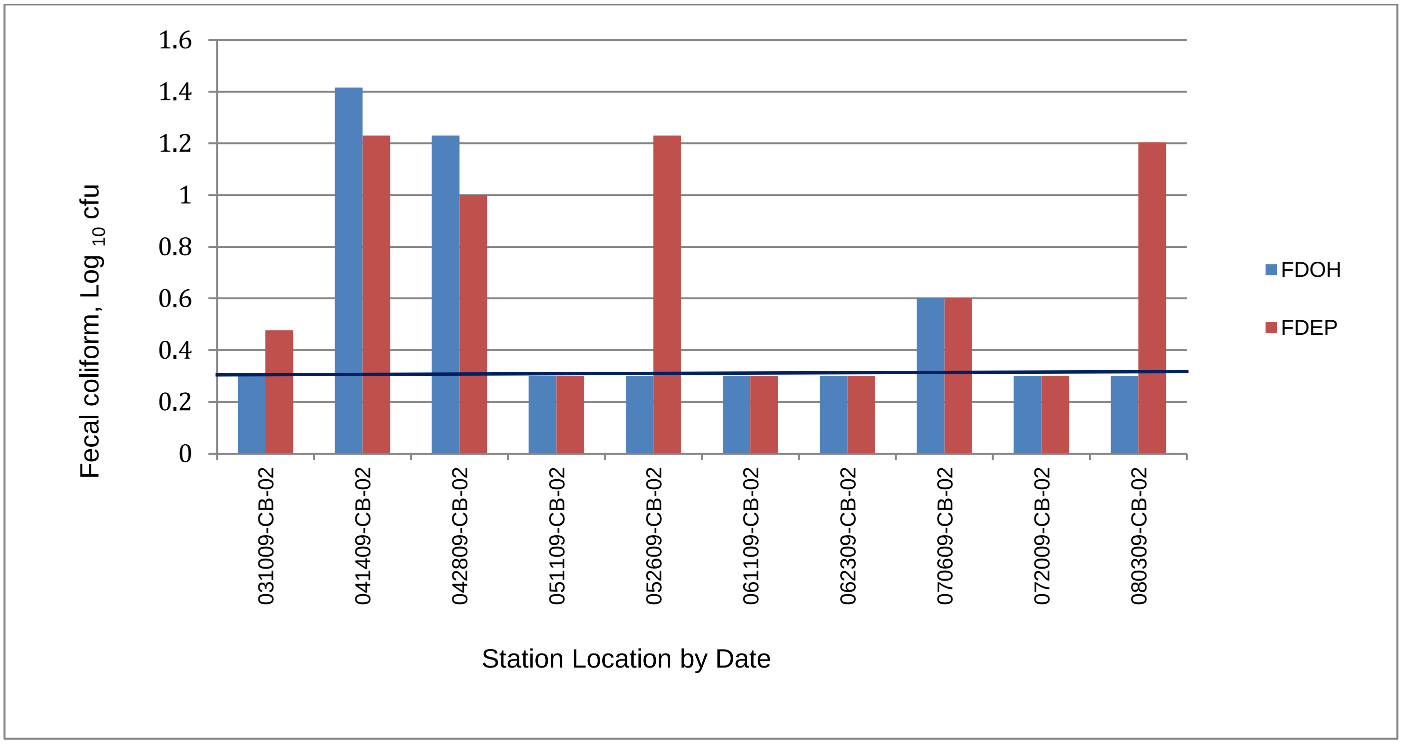


Figure 13: Comparison of fecal coliform for FDOH data with Carrabelle Beach Station CB-2 for Year 1 for all Depths by date

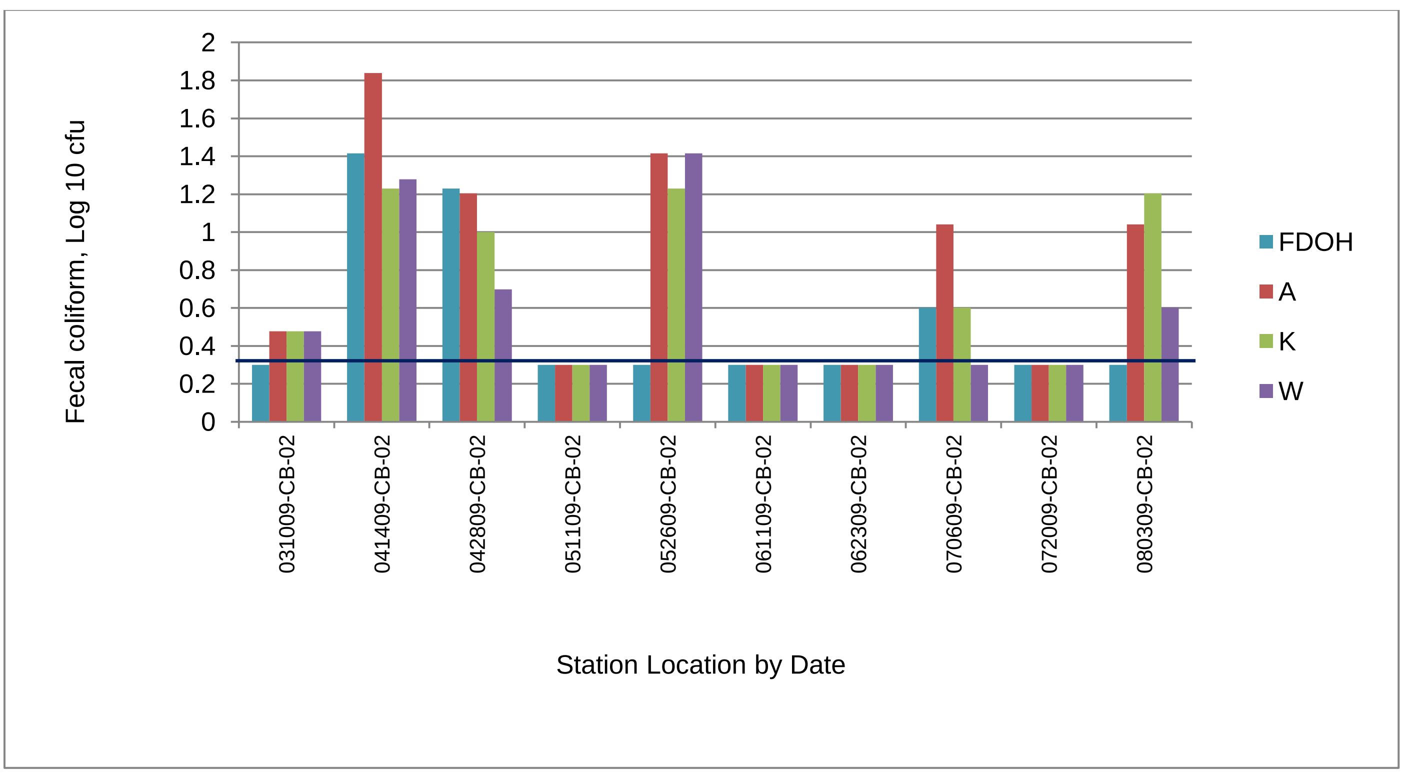


Figure 14: Comparison of fecal coliform for FDOH data with Carrabelle Beach Station CB-2 for Year 2 by date

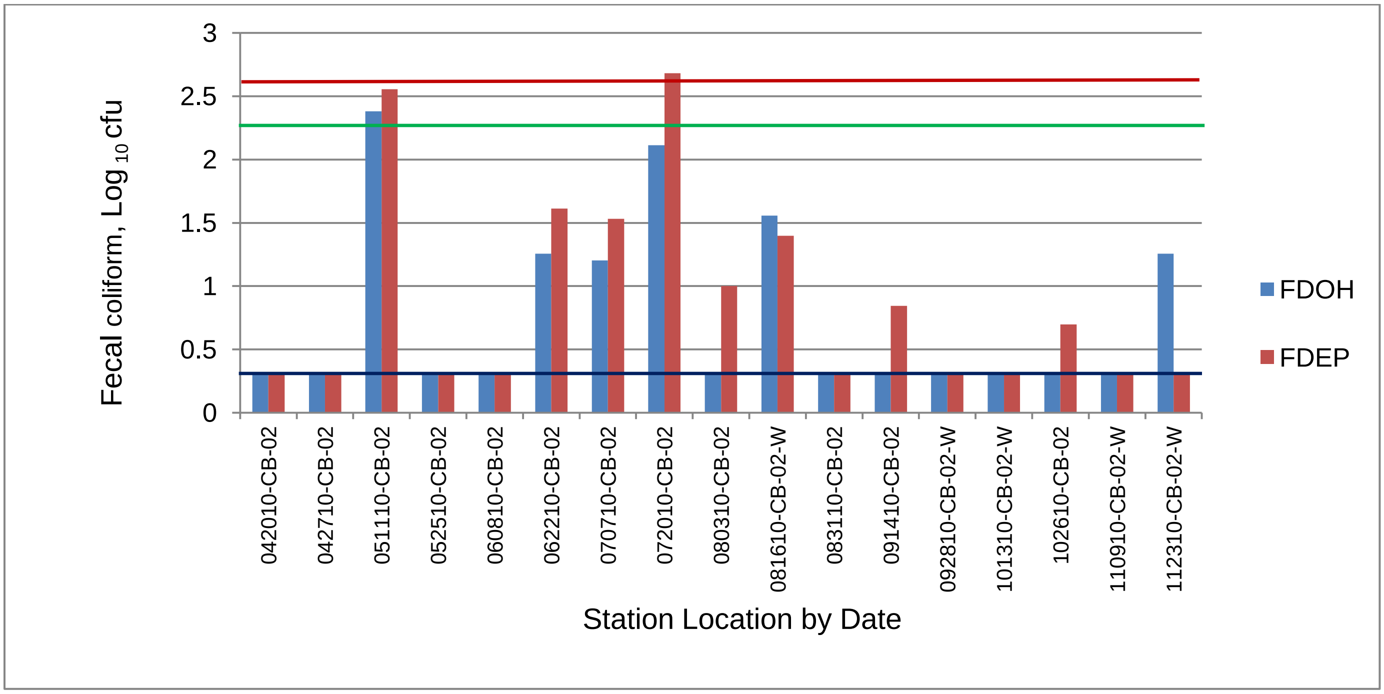


Figure 15: Comparison of fecal coliform for FDOH data with Carrabelle Beach Station CB-2 for Year 2 for all Depths by date

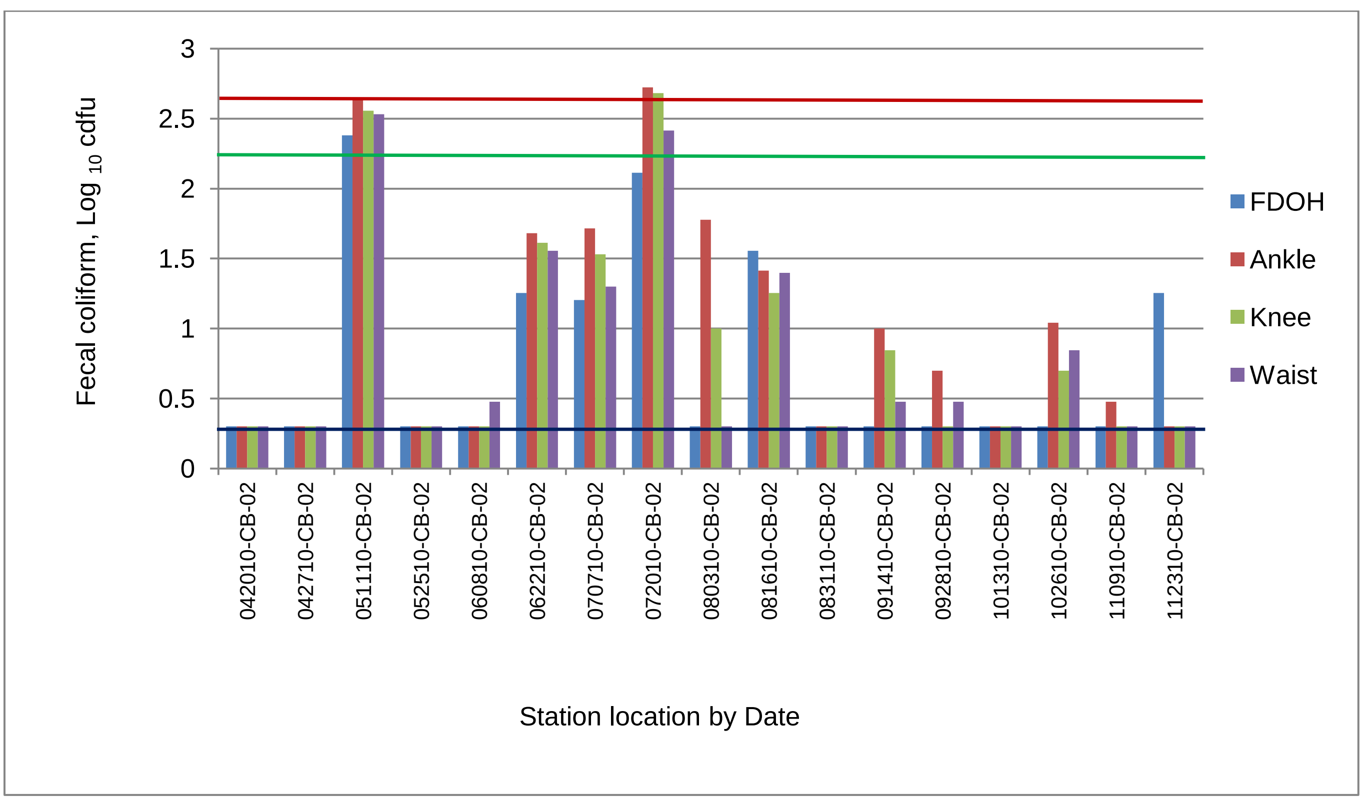


Figure 16: Comparison of fecal coliform for FDOH data with Mashes Sands Beach Station MS-2 for Year 1 by date for the Knee Depth by date

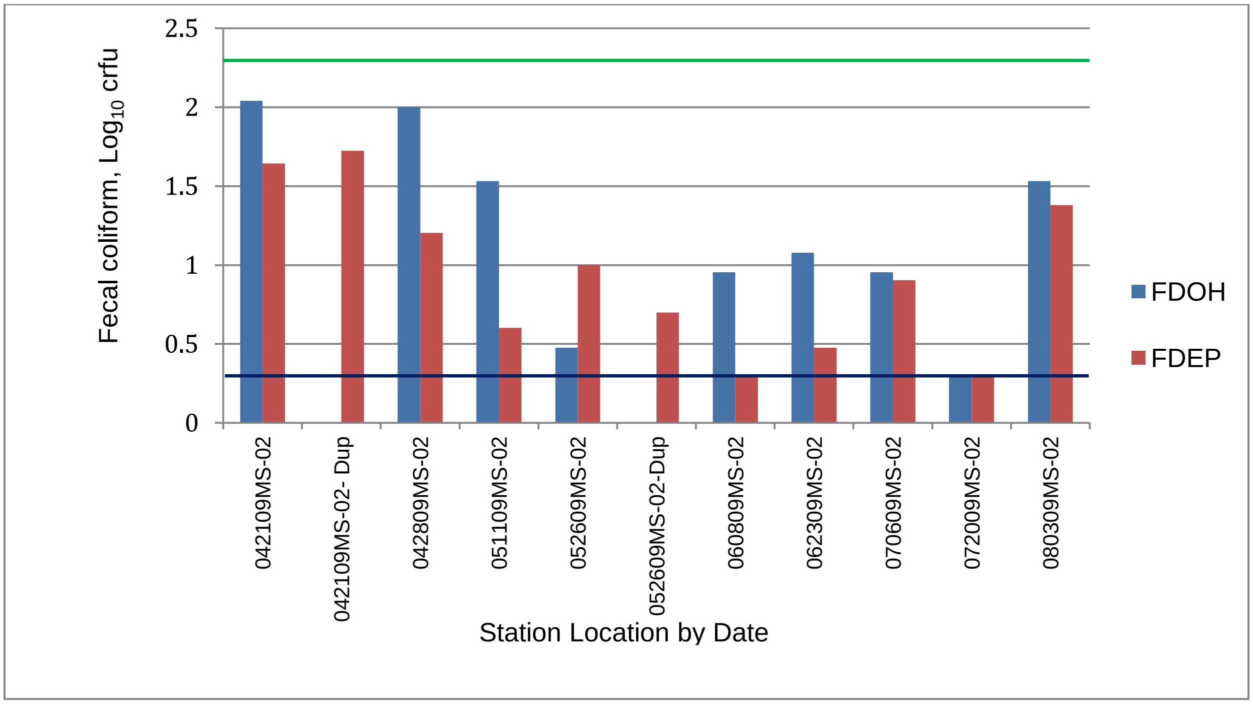


Figure 17: Comparison of fecal coliform for FDOH data with Mashes Sands Beach Station CB-2 for Year 1 by date for all Depths by date

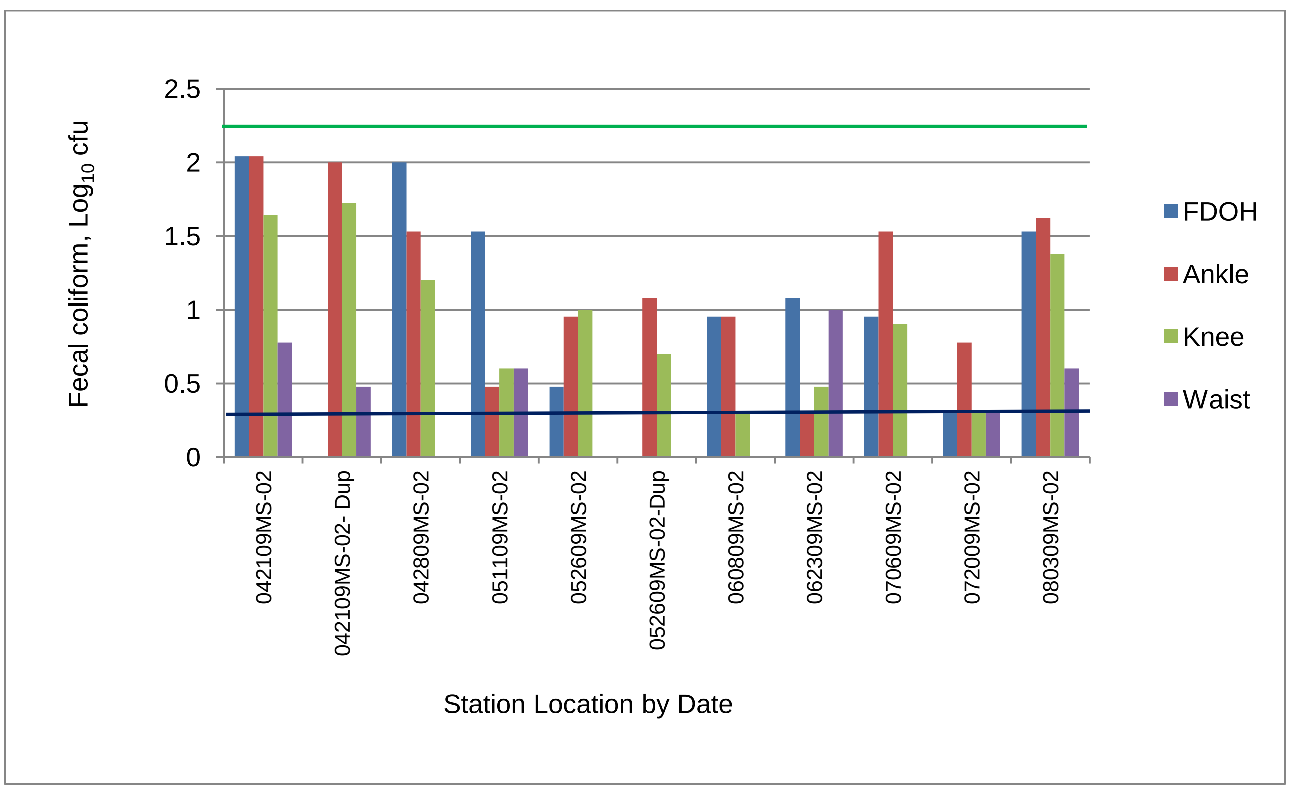


Figure 18: Comparison of fecal coliform for FDOH data with Mashes Sands Beach Station CB-2 for Year 2 by date for the Knee Depth by date

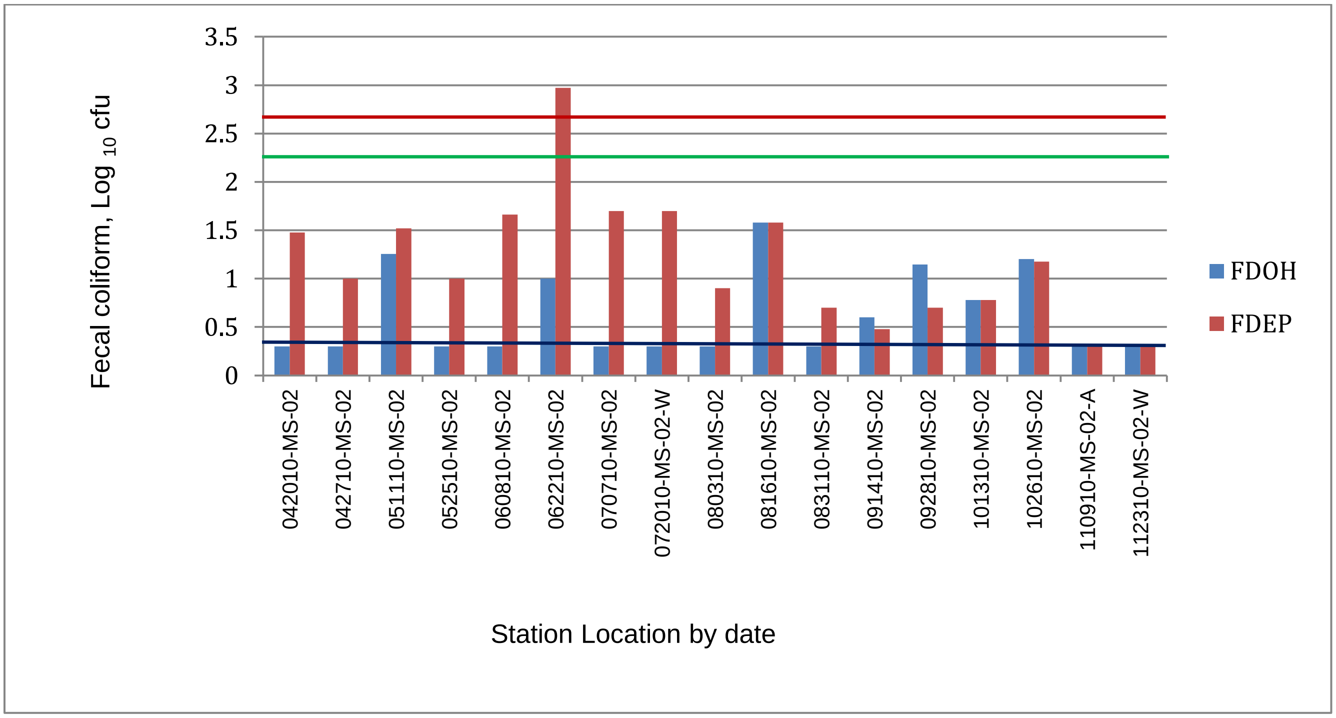


Figure 19: **Comparison of fecal coliform for FDOH data with Mashes Sands Beach Station CB-2 for Year 2 by date for all Depths by date**

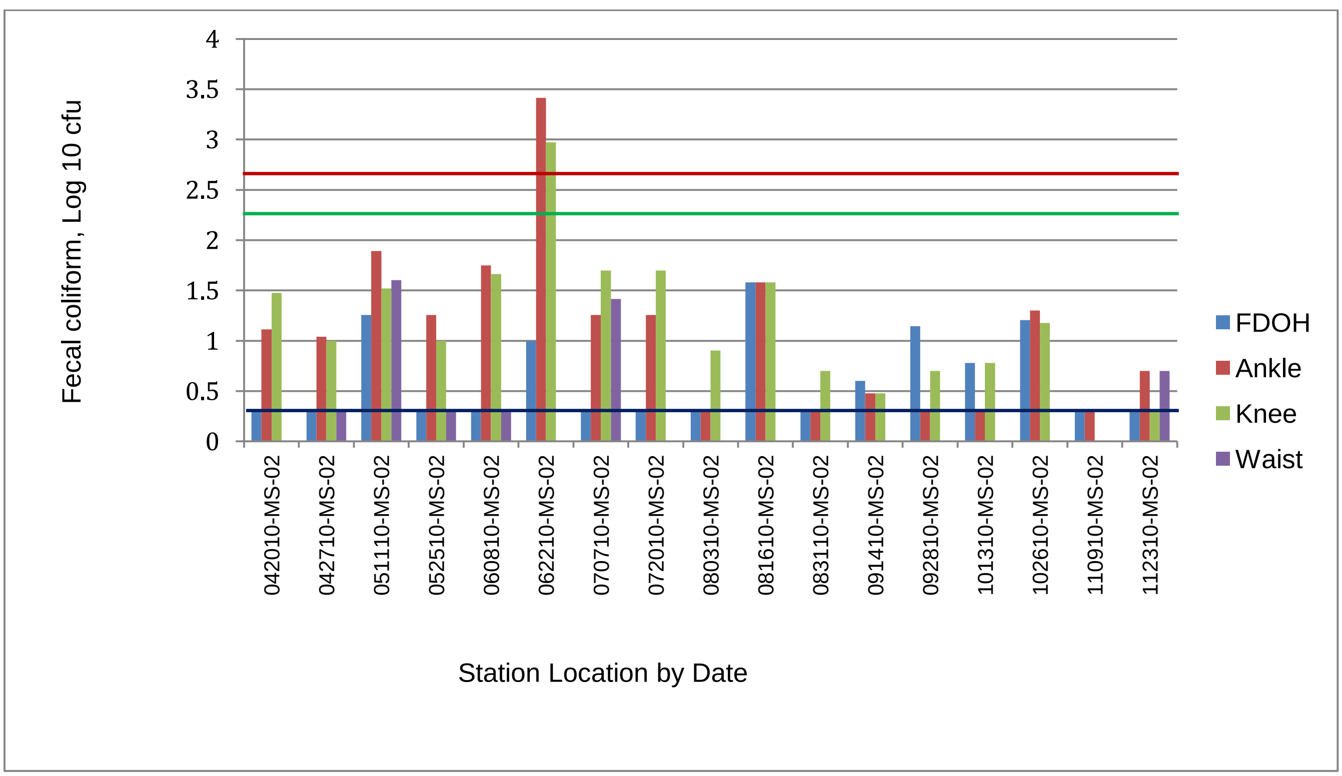


Figure 20: Comparison of enterococci for FDOH data with Carrabelle Beach Station CB-2 for Year 1 by date for the Knee Depth by date

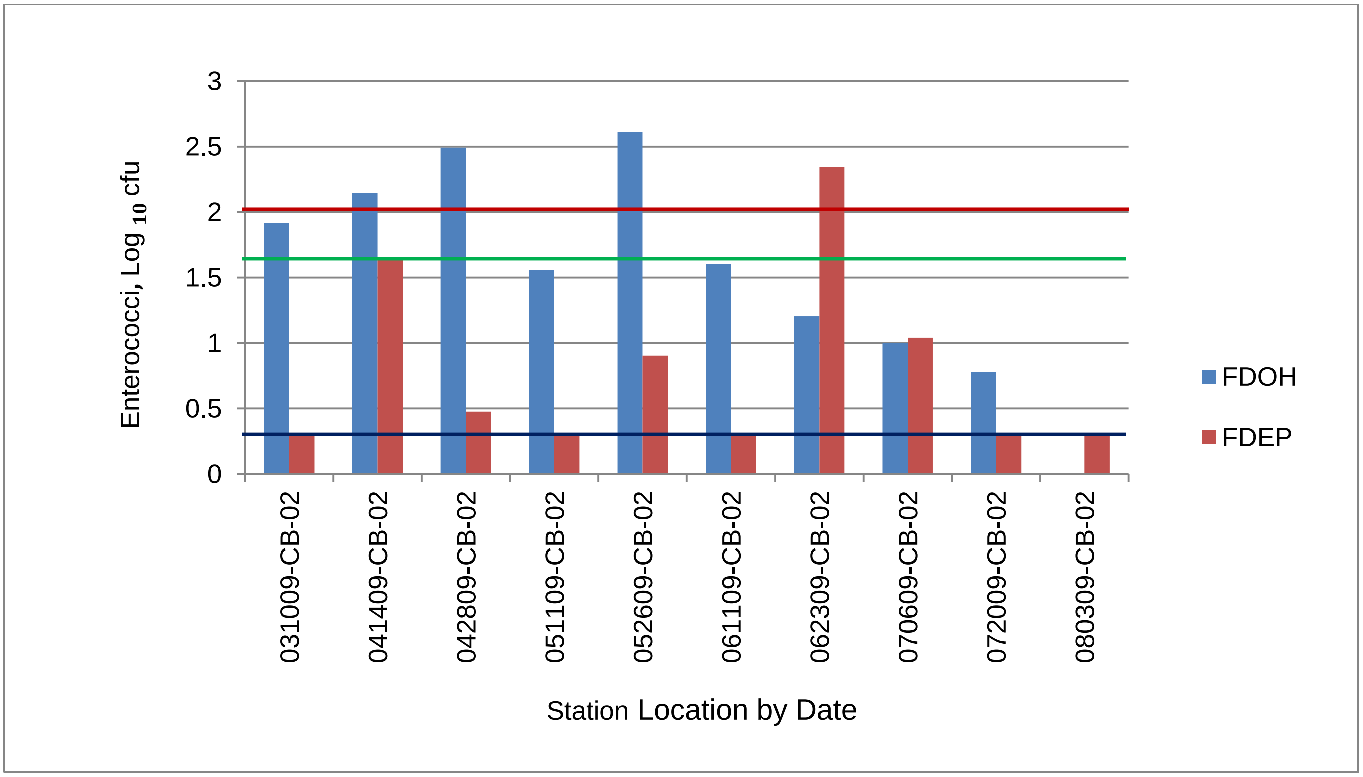


Figure 21: Comparison of enterococci for FDOH data with Carrabelle Beach Station CB-2 for Year 1 by date for all Depths by date

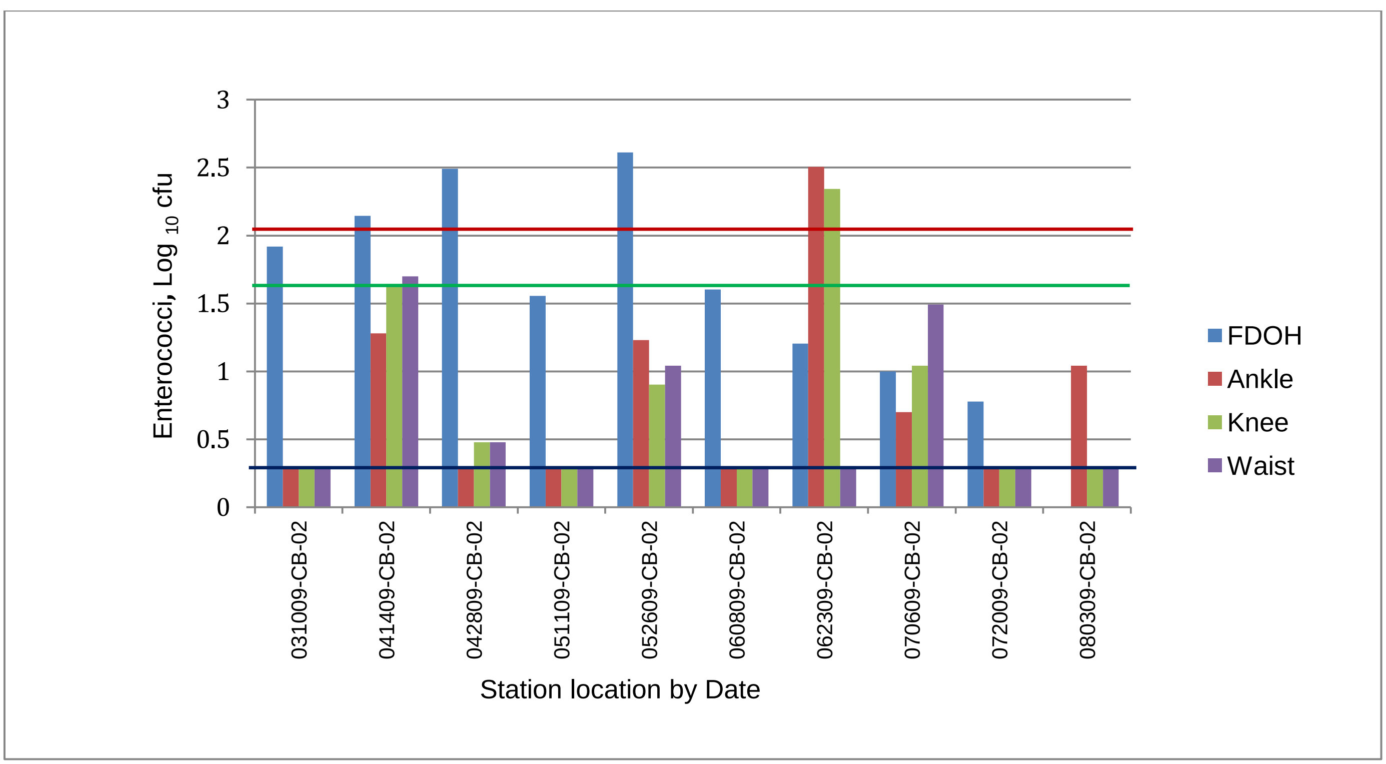


Figure 22: Comparison of enterococci for FDOH data with Carrabelle Beach Station CB-2 for Year 2 by date for the Knee Depth by date

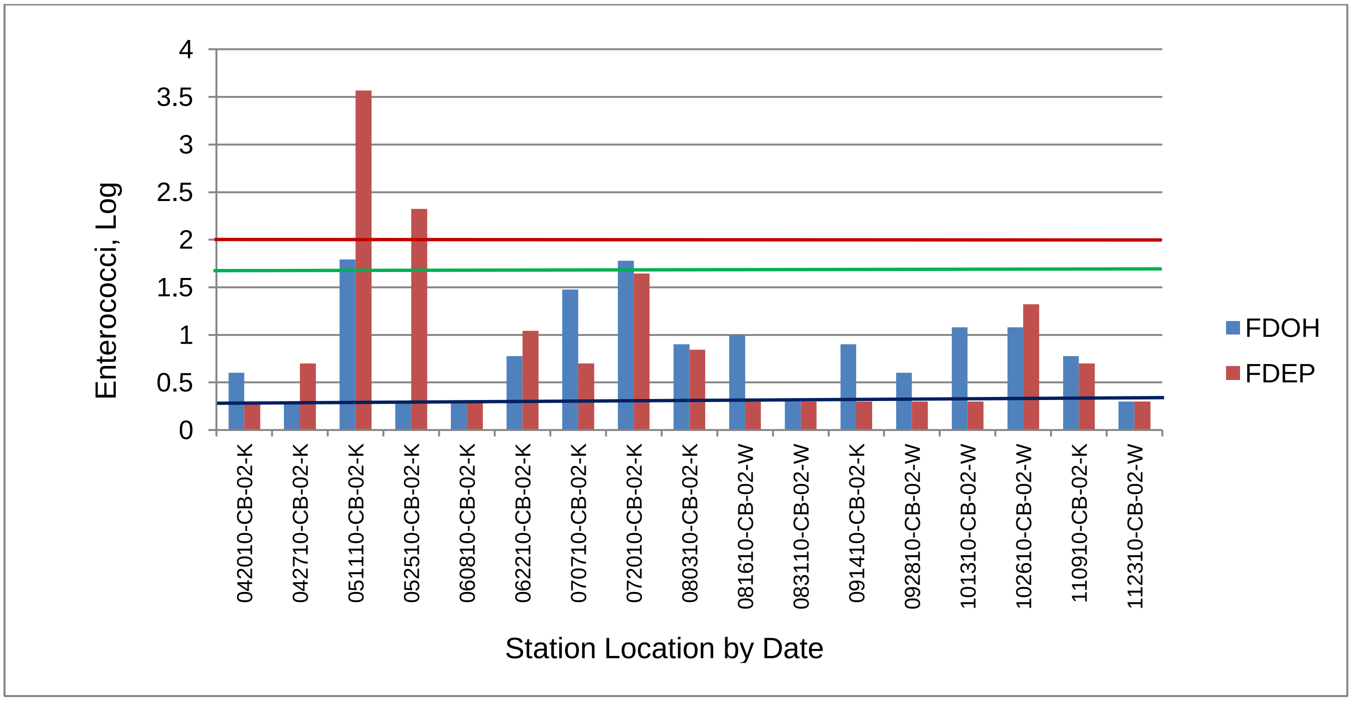


Figure 23: Comparison of enterococci for FDOH data with Carrabelle Beach Station CB-2 for Year 2 for all Depths by date

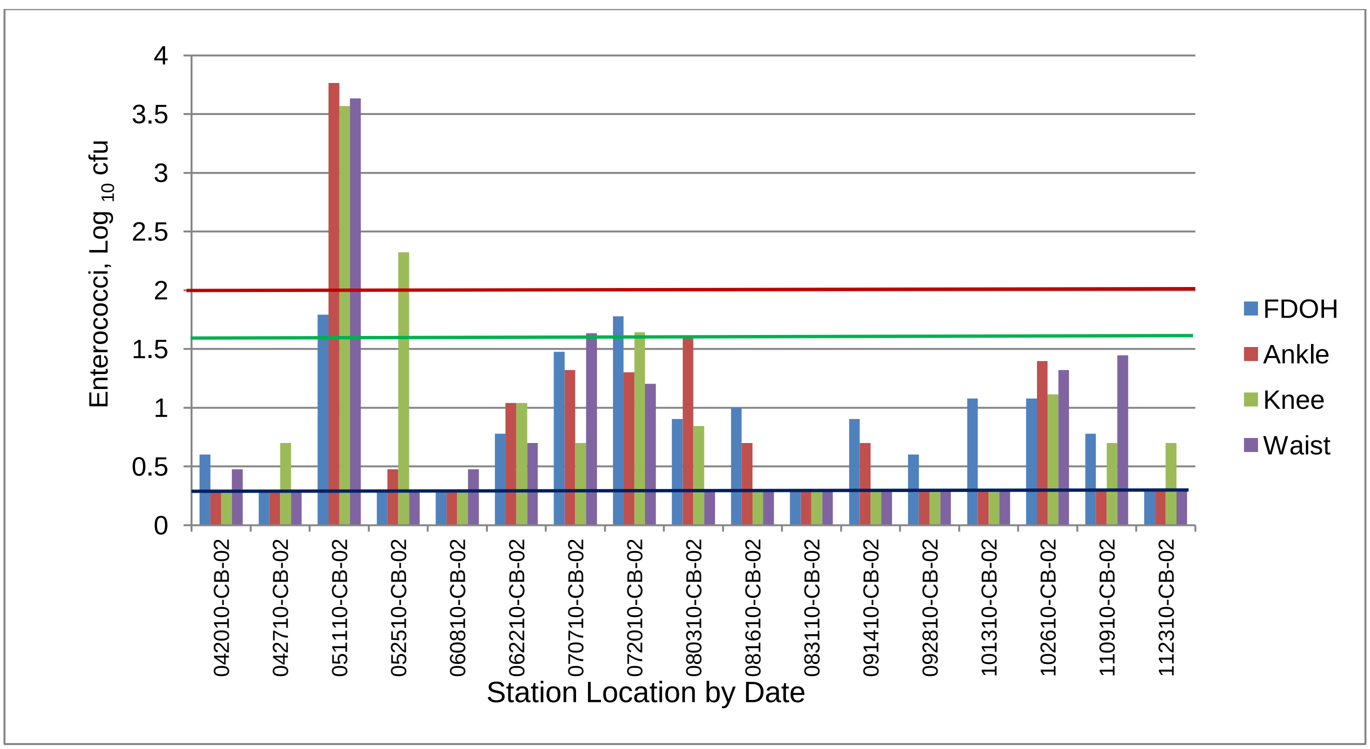


Figure 24: Comparison of enterococci for FDOH data with Mashes Sands Beach Station MS-2 for Year 1 by date for the Knee Depth by date

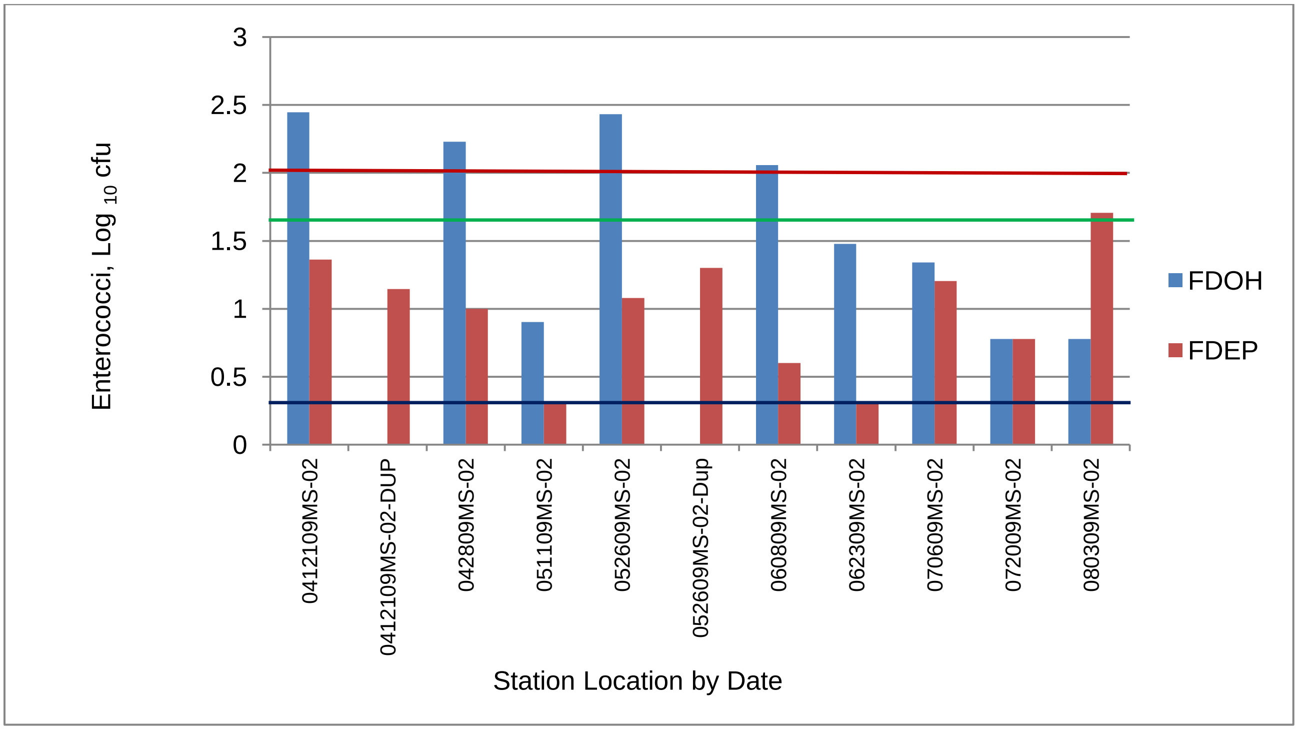


Figure 25: Comparison of enterococci for FDOH data with Mashes Sands Beach Station MS-2 for Year 1 by date for all Depths by date

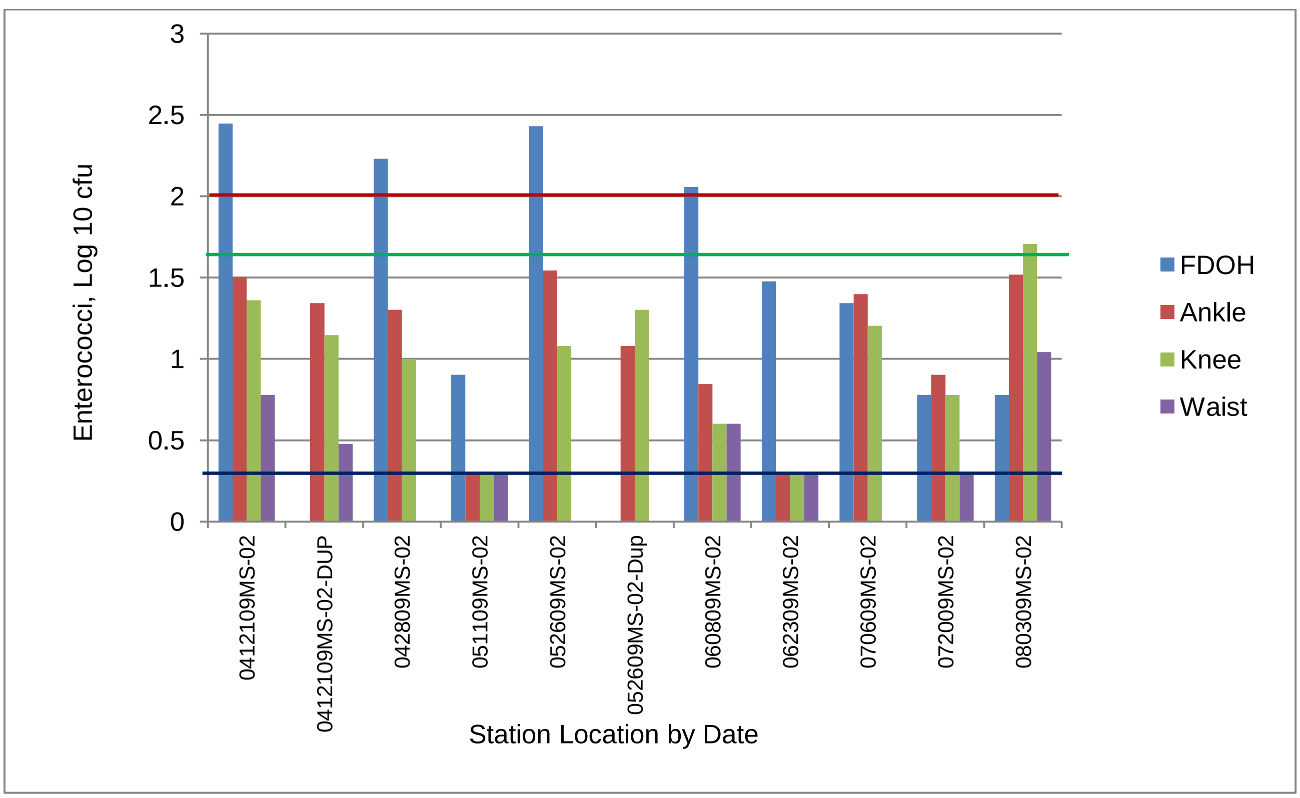


Figure 26: Comparison of enterococci for FDOH data with Mashes Sands Beach Station MS-2 for Year 2 by date for the Knee Depth by date

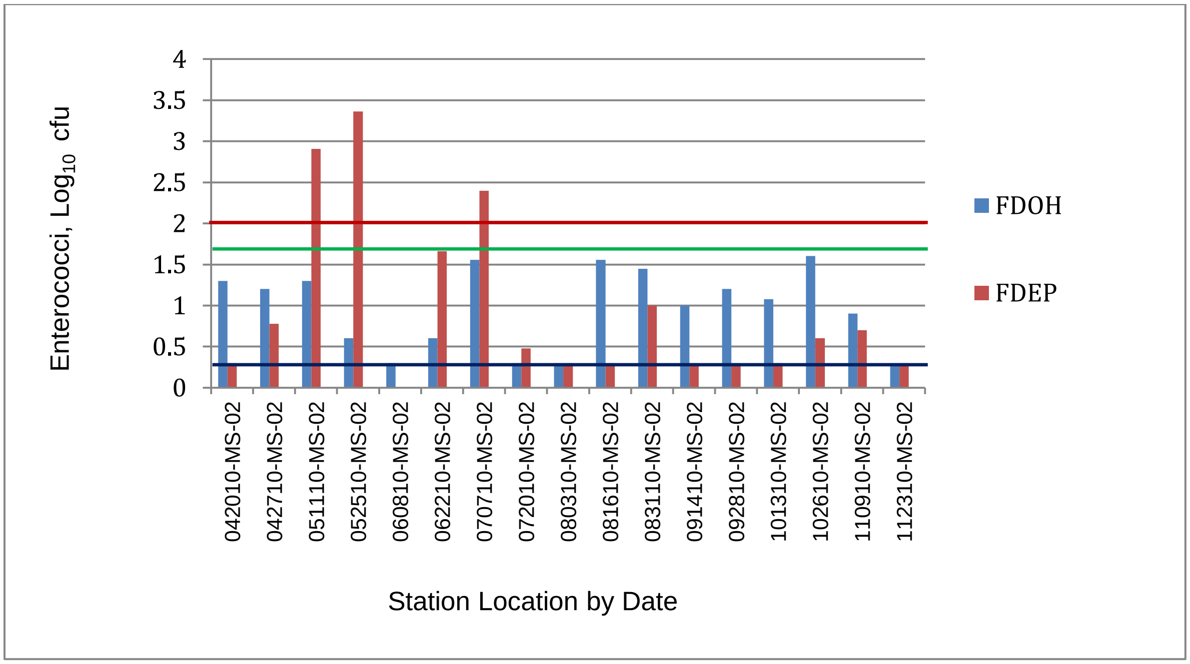
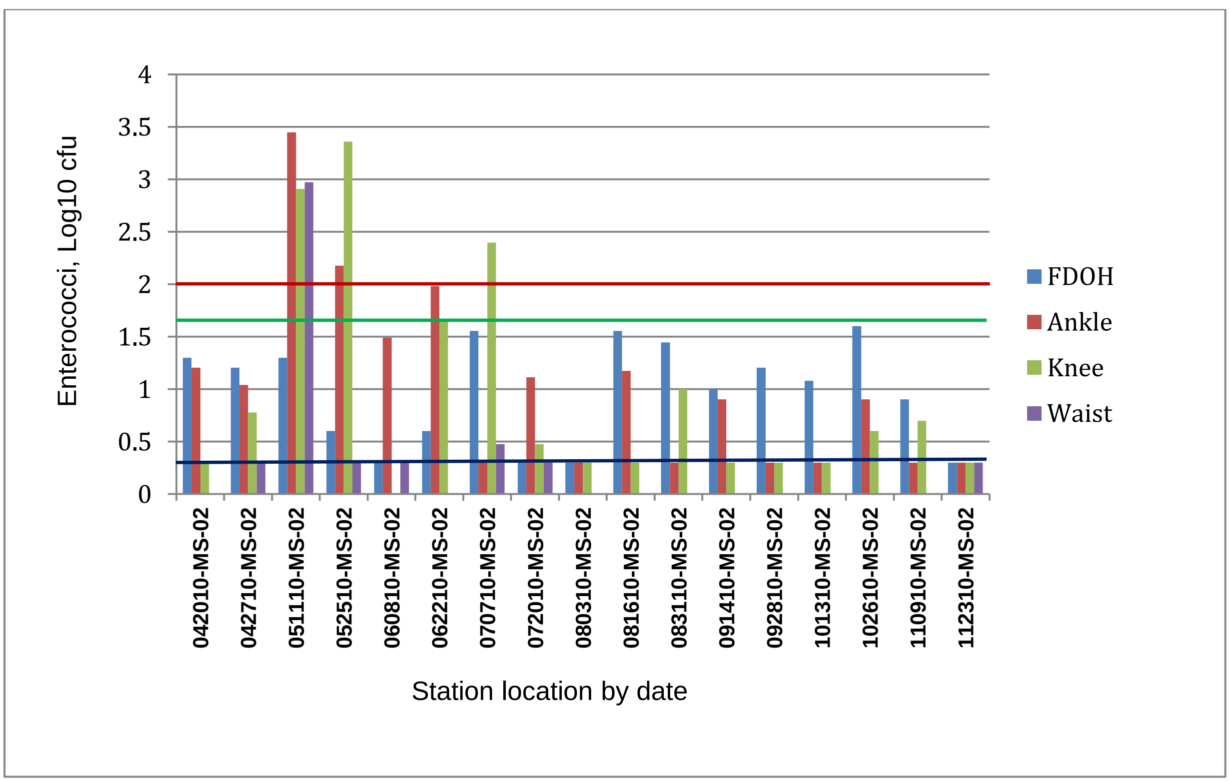


Figure 27: Comparison of enterococci for FDOH data with Mashes Sands Beach Station MS-2 for Year 2 for all Depths by date



VI. Summary and Conclusions

In 2009, the TMDL Section of the FDEP Bureau of Water Resources raised concerns that selected WBIDs were identified as impaired based largely on FDOH data for the included beach and that data, frequently a single point to characterize the entire beach collected on a weekly basis, overestimated the nature of impairment.

In response to this concern, the FDEP Biology Section, in conjunction with the FDOH's Healthy Beaches Program, initiated this study to answer the following question.

> Do the data from the single sampling location for a beach adequately characterize the water quality at that beach considering both spatial coverage and depth of sampling (ankle, knee, and waist)?

During the two year study, two beaches in north Florida, Carrabelle Beach (Franklin County) and Mashes Sands Beach (Wakulla County) were used to investigate FDEP's concern. Bi-weekly data were collected from the two beaches at four locations and at two depths (ankle and knee) and infrequently at a third depth (waist). Sampling at knee depth was done concurrently with FDOH at the designated Healthy Beaches Monitoring location at each beach. Environmental data were collected at knee depth for all stations.

Data from this study indicate that each year presented a different bacterial profile over the sampling year with greater activity in year 2. The cause for this remains undetermined particularly since the environmental data for these years were similar and no anomalous environmental events occurred in Year 2.

Data for depth of sample indicated that bacterial densities generally decreased with increasing water depth but suggesting that the source of bacteria is land based and that dilution affects the resulting bacterial densities reported. The statistical evaluation of the depth of sample, with data composited by beach by depth, indicated that depth of samples was mutually correlated indicating that the distances between the depths of samples along a transect may have been too close in proximity to discriminate independence. Data sets were insufficient to statistically evaluate correlations between individual depths.

Data for spatial differences in station location indicated that statistically significant correlations between stations varied. Correlations noted were hypothesized based on the proximity of the two stations being compared and the external influence of one of the stations (river, bay or marsh) upon the other. An evaluation of the exceedances of bacterial densities at the Poor level of beach water quality appeared to support the interpretation the spatial relationships.

For the comparison of the concurrent FDOH and FDEP data, two comparisons were reviewed. First, applying the FDOH beach classification criteria categories of Good, Moderate and Poor, the comparison between concurrent FDOH and FDEP data at the FDOH designated sampling location for the two years combined showed a high degree of agreement (96%) for fecal coliform at both Carrabelle Beach and Mashes Sands Beach. Data for enterococci indicated 65%

agreement for Carrabelle Beach and 59% agreement for Mashes Sands Beach. Discrepancies occurred in which the two agencies differed sufficiently to affect the determination of impairment (i.e., Good reclassified to Poor). Second, the data for all depths (ankle, knee, waist) at the FDEP Station 2 for each beach by parameter were graphed against the FDOH data to be able to visually compare classification agreement. The additional data did not change any of the earlier interpretations of agreement or non-agreement.

In conclusion, variability both temporally and spatially occurred in the bacterial densities for both fecal coliform and enterococci independent of the beach being studied. Variability between concurrent samples taken at the same location, specifically the designated FDOH monitoring location, was also shown. Bacterial densities were shown to generally decrease with increasing depth of sample that strongly suggests that shifting a sampling point from knee depth to waist depth will decrease the potential for identifying Poor beach water quality and reducing the determination of impairment. Although source(s) of the elevated bacterial densities in the shallow waters were not identified and therefore the potential impact on the public health of those recreating in these waters was not determined, these data underscore the need for additional study in this area of the beach since the most sensitive segments of the population, e.g., the elderly and the very young, are known to frequent these shallow waters.

Ultimately, whether the single sample taken at the beach monitoring location at a single depth is sufficient to characterize the water quality of the beach, the answer must be no.

VII. References

Anderson, K. L., J. E. Whitlock, and V. J. Harwood. 2005. Persistence and Differential Survival of Fecal Indicator Bacteria in Subtropical Waters and Sediments. Appl. & Env. Micro. 71(6): 3041-3048.

Bonilla, T., K. Nowosielski, M.Cuvelier, A. Hartz, M. Green, N. Esibou, D. S. McCorquodale, J.M. Fleisher, A. Rogerson. 2007 Prevalence and distribution of fecal indicator organisms in South Florida beach sand and preliminary assessment of health effects associated with beach sand exposure. Marine Pollution Bulletin doi:10.10016/jamrpolbul 2007.04.016.

Colford, J.M. Jr, T. J. Wade, K.C. Schiff, et al. 2005. Recreational Water Contact and Illness in Mission Bay, California, Westminister, CA: Southern California Coastal Water Research Project.

Cuvelier, M. L., K. L. Nowosielski, N. Esiobu, D. McCorquidale, A. Echeverry, R. Mohammed, and A. Rogerson. 2004. Poster. The Implication of High Numbers of Fecal Indicator Organisms in Sub-Tropical Beach Sands.

Frick, W.E., Z. Ge, and R.G. Zepp. 2008. Nowcasting and Forecasting Concentrations of Biological Contaminants at Beaches: A Feasibility and Case Study. Envir.Sci.Technol. 42:13, pp 4818-4824.

Hartz, A., M. Cuvelier, K. Nowosielski, T.D. Bonilla, M. Green, N. Esiobu, D.S. McCorquodale, and A. Rogerson. 2008. Survival Potential of *Escherichia coli* and enterococci in Subtropical Beach Sand: Implications for Water Quality Managers. J. Env. Quality. 37:898-905.

Hardina, C.M., R.S. Fujioka. 1991. The Environmental Source of Escherichia coli and enterococci in Hawaii's Streams. Env. Tox and Water Qual. 6:185-189.

Ishii, S. and M.J. Sadowsky. 2008. Minireview: Escherichia coli in the Environment: Implications for Water Quality and Human Health. Microbes Environ. 23(2):101-108.

Harwood, Valeria. 2005. "Microbial Source Tracking: Tools for Refining Total Maximum Daily Load Assessments" (September 30, 2005) Report

Harwood, 2007. Investigation of the Sources of Fecal Indicator Bacteria in Wakulla County, Florida (July 20, 2007). Report. (Harwood sampling station at Mashes Sands Beach: Lat N 29° 58.174′/Long W 84° 20.517′).

Solo-Gabriele, H.M., M.A. Wolfert, T.R. Desmaris, and C. L. Palmer. 2000. Sources of Escherichia coli in a Coastal and Subtropical Environment. Appl. & Env. Micro. 66(1):230-237.

Townsend, Jason. 2006. American Society of Microbiology. "Contributions of Sediment to High Enterococcci Counts along the Northern Gulf of Mexico". (Poster Q483)

USEPA, 2008. Great Lakes Beach Sanitary Survey User Manual. EPA-823-B-06-001.

Whitman, R. L. and M.B. Nevers. 2003. Foreshore Sand as a Source of *Escherichia col*i in Nearshore Water of a Lake Michigan Beach. Appl. & Env. Micro. 69(9):5555-5562.

Appendices

Appendix A: Pictures –Carrabelle Beach

The Pavilion at Carrabelle Beach Landward of Station CB-2

West view from Station CB-2 towards CB-1

East view from Station CB-2 towards CB-3

West view from Station CB-4

Landward view of CB-5 looking towards Gulf Beach Road

Station CB-6

Station CB-07

Station CB-7 at water's edge

Appendix B: Pictures – Mashes Sands

Landward of Station MS-1

Mashes Sands Beach at the end of the Road MS-2 off to the right of the picture.

Landward marker of Station MS-2 during Year 1 of the study; Replaced in Year 2 With a permanent structure

Landward Marker for Station MS-3

Landward marker for Station MS-4

Looking east from Station MS-4

Appendix C: FDEP Rule 62-303 - Selected Excerpts

CHAPTER 62-303 (Selected Excerpts)

IDENTIFICATION OF IMPAIRED SURFACE WATERS

62-303.100 Scope and Intent.

(1) This chapter establishes a methodology to identify surface waters of the state that will be included on the state's planning list of waters that will be assessed pursuant to subsections 403.067(2) and (3), Florida Statutes (F.S.). It also establishes a methodology to identify impaired waters based on representative data that will be included on the state's verified list of impaired waters, for which the Department will calculate Total Maximum Daily Loads (TMDLs), pursuant to subsection 403.067(4), F.S., and which will be submitted to the United States Environmental Protection Agency (EPA) pursuant to paragraph 303(d)(1) of the Clean Water Act (CWA).

(2) Subsection 303(d) of the CWA and Section 403.067, F.S., describe impaired waters as those not meeting applicable water quality standards, which is a broad term that includes designated uses, water quality criteria, the Florida antidegradation policy, and moderating provisions. However, as recognized when the water quality standards were adopted, many water bodies naturally do not meet one or more established water quality criteria at all times, even though they meet their designated use. It is not the intent of this chapter to include waters that do not meet otherwise applicable water quality criteria solely due to natural conditions or physical alterations of the water body not related to pollutants. Similarly, it is not the intent of this chapter to include waters where designated uses are being met and where water quality criteria exceedances are limited to those parameters for which permitted mixing zones or other moderating provisions (such as site-specific alternative criteria) are in effect. Waters that do not meet otherwise applicable water quality standards due to natural conditions or to pollution not related to pollutants shall be noted in the state's water quality assessment prepared under subsection 305(b) of the CWA [305(b) Report].

(3) This chapter is intended to evaluate attainment of water quality standards as set forth in Chapter 62-302, F.A.C., for the purposes of identifying water bodies or segments for which TMDLs will be established. It is the intent of this chapter to establish requirements that would apply solely for purposes of assessment and listing under CWA sections 303(d) and 305(b). However, it is not the intent of this chapter to establish requirements for other purposes under Florida law. In cases where this chapter relies on numeric indicators of ambient water quality as part of the methodology for determining whether existing narrative criteria are being met, these numeric values are intended to be used only in the context of developing a planning list and identifying an impaired water pursuant to this chapter. As such, exceedances of these numeric values shall not, by themselves, constitute violations of Department rules that would warrant enforcement action.

(4) Nothing in this rule is intended to limit any actions by federal, state, or local agencies, affected persons, or citizens pursuant to other rules or regulations.

(5) Pursuant to Section 403.067, F.S., impaired waters shall not be listed on the verified list if reasonable assurance is provided that, as a result of existing or proposed technology-based effluent limitations and other pollution control programs under local, state, or federal authority, they will attain water quality standards in the future and reasonable progress towards attainment of water quality standards will be made by the time the next 303(d) list is scheduled to be submitted to EPA.

Specific Authority 403.061, 403.067 FS. Law Implemented 403.021(11), 403.062, 403.067 FS. History–New 6-10-02, Amended 12-11-06.

62-303.360 Primary Contact and Recreation Use Support.

(1) A Class I, II, or III water shall be placed on the planning list for evaluating primary contact and recreation use support if:

(a) There is a sufficient number of samples from the water segment that do not meet the applicable water quality criteria for bacteriological quality based on the methodology described in Rule 62-303.320, F.A.C., with the exception that paragraph 62-303.320(4)(a), F.A.C., does not apply and samples collected on different days within any four day period will be assessed as daily samples, or

(b) The water segment includes a bathing area that was closed by a local health Department or county government for more than one week or more than once during a calendar year based on bacteriological data, or

(c) The water segment includes a bathing area for which a local health Department or county government has issued closures, advisories, or warnings totaling 21 days or more during a calendar year based on bacteriological data, or

(d) The water segment includes a bathing area that was closed or had advisories or warnings for more than 12 weeks during a calendar year based on previous bacteriological data or on derived relationships between bacteria levels and rainfall or flow, or

(e) The water segment includes a sampling location that has one or more monthly geometric mean values above the monthly geometric mean fecal coliform or enterococci criterion during the planning period. To calculate a monthly geometric mean, there shall be at least ten samples collected within that month, with at least one sample from each full week of the month, or

(2) When evaluating a water segment for bacteriological quality under paragraph (1)(a), the criterion used for fecal coliforms shall be that the Most Probable Number (MPN) or Membrane Filter (MF) shall not exceed 400 counts per 100 ml.

(3) For data collected after August 1, 2000, the Florida Department of Health (DOH) database shall be the primary source of data used for determining bathing area closures.

(4) Advisories, warnings, and closures based on red tides, rip tides, sharks, hurricanes, or short-term releases of pollutants, such as sewage spills that have been repaired and medical wastes, shall not be included when assessing recreation use support. However, the Department shall note for the record that data were excluded and explain why they were excluded.

62-303.460 Primary Contact and Recreation Use Support.

(1) The Department shall review the data used by the DOH as the basis for bathing area closures, advisories or warnings and verify that the values exceeded the applicable DOH thresholds and the data meet the requirements of Chapter 62-160, F.A.C. If the segment is listed on the planning list based on bathing area closures, advisories, or warnings issued by a local health department or county government, closures, advisories, or warnings based on red tides, rip tides, sewer line breaks, sharks, medical wastes, hurricanes, or other factors not related to chronic discharges of pollutants shall not be included when verifying primary contact and recreation use support. The Department shall then re-evaluate the remaining data using the methodology in paragraph 62-303.360(1)(c), F.A.C. Water segments that meet the criteria in paragraph 62-303.360(1)(c), F.A.C., shall be included on the verified list.

(2) If the water segment was listed on the planning list due to samples that do not meet water quality criteria for bacteriological quality, the Department shall, to the extent practical, evaluate the source of

bacteriological contamination and shall verify that the impairment is due to chronic discharges of human-induced bacteriological pollutants before listing the water segment on the verified list. The Department shall take into account the proximity of municipal stormwater outfalls, septic tanks, and domestic wastewater facilities when evaluating potential sources of bacteriological pollutants. For water segments that contain municipal stormwater outfalls, the impairment documented for the segment shall be presumed to be due, at least in part, to chronic discharges of bacteriological pollutants. The Department shall then re-evaluate the data using the methodology in subsection 62-303.320(1), F.A.C., excluding any values that are elevated solely due to wildlife, or for enterococci in coastal recreational waters, adjusting the values based on the human health-related risk factors for wildlife-based enterococci upon meeting the relevant requirements of 40 CFR 131.41(c)(2).

(3) Water segments shall be included on the verified list if:

(a) The number of samples that do not meet the applicable single-sample bacteriological water quality criteria meet the requirements in subsection 62-303.420(6), F.A.C., with the exception that paragraph 62-303.320(4)(a), F.A.C., does not apply and samples collected on different days within any four day period will be assessed as daily samples, or

(b) There are one or more exceedances of a bacteriological water quality criterion expressed as a monthly geometric mean during the verified period. To assess the monthly data for a sampling location, there shall be at least ten samples collected within that month, with at least one sample from each full week of the month.

(4) When evaluating a water segment for bacteriological quality under paragraph 62-303.460(3)(a), F.A.C., the criterion used for fecal coliforms shall be that the Most Probable Number (MPN) or Membrane Filter (MF) shall not exceed 400 counts per 100 ml.

(5) When assessing waters pursuant to paragraphs 62-303.460(3)(a) and (b), F.A.C., the Department shall evaluate whether the samples are representative and are not biased toward collecting samples at times either under the influence of wet weather conditions or absent the influence of wet weather conditions. Representative sampling generally reflects a consistent number of samples evenly spaced over regular intervals. Any determinations to exclude waters based on this provision shall be documented, and the documentation shall include the basis for the decision.

Specific Authority 403.061, 403.067 FS. Law Implemented 403.062, 403.067 FS. History–New 6-10-02, Amended 12-11-06, 9-4-07.

Appendix D: Florida Department of Health Rule 64E-9

Florida Department of Health

Rule 64E-9, April 10, 2009

64E-9.013 Bathing Places

(1) General – Approval for the development of a public bathing place and a permit to operate a public

bathing place shall be obtained from the department.

(2) Development – The following shall be submitted to the department for consideration towards the

development of a public bathing place:

(a) Six sets of site plans, prepared by a professional engineer or professional surveyor or mapper which detail the location, contours of the shoreline and bottom, appurtenances such as sanitary facilities, nearby boat docks/mooring facilities, diving or slide facilities, and pertinent details from the sanitary survey. Said professional engineer(s) and surveyors and mappers shall be licensed in the state of Florida under the provision of Chapter 471 or 472, F.S., and shall fulfill the requirements of Section 471.025 or 472.025, F.S.

(b) A sanitary survey identifying potential sources of contamination as exemplified by streams,

unsewered residential areas, water and wastewater treatment plants, sewage outfalls, storm drain outfalls, industrial drainage and waste outfalls, agricultural drainage, sanitary landfills, open dumps, animal enclosures, wildlife populations, and potential high erosion areas. The survey shall include consideration of present or possible future pollution of the bathing water from the above potential sources of contamination and from other forms of pollution including bottom deposits, turbidity of water, decaying vegetation, surface runoff, and the anticipated bather load. The survey shall establish that the bathing water has a flow through of a minimum of 500 gallons per anticipated bather per 24 hours, unless the water surface area of the body of water is two acres or more. The bathing load in lakes shall be based on 100 sq. ft. per bather, and only those portions of the lake within the bathing area shall be considered as the basis of the bathing load. Water currents shall not exceed three feet per second. A written report of the sanitary survey shall be submitted to the department and shall include a presentation and evaluation of the findings and a recommendation relative to the development and permitting of the bathing place.

(c) A bacteriological survey shall be submitted to the department and the fecal coliform, *E. coli,* or

enterococci densities indicated by this survey shall not exceed the standards of subsection 64E-9.013(4), F.A.C. The survey shall consist of a minimum of three bacteriological samples collected from the proposed bathing area daily for the first three days of each week for three consecutive weeks. Either MPN or MF counts may be utilized. Should the MF method results differ significantly from the MPN method results, the MPN results shall prevail. The bacteriological survey results shall be reviewed in light of the sanitary survey.

(d) Fees as per Rule 64E-9.015, F.A.C.

(e) A legal survey of the property by a registered land surveyor shall be provided.

(f) A water clarity measurement by Secchi disk reading in feet using an 8 inch diameter black and white Secchi disk. Where water clarity does not achieve four feet depth for a period of at least 5 days during the period of operation due to natural water color, the department will consider approval of the bathing area with submittal of a satisfactory lifeguard plan, patron notification plan and materials, swim zone depth demarcation for children and beginners, and other special conditions that would apply to the individual site.

(g) A lifeguard and/or safety plan shall be submitted with the application for development of all new bathing areas.

(3) Operation.

(a) The following must be submitted prior to operation:

1. Six operating permit applications, DH 917.

2. Fees as per Rule 64E-9.015, F.A.C.

(b) Operational water quality – The water shall be free of chemical and physical substances known or suspected of being capable of creating toxic reactions or skin or membrane irritations. Algae and aquatic vegetation shall be controlled so that no hazard to bathers results.

(c) Bacteriological samples shall be collected monthly. A set of two samples shall be collected for every 200 fee of shoreline, the samples shall be taken a foot below the surface in three feet of water and at least 25 feet apart. The samples shall be analyzed by a DOH certified laboratory using EPA approved methods for ambient water and the results submitted to the department within 10 days after the end of the month. Should the test results of these samples exceed the standards in 64E-9.013(4) below, the county health department shall be notified within 24 hours of receipt from the lab, and re-sampling by the permit holder shall be required within 24 hours. All sampling results shall be submitted to the county health department. If 24 hour resampling is not possible for any reason, then the bathing place shall be closed immediately to swimming based upon these initial results during the time period waiting for re-sampling results. If the 24 hour confirmation samples reveal an exceedance of the standards, the bathing place shall be closed immediately to swimming until additional testing reveals the water meets single sample standards again. If a pollution source is identified, that source shall be eliminated before reopening the bathing area.

(d) Inspections – county health departments shall perform two inspections per year which shall include:

1. A site inspection in light of the original sanitary survey, changed natural conditions, changed use conditions, and originally permitted facilities.

2. A bacteriological test consisting of the normal monthly sampling requirement. The fecal coliform, *E. coli* or enterococci density must not exceed the single sample standards of subsection 64E-9.013(4), F.A.C.

3. A water clarity test shall be performed wherein an 8'' black and white secchi disk shall be visible to a minimum depth of four feet.

4. The bathing place shall be temporarily closed or swimming prohibited, as appropriate, by the owner or the department if inspection reveals water clarity violations, unsafe bacterial test results, or immediate hazards to health or safety such as, but not limited to sewage in water, broken glass, dangerous wildlife, hazardous structural or electrical conditions, toxic algal blooms, or other serious disease agents present.

5. The bathing load shall be calculated on one bather per 25 square feet of surface area in areas of less than four feet of depth and one bather per 75 square feet of surface area where the water depth exceeds four feet,

(e) Trash and garbage receptacles shall be provided and said trash disposed of at least weekly.

(f) Muck or silt shall not be present from the shoreline to a depth of five feet and aquatic vegetation shall be controlled.

(g) Site specific signage shall be provided. The bathing load shall be posted and due consideration shall be given to safety guidelines such as steep slope, diving areas, deep water, underwater obstruction, dangerous wildlife, or lifeguard not on duty. Additional signage shall be provided if the bathing area is longer than 300 feet.

(h) Restrooms, platforms, diving boards, docks, beaches and walkways shall be kept clean and in good repair. Diving areas shall be readily identified, and shall have adequate water depth for safe diving based on the depth requirements of the FINA standards previously adopted herein. Shallow areas shall not be utilized for diving and shall be so marked.

(i) Glass items and domestic animals are prohibited in the bathing area and on the adjacent beach area.

(j) Sanitary facilities shall be provided and shall be as near to the bathing area as prudent to ensure patron use.

1. Women's restrooms shall have a fixture set including a water closet and a lavatory.

2. Men's restrooms shall have a fixture set including a urinal, a water closet and a lavatory.

3. Additional restroom fixture shall be provided based on stated usage. A second water closet, urinal and lavatory shall be provided in the men's restroom if the stated usage exceeds 50 patrons, but is less than 150 patrons. Another urinal, water closet and lavatory shall be provided for each additional 100 patrons. The number of water closets in the women's restroom shall be based on a three to two ratio with three water closets being provided in the women's restroom for every two fixtures in the men's restroom. For this purpose of establishing the men's restroom fixture count, both water closets and urinals shall be included. The number of lavatories in the women's restroom shall match the number in the men's restroom.

4. Restroom floors shall be impervious, slip resistant and slope to floor drains.

(4) Bacteriological Standards – Either fecal coliform, *E. coli, or* enterococci bacteria shall be tested for, at the option of the permit holder. All samples tested will be considered to determine compliance, unless found to b invalid by the certified lab or county health department. The enterococci density shall not exceed an average of 33 colony forming units (CFU) per 100 mL of water, nor exceed 61 per 100 mL of water in any single sample; or the *E.coli* density shall not exceed an average of 126 CFU per 100 mL of water, nor exceed 235 CFU 800 per 100 mL of water in any single sample; or the fecal coliform shall not exceed an average of 200 CFU per 100 mL of water, nor 400 per 100 mL of water in 10 percent of the samples, nor 800 CFU per 100 mL of water in a single sample. This average shall be expressed as geometric means using at least 5 samples per 30 day period.

Rulemaking Authority 381.006, 514.021 FS. Law Implemented 381.006, 514.021, 514.03, 514.031, 514.04, 514.05, 514.06 FS. History–New 10-5-93, Formerly 10D-5.142, Amended 12-27-98, 5-27-04, 5-24-09.