

Analyzing nutrient concentrations of surface soil within established switchgrass plots

By Dana Devore

Advisor Dr. Christian Krahforst

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ABSTRACT

Exponential population growth and consumption cause rippling effects on the environment worldwide. The Chesapeake Bay is an extremely productive estuary suffering from excessive nutrient runoff from agriculture, industrial, and residential areas all of which are increasing in magnitude with growth. There has been an array of regulations established to try and help with the current nutrient pollution. One of these is establishing filter strips with approved vegetation to impede runoff. Runoff occurs most often when soil is satiated with water and flow from rain or melting snow runs across the satiated area causing soil to runoff with the flow. Filters strips help to inhibit that process most often by stabilizing soil. Switchgrass is a native perennial grass studied for its efficient use of nutrients and its potential to be used in filter strips. The focus of this study is an preliminary examination assessing the capacity of switchgrass to reduce nutrients in

surrounding soil in the Chester River watershed, a tributary to the Chesapeake Bay. The hypothesis is that the presence of established stands of switchgrass reduces the amount of nutrients, specifically nitrogen and phosphorus in the surface soil. To do so, soil samples were taken at Chino Farms from within a switchgrass field, in an adjacent forested area, and an adjacent grassland area using a TrueTemper corer. After the samples were collected, they were analyzed for soil moisture content using a gravimetric soil moisture procedure, bioavailable nitrate using a KCl extraction and a HACH nitrate kit and total reactive phosphorus using a Mehlich-3 extraction. The results were generally in support of the hypothesis in that nitrate values were lowest in the switchgrass samples when compared to the forested and grassland areas. The phosphate concentrations in the switchgrass field samples were found to be in between the concentrations found in the grassland samples and the forested samples. There is an array of factors that could have played a role in the results however due to certain restraints, not all of them could be looked into.

INTRODUCTION

This study is a preliminary examination focusing on switchgrass' ability to sequester nutrients in surrounding soil. The hypothesis is that the presence of established stands of switchgrass reduces the amount of bioavailable nutrients in the surface soil. To do so, soil samples were taken from within a switchgrass field, a forested area, and a grassland area in close proximity. Nutrient levels, specifically nitrogen and phosphorous content, were measured to determine the effect of switchgrass on bioavailable nutrients in soil. The switchgrass in this study is of the Kanlow variety, which is a lowland type. The plot selected for sample extraction is located within the Chester River Field Research

Station in Chino Farms in Queen Anne's county, MD. Determining whether nutrient levels are due to infiltration into groundwater or plant uptake is beyond the scope of this study. Nonetheless, it is important to better understand the role of switchgrass in nutrient mitigation from agricultural runoff, so that informed decisions can be made to decide whether switchgrass is a plausible best management practice for the Chester River watershed, and possibly for the Chesapeake region. For the purpose of this study, it is assumed that infiltration of nutrients into groundwater has reached a steady state.

The Chesapeake Bay is the largest estuary in the United States and is of great importance to the country's economy due to the productive aquatic community it supports. It is a destination for recreation, provides a large fishery, and sustains an abundance of organisms. However, in recent years, the condition of the Bay has been in decline.

Since European arrival to the Chesapeake region, agriculture has dominated the vast watershed. The Chesapeake was first farmed by Native Americans using slash and burn farming for subsistence crops. European settlers arrived and did the same, but then the agriculture industry vastly expanded with tobacco. Timber was also very popular in the region, and left deforested land that was later used for agriculture. As years passed, tobacco growth in the region declined and more and more crops were grown in abundance for resale rather than subsistence (Wennersten, 2001).

The combination of human population growth in the watershed, the expansion of agriculture and overharvesting have been detrimental to the condition of the Chesapeake waters. The Chesapeake is very susceptible to pollution into its immense watershed at 64,000 acres crossing through New York, Pennsylvania, West Virginia, the District of

Columbia, Delaware, Maryland, and Virginia (U.S. Environmental Protection Agency, 2004). The Chesapeake's large watershed allows pollution to accumulate across great distances before reaching the Bay. Furthermore, the bathymetry of the Bay allows pollution to greatly affect its waters. It is very shallow which further heightens the effects of pollution due to the smaller volume of water but extensive surface area (Matuszeski, 2007).

One Some of the most worrisome pollutants in the Chesapeake Bay and its tributaries are nutrients. Nutrients are not harmful to aquatic communities, but excessive nutrients are, which is the key problem in the Chesapeake Region. Excessive nutrient pollution can hasten the eutrophication process in water bodies (Lim, Edwards, Workman, Larson & Dunn, 1998), and eutrophication in estuaries can cause harmful algal blooms, hypoxia, and fishery habitat decline (Simpson, Sharpley, Howarth, Paerl & Mankin, 2007). Hypoxia can lead to fish kills due to limited oxygen demand. Nutrient pollution can also lead to change in species composition and lowered health of aquatic communities depending on species' threshold for changing water quality parameters. Oysters are an example of such. Oyster populations have greatly declined throughout the Chesapeake. Part of the decline is due to disease but over harvesting as well as more turbid waters due to increased sedimentation and phytoplankton blooms do not allow for the thriving population levels the Chesapeake once supported. Decreased oysters population as well as other filter feeding organisms allows for increased effects of pollution because the organisms are not filtering the water, which helps prevent increased control turbidity which allows species sensitive to light quality turbidity to

flourish. Bacteria levels may also increase with excess nutrient concentrations (Ouyang, 2012) causing more harm to water bodies.

The two most troublesome nutrients are nitrogen and phosphorus. These two nutrients are essential for plant growth. C and so; cropland is often treated with manure and organic fertilizers that are high in nitrogen and phosphorus. Often phosphorus levels are higher than what is necessary for growth because organic fertilizers are applied to meet the nitrogen requirements, which can cause elevated phosphorus levels (Weismiller, Steinhilber & Salak, 2012). A study done in NE Mexico found that most of the heightened nitrogen concentrations were due to animal and domestic waste and ammonia fertilizers (Pasten-Zapata, Ledesma-Ruiz, Harter, Ramirez & Mahlkecht, 2014). However, there is some nitrogen in soil naturally (Pasten-Zapata, Ledesma-Ruiz, Harter, Ramirez & Mahlkecht, 2014) and some nitrogen can be from atmospheric deposition. Additionally, septic tank leaching and animal manure can cause nitrate pollution in groundwater (Pasten-Zapata, Ledesma-Ruiz, Harter, Ramirez & Mahlkecht, 2014). A study in the Lower St. John's River, FL, showed excess nutrients were coming from surface runoff from urban, rural, and agricultural land. It specifically demonstrated that nutrients collect in cropland ditches and contaminated groundwater discharge from septic tanks (Ouyang, 2012).

Sediment is another example of an important pollutant from the Chesapeake Bay watershed. It negatively affects aquatic organisms (Nelson, Ascough II & Langemeier, 2006) especially epifauna and infauna that can suffocate from increased sediment loads and runoff. Sediments affect turbidity and can also decrease the amount of light for aquatic organisms dependent on photosynthesis for survival. Sediment, nitrogen, and

phosphorus, are major components of nonpoint source (NPS) pollution, which is pollution that does not come from a single identifiable source (Blanco-Canqui, Gantzer, Anderson, Alberts & Thompson, n.d.). This is a major setback for the Bay because NPS pollution is much harder to manage since it is widely distributed; this means that one specific source for pollutants cannot be identified and hence numerous counter measures need to be developed and deployed to mitigate the effect.

Government legislation has been enacted to try and improve the condition of the Chesapeake Bay and its tributaries. In recent years, the EPA Chesapeake Bay Program has been focused on NPS pollution as what is causing the decline in Bay water quality due to its increasing impact on the quality of the Bay environment (Magette, Brinsfield, Palmer & Wood, 2003). The Bay Agreement was started to decrease nitrogen and phosphorus loading into the Chesapeake Bay. Its original goal was for a 40% reduction and for the Bay to be removed from the federal list of impaired waters by 2010. Even with the major progress made, the goals have not been met (Weismiller, Steinhilber & Salak, 2012). There is also the Water Quality Improvement Act, which states that nutrient management plans become a requirement for all farms with a gross income of twenty-five thousand dollars annually and holding at least eight “animal units” (Weismiller, Steinhilber & Salak, 2012). The Maryland Agricultural Water Quality Cost-Share Program (MACS) is a program that aids farmers with best management practices such as animal waste management systems, field borders and windbreaks, and roof runoff management, but to be eligible for MACS a farm must be determined to have “excessive levels of soil, nutrients or pollution running off into Maryland’s waters” (Weismiller, Steinhilber & Salak, 2012). Best management practices (BMP) are being set in place to

try and solve pollution issues (Lee, Isenhardt & Schultz, 2003). BMP are government-approved techniques to inhibit water pollution. Riparian buffers are an example of a recommended BMP to aid in NPS pollution especially from agricultural surface runoff (Lee, Isenhardt & Schultz, 2003). Tall, stiff-stemmed, native, warm-season, perennial grasses are thought to be most successful in mitigating pollutant runoff (Blanco-Canqui, Gantzer, Anderson, Alberts & Thompson, n.d). Riparian buffers are strips of vegetation located along small tributaries to prevent the effects surrounding land use may have on water quality. They are interchangeable with vegetated filter strips or grass barriers except that riparian specifies that it is grown along the bank of a waterway. Although these practices may differ slightly in design, species, and management (Blanco-Canqui & Gantzer, Anderson, 2006), they are all used as a BMP to stop sediment runoff and reduce nutrient loads (Lee, Isenhardt & Schultz, 2003). Filter strips can be placed along banks to help retain concentrated runoff or created within fields to prevent runoff pooling before it discharges to receiving waters (Grismer, O'Geen & Lewis, 2006). For a buffer to be successful, the plant species used must be able to assimilate contaminants through uptake during growth before they can reach groundwater or runoff (Lin, Lerch, Garrett, Jordan, & George, 2007). Various studies have shown that even low densities of buffers have the ability to remove a significant amount of runoff, which often contains contaminants (Lim, Edwards, Workman, Larson & Dunn, 1998). Many species used in vegetated filter strips also have the ability to be grown for use as a biofuel feedstock for ethanol production.

Switchgrass, *Panicum virgatum*, has been studied for a multitude of reasons including its use as a vegetated filter strip to aid in surface runoff. Switchgrass is native to the United States having migrated from the tropics across Central and North America

(Parrish & Fike, 2005). Before European arrival, one could find switchgrass across two-thirds of the United States in unforested areas (Parrish & Fike, 2005). Today, it has evolved into multiple divergent populations and can be found along the East Coast, west to Arizona and Nevada and north to parts of southern Canada (Parrish & Fike, 2005). Switchgrass is a warm season perennial grass and a C4 plant, meaning its photosynthetic pathways are adapted to be more efficient with nitrogen and water use and tolerant of heat and drought (Parrish & Fike, 2005). Due to physiological differences, C4 plants can close their stomata to prevent water loss under arid conditions unlike C3 plants. The switchgrass plots used in this study can be seen in Figure 1.



Figure 1. Mowed switchgrass field in which samples were taken within at Chino Farms in February 2014.

There are two different types of switchgrass: lowland and upland. Each have adapted to different environments. Lowland switchgrass is taller with thicker stems and

can grow better in wetter conditions than upland, which is shorter, thinner, and grows better in dry conditions (Owens, Viands, Mayton, Fike, Farris, Heaton, Bransby & Hong, 2013). Still, both forms exhibit have overlapping canopies between plants and are stiff-stemmed. Switchgrass is highly productive. It accumulates a great amount of biomass aboveground and has an extensive, deep root system below ground (Simpson, Sharpley, Howarth, Paerl & Mankin, 2007). Switchgrass leaves can grow up to three meters tall and are most often erectophile and amphistomic (Parrish & Fike, 2005). Switchgrass root systems can also extend up to three meters below the surface. This is particularly advantageous as it allows switchgrass to extract nutrients from deeper strata of soil thus improving soil conditions in a sizable surrounding area (McLaughlin & Kszos, 2005 and Maeght, Rewald & Pierret, 2013).

Much of a switchgrass plant's energy is allocated to its root system (McLaughlin & Kszos, 2005). At senescence, translocation of nutrients from the shoots to the roots occurs allowing the perennial tissues to store the nutrients needed for spring growth (Parrish & Fike, 2005). It has been observed that grasses like switchgrass can remobilize up to thirty percent of the nitrogen in its shoots to the roots and rhizomes at the end of the growing season (Owens, Viands, Mayton, Fike, Farris, Heaton, Bransby & Hong, 2013). Still, some nutrients are also removed when the biomass is harvested (Jiading, Worley, Wang, Lahner, Salt, Malay & Michael, 2009).

Switchgrass is grown for a variety of reasons. Historically, switchgrass was used and grown for forage across the Midwest (Parrish & Fike, 2005). Today, it is still being used for forage, for grazing, and for hay or silage to feed animals (Parrish & Fike, 2005). Research has shown that switchgrass can be used in papermaking, pharmaceutical

preparations, and mushroom production (Parrish & Fike, 2005). Switchgrass growth can provide microclimates creating habitat for a variety of invertebrates, reptiles, amphibians, and mammals (Parrish & Fike, 2005). This varies with plot size in that creating a monoculture would negatively impact species diversity. However, the primary focus of switchgrass growth and production in recent years has been for its use as a bioenergy feedstock (grown to be converted into biofuels) and or as a buffer for surface runoff.

The United States' current use of fossil fuels is not sustainable. Biofuels are a cost-effective, independent, and environmentally friendly way to produce energy (Jiading, Worley, Wang, Lahner, Salt, Malay & Michael, 2009). Switchgrass can be used as feedstock for cellulosic ethanol production. This would lessen the dependence on petroleum-based fuel and alleviate the issue of using corn for ethanol production, which would raise prices of corn, limit corn grown for food production, and inhibit biodiversity (Matuszeski, 2007). Unlike corn, switchgrass is efficient in its use of nutrients. It requires less fertilizer and leaves lower concentrations of nutrients in the soil after senescence (Matuszeski, 2007). There are environmental benefits of replacing fossil fuels with ethanol such as reduced emissions and a better energy balance by being carbon neutral. There is also the advantage that ethanol production would be independent of any foreign energy supply and has the opportunity to create domestic jobs and revenue (Simpson, Sharpley, Howarth, Paerl & Mankin, 2007).

Switchgrass biomass can be burned for fuel, pelleted, or converted for cellulosic ethanol (Nelson, Ascoug II & Langemeier, 2006). It has been said that biofuel demand will increase and that the market for grain-based ethanol is growing (Matuszeski, 2007 & Simpson, Sharpley, Howarth, Paerl & Mankin, 2007). The US Department of

Environment has chosen switchgrass as a bioenergy crop (Jiading, Worley, Wang, Lahner, Salt, Malay & Michael, 2009). However, multiple other factors must be considered until switchgrass harvesting is increased for biofuel production.

Switchgrass grown for biofuel may displace other crops. In this case, it must be useful and in demand (Magette, Brinsfield, Palmer & Wood, 2003). Switchgrass grown on crop reserve land, marginal soils, or other land that had not previously been used for crops has environmental consequences. Because fertilizer is normally applied each year, converting land to cropland for switchgrass production has been shown to greatly increase nitrogen and phosphorus loading (Simpson, Sharpley, Howarth, Paerl & Mankin, 2007). The release of nutrients from switchgrass biomass when burned can also have a negative environmental impact (Jiading, Worley, Wang, Lahner, Salt, Malay & Michael, 2009). The cost per milligram of production would have to be researched, but if costs were higher than other fuels environmental incentives could be looked at to drive down associated production costs (Nelson, Ascough II & Langemeier, 2006). Farmers could be paid for the use of switchgrass as a best management practice or for nutrient trading due to switchgrass' ability to reduce nutrients in the surrounding environment. Rather than direct payments, farmers can save money by reducing sediment loss and lowering nutrient and pesticides application needs since switchgrass prevents runoff and is an efficient user of nutrients (Nelson, Ascough II & Langemeier, 2006).

Switchgrass growth can provide an assortment of environmental benefits for both soil and water quality. Adding switchgrass to vegetative filter strips is an effective conservation practice (Blanco-Canqui, Gantzer, Anderson, Alberts & Thompson, n.d.). Surface runoff is slowed by switchgrass vegetation allowing particles to settle out and

increasing infiltration, which further reduces runoff of pesticides and nutrients (Lee, Isenhardt & Schultz, 2003). Surface runoff infiltrates into the soil and is buffered by switchgrass' extensive root system allowing for nutrient removal (Lee, Isenhardt & Schultz, 2003). It has been observed that filter strips of switchgrass and other similar grasses were able to reduce chemical oxygen demand and phosphorus levels in runoff through infiltration (Sanderson, Jones, McFarland, Stroup, Reed & Muir, 2000). The extensive root systems and stiff stems in switchgrass allow for decreased erosion and stabilization of soil (Parrish & Fike, 2005). Switchgrass root systems also have the ability to sequester carbon and increase soil organic matter due to their widespread growth, which can increase microbial activity and decrease the need for applied nutrients (Simpson, Sharpley, Howarth, Paerl & Mankin, 2007).

Nutrient reduction through switchgrass growth is a major point of interest and research. Studies have demonstrated that switchgrass is a much more efficient user of nitrogen than other crops such as corn (Sarkar, Miller, Frederick & Chamberlain, 2011). Nutrient reduction in surface runoff is initiated with retaining sediment and improving infiltration (Lin, Lerch, Garrett, Jordan, & George, 2007). Denitrification and plant uptake are the primary mechanisms behind switchgrass nutrient removal (Lin, Lerch, Garrett, Jordan, & George, 2007). Microbial denitrification reduces NO_3 to N_2 , NO , NO_2 , and other N gases (Lin, Lerch, Garrett, Jordan, & George, 2007).

Switchgrass is an efficient user of nutrients. This can be done effectively uses nutrients through plant use and water flow as well as remineralizing nitrogen from solid organic matter in the surrounding soil (Parrish & Fike, 2005). In some cases it has been observed that the amount of nitrogen removed can be even greater than the amount of

nitrogen applied (Parrish & Fike, 2005). Still, some studies observe setbacks with switchgrass production. One study reported that, of the applied nitrogen, removal in switchgrass was consistently below ten percent (Owens, Viands, Mayton, Fike, Farris, Heaton, Bransby & Hong, 2013). Another common hindrance with switchgrass production is its relatively long development. Switchgrass can take two years to produce a useful crop, three years before it is fully developed (Matuszeski, 2007) and may not contribute to nitrogen removal until it is established (Sarkar, Miller, Frederick & Chamberlain, 2011). In response to such, many advocates for switchgrass would emphasize that it requires both low maintenance and low production costs to establish.

To produce switchgrass, costs for seed, herbicide, insecticide, fertilizer, lime, drying, and tilling must be considered (Nelson, Ascough II & Langemeier, 2006). However, fertilizer is often not added until the second year of growth and at that time various nitrogen application ranges are recommended. Switchgrass requires less nitrogen and phosphorus than most other crops, decreasing costs for both the farmer and the environment (Simpson, Sharpley, Howarth, Paerl & Mankin, 2007). Additionally, switchgrass stands can grow for twenty plus years if managed properly (Simpson, Sharpley, Howarth, Paerl & Mankin, 2007). Another benefit for farmers is that switchgrass is able to establish under poor edaphic conditions where other vegetation may not be able to (Parrish & Fike, 2005). A study in NE Mexico demonstrated that failing crops might offer land for switchgrass production without taking away crop reserve program (CRP) or grassland areas (Sarkar, Miller, Frederick & Chamberlain, 2011). Switchgrass growth on marginal soils allows for production without impeding on cropland and with less energy input (Matuszeski, 2007).

MATERIALS & METHOD

Study Area:

The study area where soil samples were collected is located on Chino Farms in Queen Anne's county on the Eastern Shore of Maryland. More specifically, the switchgrass plots are located in the Chester River Field Station, which is comprised of 228 acres of restored grassland and home to a variety of ongoing field research projects. Researchers at Penn State University planted the switchgrass plots. The plots were established in 2003 and since then, they have received one fertilizer application of approximately 60 pounds of nitrogen fertilizer per acre every spring. The land is adjacent the Chester River, a tributary to the Chesapeake Bay used primarily by recreational boaters and local watermen. The Chester River watershed is dominated by agriculture with some urban and forested land. The population of the Chester River watershed is unique in that it has remained relatively stable since the first census in 1790 (details can be found from the United States Census Bureau data). Yet, the Chester River suffers from much of the same pollution the Chesapeake Bay does, excess nutrients and sedimentation.

Site Specifics

A total of six sites were sampled. Sites S1 and S2 were located within the switchgrass plot, sites F1 and F2 were located within an adjacent forested area, and sites G1 and G2 were located in an adjacent grassland to be used as controls (Fig 2). Sites S1, F1, and G1 ran parallel from north to south and sites S2, F2, and G2 did the same to try to evaluate variability of runoff coming from a farm located on east of the sample sites where runoff had been observed to be coming from.

Soil Sampling Sites

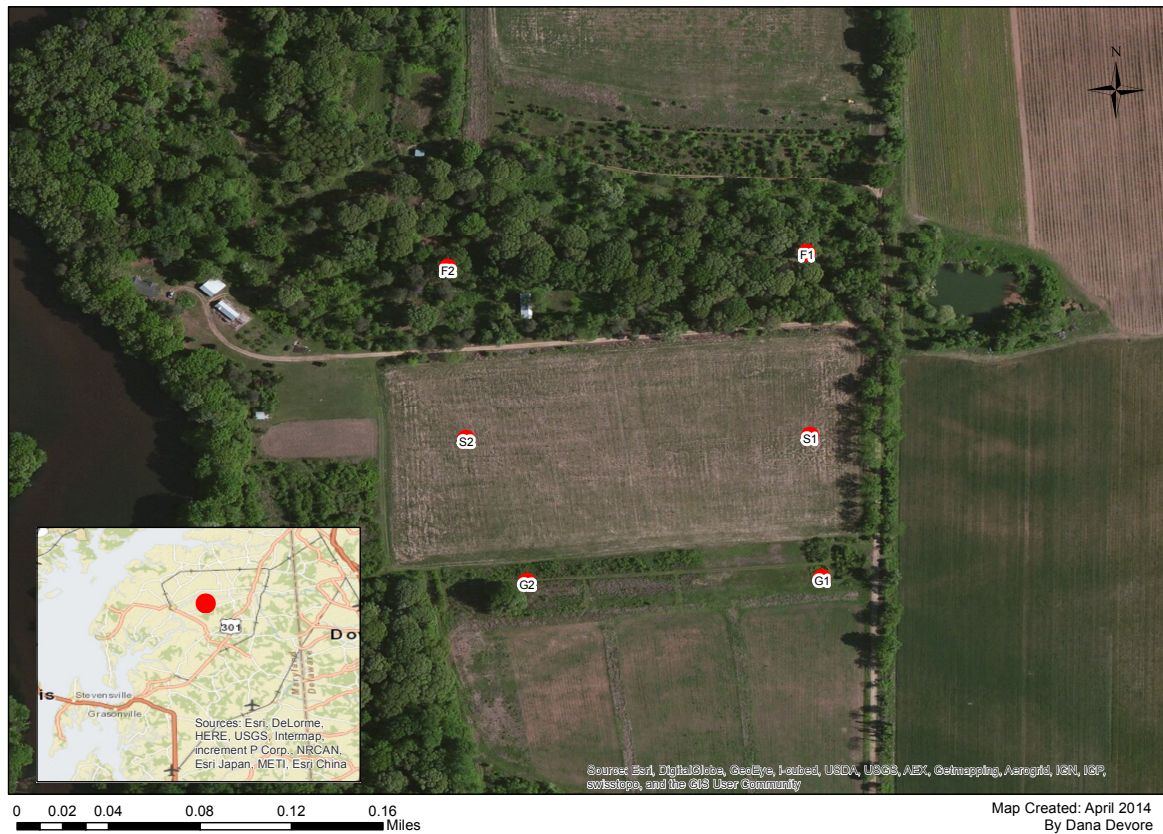


Figure 2. Map of the sample locations on Chino Farms, MD sampled in February 2014.

Sample Collection:

The sample collection took place on February 24, 2014. The ambient air temperature was 1.2°C and conditions were breezy and sunny. Before sample collection, the organic layer was removed (such as leaves, stones and other debris), and not included in the collection. Each site was sampled three times using a True Temper premium BP soil corer (Fig. 3) and compiled as one sample for a total of six samples for each testing date. The probe reached a depth of approximately nine inches, and the bottom two inches was saved for nutrient level analysis and the rest was discarded from each coring. Soil temperature was measured using a Fisher Scientific Traceable Calibration (model 02-402-0) thermometer. A 10 g subsample of soil was removed from each site and put in a

plastic bottle containing 100 mL of 2 M potassium chloride (KCl) to determine total inorganic nitrogen. Once extracted, the samples were immediately placed in plastic bags after collection to be transported back to the lab. Sample information such as sampling time, amount of sample retrieved for analysis, temperature and soil characteristics are noted in Table 1 below.

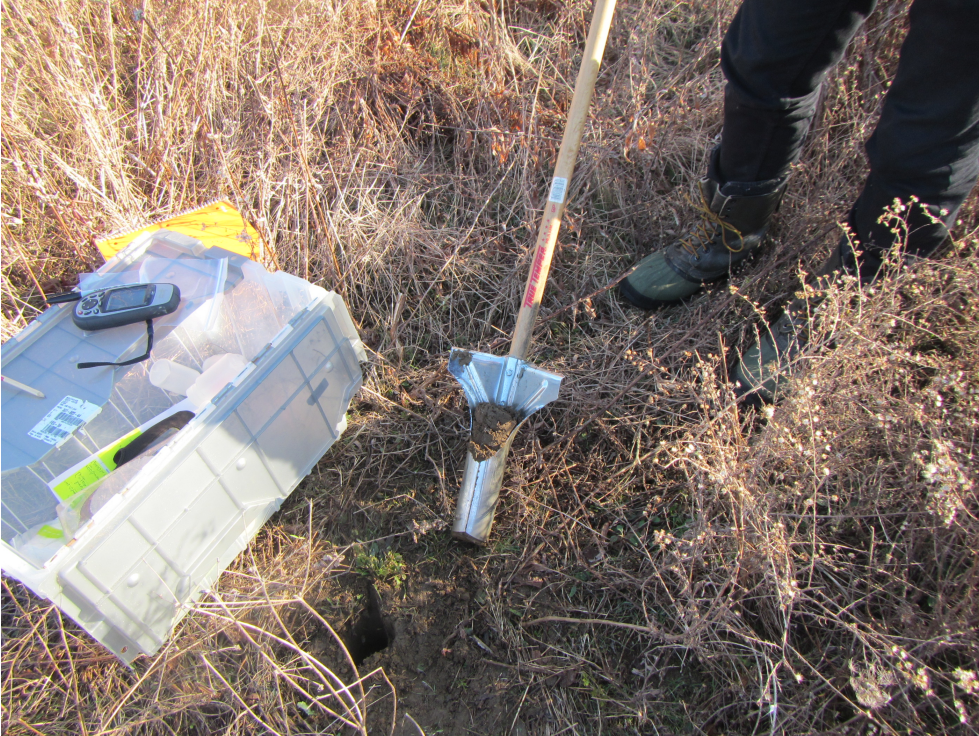


Figure 3. True Temper premium BP soil corer used to collect soil samples.

Table 1. Site and sample details for soil collected February 2014 using TrueTemper corer. Soil added is the amount of subsample weighed out for the KCl extraction.

Site	x (N)	y (W)	Sampling time	Soil temperature (°C)	Soil added (g)	Notes
S1	75.98181	39.22463	8:55 AM	2.4	9.9	Sandier, lighter
S2	75.98437	39.22438	9:28 AM	2.6	9.3	Sandier, lighter
F1	75.98173	39.22583	9:08 AM	4.5	9.9	Sandy
F2	75.98441	39.22587	9:18 AM	5.4	11.2	Sandy
G1	75.96555	39.112	8:40 AM	3.2	10.6	n/a
G2a	75.98411	39.22382	9:36 AM	3.8	11.2	Dryer soil, harder to core, found worms
G2b	75.98411	39.22382	9:36 AM	3.8	10.7	Dryer soil, harder to core, found worms

Analyzing procedure:

Gravimetric Soil Moisture:

Moisture content of each sample was determined by gravimetric analysis. Individual soil samples were dispensed in a separate, clean, dry pre-weighed beakers (one for each site). The beakers were then weighed again and the exact weight of each sample was noted. The beakers were next placed in a GREIVE laboratory oven (model L0-201C) and dried at 105°C for 48 hours. Once dried, the beakers were allowed to cool to room temperature before re-weighing. The moisture content in each sample was then determined by comparing the weight of each sample before and after heat treatment (see Table 2).

Determination of Soil Inorganic Nitrogen Content:

A KCl extraction procedure (Castle, 2005) was used to extract the inorganic nitrogen in the soil samples. All plastic bottles that were going to be used were first set in a 5% v/v HCl bath overnight to remove any possible nitrogen on the bottles. The next day the bottles were removed, rinsed with deionized water, and set to dry.

In the field, approximately 10 g of soil were subsampled from each site's sample and placed in 1.25 mL plastic Nalgene bottles. The soil was weighed using a OHAUS CL Series Portable Scale Model CL 201 Capacity 200 g x 0.1 g. 100 mL of 2 M KCl was added to each of the bottles, shaken and brought back to the lab. An additional subsample was collected to be used as a replicate. An additional bottle sample, containing only with 100 mL of KCl, was used as a reagent blank.

At the lab, the nine bottles were shaken for one hour at 200 rpm, and then left to settle for an hour after shaking. After settling, the samples were poured in Denville 14mL centrifuge tubes and spun at 4,000 rpm for ten minutes on a VWR Clinical 50 centrifuge. They were then frozen until ready for spectrophotometric analysis.

After thawing, nitrate concentrations of the samples were determined using a HACH TNT 835 Nitrate test kit where 1 mL of sample is added to the vial (containing?), followed by adding 0.2 mL of solution A (containing?), inverting the vials three times and allowing them to sit for fifteen minutes. Once the vials were ready, the outsides were cleaned and scanned in a HACH DR2800 spectrophotometer to find nitrate (NO₃-N) concentrations.

Total Nitrogen Content:

An attempt to find total nitrogen was made using the soil nitrogen Kjeldahl method. Before starting the procedure, the digestion tubes were rinsed with soap water, heated with sulfuric acid, and rinsed with deionized water to remove any absorbed contaminants that could affect the study. Once dried, 0.45 – 0.50 g of soil from each site were added to the tubes as well as an additional sample to be used as a replicate. A reference standard was made separately by adding 0.5 mL of Nitrogen Ammonia

Standard Solution to a digestion tube and subjecting it to the same procedure as the soil samples. 3.5 mL of concentrated sulfuric acid was added to each soil sample tube, the reference standard tube, and the three tubes being used as blanks. The samples were then digested by placing them in a Fisher Scientific Digester first at 160°C for twenty minutes, and then, at 380°C for 240 minutes. The tubes were then removed from the block and allowed to cool for at least fifteen minutes. Once cool, they were filled with deionized water to 50 mL (approximately 46.5 mL added).

A modified procedure for TKN was attempted but complications in reaction chemistry did not allow for spectrophotometric analysis. Thus, the steps necessary to complete this method could not be completed and therefore, nitrogen concentrations of the samples could not be determined.

Reactive Phosphorus Content:

A Mehlich-3 extraction was performed to find the total reactive phosphorus in the soil samples (NRCCST, 2003Ref.). First, the Mehlich-3 solution was prepared as follows: to prepare the solution stock, 69.4500 g of ammonium fluoride (NH_4F) was dissolved into a 500 mL volumetric flask and approximately 250 mL of water was added. The flask was then placed on a CORNING PC-420 D stirring plate and 36.5250 g of ethylenediaminetetraacetic acid disodium salt dihydrate (EDTA) was slowly added over thirty minutes using a plastic powder funnel. The solution was stirred until a clear solution was obtained. Next, DI water was added to bring the volume up to 500 mL.

To prepare the extracting solution, 20.000 g of ammonium nitrate (NH_4NO_3) was dissolved into approximately 500 mL of water in a 1000 mL volumetric flask, 4 mL of the Mehlich stock, 11.5 mL of acetic acid (CH_3COOH), and 0.82 mL of concentrated

nitric acid (HNO₃) was added, respectively. The solution was then diluted to 1000 mL and stored in a refrigerator until used.

To perform the extraction, approximately 2.5 g of soil was weighed out into plastic 50 mL centrifuge tubes, and approximately 25 mL of extracting solution was added. Each sample was shaken for five minutes on the shaker (shaker details noted above). Approximately 20 mL of sample was then filtered using Whatman GF/F 25 mm circle filters.

A HACH TNT Plus Phosphorus Total & Reactive kit was used to measure the biologically available form of phosphorus. 2.0 mL aliquat of the filtered sample was adding to a vial (containing?). 0.2 mL of (solution B) capped with a Dosi cap (containing?), inverted 3 times and set-aside for ten minutes. After sitting for ten minutes, the vials were inverted 3 more times, wiped clean using Kim wipes and PO₄³⁻ concentrations were read using a spectrophotometer (details noted above). Owing to the saturation of the kit, the results were said to be over range, so the samples were diluted 10 fold by adding 0.2 mL of sample aliquots to 1.8 mL of deionized water and re-analyzed.

RESULTS

Gravimetric Soil Moisture:

The experimentally determined soil moisture content for each cored sample is listed in Table 2 below. The soil moisture content ranged from a low value of 0.7218 g of water for sample F1, up to a high value of 3.630 g of water for sample G1. The average moisture content for the sample set was determined to be 1.80 g. Though the moisture content for sample G1 seems high as compared to the rest of the sample set, it was determined not to be an outlier using the Inter Quartile Range rule (IQR). The moisture

content was higher in S1 than S2 and G1 compared to G2 as expected due to the perceived westward flow of farm runoff but lower in F1 than F2 (Figure 4). The lowest soil moisture content was seen at site in F1 at 0.7218 g (Table 2).

Table 2. Soil moisture content (g of water) for sediment collected at each site found using a gravimetric soil moisture procedure.

Sample	Wet sediment weight (g)	Dry sediment weight (g)	Moisture (g of water)
S1	28.40	25.97	2.43
S2	18.96	17.14	1.81
F1	11.74	11.02	0.72
F2	17.11	15.81	1.29
G1	29.02	25.39	3.63
G2	11.42	10.46	0.96
Average	19.44	17.63	1.81

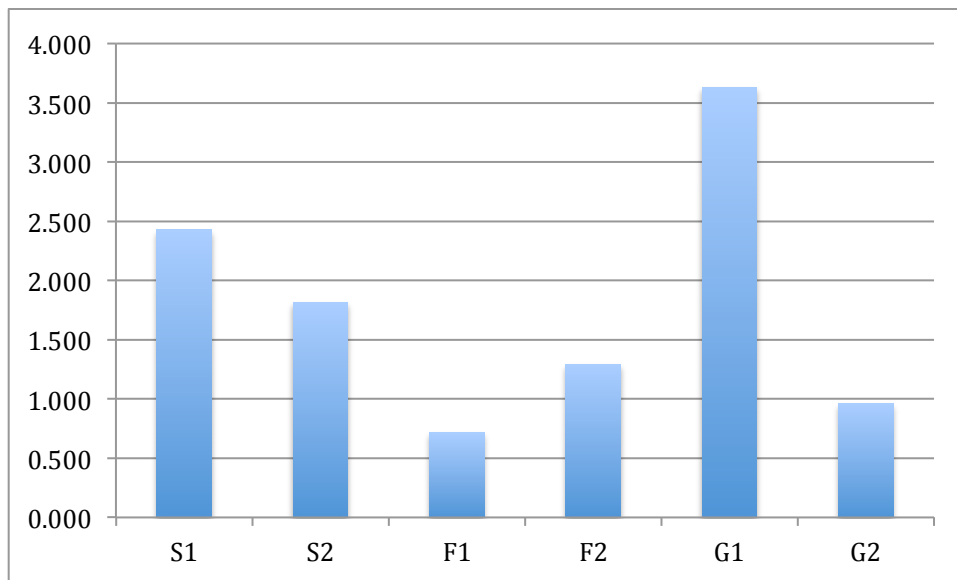


Figure 4. Soil moisture content (g of water) for sediment collected at each site found using a gravimetric soil moisture procedure.

Soil Inorganic Nitrogen:

Nitrogen concentrations ranged from 0.012 to 0.4657 mg/L (Table 3). F1 concentrations were higher than F2 and G1 higher than G2a and G2b as expected due to the direction of runoff but lower in F1 than F2 (Figure 5). Overall, the samples from the forested sites had the highest concentrations of inorganic nitrogen; the samples from the

switchgrass sites had the lowest with sums of 0.6546 mg/L and 0.292 mg/L respectively (Table 3).

Table 3. Nitrate concentrations (mg/L) for each soil sample using a KCL extraction and a HACH Nitrate kit to find the values. Average values were calculated using sites S1, S2, F1, F2, G1, and G2a.

Site	Soil added (g)	NO3-N concentration (mg/L)	NO3-N concentration (mg/L) adjusted for blank
Standard	0.00	11.70	11.30
Field Blank	0.00	0.40	0.00
S1	9.90	0.52	0.11
S2	9.30	0.58	0.18
F1	9.90	0.86	0.46
F2	11.20	0.59	0.19
G1	10.60	0.76	0.36
G2a	11.20	0.41	0.01
G2b	10.70	0.49	0.09

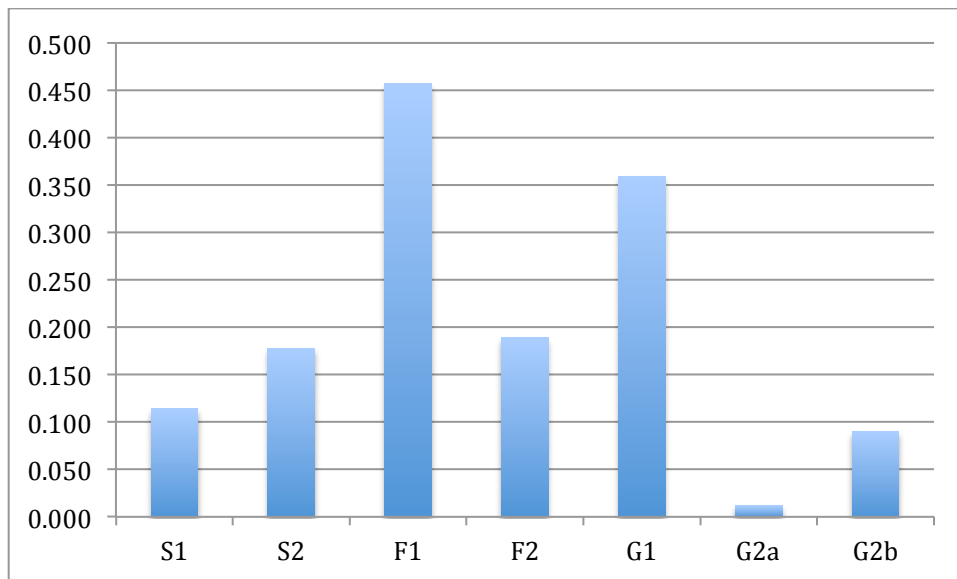


Figure 5. Nitrate concentrations (mg/L) for each soil sample using a KCL extraction and a HACH Nitrate kit to find the values.

Reactive Phosphorus:

Phosphate concentrations (H_3PO_4) ranged from 0.82 to 1.76 mg/L (Table 4). In each land use classification, the concentrations were higher in the first site compared to the second site, which was expected (Figure 6). Overall, the grassland site samples had

the highest concentrations at 3.050 or 3.120 mg/L while the forested site samples had the lowest concentration at 1.692 (Table 4).

Table 4. Active phosphate concentrations (mg/L) found for each soil sample using a Mehlich-3 extraction and a HACH phosphate kit to find the values.

Sample	Soil Added (g)	Solution added (g)	PO43- Concentration (mg/L) after dilution	PO43 (H ₃ PO ₄ ?)- Concentration (mg/L) before dilution
Sol. Blank	0.00	25.57	-0.04	n/a
Blank	0.00	0.00	n/a	n/a
Standard	0.00	0.00	0.89	8.86
S1	2.48	24.18	1.76	17.60
S2	2.48	24.34	1.19	11.90
F1	2.49	25.75	0.88	8.75
F2	2.56	23.34	0.82	8.17
G1	2.60	25.23	1.73	17.30
G2a	2.65	24.22	1.32	13.20
G2b	2.49	25.25	1.39	13.90

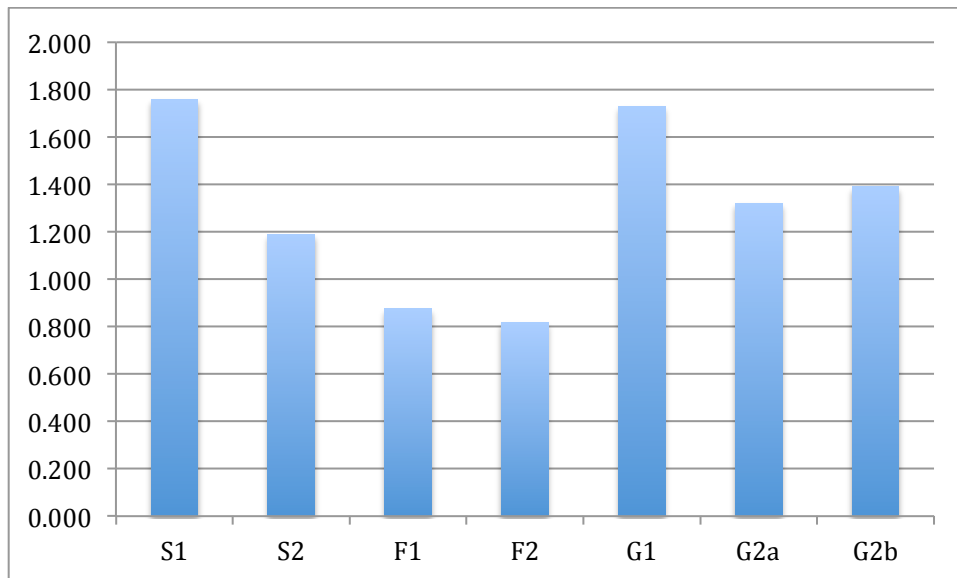


Figure 6. Active phosphorus concentrations (mg/L) found for each soil sample using a Mehlich-3 extraction and a HACH phosphate kit to find the values.

Overall Results:

The four critical parameters monitored in this short study (location/site, moisture, NO₃, and H₃PO₄ phosphorous content) are compared in Table 6 and graphically

represented in Figure 8 below. Samples S1, S2, F2, and G2a had below average nitrate concentrations (Table 6). The switchgrass sites were the only sites that had lower than average nitrate concentrations at among all sites investigated (Table 6). Samples F1 and F2 showed the lowest phosphate concentrations (Table 6). Figure 8 displays a comparison between nitrate and phosphate concentrations with very little correlation between sites and surrounding plant biology. Figure 9 displays a comparison between soil moisture and phosphate concentrations demonstrating a small correlation. The same was done with soil moisture and nitrate concentrations but no correlation was seen.

Table 6. Summary table of the four parameters examined in this study. Average values are included as a means for comparison between the sample sites.

Site	Moisture (g)	NO3-N concentration (mg/L) adjusted for blank	PO43- Concentration (mg/L) after dilution
S1	2.430	0.114	1.760
S2	1.814	0.178	1.190
F1	0.718	0.457	0.875
F2	1.295	0.189	0.817
G1	3.630	0.359	1.730
G2a	0.964	0.012	1.320
Avg.	1.809	0.218	1.282

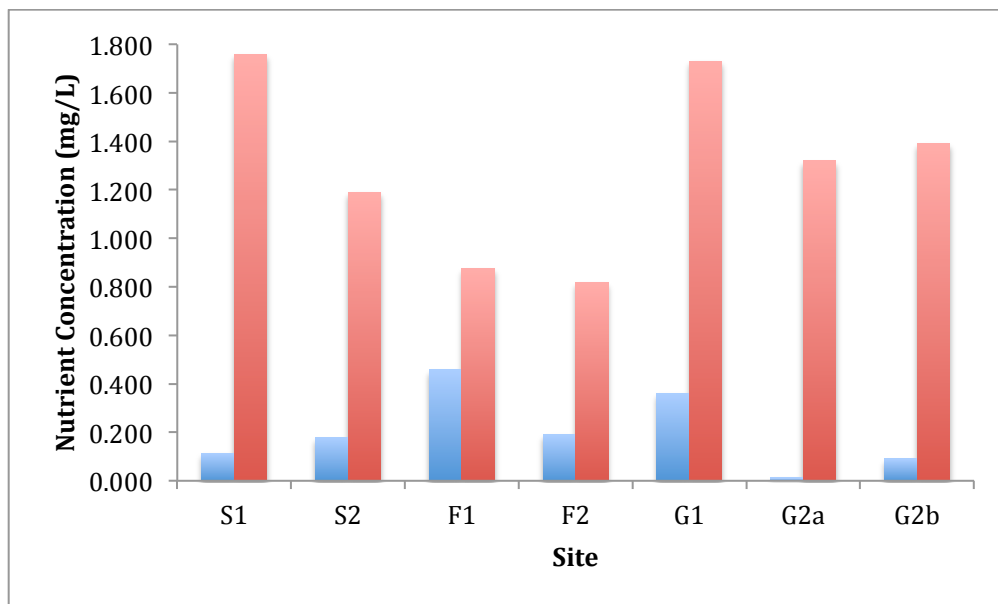


Figure 8. Comparison of NO³⁻ mg/L (blue) and PO³⁻ mg/L (red) concentrations for each sample site. Note: G2a and G2b are the same sample site but separate samples (G2b used as replicate).

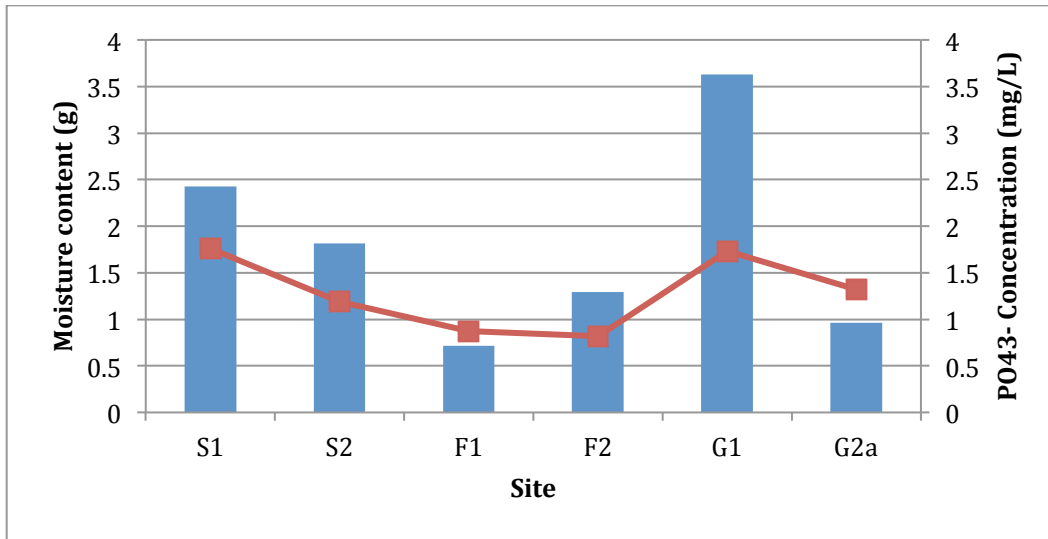


Figure 9. Comparison of soil moisture content (g) in blue and PO₄³⁻ (mg/L) concentrations in red for each sample site.

DISCUSSION

The results varied in support to the hypothesis depending upon the nutrient component under consideration. From this brief study, a conclusion can be drawn that switchgrass is indeed effective in removing nutrients from soil, showing moderate preference for nitrogen based nutrients over phosphorous based nutrients. The consistently lower nitrate concentrations observed in soil sampled around switchgrass plots supports the hypothesis explored in this study. Phosphate concentrations were lowest in forested sites and were highest in the grassland site showing that forested areas are more efficient in eliminating excess phosphorous from run-off since neither receives fertilizer applications..

Due to the expected westward flow of runoff the S1, F1, and G1 samples would be have higher nutrient concentrations than their respective site samples. However this was not the case in nitrate concentrations for the switchgrass sites. This does not disprove or support the hypothesis but may indicate a flaw in experimental design or switchgrass

may not be as effective in phosphorus removal. An additional explanation is that switchgrass roots can extend to eight feet below the surface; nutrient uptake is most likely occurring further into the soil than what was sampled in this study.

The uptake of nutrients from soil can be affected by a multitude of factors and unknowns. Ideal conditions and treatment are site specific to each switchgrass cultivar. Nutrient efficiency in switchgrass can be affected by root and shoot growth between species and within plots (Wang, Kelly, & Kovar, 2005). Age of plots is known to affect nutrient efficiency in that, plots that are not established cannot uptake nutrients as well, resulting in a higher concentration remaining in soil thus increasing the chances of the nitrogen and phosphorus leaching into water bodies through groundwater or runoff events (Sarkar, Miller, Frederick & Chamberlain, 2011). However, in this study age can be ruled out because the plots were well established.

Soil conditions also affect nutrient content. In some cases, drought conditions minimizing water availability for roots limits nutrient uptake efficiency (Sarkar, Miller, Frederick & Chamberlain, 2011). This too was most likely not an issue in this study due to the very wet growth season and apparent success of the plots. Soil condition issues are most often due to sediment size. Switchgrass' extensive root system allows for increased sediment retention, which gives the plants more opportunity to uptake nutrients bound in the soil. However, once material is infiltrated into groundwater, there is nothing left to be taken up (Magette, Brinsfield, Palmer & Wood, 2003). This could be the case in this study. The nitrate may have been able used by the plants while the phosphate remained in the sediment, or this could be because the switchgrass fields were the only sites that receive fertilizer annually, which adds phosphorus to the soil within the plots.

Additionally, phosphate concentrations often increase because fertilizer is applied to match nitrate requirements, which can lead to phosphorus being greater in excess than nitrogen.

There are other potentially contributing factors that were not part of the experimental design but also could have played a role in the results. Precipitation, which was not monitored previous to collection, directly affects runoff events. Temperature does not directly affect runoff, but it can affect growth, which affect nutrient use throughout the roots and shoots. Warmer temperatures are signals for flowering and cooler temperatures for dormancy (Parrish & Fike, 2005). It has been documented that switchgrass uses fertilizer most efficiently at elevated temperatures (Owens, Viands, Mayton, Fike, Farris, Heaton, Bransby & Hong, 2013). The plots in this study were only examined under low temperatures between growing seasons, but the effect of temperature fluctuations throughout the seasons may be an important factor to look into. Harvest time and frequency of harvest are other factors that can affect biomass accumulation and nutrient uptake efficiency but were beyond the scope of this study. Harvesting may not have played any significant role in this study since these plots were harvested once after senescence, which is said to maximize yield (Parrish & Fike, 2005). However, harvesting after senescence may increase nutrient concentrations in the soil because nutrients are translocated to root tissues after dieback (McLaughlin & Kszos, 2005).

CONCLUSIONS

Overall, the experimental data obtained from analysis of nutrient levels in field samples generally support the hypothesis that nutrients concentrations would be lower within switchgrass plots. Further study is warranted here, future projects of nutrient

analysis should encompass a larger sample set, collected and analyzed across multiple sites and at regular intervals rather than a single time point. The study would also be better by sampling deeper in the soil to better understand the role of switchgrass' extensive root system in nutrient uptake. In addition, levels of nutrients in ground water need to be monitored concurrently throughout the seasons. This would allow for a deeper understanding of the effectiveness of switchgrass in regulating nutrient levels and also provide an opportunity to study any detrimental effects on the environment.

The major setback in this study was time constraints. This study would have benefited if there were multiple sampling dates throughout the seasons, so that one could better understand the role of nutrients in the soil throughout the seasons rather than a snapshot from one date. This would also allow one to better understand the role of fertilizer in soil nutrient concentrations. Still, an overall take away is that this study did demonstrate that even with a fertilizer application switchgrass plots appear to be non-detrimental to soil nitrate concentrations but may be of no help in lowering phosphorus in the soil quality.

What needs to be further examined before concluding that switchgrass does not harm the environment overall is groundwater nutrient concentrations. This could not be looked at in this study due to logistical and monetary constraints.

Additionally, it is unclear as to whether increased switchgrass production could be beneficial to the Chesapeake region. This would be dependent on whether the plots would be on previously farmed land, whether the plots would buffer runoff, and the effect it would have on current wildlife. The results indicate that a combination of switchgrass

and forested areas would be more efficient in maintaining a healthy composition of nitrogen and phosphorous-based nutrients in the Chesapeake Bay region.

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WORKS CITED

- Blanco-Canqui, H., Gantzer, C. J., Anderson, S. H., Alberts, E. E., & Thompson, A. L. (n.d.). Grass barrier and vegetative filter strip effectiveness in reducing runoff, sediment, nitrogen, and phosphorus loss. *Soil Science Society of America Journal*, 1670-1678. doi: 10.2136/sssaj2004.1670
- Blanco-Canqui, H. & Gantzer, C. J., Anderson, S. H. Alberts, (2006). Performance of grass barriers and filter strips under interrill and concentrated flow. *Journal of Environmental Quality* , 35(6), 1969-1974.
- Castle, S. 2005. Soil Inorganic Nitrogen: KCl Extraction. Aridlands Ecology Lab Protocol, <http://www.colorado.edu/eeb/facultysites/barger/Linked%20PDFS>.
- Grismer, M. E., O'Geen, A. T., & Lewis, D. (2006). *Vegetative filter strips for nonpoint source pollution control in agriculture*. Informally published manuscript, Division of Agriculture and Natural Resources, University of California, Oakland, CA, .
- Jiading, Y., Worley, E., Wang, M., Lahner, B., Salt, D. E., Malay, S., & Michael, U. (2009). Natural variation for nutrient use and remobilization efficiencies in switchgrass. *Bioenergy Resources*, (2), 257-266. doi: 10.1007/s12155-009-9055-9
- Lee, K. H., Isenhardt, T. M., & Schultz, R. C. (2003). Sediment and nutrient removal in an established multi-species riparian buffer. *Journal of Soil and Water Conservation*, 58(1), 1-10.
- Lim, T. T., Edwards, D. R., Workman, S. R., Larson, B. T., & Dunn, L. (1998). Vegetated filter strip removal of cattle manure constituents in runoff. *American Society of Agricultural Engineers*, 41(5), 1375-1381.

- Lin, CH., Lerch, RN., Garrett, HE., Jordan, D., & George, MF. (2007). Ability of forage grasses exposed to atrazine and isoxaflutole to reduce nutrient levels in soils and shallow groundwater. *Communications in Soil Science and Plant Analysis*, 38, 1119-1136, doi: 10.1080/0010362071327976.
- Magette, W. L., Brinsfield, R. B., Palmer, R. E., & Wood, J. D. (2003). Nutrient and sediment removal by vegetated filter strips. *American Society of Agricultural and Biological Engineers*, 32(2), 663-667.
- Maeght, J., Rewald, B., & Pierret, A. (2013). How to study deep roots—and why it matters. *Frontiers in Plant Science*, 4(299), doi: 10.3389/fpls.2013.00299
- Matuszeski, B. (2007). *Biofuels and the bay getting it right to benefit farms, forests and the chesapeake*. Annapolis, MD: Chesapeake Bay Commission.
- McLaughlin, S. B., & Kszos, L. A. (2005). Development of switchgrass (*panicum virgatum*) as a bioenergy feedstock in the united states. *Biomass and Bioenergy*, 28, 515-535.
- Nelson, R. G., Ascough II, J. C., & Langemeier, M. R. (2006). Environmental and economic analysis of switchgrass production for water quality improvement in northeast kansas. *Journal of Environmental Management*, 79, 336-347. Retrieved from <http://naldc.nal.usda.gov/download/16052/PDF>
- Ouyang, Y. (2012). Estimation of shallow groundwater discharge and nutrient load into a river. *Ecological Engineering*, 38, 101-104. doi: 10.1016/j.ecoleng.2011.10.014
- Owens, V. N., Viands, D. R., Mayton, H. S., Fike, J. H., Farris, R., Heaton, E., Bransby, D. I., & Hong, C. O. (2013). Nitrogen use in switchgrass grown for bioenergy across the usa. *Biomass and Bioenergy*, 58, 286-293.
- Parrish, D. J., & Fike, J. H. (2005). The biology and agronomy of switchgrass for biofuels. *Critical Reviews in Plant Sciences*, 24(0735-2689), 423-459.
- Pasten-Zapata, E., Ledesma-Ruiz, R., Harter, T., Ramirez, A. I., & Mahlknecht, J. (2014). Assessment of sources and fate of nitrate in shallow groundwater of an agricultural area by using a multi-tracer approach. *Science of the Total Environment*, 470-471, 855-864.
- NRCCST Northeast Region Coordinating Committee on Soil Testing, Recommended Soil Testing Procedures for the Northeastern United States, 2003. Preparations of Mehlich-3 Solutions. 2(493).
- Sanderson, M. A., Jones, R. M., McFarland, M. J., Stroup, J., Reed, R. L., & Muir, J. P. (2000). Nutrient movement and removal in a switchgrass biomass–filter strip system treated with dairy manure. *Journal of Environmental Quality*, 30, 210-216.

- Sarkar, S., Miller, S. A., Frederick, J. R., & Chamberlain, J. F. (2011). Modeling nitrogen loss from switchgrass agricultural systems. *Biomass and Bioenergy*, 35, 4381-4389. Retrieved from [http://nadp.isws.illinois.edu/dl/dgay/North Carolina Info/Sarkar et al 2011 N Loss from Switchgrass.pdf](http://nadp.isws.illinois.edu/dl/dgay/North%20Carolina%20Info/Sarkar%20et%20al%202011%20N%20Loss%20from%20Switchgrass.pdf)
- U.S. Environmental Protection Agency, (2004). *A report to the citizens of the bay region* (EPA 903-R-04-009). Annapolis, Maryland: Chesapeake Bay Program.
- Wang, Z. Y., Kelly, J. M., & Kovar, J. L. (2005). Depletion of macro-nutrients from rhizosphere soil solution by juvenile corn, cottonwood, and switchgrass plants. *Plant and Soil*, 270, 213-221. doi: 10.1007/s11104-004-1538-z
- Weismiller, R. A., Steinhilber, P. M., & Salak, J. L. (2012). *Managing agricultural nutrients in maryland's chesapeake bay basin*. Informally published manuscript, Department of Natural Resource Sciences and Landscape Architecture, University of Maryland at College Park, College Park, MD, .
- Wennersten, J. R. (2001). *The chesapeake an environmental biography*. Baltimore: Maryland Historical Society.