# WATER CONTENT EFFECT ON NUTRIENT REMOVAL IN STORMWATER BIORETENTION SYSTEMS

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#### **ABSTRACT**

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Bioretention cells and constructed wetlands are both established best management practices (BMPs) for stormwater quality improvement. These systems vary in terms of hydraulic loading where processes such as retention, sedimentation, absorption, infiltration, filtration, phytoremediation, nitrification and denitrification remove waterborne pollutants. However, the boundary between bioretention and wetlands can be blurred when it comes to design and operational parameters, and it is therefore important to explore the causes and consequences of performance variability in these systems. In an experiment to observe optimum water content for treatment pathways for ecological pollutants, five bioretention bays (2-22% water content) and fifteen bioretention columns (7-47% water content, as much as complete pore space saturation) were used to run parallel tests. Pollutant concentrations were reduced in field bays for COD, TN, and total solids (TS), although there was no difference between treatment groups in terms of any pollutant concentrations. Asclepias incarnata, Carex vulpinoidea, Scirpus validus, and Juncus effusus grew slightly taller in wetter bays, although survival of Sagittaria latifolia was uniformly poor in all treatment groups. No net pollutant removal occurred in columns, although effluent concentrations and mass export were significantly lower for near-saturation treatment groups for chemical oxygen demand (COD), nitrate, and total nitrogen (TN). There was no soil moisture level in which COD, nitrate, TN, phosphate, and TS were simultaneously improved.

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# TABLE OF CONTENTS

LIST OF TABLES	vi
LIST OF FIGURES	vii
KEY TO ABBREVIATIONS	xi
1. INTRODUCTION	1
2. LITERATURE REVIEW	3
2.1. Characterization of Stormwater	4
2.2 Sediment removal	5
2.3 Phosphorus removal	
2.4. Fate of Metals and Salts	
2.5. Nitrogen removal	11
2.6. Microbial Community	
2.7. Wetland Design and Efficacy	
2.8. Bioretention Design and Efficacy	
2.9. Water Content Effects	
2.10. Vegetation in BMPs	
3. MATERIALS AND METHODS	29
3.1. Field-scale bioretention set-up and operation	29
3.2. Laboratory scale bioretention set up and operation	
3.3. Sample collection and determination	
3.4. Statistical analysis	
4. RESULTS AND DISCUSSION FROM FIELD STUDY	39
4.1. Establishment of prolonged water content	39
4.2. Water content effects	
4.2.1. Chemical Oxygen Demand Results	42
4.2.2. Nitrate Results	
4.2.3. Total Nitrogen Results	
4.2.4. Phosphate Results	
4.2.5. Total Solids Results	
4.2.6. Effects of Water Content on Plant Growth	
4.3. Sampling Methods and Technology	
5 RESULTS AND DISCUSSION FROM LABORATORY STUDY	55

5.1. Establishment of Prolonged Water Content	55
5.2. Water Content Effects	
5.2.1. Chemical Oxygen Demand Results	56
5.2.2. Nitrate Results	60
5.2.3. Total Nitrogen Results	62
5.2.4. Phosphate Results	64
5.2.5. Total Solids Results	64
5.3. Summary of Results	67
6. CONCLUSIONS	70
APPENDICES	73
APPENDIX A. Michigan State University Bioretention Field Site	
APPENDIX B. Supporting Data During Research	
APPENDIX C. Enviroscan Data from Establishment Period	
APPENDIX D. Plant Selection and Characteristics	
APPENDIX E. TDR Sensor and Data	
APPENDIX F. Stock water Design	
APPENDIX G. Standard Operating Procedure: Total Solids	
APPENDIX H. Standard Operating Procedure: Nitrate and Phosphate	
APPENDIX I. Standard Operating Procedure: Total Nitrogen and Total Phosphorus	
APPENDIX J. Examining Normality and Seasonality, QQ Plots and Residuals	
APPENDIX K. Photographs from Research	94
BIBLIOGRAPHY	97

# LIST OF TABLES

Table 1. Bioretention and Constructed Wetland Comparison	23
Table 2. Recommended Water Levels	32
Table 3. Synthetic Stormwater	33
Table 4. Synthetic Stormwater applied to Bays and Columns	34
Table 5. Plant Growth in Field Bays	53
Table 6. Statistics Results from ANOVA	68
Table 7. Nutrient Concentration and Treatment Performance	69
Table 9. Stock water design from Lucas and Greenway	80

# LIST OF FIGURES

Figure 1. Sediment circulation in wetland systems	6
Figure 2. Carbon cycling in wetland systems	8
Figure 3. The Phosphorus Cycle	9
Figure 4. Summary of Nitrogen Cycle	12
Figure 5. Treatment wetland types, including horizontal subsurface flow	17
Figure 6. Bioretention schematic	20
Figure 7. MSU Bioretention Research Site.	30
Figure 8. Bioretention field bays	31
Figure 9. Bioretention columns.	35
Figure 10. Water Content in Bays	39
Figure 11. Enviroscan Measurement Summaries in Field Bays	41
Figure 12. Concentration comparison of Effluent and Influent COD in Bays	42
Figure 13. Distribution of COD concentrations in Bays	43
Figure 14. Concentration comparison Influent and Effluent Nitrate in Bays	44
Figure 15. Distribution of Nitrate concentration in Bays	45
Figure 16. Concentration comparison Influent and Effluent TN in Bays	46
Figure 17. Distribution of TN concentration in Bays	46
Figure 18. Concentration comparisons Influent and Effluent Phosphate in Bays	48
Figure 19. Distribution of Phosphate concentration, Bays	49
Figure 20. Concentration comparison Influent and Effluent TS in Bays	50
Figure 21. Distribution of TS concentration, Bays	51

Figure 22. Summary of Plant Growth in Bays	52
Figure 23. Water Content in Columns	55
Figure 24. Influent and Effluent Volumes in Columns.	56
Figure 25. Concentration comparison of Effluent and Influent COD in Columns	57
Figure 26. Distribution of COD concentrations, Columns	58
Figure 27. Distribution of (calculated) COD Export by Mass in Columns	59
Figure 28. Concentration comparison Influent and Effluent Nitrate in Columns	60
Figure 29. Distribution of Nitrate concentration in Columns	61
Figure 30. Distribution of (calculated) Nitrate export by mass in Columns	61
Figure 31. Concentration comparison Influent and Effluent TN in Columns	62
Figure 32. Distribution of TN concentration, Columns	63
Figure 33. Distribution of (calculated) TN export by mass in Columns	63
Figure 34. Concentration comparisons Influent and Effluent Phosphate in Columns	64
Figure 35. Concentration comparison Influent and Effluent TS in Columns	65
Figure 36. Distribution of TS concentration in Columns	66
Figure 37. Distribution of (calculated) TS export by mass, Columns	67
Figure 38. Environmental Conditions, 2014	75
Figure 39. Sample Period Environmental Conditions	75
Figure 40 Enviroscan image	76
Figure 41. Enviroscan Sensor Readings	76
Figure 42. Enviroscan Measurements in Bays	76
Figure 43 Bioretention species details	77
Figure 44. TDR Readings	79

Figure 45. TDR Calibration	79
Figure 46. Bay COD Normality	86
Figure 47. Bay Nitrate Normality	87
Figure 48. Bay TN Normality	87
Figure 49. Bay Phosphate Normality	88
Figure 50. Bay TS Normality	88
Figure 51. Column COD Normality	89
Figure 52. Column Nitrate Normality	89
Figure 53. Column TN Normality	90
Figure 54. Column TS Normality	90
Figure 55. COD Residuals in Bays	91
Figure 56. Nitrate Residuals in Bays	91
Figure 57. TN Residuals in Bays	91
Figure 58. Phosphate Residuals in Bays	92
Figure 59. TS Residuals in Bays	92
Figure 60. COD Residuals in Columns	92
Figure 61. Nitrate Residuals in Columns	93
Figure 62. TN Residuals in Columns	93
Figure 63. TS Residuals in Columns	93
Figure 64. Photograph of entire Farm Lane Bioretention Site	94
Figure 65. Photograph of Hydraulically Isolated Field Bays	94
Figure 66. Photograph of Wetland Overflow Area	94
Figure 67. Bioretention Bays with white PVC water content monitoring ports installed	95

Figure 68. End of season Bioretention Plants	95
Figure 69. Photograph of Laboratory Columns.	96
Figure 70. Photograph of Sampling Bottles	96

#### **KEY TO ABBREVIATIONS**

BMP: best management practice for stormwater; other literature may use synonyms including

Sustainable Urban Development Systems (SUDS), Low Impact Development (LID), and

Stormwater Control Measures (SCMs)

COD: chemical oxygen demand; a summary measure of reagent materials present in water which may contribute to eutrophication

DNRA: dissimilatory nitrate reduction to ammonium; a process of anaerobic respiration

HRT: hydraulic residence time or hydraulic retention time; the amount of time water is within the body of the stormwater system

MSU: Michigan State University, where this research took place

NOx: nitrogen containing compounds, including nitrite (NO<sub>2</sub>) and nitrate (NO<sub>3</sub>),

TN: total nitrogen; a measure of all common nitrogen compounds, including ammonia (NH<sub>4</sub><sup>+</sup>), nitrite (NO<sub>2</sub><sup>-</sup>), nitrate (NO<sub>3</sub><sup>-</sup>), nitrous oxide (N<sub>2</sub>O), dissolved elemental nitrogen or dinitrogen gas (N<sub>2</sub>), organic nitrogen

TP: total phosphorus; a measure of all phosphorus compounds, including dissolved phosphorus and phosphate

TS: total solids; a measure of the mass of all suspended and dissolved solids in water

#### 1. INTRODUCTION

Bioretention basins and constructed wetlands are both widely accepted and utilized best management practices (BMPs) for the reduction of diffuse stormwater pollutants, including sediment, nitrogen, phosphorus, biochemical oxygen demand (a summary measure of oxygen use during decomposition of organic matter), petroleum products, fecal coliforms and metals (Ahiablame, Engel, and Chaubey 2012). These ecological systems combine sedimentation with biotic pollutant fixation and utilization by plants and microbes (Davis et al. 2001). In addition to their abiotic effects (flood mitigation, temperature moderation, etc.), water levels and hydraulic residence times in these systems factor into the composition of biotic life, and therefore sorption, nitrification and denitrification processes (Chen et al. 2013). Although both bioretention basins and wetlands may be designed for vertical, free-surface, or horizontal sub-surface flow, water flow depth and retention time are used to distinguish between bioretention basins and wetlands. There is considerable variability in soil saturation due to environmental conditions (including weather and watershed characteristics) and operation may blur the distinction between a wetland and bioretention cell. Infrastructure designers rely on treatment performance predicted from theoretical hydraulic loading, retention/residence times, evaporative potential, and vegetation density (Vacca 2011). Real-life conditions are not always consistent with hypothetical values, and therefore additional research into unsaturated flow conditions in BMPs is necessary for accurate modeling and the prediction of BMP performance (Barbu and Ballestero 2015).

Michigan State University's Farm Lane Bioretention Research Site provides a unique opportunity to study saturation effects within a bioretention system. The stormwater and groundwater directed into the bioretention site create ponding areas and algal growth similar to conditions found in constructed wetlands. Within this 0.5 hectare bioretention basin, a smaller

area of five hydraulically isolated bioretention bays (12 m<sup>2</sup> each) was modified to allow different amounts of water into each bay (i.e., 10%, 20%, 50%, 80% and 100% of original flow). Soil was removed from the site to fill fifteen replicates in laboratory columns (three at each hydraulic loading level). Both columns and bays were dosed with comparable mass of synthetic stormwater pollution on a regular basis. Bays and columns were also planted with wetland vegetation.

Stormwater treatment is increasingly a priority for both urban and rural development. It is important for designing and modeling BMPs to understand the impacts of water content and soil moisture. The Farm Lane Bioretention site performed inconsistently in its first few years (Thode 2013) and may benefit from more controlled management. This study was intended to compare water quality in bioretention systems maintaining differing water contents to identify soil moisture at which ecological pollutant removal was optimized while controlling for temperature, vegetation type, and soil media. Soil must be aerobic to allow nitrification, with more complete nitrogen removal if denitrification can also occur. Saturated soils may be more prone to mobilization of solids and sorbed pollutants than in a drier soil environment. Wetland plants in these systems were also compared to see if plant growth reflected available soil moisture. Wetland plants were expected to be most prolific in systems where roots had access to moisture, but also to air in pore space.

#### 2. LITERATURE REVIEW

Stormwater volume and quality are affected by climate, atmosphere, and land surface conditions (USEPA 2009). The magnitude and frequency of storms and melting snow loads varies with geography and season, and the resulting flows fluctuate considerably in the concentration of mobilized pollutants (Rimer, Nissen, and Reynolds 1978). As precipitation forms, atmospheric components are dissolved and transported to land. Porous, pervious landscapes allow infiltration into the ground to replenish aquifers, while un-infiltrated runoff flows into surface waters (Russo, Fisher, and Roche 2012). Increasing land disturbance and urbanization have reduced surface conditions that allow for detention and infiltration, increasing problematic stormwater runoff (USEPA 2013a). Stormwater runoff can dissolve or suspend particulates and pollutants, increasing erosion and high turbidity. Runoff from paved or other newly-impervious landscape features change the (pre-development) peak stream flow, in many cases causing habitat and infrastructure damage. Downstream water quality may suffer from increased eutrophication, oxygen depletion, direct pollutant toxicity, and long-term environmental alteration (Eriksson et al. 2007). In more than 770 communities in the United States, stormwater collection drains into the municipal wastewater system, drastically increasing the likelihood of combined sewer overflows with even greater pollution potential (ASCE 2017).

The mitigation of stormwater pollutants is an infrastructure priority since nonpoint pollution (from agriculture and diffuse runoff) is now the largest contributor to of pollution to waters of the United States (ASCE 2017, McMahon 2016). In recent years, the variety and use of BMPs for stormwater have grown nationally and internationally (USEPA 2013b). Stormwater treatment practices are diverse, designed to utilize different storage depths and shapes, various media and cover, site-specific design features (like recreational goals) and management

recommendations. Many BMPs aim to recreate the retention, infiltration, filtration, adsorption, microbial activity, and vegetation of pre-development hydrology within each watershed. Water quality improvement occurs in following processes: sedimentation and filtration of solid particulates, sorption of soluble pollutants onto soils, degradation by microbes, pollutant uptake by plants, and water storage (USACE 2013, USEPA 2013b). Many pollutants of concern have unique pathways within these ecological processes.

## 2.1. Characterization of Stormwater

Stormwater runoff analysis must consider a multitude of components: precipitation, groundwater transport, municipal wastes, animal and insect detritus, erosion sediments from natural and man-made features, nitrogen and other nutrients, and industrial wastes (Rimer, Nissen, and Reynolds 1978). Among potential pollutants, several are ranked as priorities because of their immediate and long term effects. BOD, COD, erosion and suspended solids, pH and nutrients can impact ecosystem health and risk eutrophication, oxygen depletion, aesthetic problems, direct pH toxicity effects, and long-term changes in aquatic habitability (Eriksson et al. 2007). Fecal pollution can cause intestinal distress in humans. Metals and polycyclic aromatic hydrocarbons can be acutely or chronically toxic to humans and other biota. Herbicides also have detrimental ecological impacts. Many additional industrial chemicals are soluble or semi-soluble and persistent pollutants with effects ranging from endocrine disruption to cell death; these are more often managed with specialized removal programs (Cross and Duke 2008). Water resource policies must reflect each of these and consider the magnitude, distribution, and fate of each constituent, as well as means and limits of control. This research focuses on typical ecosystem hazards rather than toxic pollutants.

Stormwater is characterized not only by its pollutant components but by the timing of their transport and concentration (Lee and Bang 2000). The 'first flush' mobilizes topical residue on surfaces, which accumulate during times of lower precipitation, and flood the system when rainfall first occurs. When it occurs, this first flush typically has the highest concentration of contaminants and therefore is a priority for capture, although indicator bacteria have proven an exception to the first-flush phenomenon (Hathaway 2010). The ability of BMPs to manage this volume depends on the time of peak pollutant concentration in the stormwater flow (which differs from the time of peak volume) and appropriate storage and treatment design.

#### 2.2 Sediment removal

Wetlands and other small water bodies are estimated to receive 30% of all eroded material in the US (Maynard, Dahlgren, and O'Geen 2011). The removal of sediment relies on well-known principles of sedimentation and filtration: slower velocities allow greater deposition than high velocities and larger particles precipitate more rapidly than smaller particles (Kadlec and Wallace 2009). These principles are true in surface flow and subsurface flow. The various forms of sediment migration are shown in Figure 1. Suspended sediment acts as a substrate for many sorbtive pollutants, contributing to transport and precipitation of heavy metals, bacteria, phosphorus, and carbon (Kadlec and Wallace 2009). Relatively fine media slows flow rates and increases the potential for sedimentation, interception, and dispersion of fine particulates (Hunt, Davis, and Traver 2012). Efficient surface filtration minimizes the importance of media depth and sediments most often accumulate in the top few centimeters of a BMP, particularly for heavy metals (Li and Davis 2008, Wang et al. 2017). Particulate carbon is usually removed in the sediment layer even if dissolved organic carbon may remain largely unaffected (Maynard,

Dahlgren, and O'Geen 2011). Macrophytes assist in the sedimentation process by reducing preferential flow and severely limiting resuspension opportunity (Baskerud 2001).

Figure 1. Sediment circulation in wetland systems

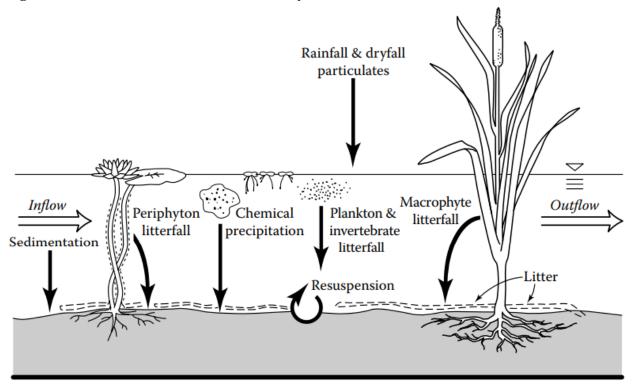


Image by (Kadlec and Wallace, 2009) page 207

Horizontal sub-surface dynamics include elements of the above figure, although sediment removal via granular bed filtration is dominated by three well-known mechanisms. In fine-grained media, inertial deposition or impaction into the media and diffusional deposition move particles to an immersed surface (Kadlec and Wallace 2009). Flow line interception is the primary removal in coarse-grained media where biofilm may cause sediments to stick.

Resuspension is much less common in low velocity sub-surface flow.

Sediments can be composed of inorganic or organic materials. Biochemical oxygen demand (BOD) and chemical oxygen demand (COD) measure the amount of organic material in wastewater. BOD is based on oxygen consumption of microorganisms during oxidation of

organic matter over the course of five to seven days. COD uses a chemical oxidant (usually potassium dichromate) to oxidize organic matter, a faster and more extensive oxidation which can yield oxygen measures that are double BOD measures in municipal wastewaters and up to 20 times BOD in more dilute systems (Kadlec and Wallace 2009). Good quality secondary effluent BOD might range from 10-20 mg/L (approximately 40-80 mg/L COD) after initial concentrations of up to 1000 mg/L BOD in raw sewage (Pescod 1992). Natural wetlands typically have considerably more organic carbon than constructed systems (15.2 vs 3.1%) (Fennessy, Rokosch, and Mack 2008) and this available carbon provides for enhanced denitrification rates (Burchell et al. 2007). Repeated dry and wet cycles increase microbial respiration and microbial biomass, increasing carbon dioxide release and carbon mineralization, although repetition and soil organic matter unavailability may stress these microbes (Xiang et al. 2008). Total carbon mobilization was generally higher in an alternating wet/dry soil than in soils with consistent moisture. This may cause leaching in bioretention systems designed for relatively rapid and frequent changes in inundation.

Carbon mineralization declines in extremely moist and anaerobic conditions in the short term, leading to carbon accumulation in soil. However, elevated moisture for weeks or months at a time in the presence of iron (Fe) reduction destabilizes carbon into CO<sub>2</sub> and CH<sub>4</sub>, releasing carbon in the long term (Huang and Hall 2017). Figure 2 details the energy exchanges which utilize carbon and other substrates. Wetlands and bioretention systems most commonly engage the processes in Zone I-III as indicated by the dotted line. A balance of oxygen availability and carbon substrate diffusion in soil water is believed to be optimal for organic matter decomposition, although mineralization is improved in saturated conditions with time.

 $O_2$ Zone I E<sub>h</sub> = > 300 mV Aerobic Sulfide Organic Methane Nitrification  $O_2$  $CO_2$ Matter oxidation oxidation respiration NH<sub>4</sub> NO: NO<sub>2</sub>  $N_2$ SO<sub>4</sub> Zone II and III N<sub>2</sub>O Dissimilatory  $E_h = -100 \text{ to } 300 \text{ mV}$ Organic nitrate matter Facultative anaerobic respiration NH4 reduction  $CO_2$ Energy H<sub>2</sub>O Mn4+ Reduction Fe3+ (Fe<sub>2</sub>O Reduction MnO Mn<sup>2+</sup> Fe<sup>2+</sup> Amino Acids Carbohydrates Organic H<sub>2</sub>S Sulfate Long chain matter fatty acids CO2  $1_2S$ Energy Zone IV and V  $E_{\rm h} = -300 \text{ to } 100 \text{ m}$ Short Anaerobic respiration CH<sub>4</sub> Acid chain Methane fatty fermentation formation Fe S  $CO_2$  $H_2$ 

Figure 2. Carbon cycling in wetland systems

Image by Kadlec and Wallace, 2009, page 241

## 2.3 Phosphorus removal

Phosphorus is present in dissolved and particulate forms. In both forms, it is an environmental concern as a limiting nutrient for algal growth in many freshwater systems. This biotic growth plays a role in oxygen-depleting eutrophication (Roy-Poirier, Champagne, and Filion 2010b, Morgan et al. 2011). Phosphorus may undergo many transformations: precipitation from a fluid, dissolution within a fluid, fragmentation in soil media, leaching out of compounds, mineralization in subsoil, or burial beneath other sediments. The most active zone of transformations in an unsaturated soil environment is indicated by the dotted line in Figure 3.

KEY PO-3 phosphates SOP soluble organic phosphorus POP particulate organic phosphorus calcium phosphates iron phosphates aluminum phosphates Inflows Air (runoff. tides, etc.) Plant/microbial uptake Sedimentation Surface water Particulate Oxidized inorganic P 3\_Adsorption 4 precipitation soil layer including Ca-P

Figure 3. The Phosphorus Cycle

Image by Mitsch and Gosselink (2015), page 202

Reduced soil layer

Filtration and sorption of phosphorus in soil, and its subsequent uptake and assimilation by microbes and plants, requires a balance of contact time and biological nutrient uptake capacity. Phosphorus precipitation may take days, while sorption may occur in a matter of hours (Li and Davis 2016). These processes can be reversed through desorption and dissolution of phosphorus in flooded soils. Repeated drying and wetting in floodplain sediments increased phosphorus release, especially when extremely dry (Schönbrunner, Preiner, and Hein 2012). Dry-out allows oxidation and mineralization, priming a system for export during the next rainfall event (Kadlec and Wallace 2009). Plants and microbes use phosphorus in their growth and

Upward diffusion

SOP

Plant uptake

POP

Anaerobic

release of P

release phosphorus in their decay. The ratio of organic carbon to organic phosphorus on a molar basis is critical to the fate of these nutrients (Li and Davis, 2016). Because of these complex interactions, phosphorus removal varies widely in BMPs, from 40-60% removal in some wetlands (Vymazal 2007) to removal in some bioretention systems of 70%-85% or, in some cases, increased concentrations up to 240% (Davis et al. 2012).

Effluent phosphorus concentrations are based on media equilibrium (Li and Davis 2016). Models predicated on soil type and retention time more accurately predict phosphorus behavior than output models based on regression alone. Amorphous iron oxide and aluminum oxide contents improve phosphorus sorption; therefore the oxalate ratio (Al<sub>ox</sub> + Fe<sub>ox</sub>, mmol/kg and P<sub>ox</sub>, mmol/kg) has also been used as a measure of adsorption/leaching potential. Iron shavings, steel wool, water treatment residuals and fly ash have been recommended as amendments for improved phosphorus removal (Zhang et al. 2008, O'Neill and Davis 2012). The sorption and exchange relationships can be highly pH dependent, as phosphorus becomes less available with increased pH.

#### 2.4. Fate of Metals and Salts

Metals undergo many of the same mechanical processes as phosphorus, settling out of stormwater at low water flow velocities to be adsorbed by soil particles and fixed within plants and microbes. Iron, aluminum and magnesium are ubiquitous in soils, but heavy metals such as lead, copper, cadmium and zinc present a toxicity hazard for humans and other biota. Soil moisture can directly affect soil pH, as water content moves acidic or alkaline soils closer to neutral (Ma et al. 2017). Most metals of interest carried by stormwater (notably lead, copper, and zinc) are primarily trapped in the top 20 cm of the soil profile (Davis et al. 2012). Metals can also then be remobilized by desorption and dissolution in continued water flow (Nichols and

Lucke 2016, Pitt et al. 1995). Bioretention systems have been found to be effective in reducing metals an average of 30-99% (Ahiablame, Engel, and Chaubey 2012), often for many years, as in an eleven year study for copper and zinc (Johnson 2016).

Road salt and other industrial chemicals can also contribute to stormwater pollution. A small amount of sodium is utilized by plants in regulating osmotic pressure, and the rest is flushed through the stormwater system (Kadlec and Wallace 2009). Fluoride adsorbs to soils while fluorine can be taken up by plants. Chloride and bromide are largely unaffected by filtration or biota, although chlorine can have toxic effects on microorganisms, sterilizing soils (Robinson, Hasenmueller, and Chambers 2017). Chlorine specifically can be converted in solution to chloramines, a more toxic pollutant, when combined with ammonia or nitrogenous compounds. Chlorine can also retard the adsorption of metals on sediment (Søberg, Viklander, and Blecken 2017). Volatilization, adsorption, chemical oxidation and photochemical oxidation can transform chlorine into peroxides or other stable compounds. These treatment means are all relatively ineffective for salt in comparison with the extent of road salt use and soil salinization and, therefore, experts strongly recommend prevention as the only reasonable approach to salt pollution (Talend 2016). Stormwater control measures may actually escalate negative impacts of salts by concentrating and distributing salt into groundwater throughout the year (Snodgrass et al. 2017).

### 2.5. Nitrogen removal

Every stage of the nitrogen cycle can be found in wetland and bioretention systems, as shown in Figure 4. Atmospheric nitrogen can be fixed by algae, cyanobacteria, leguminous bacteria and other microorganisms into organic nitrogen and ammonium. Ammonium may be used by plants and microbes, flux into ammonia and volatilize or be reduced into nitrite and

nitrate. This nitrification occurs in the soil and nitrates may then be leached into groundwater or be taken up by plants. Denitrification may occur in the absence of oxygen where carbon is present, transforming nitrates in to atmospheric carbon. Or anammox may occur without oxygen or carbon, transforming ammonium into atmospheric nitrogen.

KEY dinitrogen nitrous oxide ammonia ammonium ion nitrite nitrate soluble organic N Fixation NH<sub>3</sub> N<sub>2</sub> N<sub>2</sub>O Inflows Fixation Volatilization Air Algae NH<sub>3</sub> Surface water Organic N SON runoff, leaching Nitrification Oxidized Organic N SON-NO. NO: soil layer diffusion Upward diffusion Nitrate reduction NO. Reduced soil layer Denitrification Plant uptake No No

Figure 4. Summary of Nitrogen Cycle

Image by (Mitsch and Gosselink 2015), page 183

Both abiotic and biotic effects occur during the nitrogen cycle, but the pollutants ammonia and nitrate are mostly removed by biotic transformations within ecological systems (Kadlec and Wallace 2009). Nitrogen compounds are used in cell synthesis by aerobic bacteria, which use oxygen for metabolism, by obligate anaerobic bacteria, which grow in anoxic or

hypoxic conditions, and facultative anaerobic bacteria which can survive with or without oxygen. After fixation by plants and microbes, organic nitrogen from plant detritus, fecal matter, and other biological materials are enzymatically processed into ammonium by ammonification (Lyon, Buckman, and Brady 1952). At low hydraulic loads, organic nitrogen concentrations may remain unchanged in wetlands, but higher hydraulic loading rates can wash these compounds from soil media, causing increased effluent organic nitrogen concentrations by 22-31% (Crumpton and Goldsborough 1998).

Ammonium (NH $_4^+$ ) is the common form of ammonia in soil due to the neutral or slightly acidic nature of typical soils, but ammonium changes to free ammonia in alkaline conditions and at high temperatures (Kadlec and Wallace 2009). NH $_4^+$  diffusion from anaerobic ( $E_H = -700 \ to \ 300 \ mV$ ) soils to aerobic soils and the rate of ammonium oxidation are relatively slow processes, compared to nitrate diffusion into the anaerobic layer and NO $_3$  reduction in the anaerobic layer, and are therefore controlling transformations in the fate of nitrogen in flooded soils (Reddy, Patrick Jr., and Phillips 1980). Nitrate may also undergo dissimilatory reduction to ammonium nitrogen (DNRA), a pathway which may dominate in carbon-rich and alkaline environments (inversely to redox potential). Both plants and animals can uptake ammonium. The conjugate base form of ammonium, ammonia, is volatile and thus short-lived in soil systems. Chemoautotrophic and heterotrophic aerobic bacteria known as nitrifiers oxidize ammonium into nitrate (Kadlec and Wallace 2009). This transformation releases hydrogen ions and lowers pH.

Nitrate can be taken up by microorganisms and higher plants as a nutrient. It is highly soluble and may leach and contaminate drainage waters (Passeport et al. 2013). Unionized ammonia is the most toxic form of inorganic nitrogen compounds in an aquatic environment, while nitrate has relatively low toxicity (Camargo and Alonso 2006). In high doses, however,

nitrate can be toxic to human infants (in the case of methemoglobenemia or "blue baby" syndrome) and may produce organ damage in adults with long term exposure.

When oxygen is not present in soil environments, nitrates can be reduced to nitrogen gas by denitrifying bacteria. In order to achieve this more complete nitrogen removal, research recommends anoxic, saturated zones or internal water storage within bioretention treatment systems (Kim, Seagren, and Davis 2003, Brown and Hunt III 2011). Vertical flow wetlands and horizontal wetlands have limited denitrification or ammonia nitrification, depending on their drainage scheme, and therefore designers commonly recommend a hybrid combination (Vymazal 2007). Dissolved oxygen measurements in surface waters are not always a reflection of anoxic conditions in deeper levels where denitrification continues to take place (Bachand and Horne 1999). Research by Crumpton and Goldsborogh (1998) concludes that denitrification is essential to create net removal of nitrogen in wetlands at all.

In the case of Anammox, gaseous nitrogen can be formed from ammonium without a carbon source (Kadlec and Wallace 2009). During this transformation, ammonium and nitrite are metabolized to create nitrogen gas and water by *Plancomycetes* and *Nitrosomonas eutroph*, bacteria found in subsurface flow and free water wetlands. Compared to the conventional nitrification/denitrification relationship, the Anammox pathway requires half the amount of oxygen no carbon requirement. However, it is apparently less common in freshwater ecosystems, and is studied comparatively less. Anammox processes are dominated by denitrifiers at COD/N ratios greater than 1-6 g COD/g N, since denitrifying bacteria can multiply up to 100 times faster than *Nitrosomonas* (Hou et al. 2018). Aerobic denitrifiers are commonly found in soils, with denitrification effects equal to those of anaerobic processes in one study, but more study on

aerobic denitrifiers is needed for a complete understanding of their role in stormwater BMPs (Song et al. 2010).

Wetlands have been found to remove nitrogen, with typical values 78-95% nitrate removal and 54-74% TN removal annually (Phipps and Crumpton 1994). Total Nitrogen removal in wetland environments is estimated to be 1-34% by assimilation and 60-95% by denitrification (Lee, Fletcher, and Sun 2009). Nitrifiers function best near a pH of 7.2 and denitrifiers function best in a range of 6.5 < pH < 7.5 (Bachand and Horne 1999), although the same study found that nitrification was limited at colder temperatures regardless of pH.

## 2.6. Microbial Community

The microbes in stormwater systems perform many processes in addition to the specific nitrogen and phosphorus reactions already described. The microbes present in soils are specific communities dependent on soil type, plant type, porosity and migration, with increased diversity where there is predation opportunity and nutrient availability (Vacca et al. 2005). Drying and wetting processes alter bacterial communities composition, in part because of enzyme effects occurring with a time delay from the conditions of their induction (Banerjee et al. 2016). Most bacterial groups show a relatively small magnitude of population change in studies of seasonal drying and wetting regimes (Barnard, Osborne, and Firestone 2013). The relatively dramatic exceptions are *Actinobacteria* which increase in abundance with desiccation and decrease with rewetting, and *Acidobacteria* which decrease in dry times and increase with rewetting. These two groups are the active communities in the nitrogen cycles described previously. Multiple cycles of drying and wetting increased subsurface soil microbial biomass and activity as much as 8-fold, even while surface communities remained relatively stable (Xiang et al. 2008). The Xiang study

found a more dynamic environment than typical vertical flow profiles in which the surface is relatively diverse and resource rich while the subsurface is more consistent and resource poor.

A small fraction of the microbes present are considered pathogens, most of these brought into BMPs by stormwater runoff (Hathaway and Hunt 2010). *Escherichia coli*, total coliforms, fecal coliforms, and fecal streptococci are the most common indicators for pathogenic microorganisms. Pathogen removal efficiencies in constructed wetlands can reach 88-99% for *E. coli* and enterococci, with slightly lower numbers for streptococci, 80-95% (Vymazal 2005). Once a treatment system is built, water level and retention times are flexible operation elements in BMPs that can be modified to address specific pollutants of concern (Passeport et al. 2013).

## 2.7. Wetland Design and Efficacy

Constructed wetlands are categorized into subsurface flow wetlands or free water surface (flow) wetlands, with variations for vertical or horizontal subsurface flow, flood-pulse flow or hybrid combinations(Wu et al. 2015, Kadlec and Wallace 2009) as in Figure 5. Most bioretention basins resemble vertical or horizontal subsurface flow conditions, although flooded conditions may resemble surface flow wetlands (DEP 2007). Natural and constructed wetlands are characterized by surface or near-surface water levels that are sufficient to support vegetation adapted for saturated soils (USEPA 2017). The three parameters for wetland classification include the 'positive indicators' of hydrophytic vegetation (at least 50%), hydric soils, and wetland hydrology (Tiner 1993). During dry periods, hydrology indicators include oxidized rhizospheres, water-stained leaves, and vegetation characteristics indicating adaptation to saturated soils (such as shortened roots). Anaerobic soil conditions can occur within a day or two of flooding (Barnard, Osborne, and Firestone 2013). In a study of horizontal constructed wetland configuration, free-water surface wetlands had a tendency to short-circuit gravel layers and

reduce effective volume (Pedescoll et al. 2013) as water followed the path of least resistance. Plants were the most effective means of initiating flow subsurface flow within the gravel matrix in the Pedescoll study (2013). There are many varieties of wetlands, as shown in Figure 5, but treatment wetlands are most often designed for subsurface flow.

Treatment Wetlands Subsurface Flov Surface Flow **Floating** Submerged Emergent Horizontal Vertical Plants Plants Plants Flow Flow Top of gravel bed Water level Influent⇒∑ Water level control ⇒ Effluent Coarse media Impermeable liner Main bed media

Figure 5. Treatment wetland types, including horizontal subsurface flow

Images from (Kadlec and Wallace 2009), pages 5 and 6, modified by R. Bender

In constructed wetlands, hydrologic conditions are such that the substrate is saturated long enough during the growing season to create oxygen-poor conditions in the substrate.

Saturated or near-saturated pore space creates reducing (i.e. oxygen-poor) conditions within the substrate and limits the vegetation to those species that are adapted to low-oxygen environments (Davis 1994). Likewise, the presence of a saturated zone and an oxygen-consuming carbon source allows ion exchange for stabilization of heavy metals and phosphorus (Blecken et al. 2009). Saturated conditions physically facilitate denitrification of nitrate compounds into gaseous nitrogen, while living microbes cause ammonification and nitrification (Lee, Fletcher, and Sun 2009). Water quality modeling in constructed wetlands most often uses steady-state first-order plug-flow models for TP, ammonia, and nitrate, although length-to-width ratios and vegetation density are clearly factors in performance as well (Carleton et al. 2001).

Sizing is one of the most critical design components for stormwater treatment wetlands. Early designs were based on an empirical rule of wetland-to-watershed area ratio of 2% (Kadlec and Wallace 2009), which is simple to calculate but not meaningful for treatment optimization. A second approach specifies the capture and detention of some portion of expected runoff (usually 90%), which should incorporate rain frequency and inter-event periods. This approach targets flood mitigation and storage in wetlands and needs refinement to design for specific pollutants of concern. A third method adapts continuous-flow rate constants for event-driven systems, working backwards from required effluent water concentrations to design a sufficiently sized wetland system. These sizing criteria were developed in order to optimize the influent and effluent rates based on watershed and outlet characteristics, controlling for hydraulic residence time and flood storage capacity. Stormwater wetlands in particular are designed to undergo significant drying compared to many other wetland types. Like bioretention systems, stormwater wetlands include an important biological component compared to wet ponds or detention ponds used as stormwater BMPs, so conditions must be suitable for plant survival (SEMCOG 2008).

Guidance for stormwater wetlands from the Michigan low-impact development manual includes a minimum length-to-width ratio of 2:1 for sedimentation, side embankment slopes no steeper than 4-5:1, average depth of 1-2 meters, safety benches at greatest depths, minimum 0.3-m freeboard, limited woody vegetation (where structure includes embankments or other confining layers), and accommodations for wildlife and human access (SEMCOG 2008). Outlet controls and pretreatment areas (like forebays) are also recommended. Hydrologic soil groups "C" and "D" are suitable for confining layers beneath a basin, although soils "A" and "B" may be used with a synthetic or clay liner. Plant and microbial biomass will naturally occur and increase in wetlands, and open water zones should be maintained as 35-40% of total surface area.

The primary design parameters for detention and retention-based systems (like wetlands or detention basins) such as total storage volume, discharge rate, flow path length, control or affect hydraulic residence time (Geosyntec Consultants and Wright Water Engineers 2013). However, tightly managed hydraulics of constructed systems rarely mimic natural systems and therefore researchers struggle to identify reference conditions (Vacca 2011). Failures are often related to lack of understanding of biogeochemical processes or pollutant removal mechanisms and overreach of statistical models.

### 2.8. Bioretention Design and Efficacy

Bioretention systems, e.g. "rain gardens" or "bioinfiltration" systems, are designed to collect and filter water to moderate flow speed and volume after a rain event (Figure 6).

Bioretention is a relatively new technology, and therefore exact specifications and treatment expectations of these structures are still developing (Davis et al. 2012). Many bioretention guidelines are based on original guidelines developed in the 1990s in Maryland. Bioretention design emphasizes rapid infiltration so that complete drainage of ponding, with depths of 14-45

cm (6-18 inches), should occur within 48 hours of a rain event; designs may include an underdrain to facilitate this timeline (SEMCOG 2008, Davis et al. 2012). This drainage regime makes for a relatively dry system best suited to minor storm events (James and Dymond 2012).

Figure 6. Bioretention schematic

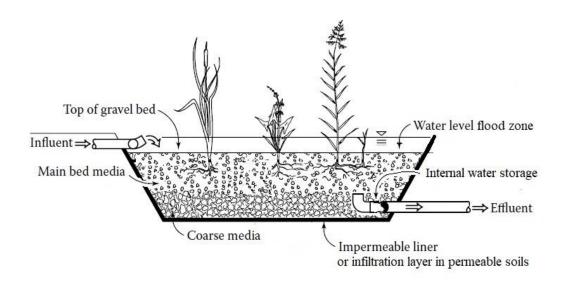


Image modified from Figure 5 by R. Bender

If bioretention is built on relatively impermeable soil, an underdrain with at least 0.5% slope should be installed and covered with a layer of gravel (MDEWMA 2000). The underdrain or underlying soil is covered with 0.3-2 m sand-based planting mix. The Maryland bioretention design manual suggests the homogenous planting mix should have pH of 5.2-7.0, 1.5-4% organic matter, and sufficient magnesium, phosphorus, and potassium to support plant life. Rapidly-draining treatment media (200 mm/h) is recommended to allow aerobic areas around plant roots and to encourage infiltration. However, increases in dissolved organic carbon availability, nitrate concentrations, and hydraulic residence time increases denitrification potential and can facilitate phosphate release (Thomas, Yeh, and Ergas 2015). Soil media should be covered by up to 5 cm shredded hardwood mulch in order to reduce preferential flow and other erosive conditions.

Uniform downward flow is ideal for maximum treatment and modeling, and bimodal pore size distribution may increase likelihood of preferential flow (Liu and Fassman-Beck 2017) The depth of the bowl (freeboard) is recommended to be 15-30 cm in order to prevent drowning vegetation, to reduce compaction beneath ponding, and to protect health and safety of humans (Hunt, Davis, and Traver 2012). Native floodplain or wet meadow plants are recommended for this dynamic habitat. A saturated anoxic zone with an overdrain (created by adding a bend to the underdrain) is proven and recommended to improve denitrification if an electron-donor substrate can be established and maintained (Kim, Seagren, and Davis 2003). Internal water storage reduces TN and TP concentrations, although soil type and respective infiltration rates should be used to determine depth and dimensions for media and underdrain (Brown and Hunt III 2011).

Designers may adjust bowl volume, engineered media composition, media depth, underdrainage and vegetation type to optimize bioretention for specific needs (Hunt, Davis, and Traver 2012). The required volume for treatment varies in different jurisdictions, but is often based on some combination of water quality volume (e.g. 90% of predicted watershed runoff from likely rain event), recharge volume, channel protection requirements, overbank flood protection, and extreme flood volumes (MDEWMA 2000). Some bioretention systems have no underdrain or confining layer, but instead allow for direct infiltration into subsoil (Davis et al. 2012). Other systems may be designed in anticipation of specific water quality concerns, such as soil media designed for greater phosphorus adsorption (Li and Davis 2016). A factor-of-safety (perhaps 10% or more) should be used in estimates of bioretention sizing and the expected effluent concentrations in order to accommodate partial failure of stormwater control measures (Blecken et al. 2017). Like wetlands, bioretention systems are known as both a source and a sink for diffuse pollutants, since pollutant settling and plant decay can provide organic and nutrient

material for large 'wash out' events (Mullane and Flury 2015, Brown, Birgand, and Hunt 2013). Infiltration basins, which have failed in their original purpose and instead taken on wetland characteristics are called "transitioned" basins (Natarajan and Davis 2016b). Clogging from sediment deposition, irregular maintenance, improper sizing or poor design can all lead to improper ponding. Clogging is unavoidable due to the necessity of settling, but it can be delayed by maintenance of surface deposits, vegetation removal and pre-treatment component design (Pedescoll et al. 2013). Even when infiltration declines, detention, retention, and evapotranspiration still occur. Thus, transitioned basins continue to remove 65-95% of total suspended solids, copper, lead, zinc, TP, dissolved phosphorous, NOx, TKN, organic nitrogen and chloride (Natarajan and Davis 2016a).

A summary of bioretention and constructed wetland characteristics explained in the literature review thus far is included in Table 1. Many similarities exist, including wide margins of effectiveness in pollutant removal.

**Table 1. Bioretention and Constructed Wetland Comparison** 

DESIGN	Bioretention	<b>Constructed wetland</b>
Loading	Minimum 20% of	Minimum 2-3% of
	watershed	watershed
Draw down	48 hours	None
Treatment layer	1-10 % organic	No organic % specified
	High sand content	Sand or gravel
Internal carbon	Substrate recommended	Plant growth and decay
Internal water storage	Optional	Required
PERFORMANCE	Bioretention	Constructed wetland
COD/BOD	22-55% OR -94%	up to 99% OR <-100%
Nitrogen	65 – 95%	54 – 74%
Nitrate	65 – 95%	78 – 95%
Phosphorus	70 – 85% OR <-240%	40 – 60%
Sediments	86-92% OR -12%	90-92%

## 2.9. Water Content Effects

Among the primary goals of bioretention and stormwater wetland construction are mitigation of peak flows, improved infiltration, pollutant removal, and controlled release of stormwater (Hunt, Davis, and Traver 2012). Modifications to achieve these effects include increased media-to-runoff volume ratios, deeper media depths, internal water storage, and deeply-rooted, transpiring vegetation. Stormwater BMP design must consider these modifications in terms of their effect on pollutant removal. Hydraulic residence or retention time has a positive linear relationship with treatment effectiveness for parameters like bacteria and sediment, although longer hydrologic residence times decrease removal of phosphorus and some metals when redox occurs (Diaz, O'Geen, and Dahlgren 2012, Thomas, Yeh, and Ergas 2015). These complex dynamics create conditions where a stormwater system can be a sink for nitrate at the same time that it is a source for phosphorus (Chang, Hossain, and Wanielista 2010, Read et

al. 2008). Likewise, stormwater BMPs can be a sink for pollutants at lower hydraulic loading, and a source for pollutants under higher hydraulic loading (Geosyntec Consultants and Wright Water Engineers 2013).

Soil moisture content profoundly affects microbial activity. Soil moisture reduces gaseous diffusion rates and increases liquid diffusion rates, transporting ammonia, nitrate, and soluble organics (Banerjee et al. 2016). A drying period without rain was critical during a comparison of two years of wetland treatment performance (Jordan et al. 2003). Pollutant removal efficiency declined 59% for phosphorus, 38% TN and 40% total organic carbon without a comparable three month drying period in the second year. Theoretically, changes to the water table during drying conditions allow more even distribution of resources within a soil system, even if bacterial diversity decreases overall (Banerjee et al. 2016). The legacy of prolonged hydration affects diversity in an ecosystem even after hydrologic conditions have changed, leaving variable ecosystems more resilient to future disruptions (Peralta et al. 2014). Microbial diversity which includes fungal community retains nutrients best of all because soil fungi are more resistant to moisture fluctuation than bacteria (Gordon, Haygarth, and Bardgett 2008). Mechanically, soil moisture fluctuation can break up soil aggregates and expose new substrate (Manka et al. 2016). Low moisture conditions and low pore connectivity has been shown to increase microbial diversity by allowing greater survival of isolated microbial prey species (Carson et al. 2010).

Stormwater systems with a drawdown time greater than the typical frequency of rainfall events will experience a greater incidence of overflow events with little or no treatment at all (Smolek, Hunt, and Grabow 2015). Considerable research has shown dry initial soil conditions increase preferential flow and decrease lateral flow due to hydrophobicity and channeling,

although some research does show deeper penetration of tracers in saturated, well-structured soils (Merdun, Meral, and Riza Demirkiran 2008). Infiltration rates are relatively high when ponding increases head or when water temperatures are relatively warm (Lewellyn et al. 2016). Macropores play a greater role in chemical transport in drier soil, while diffuse, dissolved transport may dominate in saturated conditions such as ponded wetlands. Flow models have been developed to improve predictive equations (i.e. Richard's equation) for unsaturated flow (Browne et al. 2008). These models incorporate surrounding soil moisture and ponding conditions, but these purely hydrologic models do not yet reflect moisture and infiltration effects on water quality.

## 2.10. Vegetation in BMPs

Ecosystem-based structural BMPs are designed to include vegetation in order to promote both physical and chemical processes. Nutrient removal from stormwater can be markedly improved by incorporating vegetation into infiltration systems, particularly for removal of TN (Lucas and Greenway 2008b). Studies have found linear correlations between ammonium concentration in the rhizosphere and plant transpiration, indicating that transpiration increases the efficiency of nitrogen removal (Wiessner et al. 2013). Transpiration and physiological activity (including enzymes and microbial symbiotes) decreases methane and ammonium concentrations by expanding the oxygenated zone. Plants like *Juncus effusus* were found to uptake 44.5% of the ammonium-nitrogen load during mesocosm experiments with synthetic wastewater (Wiessner et al. 2013). Specific plant genera, including *Juncus, Carex*, and *Scirpus/Schoenoplectus* (Vymazal 2013), have been shown to remove up to 80 g/m<sup>2</sup> N and 14 g/m<sup>2</sup> P more than other species (Tanner 1996), although this observation can be simplified, in part, by an inverse correlation between total biomass/root mass and effluent concentrations of

TN, total dissolved nitrogen, ammonium, total dissolved phosphorus and filterable reactive phosphorus (Read et al. 2008). These positive effects are tempered by decreased hydraulic conductivity of abundantly root-colonized pore space, which can become clogged and increase preferential flow paths (Pedescoll et al. 2013). Plant presence has been found to reduce accumulated solids by 26% and enhance the development of biofilm (Chazarenc et al. 2009).

Human impacts on hydrology and nutrients are likely to limit diversity in constructed stormwater control systems (Mensing, Galatowitsch, and Tester 1998). The extreme flooding and drying events in stormwater BMPs can stress biological systems and provide opportunities for invasive and nuisance species to establish, which complicates theoretical treatment potential and expected maintenance requirements. Standing water, lack of light during burial, and fungal suppression can reduce germination, preserving the seed bank in wetlands compared to more aerobic environments (Ma et al. 2017) A field study found that unplanned "volunteer" plants covered more than half of a wetland after less than a year without maintenance, not necessarily to the immediate detriment of treatment but to the detriment of aesthetics (Muerdter et al. 2016). Research into ecological responses to wetting and drying regimes suggests that temporal variation in water availability may encourage more temperate woody plants to encroach on semi-arid and arid climate grasses (Snyder and Tartowski 2006). In a study of wet, alternating wet/dry, and dry treatment regimes, mycorrhizal growth and mycorrhizal phosphorus uptake showed greater symbiotic benefits in dry systems than in historically wetter systems (Cavagnaro 2016).

Plant decomposition rates depend on the C: N litter ratio and the plant fiber content.

Based on Bachand research and review (1999), in organic carbon-limited free-surface wetlands,
a mixture of labile carbon from submergent or floating species and more recalcitrant carbon from

emergent and grass species are recommended for improving denitrification rates. The rate of plant detritus entering the water column depends upon the plants' physical structure, disturbances to the wetland, and grazing pressures. In both wetlands and bioretention, plants with substantial root biomass are considered necessary to survive both flooding and intermittent dry periods and to host robust microbe populations (Muerdter et al. 2016). Plants with vegetative, non-seed production (usually perennials) may be more successful in frequently flooded environments, although soil moisture does not necessarily effect seed bank diversity (Ma et al. 2017). Plants are not always proven to accomplish a net removal of pollutants, but they are relatively quick at nutrient uptake and relatively slow at nutrient release during decay (Kadlec and Wallace 2009). This slows the cycling of nutrients and assists in eutrophication prevention during peak flows for which stormwater best management practices are intended. Harvesting is recommended to reduce the reintroduction of pollutants during mineralization processes, although some studies have found little effect of harvesting on nutrient removal efficiencies (Williams, Frost, and Xenopoulos 2013).

Bioretention systems and constructed wetlands have the potential to dramatically improve stormwater quality. Existing research suggests that inundation with moderate load rates and extended retention are favored for reducing nitrate-N concentrations, while ammonium and P are best addressed in aerobic systems with lower retention times (Vacca 2011). While there are many diffuse pollutants which could be present in stormwater, the focus of this study are those which indicate ecosystem health: COD, TN and nitrate, phosphate, and TS. For these, the flow path, residence time, substrate availability and plant community will be controlled in order to isolate the effect of water content. The examination of prolonged wetness/saturation may identify

optimal conditions for the removal of the most pollution and the best survival of desirable flora in bioretention and wetland ecosystems.

#### 3. MATERIALS AND METHODS

This research was designed to examine the effect of average water content on nutrient removal in bioretention systems at the field and laboratory column scale. Stormwater application and collection differed between field and laboratory series and are detailed in the following section. Laboratory analysis of water samples were performed similarly in both experimental series and over the same timeline. Details of instrumentation calibration or exact laboratory techniques are available in the Appendix.

## 3.1. Field-scale bioretention set-up and operation

Five bioretention bays were isolated within a larger bioretention basin on the campus of Michigan State University (Appendix A). The bioretention basin was constructed in 2010 to treat stormwater runoff from an adjacent underpass that collects stormwater from a watershed area of approximately 5.2 hectares in size with approximately 40% impervious areas (see Figure 7 for flow path). Additionally, the underground stormwater collection and storage system allows substantial infiltration of groundwater that dilutes the stormwater prior to pumping of the stored water to the inlet of the bioretention basin. The unanticipated flow due to groundwater causes portions of the site to take on saturated wetland characteristics. From November 2016 to February 2017, the average rate of groundwater intrusion was approximately 2.8 L/s (0.74 gal/s) while the average runoff was 4.3 L/s (1.14 gal/s) (Banach and Reinhold 2017). The bioretention basin (0.023 hectares) contains an engineered media with 3% organic matter (from partially cured animal compost), 85% sand, and 12% fines with permeability of 12.7 cm/hr (5 in/hr) and 6A or larger pea stone surrounding underdrain (Thode 2013) to a depth of four feet. The drainage and ponding variation in the large basin provided an opportunity to examine bioretention and

wetland effects on water quality, including side-by-side trials hydraulically isolated bioretention bays.

Figure 7. MSU Bioretention Research Site



Google Earth image with modifications by R. Bender, 2016

Historically, the site has provided a large storage volume but little treatment in terms of nutrient and sediment removal (Thode 2013). Groundwater intrusion dilutes the expected runoff to relatively low concentrations of most pollutants of concern, and the initial soil mix was made with dairy manure rather than cured compost, which may have caused organics and nutrient leaching beyond expected levels in the first several years after installation. This research was motivated in part by the hypothesis that managing moisture content might be a means to control and improve nutrient removal performance in the underperforming basin itself.

The experimental design utilized an equalization basin that drained into five hydraulically isolated bays (5 m long x 2.4 m wide x 1 m deep) within the bioretention basin (Figure 8). Flow into the isolated bays was controlled by filling holes in the perforated wall between the equalization basin and each bay so that the relative water flow into each bay was 10%, 20%,

50%, 80%, and 100% of natural storm and synthetic dosing events (ranging from approximately zero to twenty holes, one centimeter in diameter). Water content was measured at three depths twice per week using an SDI-12 Enviroscan (CS 2016) in two PVC ports installed in each bay. Hydraulic isolation and plants were established for three months (starting in June) before the start of water quality sampling. Temperature and precipitation data are reported in Appendix B.

Figure 8. Bioretention field bays



Image taken by R. Bender, 2014

A variety of plants representing different plant types and water table tolerances were installed in the field-scale bays at the beginning of the first growing season (see Table 2). Plants were selected based on root zone classifications for both bioretention and wetlands in the LID Manual for Michigan in Ecoregion 56 for the Lansing area (SEMCOG 2008). Design water table depth specifications overlap for wetlands and bioretention, although dry down depth is much greater for bioretention (Table 2). Plants included *Asclepias incarnata* (swamp milkweed), *Carex vulpinoidea* (brown fox sedge), *Juncus effusus* (soft rush), *Sagittaria latifolia* (arrowhead), *and Schoenoplectus tabernaemontani – syn. Scirpus validus* (great bulrush). Forty plants were equally spaced within each bay in a grid pattern, with each plant type represented in each row and column at approximately 40 cm spacing (eight plants of each type in each bay).

**Table 2. Recommended Water Levels** 

Water Table: +10cm +5cm Surface -5cm -10cm As deep as -45 cm

Range for Wetland

Range for Bioretention

Asclepias incarnata

Carex vulpinoidea

Juncus effusus

Sagittaria latifolia

Scirpus validus

Water quality experiments were conducted from June 2014 until October 2014 (photographs of experimental setting and progress are included in Appendix K). Plant size and survival were evaluated in June 2014 and again in September 2015. In addition to storm event runoff, which flowed through the holes at the forefront of each bay, synthetic stormwater was mixed from the equalization basin and applied to bays in doses on a biweekly schedule: 5 L, 10 L, 25 L, 40 L, or 50 L of synthetic stormwater. Synthetic stormwater was prepared based on typical nutrient loads (Table 3), originally designed by Lucas and Greenway (2008a) and described in Appendix F.

**Table 3. Synthetic Stormwater** 

Pollutant	Chemical	Concentration (mg/L)		
Ortho-Phosphate	Potassium Phosphate	0.79		
<b>Total Dissolved Phosphore</b>	0.79 (mg P/L)			
Ammonia	Ammonium Chloride	0.41		
Nitrogen Oxides	Potassium Nitrate	0.97		
Org. Nitrogen	Nicotinic Acid	3.47		
<b>Total Dissolved Nitrogen</b>		4.86 (mg N/L)		
Cadmium	Cadmium Nitrate	0.003		
Cadmium Copper	Cadmium Nitrate Copper Sulfate	0.003 0.544		
Copper	Copper Sulfate	0.544		

This mix was prepared in the laboratory and kept in an airtight container in refrigerators (2 °C) during the duration of this experiment. Each bay received the same mass of synthetic pollutants poured on entire surface of soil (Table 4), although dilution varied according to water content category.

Table 4. Synthetic Stormwater applied to Bays and Columns

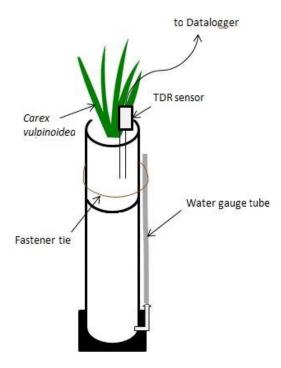
Pollutant	Stock	Column	Stormwater Stock		Dilution	(L)	Stormwater
	Concentration	or Bay	Addition (mL)		Column	Bays	Concentration
	(g/L)	number					(mg/L)
Total	0.79	1	1	10	0.5	5	1.59
Dissolved		2	1	10	1	10	0.79
Phosphorus		3	1	10	2.5	25	0.31
		4	1	10	4	40	0.19
		5	1	10	5	50	0.15
Total	4.86	1	1	10	0.5	5	9.72
Dissolved		2	1	10	1	10	4.86
Nitrogen		3	1	10	2.5	25	1.94
		4	1	10	4	40	1.22
		5	1	10	5	50	0.97
Chemical	0.165	1	1	10	0.5	5	0.33
Oxygen		2	1	10	1	10	0.17
Demand		3	1	10	2.5	25	0.07
		4	1	10	4	40	0.04
		5	1	10	5	50	0.03
Total	1.27	1	1	10	0.5	5	2.54
Metals		2	1	10	1	10	1.27
		3	1	10	2.5	25	0.51
		4	1	10	4	40	0.32
		5	1	10	5	50	0.25

The synthetic stormwater used in this study is comparable to those used in other examinations of diffuse pollution, but much less than concentrations expected even in weak domestic wastewater. For comparison, reference conditions for ambient nutrient concentrations in Ecoregion VII (which includes southern Michigan) would include TP 0.0148 mg/L and TN 0.007 mg/L for lakes and reservoirs (USEPA 2000). However, weak domestic wastewater might be expected to have 350 mg/L total solids, 20 mg/L TN, 6 mg/L TP, and 100 mg/L BOD (Pescod 1992). Regulatory intervention would not be expected for the effluent concentrations used in this study.

## 3.2. Laboratory scale bioretention set up and operation

The effects of average water content in bioretention systems were also examined in a more controllable laboratory setting. Triplicate columns (0.2 m wide x 1 m deep) for each water content level were constructed and maintained in a small greenhouse on MSU's campus (see Figure 9) where they received natural sunlight. Each column was planted with a single *Carex vulpinoidea*. Watering proportions were replicated in columns allowed to drain into collection containers twice weekly and then dosed with 0.5 L, 1.0 L, 2.5 L, 4 L, and 5 L of synthetic stormwater twice weekly. Water content was monitored continuously to a depth of 30 cm using CS616 Time Domain Reflectometry (TDR) sensors (CS 2012). Water content and plants were allowed to establish for three months before water quality sampling. Like the bays, each column received the same mass of pollutants with each dose of stormwater. Bays and columns of the same target water content category received synthetic stormwater of the same concentration.

Figure 9. Bioretention columns



### 3.3. Sample collection and determination

Bay influent samples were drawn from the shared forebay. Field bay treatment samples were drawn from the underdrain via PVC sampling ports twice weekly. Column effluent samples were drained from the water gauge tube before every new stormwater dose. All samples were stored in a refrigerator at 2 °C until analysis within 24 hours of collection in the laboratory. Total solids were measured by subtracting initial weight from final weight after evaporation of a 25 mL sample in aluminum tins according to USEPA accepted HACH Method 8271 (USEPA 2015) as described in Appendix G. COD was quantified using low range (3-150 mg/L) or high range (20-1500 mg/L) dichromate kits following USEPA Reactor Digestion Method 8000 (USEPA 1980). TN and TP were analyzed using persulfate co-digestion method 4500-NC (Aryal 2015) as described in Appendix I. This method includes a digestion reagent mix of 20.1 g potassium persulfate and 3 g sodium hydroxide dissolved in 1000 mL of e-pure water. A borate buffer solution was also prepared, composed of 61.8 g boric acid and 8 g sodium hydroxide in 1000 mL. The analysis was performed after mixing digestion reagent, standard/sample, and buffer in proportion 5:10:1. All samples and standards were autoclaved at 110°C for 30 minutes before analysis in a Dionex Ion Chromatography ICS 5000 machine (Dionex ICS 5000). Nitrates and phosphates were also analyzed using ion chromatography after preparation using USEPA method 300 (Pfaff 1993) as in Appendix H. All samples run through the ICS 5000 machine were filtered through 45 uM cellulose acetate filters prior to injection. At least five levels of standards (including blanks) were prepared with each batch of samples in order to create a linear calibration curve. Non-detects within IC data were included in statistical analysis with a value 50% of lowest recorded measure for each parameter.

### 3.4. Statistical analysis

Water quality data was analyzed separately based on field or laboratory category, in both cases the samples were grouped by watering regime (levels 1-5). Treatment group data (all Column 1 Nitrate, all Column 2 TN, etc.) were analyzed using Dixon's Q test and confirmed to have no statistically meaningful outliers assuming a normal distribution (Andale 2016). QQ plots were prepared within each group to visualize normality (Ford 2016). Non-normal patterns assisted in identifying non-detects (see Appendix J). Normal and non-normal groups were analyzed for variance and skewed treatment groups were also analyzed by Wilcoxon ranking, which are shown in results with Kruskal-Wallis summary values. Time dependence was checked by graphing the residuals (i.e. sample – group average) to identify serial correlation along a timeline (Ott and Longnecker 2001).

One-way analysis of variance (ANOVA) and the more skew-resilient Kruskal-Wallis tests (McDonald 2014) were performed to assess water content effects on reduction of the pollutant effluent concentrations. Kruskal-Wallis tests identify differences in medians between groups with differing distributions and are shown with Wilcoxon ranking box plots. Parameters for analysis of treatment groups include: chemical oxygen demand (COD), total nitrogen (TN), nitrate, phosphate and total solids (TS). ANOVA was also used to evaluate plant growth consequences of water content and plant species. Significance values of p <0.05 are considered statistically significant.

Quantification of mass export was calculated by multiplying effluent concentrations by influent volume. Effluent volume from columns was assumed to equal influent volume to compensate for disproportionate leaking in column joints. Evaporation effects were assumed to be uniform across all columns because of uniform surface area, soil type, sunlight, wind

protection, and plant type. Due to the hydraulics of the field-scale bioretention bays, effluent volumes were not measured and therefore only column data is included in figures comparing mass export.

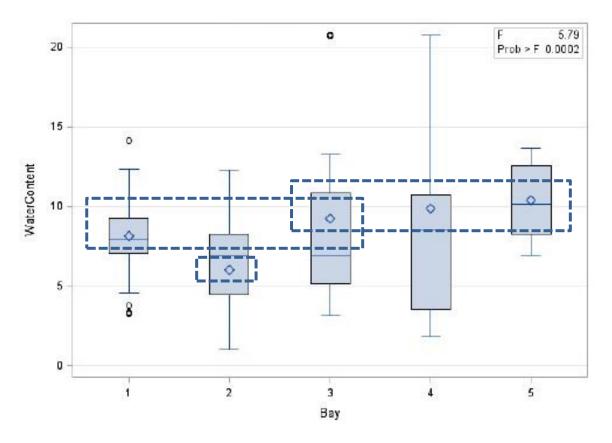
## 4. RESULTS AND DISCUSSION FROM FIELD STUDY

Field experimentation differed from laboratory experimentation in meaningful ways, particularly in set-up and water application regime. Conclusions can be drawn from data as from two independent experiments, and recommendations are made for future research.

## 4.1. Establishment of prolonged water content

In the bays, soil water content ranged from 0-20%, as shown in Figure 10, with higher variability in bays directly in line with the inflow culvert. Statistically different water content groups are outlined, showing Bay 2 as the driest bay, Bay 1 with medium wetness, Bay 3 with medium-high wetness, and Bays 4-5 as the wettest bays.

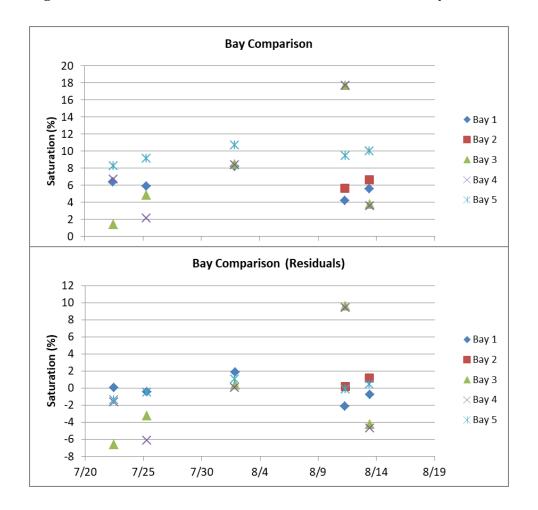
Figure 10. Water Content in Bays



Volumetric water content never exceeded 22%, probably as a result of underdrains in the field installation. This is typical of stormwater management systems, which are rarely able to recreate fully saturated models in real-time scenarios (Barbu and Ballestero 2015). Water rarely dropped below 5% even with management, probably as a result of large storm events early in the experiment and persistent ponding in the forebay. Also, the regularity of stormwater dosing required to apply consistent pollutant loading across all bays may have prevented complete dryout of bays designated for lower water content. For example, when Bay 5 received twenty gallons of water, Bay 1 would still receive one gallon of water in order to create similar hydraulic retention times in dosing for all trials, creating minimum water content.

Daily water content data was not available at sample events throughout the entire span of sampling due to technical difficulties (as shown in Figure 11 and detailed in Appendix C), but soil microbial research has shown that legacy effects of water content from an ecosystem establishment period do have effects on biotic response for several weeks afterward (Banerjee et al. 2016).

Figure 11. Enviroscan Measurement Summaries in Field Bays



# 4.2. Water content effects

The field bays in this study established fairly consistent water content differences (p<0.0002), but not enough to create differences in water treatment performance. Flooding of all bays during early establishment may have muted any long-term significant differences in microbial communities and their associated water quality improvement. Early experimental design did not fully appreciate the longevity of microbe populations which might have persisted in all bays and reduced management effect later. Generally speaking, water quality data collected in bays showed higher variability in all groups than in corresponding laboratory columns, as expected. This is consistent with other bioretention and wetland studies which are exposed to

natural weather patterns (Kearney, Zhu, and Graney 2013, Hatt, Fletcher, and Deletic 2009). Serial correlation analysis did not indicate any seasonal trend in bay or column concentrations (analysis detailed in Appendix C per sampling group).

## 4.2.1. Chemical Oxygen Demand Results

When comparing influent and effluent COD concentrations, it appears that some treatment occurred in all field bays (Figure 12). Treatment ranged from 8 mg COD/L decrease in lower water content bays to 3 mg COD/L decrease in Bay 5.

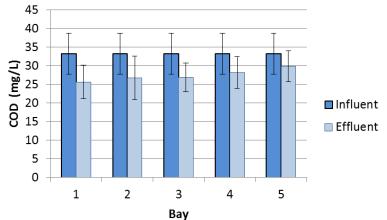
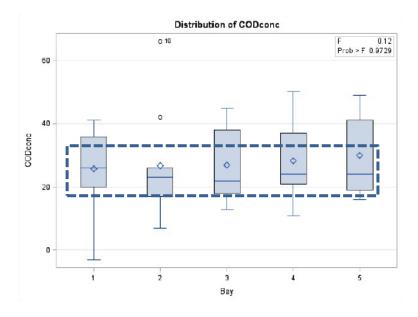


Figure 12. Concentration comparison of Effluent and Influent COD in Bays

COD concentrations in the collected leachate from the bays were relatively consistent across all five treatments (see Figure 13). No significant differences were detected between any of the treatments (n=45, p=0.9729).



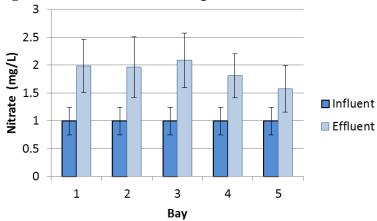


Generally the high stormwater flow in the field bioretention setting probably rinsed the soil media of much of the most available carbon. At the very least, the groundwater dilution prevents COD concentrations of concern, in the range of 100 mg/L or more (USEPA 1986). Bioretention has been shown in many cases to be most effective at removing lower hydraulic loads, when "hungry" microbes readily uptake available nutrients (Hatt, Deletic, and Fletcher 2007), but the background organic matter present in the vegetated area likely created a baseline difficult to reduce COD to zero (Huang and Hall 2017).

### 4.2.2. Nitrate Results

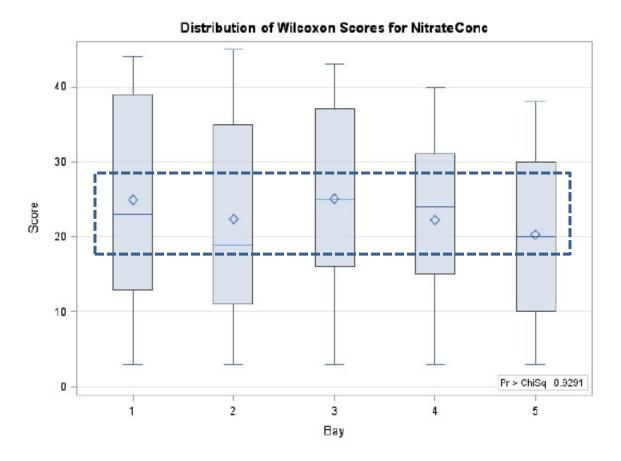
Effluent nitrate concentrations were higher than all influent concentrations in bays, as shown in Figure 14, although concentrations were very low in both cases. Typical agricultural loading rates used in a wetland pulsing study by Messer (Messer et al. 2017) ranged from 2.5-10 mg/L. The small margin of increase (0.5-1.0 mg/L) may indicate that conditions were sufficiently aerobic to sustain nitrification.





There was little difference in effluent nitrate concentrations between bay treatments (n=45, p=0.9466) as shown in Figure 15. Effluent nitrate concentrations from all treatments had comparable variability and distribution, even when controlled for non-detects by Wilcoxon ranking.

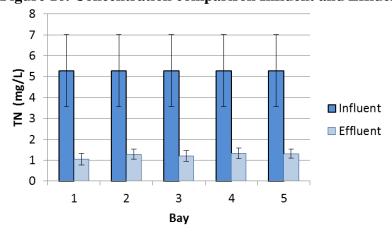
Figure 15. Distribution of Nitrate concentration in Bays



# 4.2.3. Total Nitrogen Results

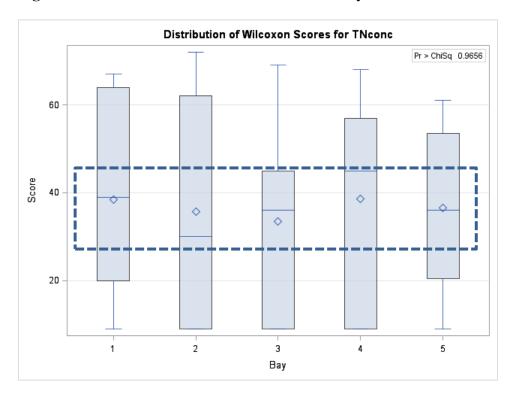
A small amount of TN treatment occurred in all field bays (Figure 16). Bays showed a reduction of approximately 4 mg/L TN in each bay (a concentration nearly equal to that applied in Bay 2).

Figure 16. Concentration comparison Influent and Effluent TN in Bays



Data from bays showed relatively high TN concentration range in all bays, with very little change between bay treatments (n = 50, p=0.99, Figure 17). This wide range reflects a high incidence of non-detects (17% in field bays) in relatively low concentrations (0-2.2 mg/L).

Figure 17. Distribution of TN concentration in Bays



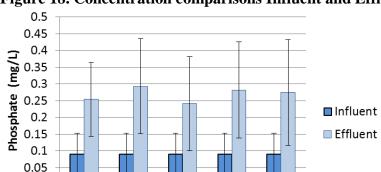
Bays showed a slight increase overall in nitrate concentrations between influent and effluent, but a reduction in TN concentrations. This may indicate a shift toward removal of non-

nitrate nitrogen compounds (like ammonia or ammonium), perhaps because of low activity in denitrifiers. TN reduction is a likely result of denitrification in the water below and up to the underdrains in all bays (Brown and Hunt III 2011), even when shallower depths showed unsaturated water content. Alternatively, microzones of anoxic conditions, most likely within the biofilms or organic matter in the media, permitted denitrification. Sorption of ammonia and plant uptake may also have occurred.

In any case, there was not a significant difference between bay treatments, probably explained by the low margin of volumetric water content from 2-20%, medians 7-11%. The cooling temperatures during the timeline for this research may have also affected nitrate and TN treatment effectiveness. Due to a cold, wet spring and an establishment period during summer (see Appendix B for details) analysis was delayed until fall, when low temperatures may have slowed microbial activity.

## *4.2.4. Phosphate Results*

Phosphate analysis had the most non-detect data of any water quality parameter (96.4% of samples). Comparisons show effluent concentrations increased approximately 0.2 mg/L from influent concentrations in all field bays (Figure 18). This is likely an effect caused the bioretention media itself, which included composted animal manure as an organic matter component (Thode 2013). In time, it is expected that an exchange equilibrium will be reached (Li and Davis 2016), although data did not reveal any expedited removal from wetter bays over the course of this intra-season study.



Bay

Figure 18. Concentration comparisons Influent and Effluent Phosphate in Bays

Effluent concentrations from bays were not significantly different between treatments including Wilcoxon rank sum analysis (n=45, p=0.99, Figure 19). Although water content could potentially affect electroconductivity of soils slightly, the most dramatic changes to soil phosphorus capacity result from soil amendments, like wood chips, or plant removal (Chang, Hossain, and Wanielista 2010). Without such material changes in bioretention media, it is not surprising that no differences arose in phosphate mobility at very low effluent concentrations.

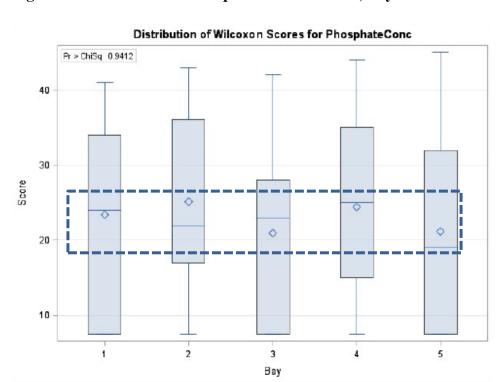
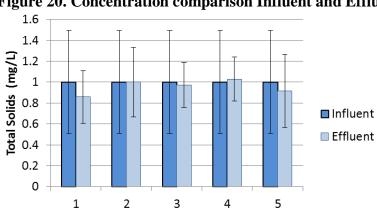


Figure 19. Distribution of Phosphate concentration, Bays

## 4.2.5. Total Solids Results

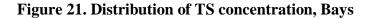
The synthetic stormwater contained no solids component, although the forebay water that entered field bays from the large bioretention basin did contribute some solids in the influent for the field experiment. In these field bays, some treatment did occur (Figure 20). Effluent TS concentrations from Bay 1 were the lowest, perhaps because lower flow rate and volume created less erosive potential. Bay 4 showed a slight increase in TS, probably because inflow velocities were highest directly in line with inflow culvert and debris from the channel may have taken the shortest path through the equalization bay.

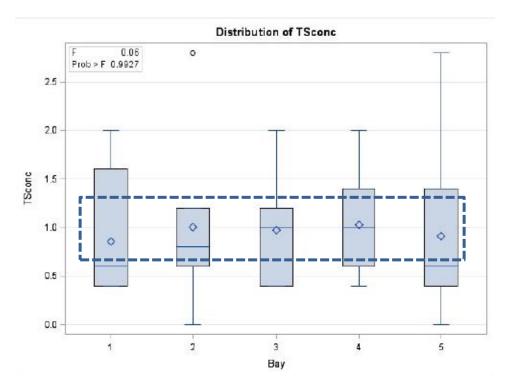


Bay

Figure 20. Concentration comparison Influent and Effluent TS in Bays

No statistically significant differences were detected between bays in terms of TS concentrations (n=35, p=0.99, Figure 21). Even in cases where TS did increase, with maximum TS concentrations less than 2.5 mg/L, data were well below MDEQ limits for dissolved solids in effluent, 750 mg/L (Quality 2006). The breakwall between the forebay and the field bays themselves may have reduced erosive velocities enough to prevent much increase in TS. The unchanged TS concentrations between influent and effluent show that fines and organics entering with the influent were either unaffected or directly replaced by internal sediments during passage through the bioretention media.





The greatest impacts on solids removal are physical ones, horizontal space for settling and obstacles which might reduce velocity and enhance precipitation. The study was designed to control for these factors. Soil cohesion may have differed between treatments with different water content, but not enough to be identified in this study.

## 4.2.6. Effects of Water Content on Plant Growth

Root architecture, plant size, and species are known to have a strong effect on pollutant removal performance in stormwater BMPs. During the eighteen months when these plants were growing in an environment with altered hydraulics, their growth and survival varied considerably. A two-way ANOVA showed plant growth (proxy: height) was significantly different based on plant species (p<0.0001), bay (p=0.0185) and the interaction between (p=0.0084). Plant growth is summarized in Figure 22, where original plant height is 100% at the

beginning of the study. A doubling is represented as 100% growth, a plant smaller in the second year than the first would be less than 0% growth, and death is shown as -100% growth.)

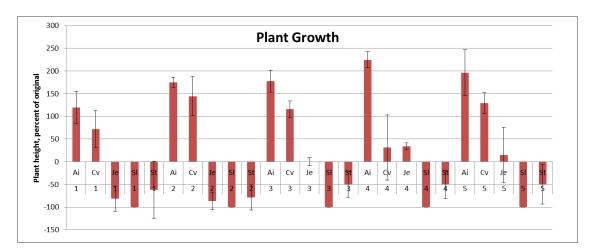


Figure 22. Summary of Plant Growth in Bays

For figure above, N =199 ANOVA: Bay P = 0.018, Species P <0.0001, Bay\*Species P = 0.008

Plants are abbreviated by initials of scientific name: *Asclepias incarnate (Ai), Carex Vulpinoidea*(Cv), Juncus effusus (Je), Sagittaria latifolia (Sl), and Scirpus validus (St, from alias

Schoenoplectus tabernaemontani).

The tall, flowering milkweed, *Asclepias incarnata*, was successful in establishing nearly every plant in every bay, despite an infestation of aphids in August 2014 that defoliated many of the plants. *Asclepias incarnata* success increased with water content until a peak in Bay 4, after which it declined. *Carex vulpinoidea*, brown fox sedge, was another very successful plant in every bay. The highest growth occurred in Bays 2 and 5, while its worse growth rates were in Bay 1 and Bay 4, suggesting a compounding variable. *Juncus effusus*, soft rush, thrived only in the two wettest bays, and showed considerable, although incomplete, mortality in the remaining bays. *Sagittaria latifolia*, the arrowhead plant, disappeared completely from every bay within the first few months of acclimation. This may have been a result of a cold spring and flooding during

early summer of 2014. *Scirpus validus*, great bulrush, also suffered nearly complete mortality, although a few plants were successful in the wetter Bays 3-5.

The four most successful plant types, Asclepias incarnata, Carex vulpinoidea, Scirpus validus and Juncus effusus, were most successful in Bay 3, Bay 4, and Bay 5. This observation is consistent with literature material which suggests that wetland environments are the most productive for biomass, reaching a threshold of soil moisture before a different plant diversity and abundance increased with depth of standing water (Ma et al. 2017). Carex and Juncus were also two of the most successful plant genera in a pollution reduction study of 20 different wetland species (Read et al. 2008), and they proved again to have the best survival in all field bays. What remains surprising is the failure of Sagittaria latifolia and Scirpus validus to survive. This may be a result of fragility during transplant of Sagittaria latifolia's delicate roots and the Scirpus validus' long hollow stem, or perhaps overexposure to the elements during establishment. Plant success between species suggests that a diverse planting scheme may ensure some amount of plant survival, and denser growth habit and stronger stems seemed most successful. Water content correlated with overall plant success (Table 5), suggesting water stress may be a factor for survival in dry or rapidly draining bioretention basins. The highest pollutant concentrations were in Bay 1, where plant growth was lowest, suggesting that low plant growth may coincide with poor treatment in some cases.

**Table 5. Plant Growth in Field Bays** 

	Bay 1	Bay 2	Bay 3	Bay 4	Bay 5
Average % growth	-10.2	10.8	28.8	28.1	38.0
Rank	Low	Low - Med	Med-High	Med-High	High

Standard error ranged from 11.5 - 11.6%. Rankings are based on significance < 0. 05. Some treatments are ranked as low-med or med-high to indicate there was no statistical difference between that specific treatment and other treatments in the high, medium, or low rankings.

# 4.3. Sampling Methods and Technology

The field bays in this study received influent from environmental stormwater as a part of the larger bioretention basin. In periods without rain, it was also supplemented with laboratory stormwater in an effort to maintain design hydraulics. The continuous underdrain prevented water retention sufficient to create saturation in the wettest bays, while a ponded equalization forebay allowed intrusion into the driest bay.

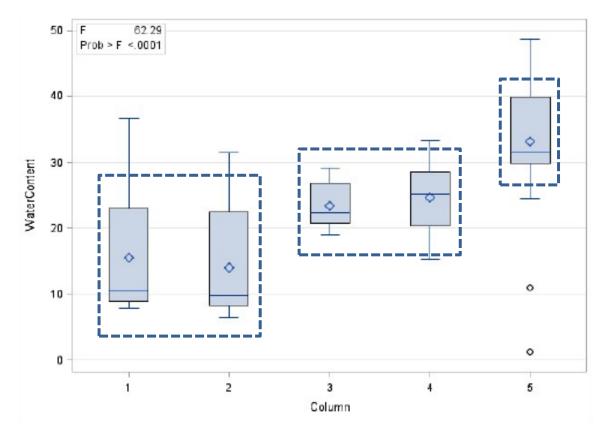
## 5. RESULTS AND DISCUSSION FROM LABORATORY STUDY

Field experiments were characterized by uncontrollable environmental conditions, leading to high variability in the range of water contents observed in each bay. To examine the effects of hydraulic loading and soil water content on treatment by bioretention under controlled environmental conditions, columns studies were also conducted. Laboratory column analyses revealed no net removal of pollutants, but comparisons in concentrations are still discussed in the following sections.

# 5.1. Establishment of Prolonged Water Content

Laboratory column water content was measured continuously in five of the fifteen columns, showing a range of 5-50% water by volume as shown in Figure 23. There were three groupings for statistically significant water content: low, medium and high.

Figure 23. Water Content in Columns



Water periodically drained from columns in sample collection was small relative to the amount of water added, as shown in Figure 24. Column materials could be improved in future research to better contain leaking effluent. To adjust for the disproportionate collection, effluent pollutant mass values were calculated assuming effluent equal to influent volume.

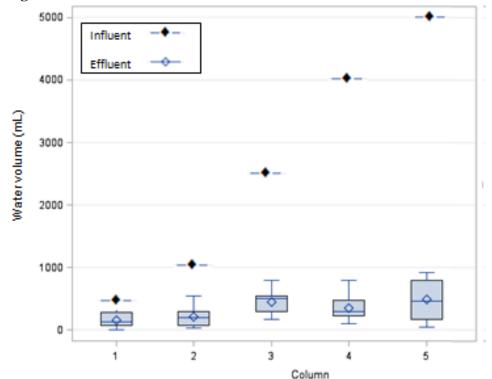


Figure 24. Influent and Effluent Volumes in Columns

# 5.2. Water Content Effects

The wide range of water content in laboratory columns led to significant differences between treatment groups than those observed in the field study. Comparisons were made between effluent concentrations regarding statistically significant differences between water content treatments.

# 5.2.1. Chemical Oxygen Demand Results

The synthetic stormwater introduced a negligible amount of organic pollution into each column, but no COD removal occurred in laboratory columns. Instead, COD concentration increased in columns, probably a result of leaching of organic carbon from compost in bioretention media (Thode 2013). Column effluent COD concentrations where much higher than those measured in the bays, probably an indication of dilution from larger bioretention basin water intrusion. Figure 25 shows average COD concentration and standard error of influent and effluent.

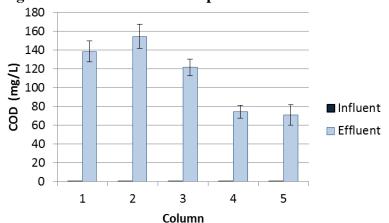
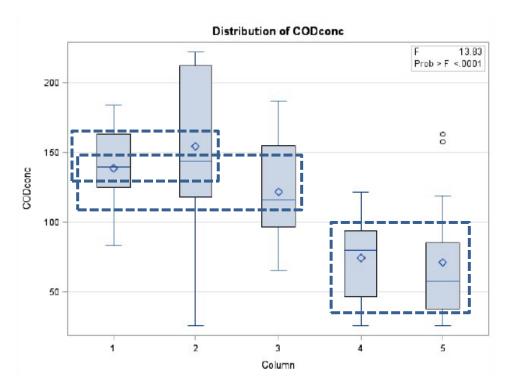


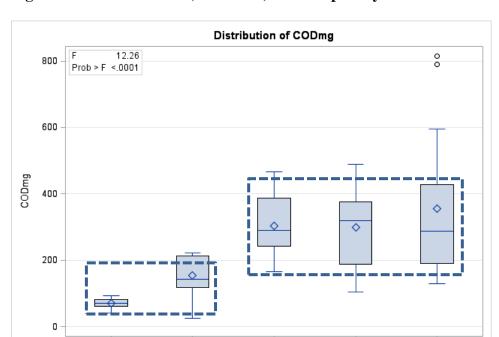
Figure 25. Concentration comparison of Effluent and Influent COD in Columns

Figure 26 reveals a significant difference in COD effluent concentrations between Columns 1-3 and Columns 4-5 (n= 79, P<.0001). Samples from columns with lower water contents (1-3) had higher concentrations of COD; effluent concentrations from Column 2 showed the highest range, up to 169 mg/L.





When concentrations were multiplied by water volume to estimate mass export, significant differences were observed between drier and wetter columns (see Figure 27). COD mass leached from Column 1 and Column 2 was significantly higher than Columns 3-5 (n=75, P = 0.0001-0007).



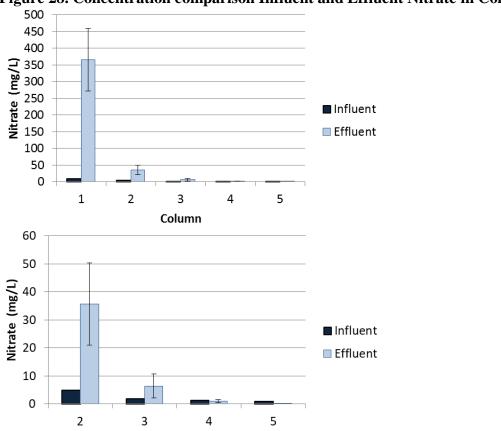
Column

Figure 27. Distribution of (calculated) COD Export by Mass in Columns

Concentrations were higher, but total mass export was lowest in drier Columns 1 and 2. Higher concentrations of COD in the effluent of the drier columns were not sufficient to compensate for smaller volumes of effluent, leading to a decrease in mass export in drier columns. Although concentrations were low in Columns 4 and 5, their net export was similar to that of Column 3. This suggests that dilution may be the dominant force controlling the relative concentration of column effluent. The enhanced decomposition in aerobic columns and denitrification in wetter columns may have balanced in terms of COD, or perhaps the carbon mobilized into solution was based on uniform factor across all columns, such as plant species or bioretention media.

### 5.2.2. Nitrate Results

Effluent nitrate concentrations were higher than or equal to influent concentrations in all columns (Figure 28) with a greater increase observed in drier Columns 1 and 2 (n=67, P<0.0001).

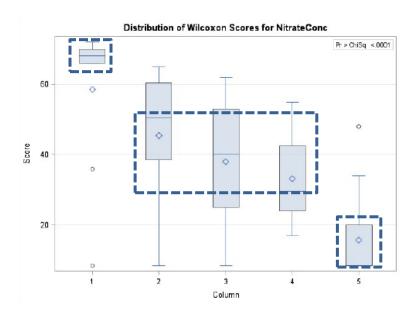


Column

Figure 28. Concentration comparison Influent and Effluent Nitrate in Columns

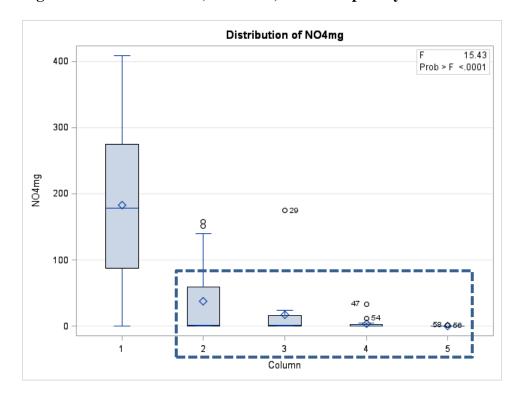
The effluent nitrate concentrations from Column 1 (the driest treatment) were significantly higher than those from all other columns (n=68, P<.0001), as shown in Figure 29. Higher water content in the columns corresponded with lower effluent nitrate concentrations, even when the effects of non-detects were minimized by use of the Kruskal-Wallis-Wilcoxon ranking.

Figure 29. Distribution of Nitrate concentration in Columns



The mass of nitrate exported from drier columns was still greater than that of the wetter column treatments, as shown in Figure 30, with significantly higher export in Column 1 (n=68, P<0.0001).

Figure 30. Distribution of (calculated) Nitrate export by mass in Columns



### 5.2.3. Total Nitrogen Results

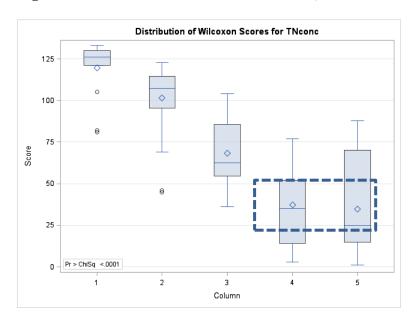
Effluent TN concentrations were greater than influent TN concentrations in all columns (Figure 31), increasing as much as 4-90 mg N/L.

140 120 100 80 60 40 20 0 1 2 3 4 5 Column

Figure 31. Concentration comparison Influent and Effluent TN in Columns

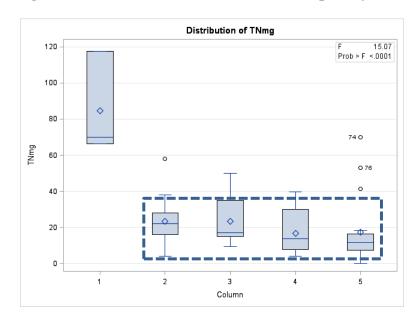
Concentration data also revealed a pattern of decreasing TN concentrations with increasing water content in the columns (Figure 32), mimicking the trend observed for nitrate Column 1 had significantly higher TN effluent concentrations than all other columns and Column 2 effluent was higher in concentration than effluent from Columns 3-5 (n=78, P<0.0001-0.0004).

Figure 32. Distribution of TN concentration, Columns



The quantified mass export of TN from the columns revealed a dramatic difference between mass export from Column 1 and all other treatments (see Figure 33).

Figure 33. Distribution of (calculated) TN export by mass in Columns



The patterns of nitrate and TN leaching were very similar. Column effluent concentrations and mass export were much higher for drier columns than wetter columns, with a nonlinear decrease in concentrations after Column 1. This confirms a pattern of increased

nitrogen removal with saturation zones established in the literature. Neither columns nor bays had net removal of nitrate or TN, showing that nutrient leaching from the soil media was still a greater impact than treatment. The autumn temperatures of the timeline for this research may have also affected nitrate and TN treatment effectiveness. A review by Lee, et al. (Lee, Fletcher, and Sun 2009) showed biological nitrogen removal is most efficient between 20-40 °C.

#### 5.2.4. Phosphate Results

Phosphate analysis had the most non-detect data of any water quality parameter. No phosphate was detected in column effluent (see Figure 34) and therefore no statistical analysis was included (n=68).

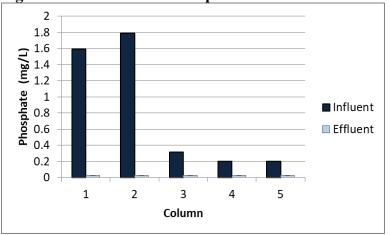


Figure 34. Concentration comparisons Influent and Effluent Phosphate in Columns

The soil and plant capacity for phosphorus sorption and uptake may have been sufficient to remove added phosphorus from the wastewater. Each *Carex vulpinoidea* filled the top several inches of soil with roots.

#### 5.2.5. Total Solids Results

The synthetic stormwater had no added solids and therefore all columns showed effluent concentration increase rather than TS treatment (Figure 35). Column 1 effluent appeared to have

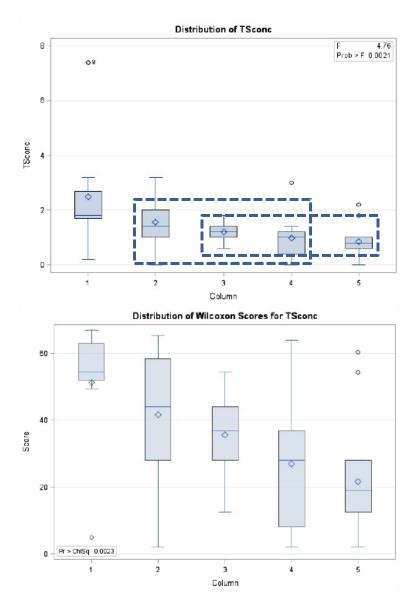
the largest increase in TS while the overall trend was a decrease in TS concentration corresponding with increasing water content.

3.5 3 Total Solids (mg/L) 2.5 2 1.5 ■ Influent ■ Effluent 1 0.5 0 2 5 3 4 1 Column

Figure 35. Concentration comparison Influent and Effluent TS in Columns

Higher concentrations of solids were exported from columns with lower water content than those with higher water content (Figure 36). Column 1 concentrations were significantly higher than all other columns (P values 0.0002-0.0269) and Column 2 was significantly higher than Column 5 (P=0.046). It is probable that the TS export was controlled by surface area, drainage area, and root volume within a column. These parameters were the same across all columns and so a similar sediment load may have been carried by a relatively smaller drainage volume, increasing concentration in smaller drainage volumes.

Figure 36. Distribution of TS concentration in Columns



When adjusted for mass export (Figure 37), the pattern of TS export was reversed. Column 1 mass export was significantly lower than Columns 3-5 (n=67, P=0.0014-0.0493) and Column 2 mass export was less than Columns 4-5 (P=0.0007-0.0017).

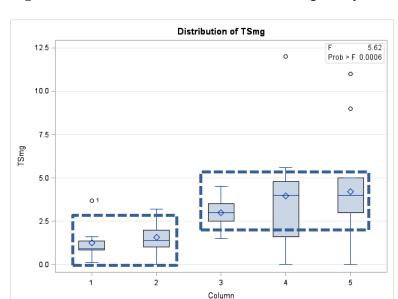


Figure 37. Distribution of (calculated) TS export by mass, Columns

Solids were exported from all columns. In the column data, effluent concentrations were greatest in drier columns even while mass export was the least. This can be understood as a function of equal surface area (where sediment is usually mobilized) and proportionally little runoff in drier columns. The most drastic pollutant reductions often occur in bioretention and wetland systems when effluent drainage is eliminated by internal storage capacity (Dumonceau, Hunt, and Winston 2012).

#### 5.3. Summary of Results

The bioretention bays and columns of this study did not prove consistently effective for improving water quality under low pollutant loading. There is substantial documentation of variable pollutant removal effectiveness of stormwater best management practices in scientific literature (Billy et al. 2010, John et al. 2010, Spatari, Yu, and Montalto 2011). Indeed, an examination of seven treatment wetlands by (Diaz, O'Geen, and Dahlgren 2012) found that nitrate and total suspended solids were the only pollutants consistently removed in these systems among a suite of contaminants analyzed (including salts, nutrients, dissolved organic carbon,

suspended solids, and bacteria). Bioretention studies have shown BMP performance ranging from nearly complete removal to significant export of nutrients (Ahiablame, Engel, and Chaubey 2012, Roy-Poirier, Champagne, and Filion 2010a). In this experiment, overall nutrient export may be a result of organic matter still curing in the compost component of the bioretention media. Organic carbon was persistent, even increased in some bioretention studies of compost leaching over two years in a bioretention setting (Mullane and Flury 2015). The low pollutant concentrations in synthetic stormwater and in the groundwater-diluted stormwater of the MSU site created a scenario in which rinsing pollutants from media was a greater effect than pollutant removal, although water content groups did have differentiated results in the column study.

Treatment groups with significant treatment differences are highlighted in Table 6. Statistical differences between treatment groups in terms of effluent concentrations of COD, nitrate, TN, and TS were detected in the columns, but not within their respective bays.

Table 6. Statistics Results from ANOVA

Parameter		F-test Value	Probability	Chi-squared	Probability
Chemical Oxygen	Columns	13.83	<0.001	32.2172	<0.0001
Demand	Bays	0.12	0.9729	0.5763	0.9657
Nitrate	Columns	24.31	<0.001	29.4199	<0.0001
	Bays	0.18	0.9466	0.8681	0.9291
Phosphate	Columns	NA	NA	0	1.0000
	Bays	0.02	0.9990	0.7792	0.9412
Total Nitrogen	Columns	50.44	<0.001	88.5	< 0.001
	Bays	0.19	0.9424	0.5766	0.9656
Total Solids	Columns	4.76	0.0021	16.56	0.0023
	Bays	0.06	0.9927	1.1189	0.8913

A complete summary of water content, pollutant concentrations, and pollutant mass is summarized in Table 7. Three categories are used to distinguish statistically significant differences in effluent concentrations: Low, Medium, and High. Field bays had less differentiation in water content (range 2-22%, median 6-9%) and little difference in pollutant concentration. Columns showed greater difference in water content (range 7-47%, median 15-34%) and greater differences in pollutant concentration. This suggests that soil moisture does have an effect on nutrient removal in bioretention systems and there may be water content thresholds for pollutant removal capacity.

**Table 7. Nutrient Concentration and Treatment Performance** 

	Bay 1	Bay 2	Bay 3	Bay 4	Bay 5
Water Content	Med	Low	Med-High	Med-High	Med-High
COD (conc.)	Med	Med	Med	Med	Med
Nitrate (conc.)	Med	Med	Med	Med	Med
TN (conc.)	Med	Med	Med	Med	Med
Phosphate (conc.)	Med	Med	Med	Med	Med
TS (conc.)	Med	Med	Med	Med	Med
Plant Growth	Low	Low-Med	Med-High	Med-High	High

	Column 1	Column 2	Column 3	Column 4	Column 5
Water Content	Low	Low	Med	Med	High
COD (conc.)	Med-High	High	Med-High	Low	Low
(mass)	Low	Low	High	High	High
Nitrate (conc.)	High	Low	Low	Low	Low
Mass	High	Low	Low	Low	Low
TN (conc.)	High	Low	Low	Low	Low
(mass)	High	Low	Low	Low	Low
Phosphate	NA	NA	NA	NA	NA
TS (conc.)	High	Med	Low-Med	Low-Med	Low
(mass)	Low	Low	High	High	High

Shaded rows indicate treatment groups where effluent concentrations exceeded influent concentrations

#### 6. CONCLUSIONS

Bioretention cells and constructed wetlands are both popular best management practices for stormwater retention, sedimentation, absorption, infiltration, filtration, phytoremediation, nitrification and denitrification. In an experiment to observe optimum water content for these treatment pathways, five bioretention bays and bioretention columns were controlled to run parallel tests of median water content ranging from 6 - 9% in field bays and 15 -34% in columns (up to complete pore space saturation).

Pollutant concentrations from bioretention bays showed influent treatment in COD, TN, and some TS, although there was no difference between treatments in terms of any pollutant concentrations. Variation in hydraulic loading between treatments did not affect pollutant concentrations, although it did correlate with differences in plant growth (as plant height correlated with design water content). The lack of effect on treatment was most likely due to the relatively small range of observed water contents. Asclepias incarnata, Carex vulpinoidea, Scirpus validus, and Juncus effusus were slightly more successful in wetter bays, while Sagittaria latifolia did not survive in any bay. Water content data in columns showed lower effluent concentrations and mass export for relatively wet treatment groups in terms of COD, nitrate, and TN; although no actual influent reduction occurred compared to influent stormwater. It appears that bioretention systems can mimic the water quality and ecological effects of wetlands if saturation conditions are maintained, although this study found no water content level in which COD, Nitrate, TN, Phosphate, and TS are simultaneously optimized. There is so much overlap between the habitat extremes and treatment effects of these practices that the treatment opportunities and pitfalls of wetlands should be considered in the operation and maintenance of bioretention systems.

Future research should continue to clarify the consequences of soil, plant, and hydration characteristics which effect pollution removal in bioretention. Closer examination of soil moisture effects within the soil profile could provide more accurate design depths in bioretention basins to validate shallow or infiltrating basins (Browne et al. 2008). In a large bioretention basin like that at MSU, soil moisture and internal water quality could be sampled from perimeter and internal points to increase the number of replicates and evaluate consistency within the BMP. Preliminary studies suggest that mycorrhiza with nutrient sequestration effects may also be more resilient to drying than other microbes (Barnard, Osborne, and Firestone 2013) and should be included in studies of soil moisture in BMPs. Atmospheric moisture and evaporation effects were not included in this study, but may be useful in designing soil media which can maintain desirable moisture qualities in a changing climate (Pyke et al. 2011). Bioretention and wetland ecosystem recovery after drought periods including a component of antecedent moisture levels, will create more accurate predictions of BMP performance (Cavagnaro 2016). Greater understanding and predictability will lead to more effective design and improved water quality in stormwater management systems.

An improvement to this study would control effluent drainage to create a sampling regime with a distinct hydraulic residence time. In some biofilter studies, outflow water improved only during the period in which resident storage water is displaced, rather than new influent introduced (Subramaniam et al. 2016). Also, distinct climatic or dry periods show a seasonal first flush, while uniform rainfall often does not, so flow-weighted composite sampling may be a consideration for quantification of mass emissions at the field scale (Lee et al. 2007). This study did not investigate seasonal or first flush effects, but water content could be maintained through an entire year to better examine seasonal effects and assimilation capacity of

first flush materials. It may be easier to evaluate differences if initial concentrations of pollutants of concern were elevated to levels of domestic wastewater rather than more diffuse stormwater, thus reducing non-detects.

APPENDICES

#### APPENDIX A. Michigan State University Bioretention Field Site

Michigan State University installed the Farm Lane Bioretention Research Site in 2009 at the northeast corner of Farm Lane and Service Road. The approximate watershed area is 12.8 acres (5.2 hectares), 40% of which is impervious. Water collects at the lowest point of the watershed, in the underpass on Farm Lane constructed also in 2009, and is pumped up to a large storage cistern adjacent to the roadway. Once the tank fills to 11,016 gallons (41,700 liters) a float switch is triggered and an additional 700 gallon (2649 liters) per minute pump moves water in a pipeline across the north edge of the bioretention basin and releases the flow on the east side.

When water enters the 2500 ft<sup>2</sup> (0.023 hectare) basin, it flows over a rocky area and disperses into the influent sampling zone or the wetland overflow. What passes through the influent sampling zone goes over a concrete equalization basin and through five bays, hydraulically isolated from each other. Water can collect in the larger body of the retention basin where native and ornamental plants are grown.

An underdrain beneath the central basin carries effluent water through the effluent sampler zone and out of the bioretention system to connect with the Michigan State University stormwater system at large.

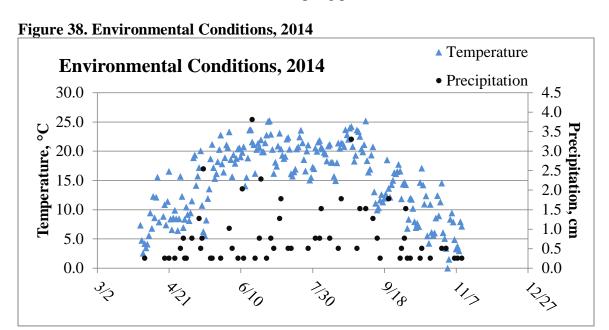
In the influent sampling zone and the effluent sampling zone, ISCO samplers have been installed. ISCO 6700FR samplers have a 24 bottle configuration with programmable draw times. Samplers are refrigerated to preserve the samples. Flow data is collected by Area Velocity Flow 750 Modules installed in each sampling zone.

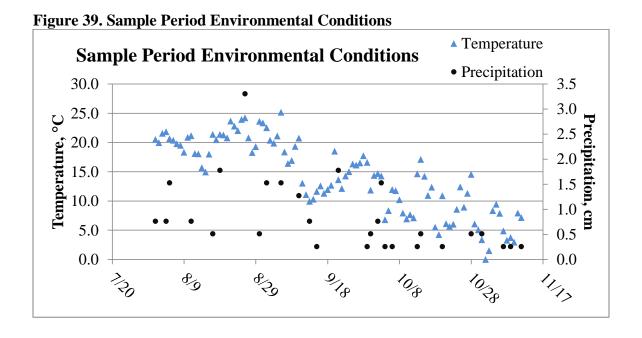
Several years of data have been collected on site performance. Some data has shown sporadic peaks in Chemical Oxygen Demand (Thode 2013).

### APPENDIX B. Supporting Data During Research

The figures below show two large rain events, one in June and another in September.

Daily average temperatures were consistent during establishment period from June to September and then declined over the course of the sampling period





# APPENDIX C. Enviroscan Data from Establishment Period

# Figure 40 Enviroscan image

The Enviroscan machine held five sensors over its

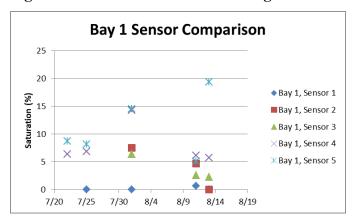
length. The testing ports could not be buried more than two

feet deep in order to protect the lining of the bioretention

basin. Therefore, only the bottom three sensors, numbers 3
5, were in contact with the soil. The additional sensor data,

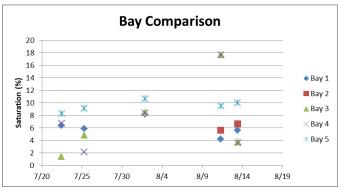
numbers 1-2, were excluded from the averages used in this analysis.

Figure 41. Enviroscan Sensor Readings



Water content was measured twice
weekly during establishment and during
the extent of water quality testing. Later
season data was lost due to failures during
transfer, and therefore establishment is
based on a smaller subset of the data.

Figure 42. Enviroscan Measurements in Bays



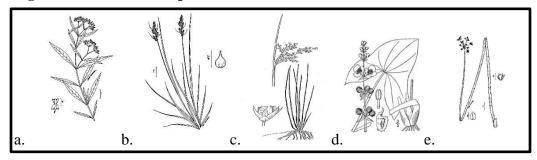
The Enviroscan measures are based on the default calibration curve for sandy loam soil.

#### APPENDIX D. Plant Selection and Characteristics

There are many suggestions for plant selection in stormwater management system and constructed wetlands. Primary guidelines include ecological acceptability (resistance to pests without invasive tendencies), tolerance of local climatic conditions, tolerance of pollutants, ready establishment and propagation, and high pollutant removal capacity (Tanner 1996). Pollutant removal is estimated based on direct assimilation and storage ability and also by microbial symbiosis, as in nitrification and denitrification.

Research has tested many different plants for these performance characteristics, but for others generalizations must be made. The plants selected for this study were chosen from the Michigan LID manual to reflect a range of water table depth and a mix of monocots and dicots. Images and basic information were taken from the PLANTS database (USDA 2018).

Figure 43 Bioretention species details



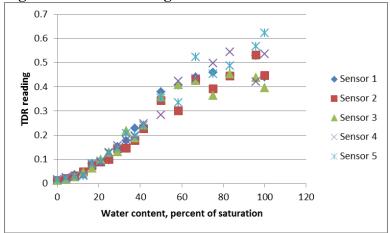
a.) Asclepias incarnata (swamp milkweed) is a perennial forb native to east and North America as far north as Hudson Bay. As an obligate wetland plant, it is recommended for stormwater management systems for hardiness and aesthetics. It increases in growth under high nutrient loading and despite aggressive competition from species such as canary reed grass (Green and Galatowitsch 2002).

- b.) *Carex vulpinoidea* (fox sedge) is a prolific, monocot perennial native to all of North America where it is a facultative or obligate wetland plant. It has been successful in flood-pulse wetlands where it also serves as a wildlife food plant (Drinkard et al. 2011).
- c.) *Juncus effusus* (common rush) is a perennial grass native to all non-desert areas of North America, primarily as an obligate wetland plant. A thin stemmed and densely-growing plant, it has been used extensively in pollutant uptake research where it shows good hydraulic performance, allowing mixing while resisting erosion damage with a high effective volume ratio in its growth habit (Guo et al. 2017).
- d.) *Sagittaria latifolia* (broadleaf arrowhead) is a perennial forb native to all but the northernmost region of North America, exclusively as an obligate wetland plant. This species was found to be successful (although not prolific) in several urban wetland sites in New York, despite the pressures of invasive species and elevated pollution (Larson et al. 2016),
- e.) Schoenoplectus tabernaemontani aka. Scirpus validus (soft stem bulrush) is a perennial grass native to all of North America as an obligate wetland plant. Research by G.S. Edwards (1992) found that most roots occurred in the upper 12-15 cm of soil media, filling approximately 5% of substrate with roots. As in this study, Edwards witnessed poor survival of plants after long periods of submergence. Their growth habit may limit the clogging and hydraulic limitations of roots that proliferate the soil profile completely (Pedescoll et al. 2013).

### APPENDIX E. TDR Sensor and Data

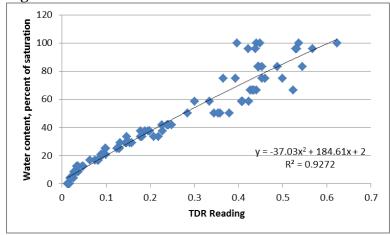
Time domain reflectometer (TDR) sensors were calibrated in the laboratory before installation in the columns. A five gallon bucket was filled with soil dried in the greenhouse. Water was added in 0.2 L increments until it pooled on the soil surface. With each addition of water, the TDR reading was recorded.

Figure 44. TDR Readings



Calibration recordings were then inverted to establish an equation converting TDR readings to water content. All data was converted before statistical analysis.

Figure 45. TDR Calibration



# APPENDIX F. Stock water Design

Table 8. Stock water design from Lucas and Greenway

Pollutant	Chemical	Stock	Stormwater	Stormwater
		Concentration	Stock Addition	Concentration
		(g/L)	(uL/L)	(mg/L)
Ortho-	Potassium Phosphate	7.97	100	0.79
Phosphate	K <sub>3</sub> PO <sub>4</sub> (mm 104g)			
Total Disso	lved Phosphorus			0.79
Ammonia	Ammonium Chloride	4.12	100	0.41
	NH <sub>4</sub> CL (mm 28g)			
Nitrogen	Potassium Nitrate	8.69	102	0.97
Oxides	KNO <sub>3</sub> (mm 50g)			
Org.	Nicotinic Acid	6.62	365	3.47
Nitrogen	$C_6H_5NO_2 \ (mm\ 64g)$			
Total Disso	lved Nitrogen			4.86
Cadmium	Cadmium diNitrate	0.26	10	0.003
	$CdN_2O_6$ (mm110g)			
Copper	Copper Sulfate	54.4	10	0.544
	CuO <sub>4</sub> S (mm 77g)			
Lead	Lead Nitrate	15.0	10	0.150
	Pb(NO <sub>3</sub> ) <sub>2</sub> (mm 114g)			
Zinc	Zinc Chloride	57.7	10	0.578
	Cl <sub>2</sub> Zn (mm 64g)			
Total Metal	S			1.27

Anticipated dissolution in stormwater includes the following reactants and products:

$$K_3PO_4 \rightarrow 3 K^+ + PO_4$$
  $CdN_2O_6 \rightarrow Cd + 2 NO3$   $NH_4Cl \rightarrow Cl^- + NH_4^+$   $CuO_4S \rightarrow Cu^{+2} + SO_4$   $KNO_3 \rightarrow K^+ + NO3$   $Pb(NO_3)_2 \rightarrow Pb^{+2} + 2 NO_3$   $C_6H_5NO_2 + 5.5 O_2 \rightarrow 6 CO_2 + H_2O + NH_3$   $Cl_2Zn \rightarrow 2 Cl^- + Zn^{+2}$ 

Nicotinic acid is responsible for theoretical chemical oxygen demand. Expected concentration is calculated based on molar mass below. However, nicotinic acid has proven resistant to dissolution even during digestion with dichromate COD, yielding as only 60 mg/L in experiments with theoretical COD of 500 mg/L, therefore adjustments have been made.

$$C_6H_5NO_2 + 5.5 O_2 \rightarrow 6 CO_2 + H_2O + NH_3$$

Molar mass: 
$$64g + 5.5(16) g \rightarrow 6(22) g + 10g + 10g$$

 $1 \text{g C}_6 \text{H}_5 \text{NO}_2 \Rightarrow$  theoretical 1.375 g O<sub>2</sub>, actual 0.165 g/L O<sub>2</sub> demand

The stormwater components designed by (Stuber 2012) based on nutrient loads from (Lucas and Greenway 2008b) provided the basic stormwater mix. The stock concentration was then diluted in each column and bay according to the prescribed water content. The dilution was designed so

that each trial column and bay received the same mass of pollutants as the other columns or bays.

# APPENDIX G. Standard Operating Procedure: Total Solids

#### USEPA Gravimetric Method 8271 (USEPA 2015)

Samples are collected using identifiers O for the orange series of columns, G for the green series of columns, B for the blue series of columns, and Bay for the field experiments, as well as a number 1-5 to indicate water content classification. The label "source" indicated influent samples.

Aluminum drying dishes are labeled for each sample with duplicates "a" and "b."

The initial weight of empty labeled dish is measured on a balance and recorded.

Sample collection bottles are mixed by inversion up to three times to ensure mixing.

Using a graduated cylinder, 25 mL of each sample was measured and poured into the respective dish.

Full dishes are placed on metal tray.

Once all samples have been prepared in this way, tray is placed in drying oven.

Oven is heated to approximately 105 °C for at least 6 hours.

Samples are checked to ensure complete drying, and then oven is turned off and allowed to cool.

Once cooled to room temperature, the aluminum dishes are individually weighed and their mass

is recorded.

Total solids measurement is calculated by subtracting initial weight from the final weight.

### APPENDIX H. Standard Operating Procedure: Nitrate and Phosphate

Determination of Inorganic Anions by Ion Chromatography Method 300(Pfaff 1993)

Samples are collected using identifiers O for the orange series of columns, G for the green series of columns, B for the blue series of columns, and Bay for the field experiments, as well as a number 1-5 to indicate water content classification. The label "source" indicated influent samples.

Stock solutions for standards were purchased from chemical supplier.

Dionex Ion Chromatography ICS 5000 was used for anion analysis, separated on AG 22 guard column and AS22 analytical columns with a mobile phases of 4.5 mM sodium carbonate and 1.4 mM sodium bicarbonate at a flow rate of 1.2 mL/min and a conductivity detector. Standards with at least five levels (i.e. 0.1 ppm, 1 ppm, 10 ppm, 50 ppm, 100 ppm, 200 ppm) were used to make linear calibration curves. Non-detect values for samples were replaced with a value 50% of the lowest recorded concentration for the parameter of concern.

### APPENDIX I. Standard Operating Procedure: Total Nitrogen and Total Phosphorus

Persulfate digestion method 4500-N C and 4500-P J. for simultaneous determination of Total Nitrogen and Total Phosphorus(De Borba, Jack, and Rohrer 2016)

Persulfate digestion method is as described in *Methods and Standards for Examining Water and Wastewater* and Thermo Fisher Scientific guidance with modifications based on the work of N. Aryal (Aryal 2015).

Samples are collected using identifiers O for the orange series of columns, G for the green series of columns, B for the blue series of columns, and Bay for the field experiments, as well as a number 1-5 to indicate water content classification. The label "source" indicated influent samples.

A digestion reagent was prepared and stored at room temperature in the laboratory. 20.1 g low nitrogen potassium persulfate ( $K_2S_2O_8$ ) and 3 g sodium hydroxide (NaOH) were dissolved in 1000 mL e-pure water. Borate buffer was prepared using 61.8 g boric acid and 8 g NaOH in 1000 mL water. 5mL of digestion reagent was added to 10 mL of standard solution or water sample in labeled glass vials. After inverting several times and securing in a wire rack, the standards and samples were heated at 110 °C in an autoclave for 30 minutes. Once cooled, 1mL of borate buffer was added. The complete solution was filtered into a 10 mL IC vial using 45 uM cellulose acetate filters. This procedure always maintained the same dilution ratio (digestion reagent: sample: borate buffer = 5:10:1).

Dionex Ion Chromatography ICS 5000 was used for anion analysis, separated on AG 22 guard column and AS22 analytical columns with a mobile phases of 4.5 mM sodium carbonate and 1.4 mM sodium bicarbonate at a flow rate of 1.2 mL/min and a conductivity detector. Standards with at least five levels (i.e. 0.1 ppm, 1 ppm, 10 ppm, 50 ppm, 100 ppm, 200 ppm) were used to make

linear calibration curves. Non-detect values for samples were replaced with a value 50% of the lowest recorded concentration for the parameter of concern.

The method described above requires 30 minutes of digestion in an autoclave, while the official method recommends at least 55 minutes for complete separation of phosphate components. This discrepancy was not identified until all samples had been processed; therefore TP data were not included in qualitative or quantitative analyses.

# APPENDIX J. Examining Normality and Seasonality, QQ Plots and Residuals

The Q-Q plot is a graphical tool for visual evaluation of distribution normality (Ford 2016). Quantile data is plotted against the predicted quantiles of a theoretical normally distributed data set based on the same mean and standard deviation. A high R<sup>2</sup> value with a line near 1:1 would prove distribution normality. Plots below show each bay and column series (1, 2, 3, 4, and 5) in terms of each water quality parameter. Non-linear QQ plots are identified as "NON NORMAL" and included in results and discussion section in terms of their Wilcoxon rank sum analysis of statistically significant variance.

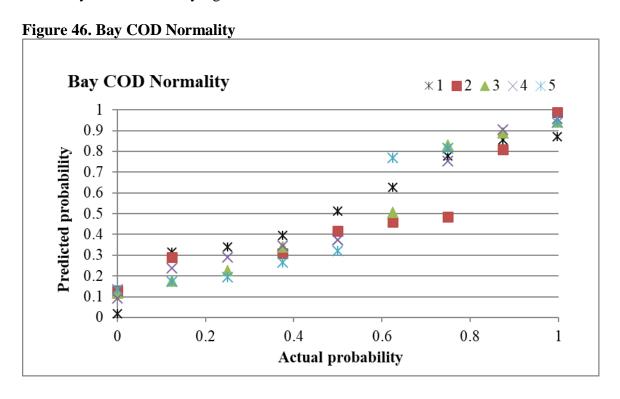
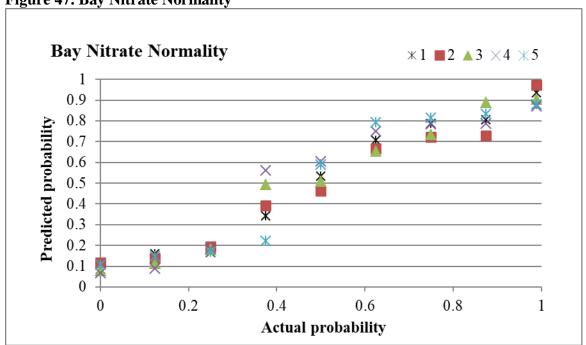


Figure 47. Bay Nitrate Normality





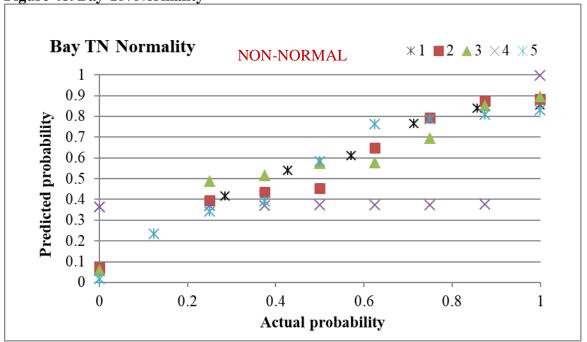
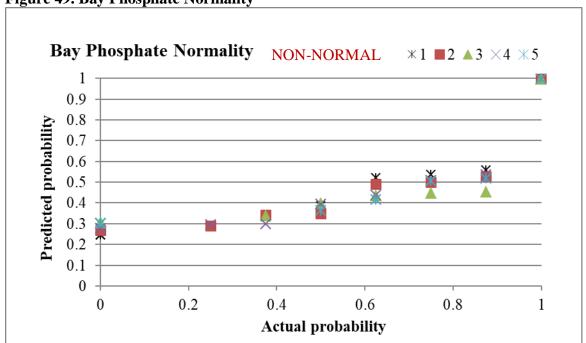


Figure 49. Bay Phosphate Normality





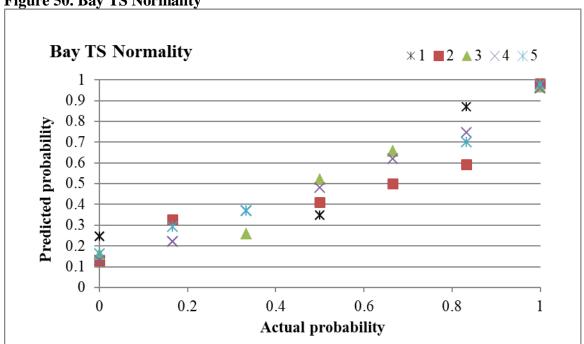
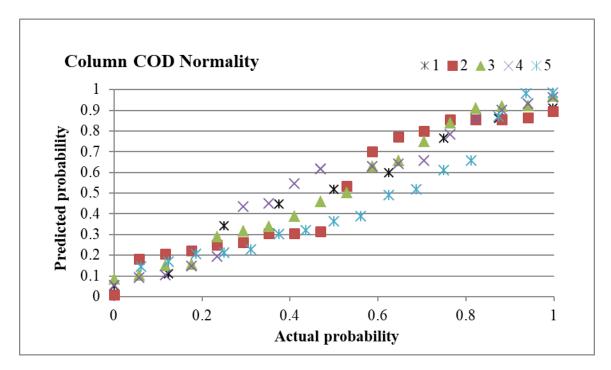


Figure 51. Column COD Normality





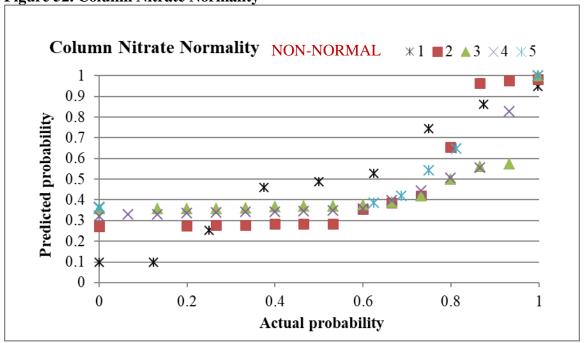
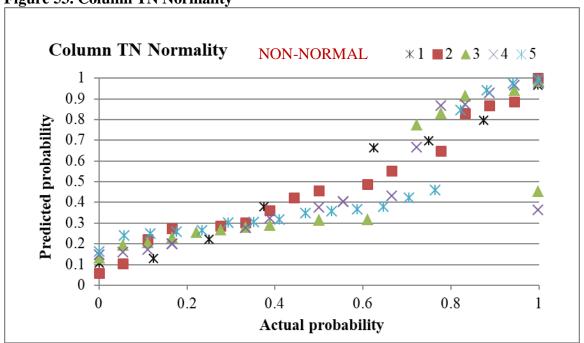
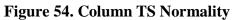
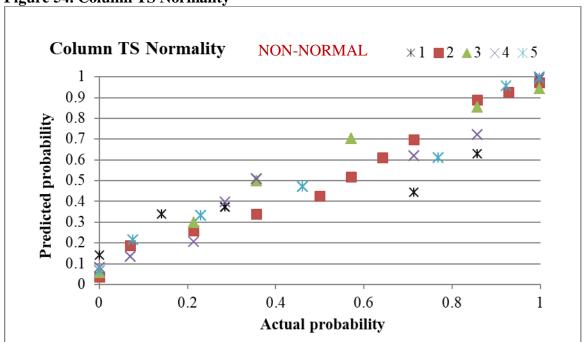


Figure 53. Column TN Normality

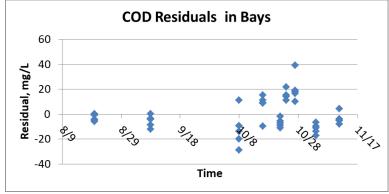






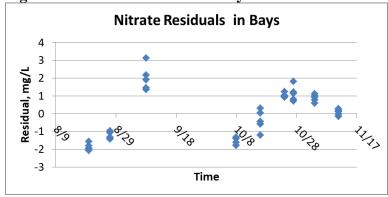
After a review of QQ Plots, residuals were examined to double-check normality and identify seasonal trending. Graphs did not indicate positive serial correlation (concentrations trending in line with previous point) or negative serial correlation (concentrations trending opposite previous point) and therefore seasonality is not an apparent determiner (Ott and Longnecker 2001).

Figure 55. COD Residuals in Bays



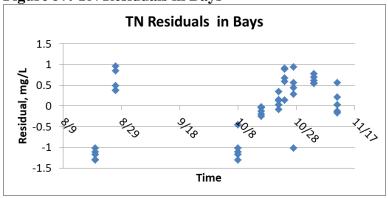
No apparent serial correlation

Figure 56. Nitrate Residuals in Bays



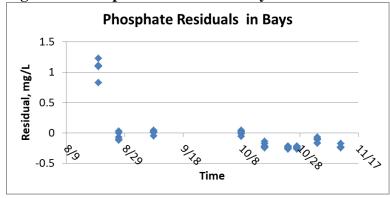
No apparent serial correlation

Figure 57. TN Residuals in Bays



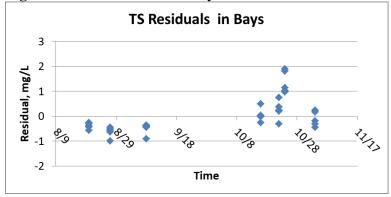
No apparent serial correlation

Figure 58. Phosphate Residuals in Bays



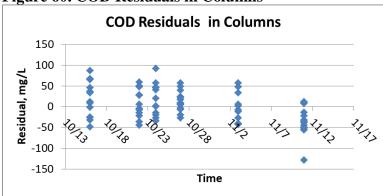
No apparent serial correlation

Figure 59. TS Residuals in Bays



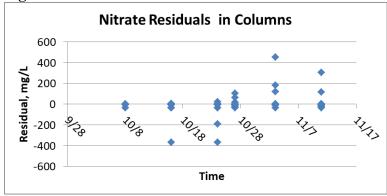
No apparent serial correlation

Figure 60. COD Residuals in Columns



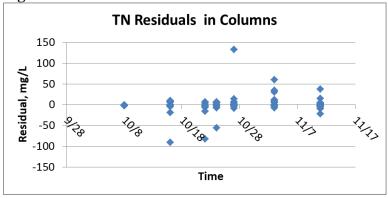
No apparent serial correlation

Figure 61. Nitrate Residuals in Columns



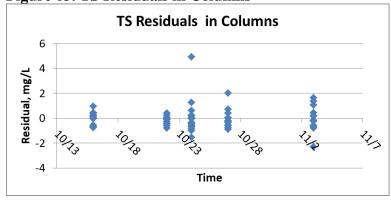
No apparent serial correlation

Figure 62. TN Residuals in Columns



No apparent serial correlation

Figure 63. TS Residuals in Columns



No apparent serial correlation

# APPENDIX K. Photographs from Research

Figure 64. Photograph of entire Farm Lane Bioretention Site



Figure 65. Photograph of Hydraulically Isolated Field Bays



Figure 66. Photograph of Wetland Overflow Area



(adjacent to bioretention field bays)

Figure 67. Bioretention Bays with white PVC water content monitoring ports installed



Above, plants have just been put in place and are beginning establishment period.

Figure 68. End of season Bioretention Plants



Several species of plants thrived in bioretention field bays. Periodic weeding was necessary.

Figure 69. Photograph of Laboratory Columns



Laboratory columns were installed in greenhouse behind Farrall Hall. Disassembled column to left shows how modifications allowed different levels of observation.

Water samples were labeled with color, number, and location as necessary to identify different trials. They were examined bi-weekly along with larger stormwater monitoring in Farm Lane utilities.

Figure 70. Photograph of Sampling Bottles



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