2950 Niles Road, St. Joseph, MI 49085-9659, USA
269.429.0300 fax 269.429.3852 hq@asabe.org www.asabe.org

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Indicator Bacteria Removal by Bioretention in North Carolina

Jon M. Hathaway

NC State University, Campus Box 7625, Raleigh, NC 27695, jon_hathaway@ncsu.edu

Next Author (repeat for each author)

NC State University, Campus Box 7625, Raleigh, NC 27695, bill_hunt@ncsu.edu

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Abstract. Stormwater runoff is a transport mechanism for indicator bacteria to receiving waters, resulting in an increased risk to public health through consumption of contaminated shellfish or ingestion by swimmers. The resulting economic and public safety concerns are common throughout the United States, particularly in coastal areas. Urban stormwater is commonly treated by stormwater Best Management Practices (BMPs), each of which provides some combination of natural treatment mechanisms and fosters certain environmental conditions. Although BMPs have been studied in detail for many pollutants, there is still a relatively limited understanding of their ability to remove or inactivate indicator bacteria. The North Carolina State University Biological and Agricultural Engineering Department evaluated bioretention areas in Wilmington, NC, to evaluate their efficiency with respect to indicator bacteria removal. Data collected from these studies indicates that positive removal of indicator bacteria is possible in bioretention areas; however, removal can be highly variable from BMP to BMP and from storm to storm. Results also indicated the bioretention cell depth and soil type may influence the effectiveness of these systems.

Keywords. Stormwater, Indicator Bacteria, Pathogens, Runoff, BMP, bioretention

Introduction

In the United States Environmental Protection Agency's (USEPA 2008) National Water Quality Inventory in 2006, approximately 12% of the river and stream miles that were surveyed were impaired by indicator bacteria. Numerous studies have indicated that development in watersheds leads to increased export of indicator bacteria. In a study of 18 mixed land use watersheds in West Georgia, Schoonover and Lockaby (2006) determined that watersheds consisting of greater than 24% imperviousness exhibit higher fecal coliform concentrations than watersheds with impervious percentages less than 5% during both base and storm flow. Studies by Line et al. (2008) and Mallin et al. (2000) conclude similarly that urbanization in watersheds leads to increases in indicator bacteria export. In light of the negative impact that indicator bacteria have on surface waters in the United States (indicating the possible presence of pathogens), TMDLs have been established for impaired water bodies. Municipalities across the country are exploring options to reduce indicator bacteria inputs from point and non-point sources.

To test for the presence of harmful pathogens in surface waters, indicator species are used. In 1986, the EPA's Ambient Water Quality Criteria for Bacteria report (USEPA, 1986) discussed the merits of these various indicator species. Criteria were established whereby *E. coli* and enterococci were suggested as indicators in freshwater environments and enterococci was suggested as an indicator in marine environments. This criteria states that for fresh waters designated for use as full body contact recreational waters, the geometric mean over a 30 day period should not exceed 126 col/100 ml for *E. coli* and should not exceed 33 col/100 ml for enterococci. For marine waters designated for use as full body contact recreational waters, the geometric mean over a 30 day period should not exceed 35 col/100 ml for enterococci.

Indicator bacteria (and pathogens) can be removed from surface waters and stormwater through a number of natural treatment processes, such as photo-degradation (ultraviolet light from sun), sedimentation, and filtration. They also have varied susceptibility to environmental conditions, such as temperature, moisture, and salinity (USEPA 2001; Schueler 2000; Arnone 2007; Davies-Colley et al. 1994). Urban stormwater is commonly treated by stormwater Best Management Practices (BMPs), each of which provides some combination of natural treatment mechanisms and fosters certain environmental conditions.

Bioretention is increasingly being used as part of watershed management strategies in urbanizing watersheds. Bioretention performance for indicator bacteria has not been evaluated in depth, and studies that have been performed were primarily for systems constructed with media consisting of some combination of organic matter, fine particles, and sand or expanded slate fines. However, design specifications for bioretention fill media are typically focused on hydraulic efficiency (i.e., infiltration rate). Thus, it is possible that in-situ soils would be used as bioretention fill media in watersheds containing sandy soils. These potential fill soils have not been tested for indicator bacteria removal when used in bioretention designs. Although there has been some field evaluation performed on bioretention areas for indicator species removal by Hathaway et al. (2009), Li and Davis (2009), Dietz and Clausen (2005), and Passeport et al. (2009), no field evaluation has been performed on bioretention for enterococcus other than a study in New England by Jones et al. (2008).

Due to the limited amount of literature pertaining to indicator bacteria removal by stormwater BMPs, particularly bioretention, more research is needed to aid communities throughout the United States in reaching their indicator bacteria TMDLs. Determining if bioretention areas are capable of efficient indicator bacteria reduction will result in more effective watershed restoration programs. This research is intended to examine indicator bacteria removal in bioretention areas studied in North Carolina.

Materials and Methods

Site Descriptions

The experimental sites were located in Wilmington, North Carolina. Samples were collected between February 2008 and February 2010. The two bioretention areas were located within the same parking lot which serviced a coffee shop (Figure 1). A paired watershed design was sought, with each bioretention having a similar footprint, but watershed area differed due to microtopography within the parking lot. One bioretention was constructed with a soil depth of approximately 60 cm (Bioretention-D), one with a soil depth of 30 cm (Bioretention-S). All fill soil for the bioretention areas came from on site sandy soils. Each cell was constructed with a 10-cm underdrain to facilitate sample collection. It should be noted that underdrains are not typically required for bioretention areas in the sandy soils of coastal areas, thus this design differs from standard practice in the region. Runoff entered each bioretention cell as sheet flow. A small flume was installed at the pavement edge in a location presumed to be representative of the entire watershed. This allowed some pooling of runoff as it entered the bioretention cell, facilitating sampling of the inlet. The bioretention areas were covered with turf grass and had a small number of shrubs. General BMP characteristics are given in Table 1.

Table 1: General characteristics of Wilmington bioretention areas

Characteristic	Bioretention-D	Bioretention-S
Approximate Year Constructed	2006	2006
Drainage Area (ha)	0.10	0.05
Watershed Composition	Commercial (parking lot)	Commercial (parking lot)
Estimated Imperviousness	100%	98%
Primary Surrounding Soil Type (hydrologic group)[1]	Baymeade fine sand (A)	Baymeade fine sand (A)
Surface Area (ha)	0.006	0.006
Surface Area: Drainage Area Ratio	0.06	0.12
Storage Depth (cm)	28	28
Estimated Average Soil Depth (cm)	30	60

1. NRCS 2010 – Soil Data Mart (http://soildatamart.nrcs.usda.gov/)

Figure 1: Illustration of Bioretention-D in Port City Java parking lot.

Monitoring Methods

Short hold times and the increased man-hours and technical difficulty of using automatic samplers for microbial analyses led to the use of grab samples for BMP evaluations. This is a common methodology for sampling surface waters for indicator bacteria (USEPA 2002, Burton and Pitt 2002). One sample set was collected from the inlet and outlet of each BMP for each storm event. Inlet samples were collected for both bioretention areas from the inlet flume mentioned previously. Each sample set consisted of two sterile bottles to facilitate two bacterial analyses (*E. coli* and enterococcus). Outlet samples were collected from each respective bioretention cell's underdrain. There are valid concerns over the use of grab samples, as concentrations of a given pollutant may vary during the course of the storm. However, use of grab samples was necessary in this study, and studies such as McCarthy et al. (2008) have illustrated the uncertainties present in indicator bacteria field monitoring, which potentially overshadow the negative impacts of using single grab samples to some degree.

Samples were transported to Tritest, Inc for analysis. Hold times were generally less than 6 hours. Samples were analyzed for both *E. coli* and enterococcus. *E. coli* was enumerated using Colilert® and enterococcus was enumerated using Enterolert®. Each methodology is based on the use of a defined substrate media (IDEXX Laboratories Inc., Westbrook, Maine). Sample dilutions were performed as needed to adequately characterize bacteria concentrations. The Limit of Detection (LOD) was typically either 2 or 10 MPN / 100 ml depending on the dilution used. The Maximum Reporting Limit (MRL) was typically 24,196 MPN / 100 ml. The MRL for *E. coli* was typically higher than that of Hathaway et al. (2009), allowing a better overall estimation of functionality. Data are analyzed herein using the values at the reporting limit without adjustment.

Statistical Evaluations

Statistical analyses were used to evaluate the performance of each BMP for indicator bacteria. Removal percentages (Concentration Reduction "CR") were calculated for each BMP using a similar methodology to that used to generate efficiency ratios (USEPA, 2002); however, event mean concentrations are necessary to generate efficiency ratios. This was not possible due to the use of single grab samples in this study, leading to the use of Equation 1.

$$\text{[illegible]} = \left(1 - \frac{\text{[illegible]}}{\text{[illegible]}}\right) \times 100\% \qquad (1)$$

Microbial water quality standards are concentration based. Thus, geometric mean effluent concentrations from each BMP were compared to water quality standards for *E. coli* and enterococcus. Based on USEPA recommendations, geometric mean *E. coli* concentrations should not exceed 126 organisms / 100 ml over a 30-day period for fresh water designated as full body recreational waters (USEPA 1986). Similar recommendations exist for enterococcus, whereby geometric mean concentrations should not exceed 33 organisms / 100 ml for fresh waters or 35 organisms / 100 ml for marine waters over a 30-day period (USEPA 1986). A non-parametric Wilcoxon Signed Rank test was used to determine differences among influent and effluent concentrations. Non-parametric analyses also lessen the influence of high and low concentrations, which is important when data sets contain values below the MDL or above the MRL.

Results and Discussion

Summary Statistics

Twenty storms were sampled from each bioretention area between February 2008 and February 2010. Summary statistics for these data are presented in Table 2. Samples were fairly well distributed throughout the seasons, with storm sizes ranging from 0.8 to 12.8 cm. The geometric mean of the influent *E. coli* was 130 MPN / 100 ml for Bioretention-D and Bioretention-S. This was slightly lower than the 241 MPN / 100 ml reported for *E. coli* entering a bioretention area in Charlotte, NC, by Hathaway et al. (2009). The influent geometric mean enterococcus concentration was 375 MPN / 100 ml, similar to influent enterococcus concentrations for BMPs studied by Jones et al. (2008).

Table 2: Summary statistics for monitored storm events

Location	Number of Samples	Statistic	*E. coli* MPN / 100 ml		enterococcus MPN / 100 ml	
			inlet	outlet	inlet	outlet
Bioretention-D	20	geometric mean	130	39	375	39
		maximum	7701	8164	4839	1454
		minimum	2	2	30	2
		standard deviation	2106	1959	1355	323
Bioretention-S	20	geometric mean	130	284	375	378
		maximum	7701	19863	4839	4839
		minimum	2	2	30	20
		standard deviation	2106	5632	1355	1536

Concentration Reduction

Concentration reductions for each bioretention area are documented in Table 3. Bioretention-D performed well with a concentration reduction of 70% for *E. coli* and 89% for enterococcus. However, poor performance was noted for Bioretention-S. For each BMP, removal performance was variable from storm to storm. Individual event concentration reductions varied from greater than 90% to an *addition* of both *E. coli* and enterococcus for both BMPs. Similar inter-event variations in BMP performance for indicator bacteria were showed for bioretention areas by Li and Davis (2009) and for a stormwater wetland by Birch et al. (2004). Statistically significant reductions were only noted for enterococcus in Bioretention-D ($p < 0.05$). Significant relationships can be difficult to find in microbial data sets given the inter-storm performance variability noted in these data and in other studies. Statistical analyses generally support the concentration reduction data, as Bioretention-D was found to perform well.

Table 3: Indicator bacteria concentration reductions for Wilmington bioretention areas

Parameter		Bioretention - D	Bioretention – S
E. coli	Geometric Mean Influent (MPN / 100 ml)	130	130
	Geometric Mean Effluent (MPN / 100 ml)	39	284
	Concentration Reduction (%)	70	-119
Enterococcus	Geometric Mean Influent (MPN / 100 ml)	375	375
	Geometric Mean Effluent (MPN / 100 ml)	39	378
	Concentration Reduction (%)	***89****	-1

* Significant Differences Bold and Italicized

Few field evaluations of indicator bacteria removal have been performed for bioretention, particularly for enterococcus. Studies by Hathaway et al. (2009) on a bioretention area in Charlotte, NC, and Passeport et al. (2009) on two bioretention cells in Graham, NC, indicated high fecal coliform concentration reductions, with all three cells having concentration reductions above 85%. Conversely, evaluations by Li and Davis (2009) on two bioretention areas in Silver Spring and College Park, MD, showed relatively poor performance for *E. coli* (median removal of 0% and 57%, respectively) and fecal coliform (median removal of 50% and 0%, respectively). Likewise, there was a substantial difference in functionality between the two bioretention areas studied in Wilmington, NC. The differing depth of media, nominally 60 cm for Bioretention-D and 30 cm for Bioretention-S, appeared to result in varied performance. Further investigation is planned to explore possible explanations for the difference in performance between cells.

Analysis of Effluent Concentrations

Microbial contamination is regulated by target concentrations established by the USEPA (1986). Thus, effluent BMP concentrations can be compared to these values to determine how they will affect concentrations in receiving waters. Obviously, mass balances would be required to evaluate the full impact of these practices on targeted watersheds. Median effluent indicator bacteria concentrations are shown in Tables 4 and 5.

Table 4: Median effluent *E. coli* concentrations

BMP Type	*E. coli* Concentrations (MPN/100ml)			
	Geometric Mean Influent	Geometric Mean Effluent	Number of effluent samples less than 126 MPN / 100 ml	number of effluent samples less than 126 MPN / 100 ml (percentage)
Bioretention-D	130	39	13	13 of 20 (65%)
Bioretention-S	130	284	7	7 of 20 (35%)

Table 5: Median effluent enterococcus concentrations

BMP Type	Enterococcus Concentrations (MPN/100ml)			
	Geometric Mean Influent	Geometric Mean Effluent	Number of effluent samples less than 33 MPN / 100 ml	Number of effluent samples less than 33 MPN / 100 ml (percentage)
Bioretention-D	375	39	10	10 of 20 (50%)
Bioretention-S	375	378	3	3 of 20 (15%)

The median effluent *E. coli* concentration was below USEPA target concentrations for Bioretention-D. For enterococcus, neither BMP had median effluent concentrations below USEPA targeted values, although Bioretention-D approached the targeted value. Further, neither BMP consistently provided *E. coli* or enterococcus concentrations lower than USEPA targeted values. Bioretention-D provided the highest percentage of effluent *E. coli* and enterococcus samples below target value (65% and 50%, respectively).

Conclusion

Although good performance was noted for Bioretention-D, these data suggest some bioretention areas may export indicator bacteria, as noted with Bioretention-S. Similar results for stormwater BMPs have been seen in such studies at Krometis et al. (2009), Li and Davis (2009), Jones et al. (2008), and Hathaway et al. (2009). These results are not illogical, as indicator bacteria have been shown to persist in sediments of streams and estuaries (Sherer et al. 1992, Jeng et al. 2005).

Bioretention-D showed promise in meeting USEPA target *E. coli* concentrations, but enterococcus geometric mean effluent concentrations were slightly higher than target concentrations. Bioretention-S did not approach target concentrations for either indicator bacteria. Although this creates some concern as to the benefit of stormwater BMPs in watersheds impacted by microbial pollution, a stormwater BMP's contribution to watershed restoration cannot be evaluated based on concentration reduction alone. Reductions in indicator bacteria mass entering surface waters may be achieved through such mechanisms as infiltration. Evaluation of the impacts of infiltration on groundwater microbial quality represents a need within the field of stormwater management, particularly as infiltration-based BMPs become increasingly implemented.

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