STORMWATER MANAGEMENT MEASURES
AND FECAL INDICATOR BACTERIA

BY

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THESIS

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A heartfelt thanks goes out to the fine gentlemen of the UNH Stormwater Center: Pedro Avellaneda-Lopez, Joshua Briggs, and James Houle. A great team in any weather.

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# TABLE OF CONTENTS

ACKNOWLEDGEMENTS ........................................................................................................................ iii

TABLE OF CONTENTS ................................................................................................................................ iv

LIST OF TABLES ........................................................................................................................................ vii

LIST OF FIGURES .................................................................................................................................... viii

ABSTRACT ............................................................................................................................................... ix

CHAPTER 1: INTRODUCTION AND PURPOSE OF STUDY ................................................................. 1

  Introduction .............................................................................................................................................. 1
  Regulatory Limits for Indicator Bacteria ............................................................................................... 2
  Indicator Bacteria in the Environment ................................................................................................. 3
  Indicator Bacteria and Stormwater Management Measures .............................................................. 6
  Purpose of Current Study ....................................................................................................................... 9

CHAPTER II: STUDY AREA AND STORMWATER MANAGEMENT MEASURES ....................................... 11

  Research Facility .................................................................................................................................. 11
  Influent Characterization ....................................................................................................................... 12
  Stormwater Management Measures ...................................................................................................... 13
    Device Sizing ....................................................................................................................................... 13
    Wet Detention Pond (WDP) .................................................................................................................... 14
    Rock-Lined Swale (RLS) ......................................................................................................................... 14
    Surface Sand Filter (SF) ....................................................................................................................... 15
    Bioretention Area (BA) .......................................................................................................................... 15
    Subsurface Gravel Wetland (GW) ........................................................................................................... 15
    Stormwater Filtration Chamber (SFC) ................................................................................................... 16
LIST OF TABLES

Table 1. Survey of reported values for fecal indicator bacteria........................................5
Table 2. Meteorological Data for Sampled Rainfall-Runoff Events ...............................28
Table 3. Percentage of rainfall-runoff events that devices exported bacteria (effluent >
influent) and significance level at which device effluent can be distinguished from
influent. ........................................................................................................................... 35
Table 4. Comparison of dry weather and storm counts of ENT and EC. .......................43
Table 5. ENT concentrations in device sediments.........................................................46
LIST OF FIGURES

Figure 1. Bacterial counts at the influent during the sampling period (9/28/2004 to 10/10/2005).......................................................................................................................... 31

Figure 2. Variability of storm event ENT counts by device............................................. 35

Figure 3. Non-exceedance Probability Plots For ENT concentrations in Device Influent and Effluent......................................................................................................................... 38

Figure 4. Variability of dry weather ENT counts by device............................................. 40
ABSTRACT

This study was undertaken to address concerns that current stormwater management practices could be degrading water quality by inadvertently providing habitat, nutrients, and transport mechanisms for fecal indicator bacteria. Concentrations of *Escherichia coli* (EC) and *Enterococcus* (ENT) were evaluated in the influent, effluent, and retained water of 11 stormwater management devices. All devices received equal portions of influent from a single stormwater outfall that handled runoff from a 3.55-hectare asphalt parking lot. During storm events, influent and effluent water samples were obtained from flow-weighted composite samples, thereby representing the microbial event mean concentration. Retained water samples were obtained from grab samples taken during dry weather periods between rainfall events. Bacteria were enumerated using membrane-filtration techniques.

EC concentrations were below the EPA’s primary contact recreational water standard of 235 cfu/100mL in 98% of all samples collected in the influent and effluent. Concentrations in the influent exceeded the equivalent ENT standard of 104 cfu/100mL in 67% of the events, with a median concentration of 560 cfu/100mL. Effluent concentrations of ENT exceeded the limit in at least 25% of events for all devices. Slight statistical differences (p<0.15) were detected between ENT concentrations in the influent and three devices. The rock-lined swale had effluent ENT concentrations that were regularly higher than the influent concentrations (median concentration of 3520 cfu/100mL) and the gravel wetland and infiltration drainfield had effluent ENT
concentrations that were regularly lower than the influent concentrations (median concentrations of 44 and 54 cfu/100mL, respectively). None of the other devices were found to have effluent concentrations of ENT that were significantly different from the influent concentrations above the 85% confidence level. These results suggest that some stormwater management measures are increasing bacterial concentrations in stormwater runoff. Further research is necessary to determine mechanisms by which this apparent loading is occurring.
CHAPTER 1: INTRODUCTION AND PURPOSE OF STUDY

Introduction

Fecal indicator bacteria, specifically *Escherichia coli* (EC) and bacteria of the genus *Enterococcus* (ENT), were recommended by the United States Environmental Protection Agency (EPA) to identify potential contamination of waterbodies by pathogenic organisms (EPA, 1986). The successful use of indicator species is dependent on those species being “associated with sources of human pathogens, equally or more resistant to disinfection than are pathogens, unable to grow in aquatic environments, applicable to all types of water, and remain quantifiable after infectious levels of pathogens have disappeared” (Cude, 2005). In accordance with EPA’s 1986 recommendation, community and state agencies have been using these indicators to evaluate waterbodies in their jurisdictions. Protection of primary contact users of waterbodies is the dominant public health goal addressed by monitoring for microbial pollutants. A recurring concern about using EC and ENT as surrogates for all water-borne pathogens is the imperfect epidemiological relationship of EC and ENT to gastrointestinal illness among swimmers (Calderon, 1991; Field, 1993; EPA, 2003). In 2000, the EPA published a summary of research conducted since 1986 that further examined the relationship of indicator bacteria to gastrointestinal illness in swimmers (EPA, 2000). Although some studies cited in the EPA review did not support the continued use of these bacterial indicators, the bulk of the studies examined suggested
that the policy was sound. Other research examining the persistence of waterborne pathogens, such as *Vibrio cholerae* (Hood, 1982), *Cryptosporidium parvum* and *Clostridium perfringens* (Medema, 1997), and *Salmonella* (Evison, 1988) and (Pommepuy, 1992), has found that EC and ENT were sometimes less long-lived in the environment than the potentially pathogenic species they were representing. However, until such time as better indicators can be identified, EC and ENT will continue to be recommended as bacterial indicators for contamination of surface waters and were examined in the current study.

**Regulatory Limits for Indicator Bacteria**

For non-point source discharges, regulatory guidance for ENT and EC are predicated on the use of the receiving waterbody for recreation or shellfishing. Federal guidance for freshwater recreation waters uses a sliding scale of 61 to 151 cfu/100mL for ENT and 126 to 575 cfu/100mL for EC, with waters designated for higher levels of use receiving the lower limits. Because EC is not recommended for marine waters, commonly prescribed guidance for marine waters is limited to ENT, with a value of 104 cfu/100mL for a “designated bathing beach” (EPA, 2005). For ENT, the State of New Hampshire posts beach advisories when a water sample at “a marine beach exceeds the state standard of 104 counts of ENT per 100 mL of water, or ENT levels exceed the geometric mean of 35 counts of ENT per 100 mL of water in at least three samples collected over a 60-day period” (NHDES, 2005). For EC, the corresponding limits at swimming beaches are 88 cfu/100mL for single sample counts and 47 cfu/100mL for a 3-day average count. For other recreational waters, these limits are raised to 406 cfu/100mL
for single samples and 126 cfu/100mL for 3-day average (NHDES, 2005). A pilot study conducted previously at this site examined EC and ENT at the location that became the influent monitoring point for the research facility. In that study, 2 out of 9 storm samples exceeded the recreational water standard for ENT and 1 out of 8 storm samples exceeded the recreational water standard for EC (Eckman, 2000). With the exception of a single high count of EC (1030 cfu/100mL) in the previous study, the remainder of EC samples were well below the recreational water standard and similar to the counts observed during the present study.

National Pollutant Discharge Elimination System (NPDES) permits regulate ENT discharges in municipal wastewater treatment plant (WWTP) effluent. As a potential source of bacterial contamination to recreational waters, WWTP permits may use the geometric mean limits of 33 cfu/100mL for discharges to fresh waters and 35 cfu/100mL for discharges to marine waters (EPA, 2004).

**Indicator Bacteria in the Environment**

Since the EPA promulgated the indicator bacteria rules, numerous agencies and municipalities responsible for protecting public health in recreational waterbodies have been required to issue advisories or warnings following rainfall events (EPA, 2005; Trowbridge, 2006). In attempts to address bacterial water quality violations, initial hypotheses for explaining elevated levels of indicator bacteria pointed to combined-sewer overflows (CSOs), leaking septic systems, illicit discharges, and pet waste (Lijklema, 1987; Jones, 1999). While major efforts have been made to control the obvious sources of pollution, it has become apparent that the problem is more complex than simple point-
source pollution models predict. Attention has now turned to the indicator organisms themselves and their population dynamics in the environment. The result is an expanding body of literature demonstrating the ability of EC and ENT to persist and/or multiply in the environment. Table 1 presents a summary of concentrations of indicator bacteria reported in the literature. With the exception of researchers who examined sites impacted by CSO outfalls, much of the literature provides evidence of persistent bacterial concentrations unrelated to storm events or point-source discharges. If concentrations of indicator bacteria do not correlate with contamination by recent pollution events because the bacteria are reproducing in the environment, then it may be difficult to justify their continued use as a water quality metric. Further epidemiological research may be warranted to determine if the presence of such bacteria is truly indicative of potential health hazard.
Table 1. Survey of reported values for fecal indicator bacteria

<table>
<thead>
<tr>
<th>Reference</th>
<th>Indicator</th>
<th>Location</th>
<th>Concentrations</th>
</tr>
</thead>
<tbody>
<tr>
<td>An, 2002</td>
<td>EC</td>
<td>Near-shore lake water, Texas/Oklahoma border</td>
<td>&lt;1 to 179 MPN/100mL</td>
</tr>
<tr>
<td>An, 2002</td>
<td>EC</td>
<td>Lake bottom sediment, Texas/Oklahoma border</td>
<td>3.5x10⁴ to 5x10⁵ cfu/g</td>
</tr>
<tr>
<td>Anderson, 1997</td>
<td>ENT</td>
<td>Leaf litter, New Zealand</td>
<td>Up to 3.3x10² cfu/g</td>
</tr>
<tr>
<td>Characklis, 2005</td>
<td>EC</td>
<td>Creek water (storm event), NC</td>
<td>Up to 8.7x10⁴</td>
</tr>
<tr>
<td>Characklis, 2005</td>
<td>ENT</td>
<td>Creek water (storm event), NC</td>
<td>Up to 4.4x10⁴</td>
</tr>
<tr>
<td>Corbett, 1993</td>
<td>FC</td>
<td>Beach water samples, Sydney, Australia</td>
<td>Up to 2.6x10⁴ cfu/100mL</td>
</tr>
<tr>
<td>Corbett, 1993</td>
<td>FS</td>
<td>Beach water samples, Sydney, Australia</td>
<td>Up to 1.4x10⁴ cfu/100mL</td>
</tr>
<tr>
<td>Davies, 2000</td>
<td>ENT</td>
<td>Suburban runoff, Sydney, Australia</td>
<td>76 to 8.5x10⁴ cfu/100mL</td>
</tr>
<tr>
<td>Davies, 2000</td>
<td>ENT</td>
<td>Suburban stormwater wetland outflow, Sydney, Australia</td>
<td>8 to 2.4x10⁴ cfu/100mL</td>
</tr>
<tr>
<td>Davies, 1995</td>
<td>FS</td>
<td>Marine and freshwater sediments, Australia</td>
<td>18 to 10³ cfu/g</td>
</tr>
<tr>
<td>Desmarais, 2002</td>
<td>EC</td>
<td>Tidal river sediment, Florida</td>
<td>30 to 1.6x10⁴ MPN/g</td>
</tr>
<tr>
<td>Desmarais, 2002</td>
<td>ENT</td>
<td>Tidal river sediment, Florida</td>
<td>Up to 1.2x10⁴ MPN/g</td>
</tr>
<tr>
<td>Ellis, 1995</td>
<td>FS</td>
<td>CSO runoff, urban catchment, London</td>
<td>40 to 4.5x10³ MPN/100mL</td>
</tr>
<tr>
<td>Ellis, 1995</td>
<td>FC</td>
<td>CSO runoff, urban catchment, London</td>
<td>8.0x10² to 8.1x10⁵ MPN/100mL</td>
</tr>
<tr>
<td>Ellis, 1995</td>
<td>FS</td>
<td>CSO sediment, urban catchment, London</td>
<td>1.4x10³ MPN/g</td>
</tr>
<tr>
<td>Ellis, 1995</td>
<td>FC</td>
<td>CSO sediment, urban catchment, London</td>
<td>2.8x10³ MPN/g</td>
</tr>
<tr>
<td>Fujioka, 2001</td>
<td>EC</td>
<td>Urban stream, Hawaii</td>
<td>27 to 10³ cfu/100mL</td>
</tr>
<tr>
<td>Fujioka, 2001</td>
<td>ENT</td>
<td>Urban stream, Hawaii</td>
<td>Up to 3.4x10³ cfu/100mL</td>
</tr>
<tr>
<td>Grant, 2001</td>
<td>ENT</td>
<td>Tidal marsh waters, Santa Barbara, CA</td>
<td>&lt;10 to 2142 MPN/100mL</td>
</tr>
<tr>
<td>Grant, 2001</td>
<td>ENT</td>
<td>Tidal marsh sediments, Santa Barbara, CA</td>
<td>&lt;10 to 5 x10³ MPN/100g</td>
</tr>
<tr>
<td>Grant, 2001</td>
<td>ENT</td>
<td>Storm drain dry weather flow, Santa Barbara, CA</td>
<td>23 to 3.5x10³ MPN/100mL</td>
</tr>
</tbody>
</table>
Table 1. (cont.) Survey of reported values for fecal indicator bacteria

<table>
<thead>
<tr>
<th>Reference</th>
<th>Indicator</th>
<th>Location</th>
<th>Concentrations</th>
</tr>
</thead>
<tbody>
<tr>
<td>Jones, 1996</td>
<td>EC</td>
<td>Commercial parking lot runoff, New Hampshire</td>
<td>&lt;1 to 4.8x10⁴ cfu/100mL</td>
</tr>
<tr>
<td>Kinzelman, 2003</td>
<td>EC</td>
<td>Lake Michigan recreational waters</td>
<td>&lt;1 to 2.4x10⁴ MPN/100mL</td>
</tr>
<tr>
<td>Kinzelman, 2003</td>
<td>ENT</td>
<td>Lake Michigan recreational waters</td>
<td>&lt;1 to 5.5x10³ MPN/100mL</td>
</tr>
<tr>
<td>Lijklema, 1987</td>
<td>FS</td>
<td>Detention pond receiving CSO inputs, Netherlands</td>
<td>0 to 10⁶ FS/100mL</td>
</tr>
<tr>
<td>Lijklema, 1987</td>
<td>EC</td>
<td>Detention pond receiving CSO inputs, Netherlands</td>
<td>0 to 10⁶ FS/100mL</td>
</tr>
<tr>
<td>Martin, 2005</td>
<td>FC</td>
<td>Beach wrack vegetation, San Diego, CA</td>
<td>Up to 1.4 x10⁵ MPN/g</td>
</tr>
<tr>
<td>Martin, 2005</td>
<td>ENT</td>
<td>Beach wrack vegetation, San Diego, CA</td>
<td>Up to 4 x10⁴ MPN/g</td>
</tr>
<tr>
<td>Muirhead, 2004</td>
<td>EC</td>
<td>Stream in agricultural area, New Zealand</td>
<td>10² to 10⁶ cfu/100mL</td>
</tr>
<tr>
<td>Reeves, 2004</td>
<td>ENT</td>
<td>Urban dry weather runoff, Orange County, CA</td>
<td>1.8x10³ MPN/100mL</td>
</tr>
<tr>
<td>Reeves, 2004</td>
<td>ENT</td>
<td>Urban storm drain sediments, Orange County, CA</td>
<td>3x10³ MPN/100g</td>
</tr>
<tr>
<td>Solo-Gabriele, 2000</td>
<td>EC</td>
<td>Urban tidal river, Ft. Lauderdale, Florida</td>
<td>Up to 3.6x10³ MPN/100mL</td>
</tr>
<tr>
<td>Solo-Gabriele, 2000</td>
<td>EC</td>
<td>Urban tidal river sediments, Ft. Lauderdale, Florida</td>
<td>Up to 2.0x10² MPN/g</td>
</tr>
<tr>
<td>Whitman, 2003</td>
<td>EC</td>
<td>Algal mats at freshwater beaches, Lake Michigan</td>
<td>Up to 1.6 x10⁶ cfu/g</td>
</tr>
<tr>
<td>Whitman, 2003</td>
<td>ENT</td>
<td>Algal mats at freshwater beaches, Lake Michigan</td>
<td>Up to 10⁶ cfu/g</td>
</tr>
<tr>
<td>Zhang, 2005</td>
<td>EC</td>
<td>Urban structural BMP sump water, Providence, RI</td>
<td>Up to 3.5 x10⁷ cfu/100mL</td>
</tr>
<tr>
<td>Zhang, 2005</td>
<td>ENT</td>
<td>Urban structural BMP sump water, Providence, RI</td>
<td>Up to 2.9 x10⁷ cfu/100mL</td>
</tr>
</tbody>
</table>

**Indicator Bacteria and Stormwater Management Measures**

Published values of ENT concentrations in stormwater management measures are currently uncommon. In part, this is because ENT were enumerated as a group with other fecal streptococci (FS) before being reclassified in 1984 (Committee on Indicators for
Waterborne Pathogens, 2004) and many reports even after 1984 have published values of FS without separately quantifying ENT. In dry-weather flows in a stormwater drainage system Grant (2001) reported ENT values of 23 to 3.5x10^3 MPN/100mL. In a study conducted at commercial parking lots in NH, Jones and Langan reported EC values in runoff entering stormwater management measures from <1 to 4.8x10^4 cfu/100mL, with a median value of 40 cfu/100mL (Jones, 1996). Zhang et al (2005) reported ENT concentrations of up to 2.9 x10^4 cfu/100mL in the sump of an urban BMP.

For many stormwater treatment devices, the mechanism for pollutant removal relies primarily on the capture of gross particles through settling, owing to the primary stormwater pollutant concern traditionally being sediment. Increasingly, other chemical or physical methods are being employed to treat stormwater, such as filtration. As a result, stormwater devices often contain a substantial amount of organic debris and sediment that is kept submerged or is repeatedly wetted by runoff events. As shown by several researchers (Lijklema, 1987; Marino, 1991; Sherer, 1992), this sediment can act as a refuge for indicator bacteria that would normally die off if left in the water column alone. Through a number of experiments performed in a Paris storm sewer system, Ahyerre and colleagues examined the impact of rainfall and sewer flushing events on the layers of settled particles in sewer pipes (Ahyerre, 2000; Gromaire, 2000; Ahyerre, 2001). Their work found that the erosion of sediments within the sewer mobilized fine-grained material that was high in organic matter. Other researchers have shown that fecal indicator bacteria are frequently attached to such materials and that attached bacteria can persist longer in the environment than if they were free (unattached) organisms (Pommepeuy, 1992; Howell, 1996; Characklis, 2005). Working in freshwater streams,
Wilkinson found that an initial pulse of fecal coliforms were entrained in the water column at the onset of flow, followed by a reduced concentration if flow continued at the same rate, and that additional organisms were subsequently entrained only by increasing the flow rate (Wilkinson, 1995). Similar studies by Muirhead in New Zealand and by Jamieson in Canada found that EC persisted in stream sediments and were washed into the water column with smaller flows than might be predicted by particle settling theory alone (Muirhead, 2004; Jamieson, 2005).

A number of similarities exist between the various stormwater management measures being examined and their counterparts in the natural and built environments. A wet detention pond, for example, is modeled after naturally occurring wetlands that have been shown to have some capacity to treat stormwater contaminants (Mays, 2001). Gravel wetlands similar to the one evaluated here have been used for wastewater treatment in other settings and are reportedly capable of a one to two-log removal of fecal coliforms (EPA, 1993). However, influent from a wastewater source is likely to have much higher concentrations of bacteria than typically observed stormwater influent, so the same removal efficiency may be difficult to achieve in a stormwater system. The EPA fact sheet also notes that a secondary form of disinfection would be necessary to consistently meet a target limit of 200cfu/100mL, whereas most stormwater installations are assumed to discharge directly without further treatment. Manufactured devices share materials and basic operation with counterparts in the built environment that have also been studied by others (Ahyerre, 2000). The goal of the present study was to provide specific baseline data about fecal indicator bacteria in a variety of stormwater management measures. The impetus for this study was the results of earlier research at
UNH (Eckman, 2000) that observed concentrations of indicator bacteria frequently increased after runoff passed through stormwater management measures in New Hampshire’s seacoast region. That study examined a variety of common stormwater pollutants and screened for several types of bacteria that are considered indicator organisms (*Pseudomonas aeruginosa*, fecal coliforms, EC, ENT). For this study, EC and ENT were selected for monitoring because the EPA continues to recommend their use as indicator organisms and because they were regularly found in stormwater runoff from the West Edge parking lot during the previous study (Eckman, 2000).

**Purpose of Current Study**

This study was driven by concerns that current stormwater management measures may be degrading water quality with respect to bacterial indicators. There is a paucity of data on the subject, and a wide-matrix sampling approach was selected to collect the maximum amount of information. One hypothesis is that these systems inadvertently provide habitat and nutrients conducive to the growth of indicator bacteria. If large populations of such bacteria are resident in stormwater systems, it is likely that they will be discharged to receiving waters during rainfall runoff events. This study was undertaken to answer two key questions: “Do stormwater management measures have any effect on the concentration of indicator bacteria in the runoff that passes through them?” and, if so, “What factors can be identified to explain these effects?”

The results of this study will have implications for stormwater management in watersheds that are considered impaired with respect to fecal indicator bacteria and for design recommendations for construction in watersheds with high-quality or protected
receiving waters. This study examines the potential of these devices to act as sources or reservoirs of fecal indicator bacteria. If these devices act to further degrade the receiving water with respect to fecal indicator bacteria, it may be necessary to restrict certain types of stormwater management practices in watersheds with total maximum daily loads (TMDLs) for bacteria.
CHAPTER II: STUDY AREA AND STORMWATER MANAGEMENT MEASURES

Research Facility

This study was conducted at the UNH Stormwater Center’s research facility in Durham, NH. The facility was a testing and verification center for the evaluation of stormwater management measures and consists of a network of stormwater treatment devices that received uniform fractions of stormwater inflow from the adjacent 3.55-hectare commuter parking lot.

The parking lot was standard dense mix asphalt and was initially constructed in 1996. Parking lot activity was a combination of passenger vehicles and routine bus traffic. During the academic year (September through May), the lot was regularly used near its full capacity of 786 parking spaces. During the warmer summer months when the bulk of students were not on campus, the lot was largely unused and the regular bus services were suspended. Contaminant loading on the parking lot surface related to those activities was therefore reduced. However, higher temperatures may have resulted in increased loading from other sources (such as wildlife) and also may have enhanced bacterial growth and survival.

Construction of the stormwater management measures at the research facility was completed in June 2004 and collection of storm data began in September 2004. The system of parallel stormwater management measures at the UNH Stormwater Center ensures parity between the treatment devices being examined because all devices receive
the same inflow hydrograph and contaminant concentrations. This fundamental boundary condition is difficult if not impossible to achieve when monitoring devices at different locations because each site possesses unique influent loading and physical conditions. Additional information about the facility is provided in Appendix A: UNH Stormwater Center Site Layout and Device Descriptions and on the Center’s website: http://www.unh.edu/erg/cstev. Because influent loads were equal, results from each device may be compared directly with the results obtained from other devices located at the facility.

**Influent Characterization**

By sampling the influent before the flow was distributed to the various devices, a boundary condition was assigned to the facility without having to evaluate the wash-off function for bacteria on the surface of the parking lot or the resuspension of settled particles in the catchbasins and pipes upstream of the research facility. Runoff from the parking lot entered a network of catchbasins and pipes that discharged water through a single 0.91-meter diameter reinforced concrete pipe (RCP) backbone. This 0.91-meter RCP provided the source of influent to the stormwater testing facility. An automated sampler at this location allowed programmed sampling of the influent hydrographs. Just downstream of the influent sampling point was a distribution chamber that split the flow and provided equal fractions to each device via parallel, gravity-flow piping networks.
**Stormwater Management Measures**

Three classes of stormwater management measures were evaluated at the facility. Conventional structural devices included a wet detention pond and a rock-lined swale. Low-Impact Development (LID) devices included a surface sand filter, a bioretention area, and a horizontal flow, subsurface gravel wetland. Manufactured devices included several commercially available hydrodynamic separators, a stormwater filtration chamber, an internally baffled sediment trap/oil-and-grease separator (Water Quality Inlet), and a subsurface volume infiltration unit (Infiltration Drainfield). After the flow passed through a device, it was routed to a sampling vault for discharge measurement, sampling, and subsequent offsite discharge. For each device, storm effluent concentrations were compared with influent concentrations for 4 to 9 storms recorded between September 2004 and October 2005.

**Device Sizing**

Devices at the site were designed according to manufacturers’ reference material or to standard stormwater design guidelines. The primary sizing criteria for each device was the water quality volume (WQV) concept, which uses the site area, the percentage of impervious cover and the depth of the 90% rainfall event to determine the volume of stormwater that must be captured and treated. The equation takes the form

\[ WQV = PRvA/12 \]

where \( P \) is the 90% rainfall depth, \( Rv \) is \( 0.05 + 0.009I \), where \( I \) is the percent impervious cover, and \( A \) is the site area. Following sizing criteria in the New York State Manual (Center for Watershed Protection, 2001), the WQV for each device was found to be 92.4 cubic meters, with a peak flow of 0.03 cubic meters per second.
(cms). With the exception of the rock-lined swale, all conventional structural and LID devices were constructed with a pretreatment sedimentation basin (or forebay). These basins are commonly specified for removal of easily settleable solids and are slowly drained into the primary device via 0.15 m perforated pipes. During larger rainfall events, runoff in excess of each device’s forebay capacity bypassed over a weir directly into each primary treatment device.

**Wet Detention Pond (WDP)**

One of the most common stormwater management measures is a wet detention pond or wet pond. By retaining the WQV, it is designed to reduce peak flows during storm events and provide treatment between storms. Treatment is predominantly due to settling of suspended solids. Subsequent storms deliver a fresh influx of stormwater and force some of the retained water out of the system. During the summer months, the exposed surface area of the device results in warmer effluent temperatures and increased biomass in the form of algae and aquatic plants.

**Rock-Lined Swale (RLS)**

Another common stormwater management measure and conveyance structure is a rock-lined swale. Treatment in a swale is predominantly in the form of settling in the interstices of the rock. The swale at this site was 85 m in length and was lined with geotextile and 0.15 m crushed stone riprap. There was no pretreatment structure. Due to site constraints, the swale was sampled at its downstream end and was not conveyed by the piping network to the sampling gallery.
Surface Sand Filter (SF)

The surface sand filter was a constructed stormwater management measure that utilized a 0.5 m layer of medium sand as a filter media. The sand bed was sub-drained by a 0.15 m perforated pipe bedded in a 0.20 m layer of crushed rock. During smaller events, runoff infiltrates through the sand bed without ponding. Larger events with sufficient volume caused temporary ponding above the filter bed that was drained within 24 hours. Flows greater than the design volume bypassed the filter bed via an emergency spillway. The filter bed and sedimentation forebay were sized using criteria published in Design of Stormwater Filtering Systems (Claytor, 1996).

Bioretention Area (BA)

The bioretention area was a stormwater filtration system designed using the Center for Watershed Protection’s guidelines (Claytor, 1996). The filter media within the bioretention area was a 1.5-meter thick layer of amended soil consisting of: sand, compost, and native soils. Soils were designed for a moderately high infiltration rate (0.3 m/day). For larger storm events, 0.2 m of above-ground ponding provided additional storage before overflow bypass.

Subsurface Gravel Wetland (GW)

The subsurface gravel wetland was recently developed as an LID system designed to attenuate peak flows and provide subsurface anaerobic treatment. The gravel wetland consisted of a sediment forebay followed by a series of horizontal-flow treatment cells.
Each cell consisted of a 0.6-meter thick bed of gravel or crushed rock with a perforated pipe header to distribute incoming flow across the width of the gravel bed. Water traveled horizontally through the gravel bed and was collected by subdrains on the far side of the cell. The process was repeated in the second cell before being discharged. During high intensity events the WQV was stored above the beds and drained via a perforated riser on the upstream end of each cell to access the substrate.

**Stormwater Filtration Chamber (SFC)**

This system consisted of two manufactured devices arranged in series. The first device was a 1.2-meter diameter manhole with an internal baffle that deflected incoming flows to create a hydraulic vortex that facilitated particulate removal. The second device was a subsurface chamber with an internal spillway that directed effluent from the first unit across a suspended platform and through a perlite filter media. Both components were constructed of high-density polyethylene (HDPE). Once the influent passed through the filter media by gravity flow, it collected in the lower half of the filtration chamber, exiting when sufficient volume was accumulated to reach the outlet.

**Hydrodynamic Separators (HS1, HS2, HS3)**

The hydrodynamic separators were manufactured devices that can be used in place of traditional manholes. They were constructed of concrete or HDPE manholes modified with additional internal baffles, directional flow plates, or screens that were designed to remove particulate matter. Devices from three manufacturers were included in the current round of testing.
Water Quality Inlet (WQI) and Infiltration Drainfield (ID)

This system is comprised of two manufactured devices in series: a pretreatment water quality inlet and a larger unit that provided detention and infiltration. Both units were constructed of HDPE. The water quality inlet consisted of two baffles in series (one to capture settled particles, the other to capture oil and grease) welded inside a horizontal 1.5 m diameter pipe, with a 0.3 m pipe that bypassed larger flows directly into the infiltration drainfield. The infiltration drainfield consisted of three 12 m sections of horizontal 1.2 m diameter perforated pipes connected by headers. The perforated pipes were bedded in crushed rock and bank-run gravel. The top and sides of the infiltration unit were wrapped in a geotextile to prevent the migration of fines into the perforated pipes and the infiltration bed.
CHAPTER III: METHODS AND MATERIALS

Rainfall Runoff Events

Several criteria were used to determine if a rainfall runoff event was sampled and processed. The minimum rainfall depth was 2.54 mm following 72 hours of dry weather, with sufficient flow to trigger influent sampling and at least 10 samples over the hydrograph. Events that were not captured at the influent sampling location were not processed.

Storm Sampling Equipment

Each device in the facility was plumbed such that treated effluent from the device was delivered to a sampling port with sampling and monitoring equipment. Real-time parameters that were monitored include flow, temperature, specific conductivity, pH, DO, and rainfall. Refrigerated automated samplers were used for data collection and sampling (Isco Model 6712 SR). Flow was measured by Isco 730 Bubbler Flow Modules attached to Thel-mar compound weirs (Thel-mar Co.) and recorded by the 6712 data loggers. Due to difficulties in fitting weirs to the influent pipe and the swale, flow was monitored at those locations with Isco 720 Submerged Probe Flow Modules. Rainfall depth was monitored with an Isco 674 tipping bucket rain gage connected to the data logger at the influent monitoring point. The minimum depth that the rain gauge could record was 0.254 mm. The remaining real-time water quality parameters (temperature,
specific conductivity, pH and DO) were measured with multi-parameter YSI 600XL probes attached to the 6712 data logger for each treatment device.

The 6712 data loggers also act as the control head for the automated refrigerated samplers. The sampler was programmed to take up to 24 discrete samples over the expected hydrograph flow duration and maintain them at 4°C. In order to better capture representative flows from each device, sampling programs for each device were adjusted for the flow routing characteristics for that particular device. For example, volume devices (e.g., wet detention pond, bioretention area, and gravel wetland) had extended draining periods that lasted several hours to two days after rainfall ended. In contrast, non-storage devices (e.g., stormwater filtration chamber and hydrodynamic separators) stopped flowing within a few minutes after rainfall ended. Each 6712 sampler was programmed to capture 5 samples within the expected “first-flush” period of each storm (sampling program “A”), and then the remainder of samples over a longer time interval to capture the receding limb of the hydrograph (sampling program “B”).

A key component of timing stormwater sampling programs is recognizing the lag time of runoff traveling from the furthest point of the watershed to the watershed’s outlet, known as the time of concentration. Using a formula derived by the Soil Conservation Service (SCS), this is calculated as 

\[ t_c = 100L^{0.8} \left[ \left( \frac{1000}{CN} \right) - 9 \right]^{0.7} / 1900S^{0.5} \]

where \( t_c \) is the time of concentration in minutes, \( L \) is the length of the longest flowpath, \( CN \) is the SCS curve number that describes the composition of the watershed’s surface, and \( S \) is the average slope of the flow path (Viessman, 2003). By this method, the watershed time of concentration was estimated to be 22 minutes.
Because it was impossible to know, *a priori*, what type of rainfall event to expect and how much volume was required to fill volume devices before they began to release water, sampling programs at each device were triggered by effluent flow at the outfall of that device. The 6712 sampling programs were also adjusted seasonally to account for differences in rainfall patterns (high intensity, short-duration bursts in the summer and fall and lower intensity, long-duration storms and snow-melt events in the winter and spring).

The automated samplers were equipped with 24 bottle cages that hold Isco ProPak disposable 1-liter LDPE sampling bags. Machine blanks were periodically drawn and analyzed for bacteria to ensure that the sampling equipment was not contaminating the samples.

**Composite Sample Generation**

Before the discrete samples were composited, each device hydrograph was reviewed to verify that the samples were representative of the storm. In some cases, automated samplers accidentally continued to collect samples after the direct rainfall runoff had ended. The effluent hydrograph for non-storage devices was expected to closely follow that of the rainfall intensity and influent hydrographs. Any samples collected after the initial runoff hydrograph had passed were therefore collected from standing water in the sampling port, were not representative of storm effluent, and were discarded. Volume devices programmed to continue sampling for an extended duration occasionally collected samples in response to a secondary rainfall burst that was not characterized at the influent. Those samples were also discarded.
The discrete 1-L samples collected over the hydrograph were converted into a single 200-mL flow-weighted composite for analysis by using a spreadsheet to calculate the aliquot volume that each bottle was to contribute. The formula for determining the volume of the aliquot \( V_i \) removed from each sample bottle was determined as follows:

\[
V_i = M_i Q_i \Delta t_i,
\]

where

\[
M_i = 200mL \left[ \frac{\sum_{j=1}^{N} Q_j \Delta t_j}{\sum_{j=1}^{N} Q_j \Delta t_j} \right] * Q_i \Delta t_i
\]

\( M_i \) represents the multiplier that was used to create a single 200 mL sample for processing, \( Q_i \) represents the flow rate at the time the sample was collected, and \( \Delta t_i \) represents the time interval of the sampling program in effect when the sample was collected. Aliquots were removed from each 1-L sample bottle with sterile pipettes and were combined to form a 200-mL flow-weighted composite sample.

Composite samples were refrigerated at 4°C and delivered to the lab for analysis within 24 hours of the storm. Although EPA methods for indicator bacteria specify a 6-hour holding time for microbial samples, the extended holding times were necessary to capture the full extent of runoff hydrographs from volume devices. To verify that results were not affected by the extended holding times, duplicate samples were enumerated at 6, 24, and 48 hours as part of a dry weather sampling event. Those samples demonstrated that holding samples at 4°C provided satisfactory preservation to maintain viable populations of indicator bacteria for up to 48 hours. Results from the holding time evaluation are presented in Appendix C: Notes on Sampling and Analysis.
**Enumeration Methods**

EC were enumerated using EPA Method 1103.1: “*Escherichia coli* in Water by Membrane Filtration Using membrane-Thermotolerant *Escherichia coli* Agar (mTEC).”

ENT were enumerated using EPA Method 1106.1: “*Enterococci* in Water by Membrane Filtration Using membrane-Enterococcus-Esculin Iron Agar (mE-EIA).”

Samples were processed by Dr. Steve Jones and Mr. Colin Edwards at UNH’s Jackson Estuarine Laboratory. Following standard censoring practice for non-detect occurrences, a value of one-half of the detection limit (most often 2 cfu/100mL) was used for data analysis.

**Dry Weather Sampling**

Most stormwater treatment devices maintain some volume of resident water that is not released between storms. This water is typically in close contact with any debris or sediment that the device has captured, as well as the wetted walls of the device that may support a biofilm. As a result, the device provides a possible refuge for indicator bacteria that may then be flushed out of the device during the next flow event. Resident water was sampled periodically throughout the year to examine long-term trends that may be related to changes in temperature and nutrient inputs. Samples were also taken in expectation of coming storms in order to provide an indication of the resident population of indicator bacteria as close as possible to the beginning of a storm event. Additional samples were taken 1 to 3 days following a storm event to examine post-storm trends. For dry weather samples, devices with a standing pool of water were sampled using 1-L HDPE ball-valve bailers (Ben Meadows Co.). Bailers were sterilized using a dilute bleach solution and
then rinsed with DI water (Scalf, 1981). At the time of sampling, an initial bailer volume of water was drawn from the device being sampled and was used to further rinse the bailer. This first sample was discarded and then a water sample was taken for analysis.

The influent pipe and infiltration devices lacked any standing pool of water that could be reliably sampled between storm events. A seasonal dry weather baseflow (less than 5 m³/day) was observed at the distribution box and this water was sampled using the grab sample function of the influent automated sampler. This flow was attributed to infiltration and inflow into the RCP that conveys runoff from the parking lot to the research facility. Because of the orientation of the pipe network leading from the distribution box, such extremely low flows preferentially enter the infiltration drainfield, the sand filter, and the bioretention area. As a result, the outfall of these devices typically exhibited a small amount of dry weather flow. This water was sampled using the grab sample function of the automated samplers and was assumed to be representative of the conditions of water that was draining from the media in each device.

**Sediment Sampling**

For the wet detention pond and subsurface devices where a layer of sediment was consistently submerged, a stainless steel Petite Ponar grab sampler (Wildco Model 1728-G40) was used to sample sediment on the bottom of the unit without having to drain the water from the device. For devices with a soil media, a soil sample was taken at the surface of the media using a trowel. Sampling equipment was decontaminated between different devices by washing with Alconox cleanser and thoroughly rinsing with de-ionized water.
For each device, a 1-gram aliquot was vortexed in sterile buffered peptone water. This tube was decimally diluted and 2.5 mL of each dilution was filtered onto mTEC and mE plates. Dry weight bacterial concentrations were calculated using the average dry weight of each sample. One kilogram of sediment was dried and sieved for particle size analysis using ASTM Method D422 (American Society for Testing and Materials, 1998). After separating the different sediment particle sizes by sieving, the weight of materials retained on each sieve was calculated as a fraction of the total sample weight. From these fractions, a particle size distribution was generated (% material finer by weight versus log particle size) and used to evaluate the type of material in the sample. Particles less than 0.075 mm are considered fines (silt and clay), particles between 0.075 and 4.75 mm are described as sands, and particles between 4.75 and 75 mm are described as gravels. In many engineering calculations, the diameter of particles representing sample fractions 10, 30, 50, and 60% finer by weight are commonly referred to as the D10, D30, D50 and D60 of the sample and are used to describe the physical characteristics and behavior of the sample.

Data Analysis

Two basic types of analysis were performed using the ENT and EC counts that were gathered by the water and sediment sampling program. Device performance analysis aimed to evaluate the effect of stormwater management measures on fecal indicator bacteria by comparing concentrations of water samples taken at the influent with concentrations at the effluent of each device. Monitoring Parameter correlations
compared the concentrations of bacteria to other parameters that were monitored at the site in an attempt to identify explanatory factors.

**Device Performance Evaluation**

The basic question that this research sought to answer was what effect stormwater management measures had on the concentration of fecal indicator bacteria in stormwater runoff. The primary means of evaluating this question was to compare stormwater samples taken at the shared influent location to samples taken at the individual device effluent locations. If the device effluent had lower concentrations of bacteria than the influent, the device was determined to be removing bacteria, at least in the context of that particular storm event. Conversely, if the device effluent had higher concentrations of bacteria than the influent, it was determined to be exporting bacteria.

In many areas of research, the comparison of paired data sets (pre-treatment vs. post-treatment) would employ the Student’s t-test to evaluate the statistical significance of observed differences. Because bacterial data did not meet the t-test’s assumption of normal distribution, even after log transformation, equivalent non-parametric tests were employed instead. Two nonparametric tests, the Sign Test and the Wilcoxon Rank Sum Test, were performed to test the null hypothesis (H₀) that the mean bacterial concentration in the effluent of each device was not significantly different from the mean bacterial concentration in the influent.

The sign test is a robust, non-parametric test that evaluates the likelihood that samples drawn at random from two paired populations are, in fact, drawn from the same population. If the values from one population are consistently higher than that from the
other, the sign test provides a measure of the significance of that finding (Helsel, 1992). Sign test analyses were performed using a spreadsheet and tables populated with published values for the binomial distribution (Lowry, 2005).

The second test employed to evaluate the difference between influent and effluent bacterial counts and between storm and dry weather sample bacterial counts was the Wilcoxon Rank Sum test. The rank sum test is a non-parametric test that was used to compare the distribution of all events in the influent dataset with the distribution of all events in the effluent dataset from each device. This test complements the sign test because it examines the central tendency of the entire dataset being examined rather than the effects on specific event-paired comparisons. The rank sums analyses were performed using JMP™ software, version 5.1 (SAS Institute).

In addition to the statistical tests, exceedance probability plots were generated as a visual representation of the rank sums analysis. This technique rank-orders the available data and uses the Weibull plotting position to generate a probability of exceedance for each observed value. The resulting curve represents the relative frequency of each concentration in the influent or effluent. Devices with effluent bacterial concentrations lower than that of the influent will have a probability curve that plots to the left of the influent probability curve and vice versa. Although there is not a statistical test associated with this type of analysis, the format can be useful for visualizing trends in the data and targeting additional research opportunities.
Monitoring Parameter Correlations

In an effort to build an explanatory model of bacterial variation using other quantified parameters, correlations were sought between ENT counts and Julian Date, Daily Average Air Temperature, Daily Average Water Temperature, Peak Rainfall, Total Rainfall, and Peak Shear Stress in the Influent Pipe. ENT counts for each device were correlated to the water quality data (conductivity, pH, DO, and temperature) specific to that device, along with a uniform dataset for parameters shared by all devices (such as date, antecedent dry days, etc). Flow-weighted averages of water quality parameters were used to represent the conditions in the effluent of each device during the storm event sampling period. The data were initially plotted and examined visually before the correlation analysis was run. $R^2$ value of 0.5 or greater were considered significant.

Correlation analyses were performed using the CORREL function of Microsoft ® Excel 2000.
CHAPTER IV: RESULTS AND DISCUSSION

Rainfall – Runoff Events

During the study period, total daily rain depths of a minimum 2.54 mm were recorded an average of once every four days. The largest 24-hour precipitation event occurred on October 8, 2005 (104 mm), while the smallest event that could be recorded (0.254 mm) occurred 24 times. Average daily air temperature varied from -17°C to 29°C, with an annual average of 8°C. Nine rainfall-runoff events were examined at the influent and are summarized in Table 2. Some devices captured fewer events due to the device sampler failing to trigger or to the device being off-line for maintenance.

Table 2. Meteorological Data for Sampled Rainfall-Runoff Events

<table>
<thead>
<tr>
<th>Event Date</th>
<th>Antecedent Dry Days (&lt;2.54 mm rainfall)</th>
<th>Peak Intensity (mm/hr)</th>
<th>Total Rainfall during sampling period (mm)</th>
<th>Average Temperature of Runoff (°C)</th>
</tr>
</thead>
<tbody>
<tr>
<td>28-Sep-2004</td>
<td>9.8</td>
<td>0.94</td>
<td>0.55</td>
<td>18</td>
</tr>
<tr>
<td>10-Feb-2005</td>
<td>5.9</td>
<td>0.94</td>
<td>0.94</td>
<td>2</td>
</tr>
<tr>
<td>8-Mar-2005</td>
<td>6.9</td>
<td>0.47</td>
<td>1.02</td>
<td>3</td>
</tr>
<tr>
<td>28-Mar-2005</td>
<td>3.9</td>
<td>1.9</td>
<td>7.64</td>
<td>4</td>
</tr>
<tr>
<td>20-Apr-2005</td>
<td>17.2</td>
<td>1.9</td>
<td>2.24</td>
<td>13</td>
</tr>
<tr>
<td>15-Jul-2005</td>
<td>5.3</td>
<td>2.4</td>
<td>0.91</td>
<td>25</td>
</tr>
<tr>
<td>13-Aug-2005</td>
<td>10.8</td>
<td>3.8</td>
<td>1.85</td>
<td>22</td>
</tr>
<tr>
<td>15-Sep-2005</td>
<td>10.7</td>
<td>2.8</td>
<td>0.78</td>
<td>24</td>
</tr>
<tr>
<td>8-Oct-2005</td>
<td>8.3</td>
<td>0.47</td>
<td>0.71</td>
<td>20</td>
</tr>
</tbody>
</table>
**Influent Characterization**

During the sampling period, nine storm events and 17 dry weather (non-storm) samples were evaluated for ENT and EC at the influent monitoring location. A summary of influent data collected during this study is presented in Figure 1. As mentioned previously, dry weather samples were collected from the small, persistent baseflow that results from infiltration and inflow to the RCP pipes upstream of the distribution chamber. During the driest part of the year, (typically September and October) this baseflow ceased when groundwater levels in the vicinity of the pipe network were at their lowest (data not show). As a result, there were no dry weather influent samples taken during these periods. Full results of ENT and EC counts are also presented in APPENDIX B: RAW DATA TABLES. Several trends were apparent in the plots of the influent data and are discussed below.

The observed concentration of ENT was higher than the concentration of EC in all samples where both were detected. In 14 samples, EC concentrations were below the detection limit of 4 cfu/100mL, whereas ENT concentrations were below the detection limit only four times. As previously described in Methods & Materials, non-detect values were replaced a value corresponding to half of the detection limit and are plotted at 2 cfu/100mL. Both types of bacteria were examined in the study because of their use in regulatory standards. However, it appears that this site did not have significant sources of EC loading or that there were factors present which prevented the survival of EC once it entered the stormwater system. One factor that was examined and rejected was the possibility that high chloride levels from ice control activities were limiting EC survival.
However, after comparing information in the literature regarding EC’s chloride tolerance with concentrations of chloride as determined by a site-specific conductivity regression equation, it was apparent that chloride concentrations were insufficient to inhibit EC growth (Bromley, 2003).

Nearly all samples collected during storm events had higher concentrations of bacteria than dry weather samples collected around the same time. Six of the nine storms sampled at the influent occurred 1 to 5 days after a dry weather sample was taken. In 83% of these, ENT counts were lower in the dry weather sample than in the subsequent storm sample (median storm concentration = 400 cfu/100mL; median dry weather concentration =20 cfu/100mL). Of the influent storm samples where EC was detected and a dry weather sample had been previously obtained (4 storms), all had higher counts than were found in the dry weather sample that preceded the storm. A Wilcoxon rank sums analysis of storm and dry weather bacterial counts found that the two types of samples were statistically different (p=0.0069 for ENT and p=0.0490 for EC), with storm samples generally containing significantly more bacteria than dry weather samples.

A temporal trend was evident, with higher concentrations of both bacteria being observed in the summer and fall than in the winter and spring. Whether this was a seasonal trend or a coincidental phenomenon cannot be determined without multiple years of data. However, the parking lot and the stormwater collection network upstream of this sampling location were constructed 9 years prior to this study and have not been significantly modified since then. The parking lot is most heavily used during the academic year (September through May), corresponding at least partially with the bacterial trend.
Figure 1. Bacterial counts at the influent during the sampling period (9/28/2004 to 10/10/2005)
**Effluent Characterization (Storm Samples)**

ENT were detected in effluent from all devices throughout the year and frequently at levels in excess of the 104 cfu/100mL regulatory limit set for single-sample counts at designated bathing beaches (EPA, 2004).

Figure 2 displays the variability of the ENT counts from each device. In the box-and-whiskers format, the uppermost value represents the maximum observed concentration, the top of the box represents the 75th percentile, the mid-line represents the median concentration, the bottom of the box represents the 25th percentile, and the lowest value represents the lowest observed concentration. The INF distribution is presented to the far left of both series for comparison. The highest observed storm concentration for any device was 6653 cfu/100mL, observed at the RLS. The ID and WQI each experienced one storm where ENT was not detected. Following standard censoring practice for non-detect occurrences, a value of one-half of the detection limit, or 2 cfu/100mL was entered instead. The complete dataset for each device is presented in Appendix B: Raw Data Tables.

Three devices (BA, GW, and ID) exhibited median concentrations that were an order of magnitude lower than that of the influent, while one (RLS) exhibited median concentrations that were an order of magnitude higher than that of the influent. The remainder of devices exhibited medians that were at approximately the same order of magnitude. For the overall dataset, the Kruskal-Wallis test did not detect significant differences among the influent and the devices (p=0.3409).

Concentrations of EC were regularly low or below the detection limit in influent and device effluent samples. During the sampling period, EC counts were not detected
(i.e., were less than 4 cfu/100mL) in 39% of storm samples. In the remainder of samples, EC counts were regularly below single-sample regulatory limits, exceeding the New Hampshire limit of 88 cfu/100mL for freshwater swimming beaches only eight times and the EPA “designated bathing beach” limit of 235 cfu/100mL only four times out of 85 storm samples site-wide (EPA, 2004; NHDES, 2005). The highest observed storm concentration of EC was 8000 cfu/100mL, observed at the RLS on 10 October 2005. The highest observed EC concentration in the influent was 880 cfu/100mL, observed on 15 July 2005. Further statistical analysis of ENT and EC data was performed and is presented below. For space considerations, only ENT data is presented graphically.

As can be seen in Table 3, all devices exported ENT (effluent concentration exceeding influent concentration) at least some of the time. Notable devices were the WDP and RLS, which exported ENT in 75% and 83% of events and the BA, which exported ENT in only 25% of events. In contrast, no device exported EC in more than a third of the monitored events, and effluent concentrations at three devices (HS3, ID, and SFC) never exceeded those of the influent. As has been noted, EC was only detected in a limited number of samples (both influent and effluent).

The influent concentration was used to provide a baseline against which to directly compare the effect of the treatment devices on the stormwater effluent. Because of physical constraints at the facility, the devices were located unequal distances from the influent sampling point. As a result, each device was connected to the influent with different lengths of 0.3m HDPE pipe. Although the distribution system was designed with continuous slopes from the influent to each device, it is possible that the distribution
network had some effect on bacterial concentrations that was not accounted for by the sampling strategy.
Figure 2. Variability of storm event ENT counts by device. Influent (INF), Bioretention Area (BA), Gravel Wetland (GW), Hydrodynamic Separators (HS), Infiltration Drainfield (ID), Rock-lined Swale (RLS), Sand Filter (SF), Stormwater Filtration Chamber (SFC), Wet Detention Pond (WDP), Water Quality Inlet (WQI)

Table 3. Percentage of rainfall-runoff events that devices exported bacteria (effluent > influent) and significance level at which device effluent can be distinguished from influent.

<table>
<thead>
<tr>
<th>Stormwater Management Measure</th>
<th>Number of events sampled</th>
<th>% of events device exported ENT</th>
<th>p-value based on ENT (Rank Sums)</th>
<th>p-value based on ENT (Sign Test)</th>
<th>% of events device exported EC</th>
<th>p-value based on EC (Rank Sums)</th>
<th>p-value based on EC (Sign Test)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bioretention Area (BA)</td>
<td>5</td>
<td>25%</td>
<td>0.1615</td>
<td>0.313</td>
<td>25%</td>
<td>0.4448</td>
<td>0.5</td>
</tr>
<tr>
<td>Subsurface Gravel Wetland (GW)</td>
<td>8</td>
<td>43%</td>
<td>0.1019</td>
<td>0.5</td>
<td>14%</td>
<td>0.2992</td>
<td>0.313</td>
</tr>
<tr>
<td>Hydrodynamic Separator 1 (HS1)</td>
<td>7</td>
<td>29%</td>
<td>0.5964</td>
<td>0.5</td>
<td>29%</td>
<td>0.5863</td>
<td>0.5</td>
</tr>
<tr>
<td>Hydrodynamic Separator 2 (HS2)</td>
<td>8</td>
<td>57%</td>
<td>0.8474</td>
<td>0.5</td>
<td>14%</td>
<td>0.8837</td>
<td>0.313</td>
</tr>
<tr>
<td>Hydrodynamic Separator 3 (HS3)</td>
<td>7</td>
<td>43%</td>
<td>0.7506</td>
<td>0.5</td>
<td>0%</td>
<td>0.4461</td>
<td>0.063</td>
</tr>
<tr>
<td>Infiltration Drainfield (ID)</td>
<td>6</td>
<td>40%</td>
<td>0.1255</td>
<td>0.344</td>
<td>0%</td>
<td>0.1353</td>
<td>0.063</td>
</tr>
<tr>
<td>Rock-lined Swale (RLS)</td>
<td>7</td>
<td>83%</td>
<td>0.1527</td>
<td>0.109</td>
<td>33%</td>
<td>0.6671</td>
<td>0.5</td>
</tr>
<tr>
<td>Surface Sand Filter (SF)</td>
<td>7</td>
<td>67%</td>
<td>0.9578</td>
<td>0.344</td>
<td>33%</td>
<td>0.7052</td>
<td>&gt;0.500</td>
</tr>
<tr>
<td>Stormwater Filtration Chamber (SFC)</td>
<td>8</td>
<td>50%</td>
<td>0.8852</td>
<td>&gt;0.500</td>
<td>0%</td>
<td>0.4537</td>
<td>0.031</td>
</tr>
<tr>
<td>Wet Detention Pond (WDP)</td>
<td>9</td>
<td>75%</td>
<td>0.6911</td>
<td>0.227</td>
<td>13%</td>
<td>0.4944</td>
<td>0.109</td>
</tr>
<tr>
<td>Water Quality Inlet (WQI)</td>
<td>8</td>
<td>43%</td>
<td>0.9233</td>
<td>0.5</td>
<td>14%</td>
<td>0.5873</td>
<td>0.188</td>
</tr>
</tbody>
</table>
Sign Test Analysis

The frequency of bacterial export was used as an initial analytical approach to identify devices that may be affecting bacterial counts in stormwater. Assuming that the device had no effect on the bacterial concentrations, sampling variability would be expected to account for any differences between counts at the influent and effluent of the device. In recognition of this variability, the null hypothesis predicts that 50% of events would show bacterial export and 50% would show bacterial removal. As the frequency of export (or, conversely, removal) increased for a particular device, it was increasingly likely that the device was having some effect on the bacterial concentration. The sign test was used to evaluate the significance of this variation.

The power of the sign test was limited by small sample sizes and was not able to detect any significant differences. For ENT, the most significant finding was for the RLS (p=0.109), which exported ENT in 83% of monitored events. All devices removed EC more often than they exported it, although the sign test only detected highly significant effects (p<0.05) at HS3, ID, and SFC.

Rank Sums Test

Comparing storm event counts of ENT and EC in the devices’ effluent to counts at the influent using the rank sums test, it was not possible to detect significant effects of the devices on fecal indicator bacteria in stormwater runoff. Using an expanded confidence interval for screening purposes, differences were seen in the GW, ID and RLS effluent data, all of which could be distinguished from the influent at the 85% confidence
interval. The central tendency of the RLS was to have effluent concentrations of ENT that were higher than those of the influent, whereas the other devices had effluent concentrations of ENT that were lower than those of the influent. For EC, the ID always had effluent concentrations lower than those in the influent and it was the only device that could be distinguished from the influent using the rank sums test on the EC data.

**Probability Distribution Analysis**

For each of the plots in Figure 3, the influent probability distribution is the same for each device, while the effluent distribution varies according to the concentrations recorded at each device. Although the probability distribution format does not allow a one-to-one comparison of removal efficiency for each runoff event, it demonstrates the overall performance of each device relative to the influent loading during the period of record. For example, BA, GW, and ID all have effluent distributions that plot to the left of the influent distribution, leading to the general conclusion that they were removing ENT over the period of record. Conversely, RLS and WDP regularly plot to the right of the influent distribution, leading to the general conclusion that they were exporting ENT. The effluent plots for the remainder of the devices closely resemble that of the influent plot, leading to the general conclusion that the devices were having little impact (positive or negative) on the populations of indicator bacteria in stormwater runoff as it moves through them.
Figure 3. Non-exceedance Probability Plots For ENT concentrations in Device Influent and Effluent
Retained Water Characterization (Dry Weather Samples)

Most stormwater management devices retain some amount of water that was not fully drained between storm events, either in an impermeable chamber or in the pore spaces of a filter media. When a storm event occurs, retained water may mix with fresh influent before being expelled or may be forced from the unit without mixing with the incoming water. During the interim period between storms, interactions occur between settled materials, filter media, bacteria, and the retained water that may create conditions favorable for the survival of indicator bacteria. Other researchers (Butler, 1995; Morrison, 1995) have examined the biochemical changes that occur in gully pots (also known as catchbasins) such as the weathering of sediments, changes in pH and dissolved oxygen levels, and the associated release of soluble organics and metals. Indicator bacteria in the water column may stay in suspension or settle out with other particulate matter. Once bacteria are adsorbed to sediments and have settled out of the water column, their survival may be increased by the greater availability of nutrients and protection from predation (Schillinger, 1985; Marino, 1991). If bacterial concentrations are increasing in the devices between storm events, subsequent storm flows may experience additional loading from sources within the device.

Figure 4 displays the range of values observed in each device from dry weather samples. As noted previously, values of 2 indicate that no ENT were detected in a 25-mL sample. Median concentrations for all devices except the SFC were below the single-sample limit of 104 cfu/100mL. Although the general trend was for dry weather samples to have lower ENT concentrations than storm samples, the highest observed ENT concentration was 7240 cfu/100mL, taken from HS3 during dry weather in August 2005.
Figure 4. Variability of dry weather ENT counts by device. Influent (INF), Bioretention Area (BA), Gravel Wetland (GW), Hydrodynamic Separators (HS), Infiltration Drainfield (ID), Rock-lined Swale (RLS), Sand Filter (SF), Stormwater Filtration Chamber (SFC), Wet Detention Pond (WDP), Water Quality Inlet (WQI)
Comparison of Dry Weather Samples to Storm Samples

Comparisons of dry weather samples to storm samples at the influent and each of the devices are presented in Table 4. For samples taken from device effluent during storm events, 92% exhibited higher counts of ENT than the dry weather sample taken at the device one to five days prior to the storm event. This finding is consistent with the hypothesis that storm events result in a surge of bacteria in the stormwater system and is similar to the findings reported at the influent. Examining the ENT counts for all samples over the study period, highly significant differences (p<0.01) were detected between storm and dry weather samples in the ID, SF, and BA. Significant differences (p<0.05) were also detected at the influent and the WDP. For EC, the high number of non-detect samples makes any assessment of differences between storm and dry weather samples unreliable, however, median and peak values of EC were lower in the dry weather samples of most devices.

All of the devices where significant differences were observed exhibited a persistent baseflow (less than 5 m³/day). In part, this baseflow was due to the orientation of the pipes at the influent structure that preferentially delivered non-storm low flows to them. During the driest part of the year, baseflow ceased at the influent structure when groundwater levels drop below the RCP leading to it. However, even after the influent baseflow had ceased, a small amount of baseflow continued from the effluent of the devices where significant differences were detected. Although the native soils in which the devices were constructed were generally tight and contained a large clay fraction, there was a groundwater gradient from the parking lot above the site to the wetland area beyond the site, and the high conductivity crushed rock used to bed the sub-drains below.
the infiltration devices (BA, ID, SF) may have inadvertently collected groundwater during non-storm periods. If this were the case, the low dry weather counts were not necessarily indicative of bacterial removal by those devices. Between storms, the WDP may have successfully reducing bacterial concentrations in the retained water through sedimentation, UV disinfection, and grazing.
Table 4. Comparison of dry weather and storm counts of ENT and EC.

<table>
<thead>
<tr>
<th>Stormwater Management Measure</th>
<th>ENT (cfu/100mL)</th>
<th>EC (cfu/100mL)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Storm Median (Peak)</td>
<td>% of samples above limit</td>
</tr>
<tr>
<td>Influent (INF)</td>
<td>400 (1800) 63%</td>
<td>20 (1080) 41%</td>
</tr>
<tr>
<td>Water Quality Inlet (WQI)</td>
<td>232 (5040) 57%</td>
<td>24 (6720) 29%</td>
</tr>
<tr>
<td>Infiltration Drainfield (ID)</td>
<td>54 (400) 33%</td>
<td>6 (252) 10%</td>
</tr>
<tr>
<td>Surface Sand Filter (SF)</td>
<td>434 (2440) 50%</td>
<td>8 (72) -</td>
</tr>
<tr>
<td>Wet Detention Pond (WDP)</td>
<td>820 (2720) 75%</td>
<td>38 (1560) 36%</td>
</tr>
<tr>
<td>Bioretention Area (BA)</td>
<td>20 (600) 25%</td>
<td>4 (222) 18%</td>
</tr>
<tr>
<td>Stormwater Filtration Chamber (SFC)</td>
<td>372 (3280) 63%</td>
<td>106 (4360) 50%</td>
</tr>
<tr>
<td>Hydrodynamic Separator 1 (HS1)</td>
<td>208 (3920) 57%</td>
<td>88 (7240) 45%</td>
</tr>
<tr>
<td>Hydrodynamic Separator 2 (HS2)</td>
<td>80 (3880) 43%</td>
<td>28 (5680) 36%</td>
</tr>
<tr>
<td>Hydrodynamic Separator 3 (HS3)</td>
<td>204 (5760) 57%</td>
<td>85 (7120) 50%</td>
</tr>
<tr>
<td>Gravel Wetland (GW)</td>
<td>60 (2160) 29%</td>
<td>80 (2960) 43%</td>
</tr>
<tr>
<td>Rock-lined Swale (RLS)</td>
<td>2136 (5440) 67%</td>
<td>60 (4360) 33%</td>
</tr>
<tr>
<td>Single sample limits for designated bathing beach (EPA, 2004)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Correlation With Other Parameters

In cases where trends were visible among the plotted data, the results were varied among the devices. For example, samples with higher levels of dissolved oxygen corresponded to lower ENT counts in GW, SFC, and WQI but not in HS1, ID, RLS, and WDP where the plots appear scattershot. During most sampling events (storm and dry weather) the pH in the sampled water was between 6 and 7, resulting in vertical plots that exhibit little or no trend. For the influent and devices without storage, higher total rainfall depths corresponded with lower ENT concentrations, a reasonable finding if the source of ENT was washoff from the watershed. During larger storm events, available sources of ENT may be the limiting factor. If that occurs, the resulting ENT concentrations would be diluted by excess water, except in units with storage that may already have had elevated bacterial concentrations or which may have had internal sources.

For the influent dataset (storm and dry weather), Julian date and average water temperature yielded the highest $R^2$ values of 0.62 and 0.59, respectively. Both correlations are in agreement with the finding that ENT counts were lower in the winter and spring and were higher in the summer and fall. For storm samples in manufactured devices, the influent concentration was the best predictor of effluent concentration (positive correlation, $R^2$ ranging from 0.84 to 0.92). This result further supports the finding that manufactured devices were unable to provide treatment for fecal indicator bacteria. For traditional and LID devices, no clear trends emerged that consistently explained more than 50% of the variance. Complete tables of correlations by device are presented in Appendix D – Results of Statistical Testing.
**Sediment Characterization**

Sediment samples from several devices were taken during the study period, analyzed for bacterial counts, and sieved for particle size distribution. The results are presented in Table 5. It should be noted that the devices were built in the first half of 2004 and were brought online in June 2004. Furthermore, the manufactured devices (WQI, SFC, HS0-3) were thoroughly cleaned at the beginning of the monitoring period in September 2004 and that any sediment in the devices had accumulated during the following year. GW samples were collected from the soil lining the sedimentation forebay and WDP samples were collected from the soil just above the edge of the permanent pool.

Bacterial counts in the sediment were lower than were expected, based on reports in the literature of EC and ENT counts several orders of magnitude greater in estuarine sediment (Pommeuy, 1992), lake bottom sediment (An, 2002), stream sediment (Jamieson, 2004), and storm drain sediment (Marino, 1991). It should be noted that EC and ENT counts were approximately equal in sediment samples, whereas EC was always detected at lower concentrations in water samples. The particle size distribution data provides a possible cause for the relatively low counts that were observed. The dominant particle size of the captured sediment would be described as a fine to medium sand, with less than 10% by weight being classified as fines (particle diameter <0.075mm). Research by Auer and Niehaus found that fecal coliform bacteria were most often associated with particles less than 0.06mm, with up to 90% of bacteria being associated with particles less than 0.01mm (Auer, 1993). The low counts of bacteria in the sediment samples may
simply reflect the relative absence of appropriately sized particles in the captured sediments.

Table 5. ENT concentrations in device sediments

<table>
<thead>
<tr>
<th>Stormwater Management Measure</th>
<th>EC (cfu/gram)</th>
<th>ENT (cfu/gram)</th>
<th>$D_{10}$ (mm)</th>
<th>$D_{50}$ (mm)</th>
<th>$D_{60}$ (mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Water Quality Inlet (WQI)</td>
<td>87</td>
<td>87</td>
<td>0.187</td>
<td>0.908</td>
<td>1.226</td>
</tr>
<tr>
<td>Wet Detention Pond (WDP)</td>
<td>nd</td>
<td>nd</td>
<td>No sample retained for sieve analysis</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Stormwater Filtration Chamber (SFC)</td>
<td>nd</td>
<td>nd</td>
<td>0.111</td>
<td>0.296</td>
<td>0.358</td>
</tr>
<tr>
<td>Hydrodynamic Separator 0 (HS0)</td>
<td>177</td>
<td>59</td>
<td>0.191</td>
<td>0.734</td>
<td>0.945</td>
</tr>
<tr>
<td>Hydrodynamic Separator 1 (HS1)</td>
<td>70</td>
<td>139</td>
<td>0.133</td>
<td>0.477</td>
<td>0.613</td>
</tr>
<tr>
<td>Hydrodynamic Separator 2 (HS2)</td>
<td>nd</td>
<td>nd</td>
<td>0.150</td>
<td>0.585</td>
<td>0.758</td>
</tr>
<tr>
<td>Hydrodynamic Separator 3 (HS3)</td>
<td>302</td>
<td>452</td>
<td>0.153</td>
<td>0.601</td>
<td>0.738</td>
</tr>
<tr>
<td>Subsurface Gravel Wetland (GW)</td>
<td>nd</td>
<td>nd</td>
<td>No sample retained for sieve analysis</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Nd = not detected

A preliminary explanation for these results is that sediments in the devices were not a significant source of indicator bacteria in these systems. Water quality samples from the rainfall runoff event less than 24 hours after these samples were taken had bacteria counts 2 to 3 orders of magnitude higher than those in the water used to wash bacteria from the sediment samples. Assuming that the sediment was a major contributor of bacteria in the device effluent, processing the sediment samples by adding sterile water and vortexing should have served as a proxy for the potential resuspension of settled sediment and bacteria that might occur during a storm event. If the sediment source hypothesis were correct, counts in the processed sample water should be elevated above levels observed in dry weather samples. However, this was not observed.
To date, little information has been reported in the literature that specifically addresses bacterial counts in the sediments of stormwater management measures. However, at least one investigator (Zhang, 2005) identified counts of EC and ENT in a manufactured stormwater unit that were several orders of magnitude higher than those found in similar devices at the UNH site. In a Michigan stream whose flow regime includes a major stormwater component, Marino (1991) reported that FS and FC counts in sediment were stable at $10^5$ cfu/100mL. In that study, as in ours, sediment samples were diluted, centrifuged, and processed as water samples. Similarly, in sediments at CSO sites, Ellis and Yu (1995) reported $2.8 \times 10^3$ MPN/g for FC and $1.4 \times 10^3$ MPN/g for FS. In other types of aquatic systems (lakes, beaches, streams), there is a general consensus among researchers that sediments can act as reservoirs of fecal indicator bacteria that can subsequently be released when the sediments are disturbed (for a summary, see Jones (Jones, 1999). Further study may be warranted to expand these preliminary results, especially as the systems age and accumulate additional sediment.
CHAPTER V: SUMMARY AND RECOMMENDATIONS FOR FUTURE RESEARCH

The increased storm concentrations could be the result of the wash-off of fresh bacteria in the watershed, or from scouring within from the piping network, or the resuspension of previously settled bacteria within the devices, or from rewetting soils at the margins of constructed systems.

ENT Summary

The pilot study performed at the influent location reported lower ENT counts than were observed during the present study. One possible explanation for the increase may be that the parking lot and its drainage network have aged during the intervening five years and may now be supporting conditions favorable to a larger bacterial population. While interesting to note, conditions upstream from the stormwater facility were considered beyond the scope of the current study and were not examined.

ENT counts in both the influent and the device ef fluent s regularly exceeded EPA standards for primary contact recreational water quality of 104 cfu/100mL, the standard that would be applied if the sample had been collected as a single grab sample at a marine “designated bathing beach” (EPA, 2005). None of the stormwater management measures investigated during this study were 100% successful in reducing the ENT load that was received from the influent. The majority of devices examined (HS, SF, SFC, WDP, and WQI) exhibited no clear trend with regard to ENT and should be considered to have no
effect unless additional data becomes available. The BA, GW, and ID were the only
devices that exhibited median ENT counts less than the primary contact limit, with
median values of 24, 44, and 54 cfu/100mL respectively. These median ENT
concentrations were 1-log lower than that of the influent during storm events (median
concentration of 560 cfu/100mL), an indication that bacterial removal was occurring. The
RLS was the only device that appeared to increase the concentration of ENT and
exhibited a median concentration (3520 cfu/100mL) that was 1-log higher than that of the
influent. Because of the small sample sizes and the large amount of variation inherent in
stormwater sampling, statistical tests were unable to determine if any of these findings
were statistically significant.

EPA guidance for recreational bathing waters also specifies a geometric mean
limit of 33 cfu/100mL for fresh waters and 35 cfu/100mL for marine waters. For storm
samples, neither the influent nor any of the devices met those limits for the entire year.
The three devices that reduced ENT concentrations are relatively recent additions to the
stormwater manager’s toolbox. If continued sampling provides further support for the
conclusion that they are reducing bacteria and other contaminants, it may encourage more
widespread installation of these devices in the future. In contrast, swales are already in
widespread use and are likely to continue to be used in the future. The RLS was one
variant of this type of stormwater management measure and may not be representative of
all types of swale. Additional research is warranted to determine what changes can be
made to reduce the apparent impact that they have on bacterial loading.
EC Summary

During the sampling period, EC counts were not detected (i.e., less than 4 cfu/100mL) in 39% of storm samples and 68% of dry weather samples. In the remainder of samples, EC counts were regularly below the freshwater “designated bathing beach” limit of 235 cfu/100mL, exceeding it only four times (out of 85 samples site-wide) in the storm samples, and twice (out of 238 samples site-wide) in the dry weather samples. Regulators selected EC as an indicator species to represent fresh sources of fecal contamination. During dry weather periods, no fresh fecal material is being washed off of the watershed. By that logic, low or non-detect counts were expected in dry weather samples and were, in fact, observed.

Although it is difficult to build a strong statistical argument with these numbers, it may be reasonable to use them to make some general statements about the facility. First, it seems likely that this site did not receive a significant loading of fresh fecal material, a finding that warrants a closer scrutiny of the higher counts of ENT that were observed. Secondly, the samples where EC counts were above the regulatory threshold were all in the summer or early fall months. Possible factors that may explain this finding include a warm-weather increase in animal activity that leads to additional EC loading on the watershed or the increased survivability and detectability of EC at warmer temperatures (Solic, 1992).

Recommendations For Future Research

Based on the storm event data gathered in this study, several devices were close to achieving statistically significant measurements of their effect on concentrations of fecal
indicator bacteria. Additional storm samples of the bioretention area, infiltration drainfield, wet detention pond, gravel wetland, and rock-lined swale would be useful to validate the findings made thus far. Conversely, it does not seem likely that additional samples will yield significant results for the sand filter, water quality inlet, stormwater filtration chamber, or the hydrodynamic separators.

Bacterial counts were low during the spring and winter and higher in the summer and fall. Counts at both the influent and effluent were affected by these seasonal trends, making it difficult to evaluate treatment effects during the spring and winter when non-detect samples were frequent. Standard practice in water quality monitoring for beach recreation waters is to limit sampling to warmer months when recreational usage is expected to be high. Based on the low concentrations observed in spring and winter, it may be cost effective to focus further research efforts by limiting the sampling period from May to December.

Dry weather sampling provided information about bacterial concentrations in the water column of devices with retained water and in the draining water of devices with a baseflow. Comparing dry weather data to storm data demonstrated that bacterial concentrations increased as a result of storm events rather than as a result of regrowth in the water column or in the draining water of the devices. Although this finding was a useful component of this study, it is separate from the evaluation of treatment effects that are assumed to dominate during storm events and may be omitted in future research. Periodic dry weather screening may be warranted to justify that assumption.

Instead of dry weather sampling, additional sediment sampling may be a more fruitful area for further investigation, as it may provide better information about bacterial
dynamics that ultimately affect bacterial concentrations in the effluent. There is some evidence in the literature that in-stream scour or the rewetting of soils at the margin of a permanent pool may contribute to high counts during storm events (Solo-Gabriele, 2000; Muirhead, 2004). Limited sediment data was gathered during this research and was insufficient to make that determination. Of particular interest would be bacterial counts and particle size distributions from the RLS and the other constructed devices.

This study site does not appear to have significant loading of EC. Although EC is EPA’s preferred water quality standard for fresh water sites, continuing to gather non-detect findings may not be an advantageous allocation of resources (EPA, 2004). ENT limits were also included in the EPA’s rule and may continue to be used for comparative purposes. Although much of the work done so far on microbial source tracking has made use of EC, the infrequent detection of EC at this site may make these methods difficult to apply successfully. Some methods are available for microbial source tracking using ENT, such as evaluating carbon source utilization, ribotyping, or antibiotic resistance.

Finally, as the stormwater facility has the capability to swap out treatment devices, there are some new technologies that claim to remove bacteria from stormwater effluent and may be useful to test them in a field facility. Examples include devices with microbicidal surfaces or filter media. Such products are being marketed, often at high cost, to municipalities and other consumers as solutions to water quality problems. These consumers need unbiased, third-party data to make informed decisions about how best to spend their limited resources in pursuit of water quality goals.
Conclusion

The protection of public health is the dominant goal of water quality regulations related to fecal indicator bacteria. This research adds to the body of knowledge that is available to decision-makers when drafting legislation in support of that goal. As the body of knowledge expands, it may become apparent that earlier strategies, such as the use of EC and ENT as indicator bacteria, may not provide satisfactory outcomes or that certain types of management measures result in unintended consequences that negatively impact other goals. Devices that appear to reduce ENT counts should continue to be studied in order to ascertain if those findings are significant, and the devices put into wider practice if they are found to be effective at meeting water quality goals. Devices that appear to increase ENT counts should continue to be investigated to verify this finding and their use curtailed if proven to do so. As a point of reference, it is important to note that certain conveyance structures may adversely affect runoff water quality, even if it has been effectively treated upstream. For receiving waters that are impaired with regard to indicator bacteria, inputs from stormwater flows are becoming a major concern. It is important for planners to know what effect stormwater management measures have on bacterial concentrations, and this research has attempted to provide useful information in that regard.
REFERENCES


APPENDIX A: UNH STORMWATER CENTER SITE LAYOUT AND DEVICE DESCRIPTIONS
APPENDIX B: RAW DATA TABLES
APPENDIX D: RESULTS OF STATISTICAL TESTING