ABSTRACT

Title of Document: GRAZING AS A MANAGEMENT TOOL FOR

CONTROLLING PHRAGMITES AUSTRALIS

AND RESTORING NATIVE PLANT BIODIVERSITY IN WETLANDS

Jennifer Brundage, Master of Science, 2010

Directed By: Dr. Andrew Baldwin, Department of

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This study examined the feasibility of grazing as a sustainable and low-impact means of controlling *Phragmites*. In addition, this study examined whether grazing of *Phragmites* by large herbivores (goats) in a wetland affects soil and soil water nutrient pools, and thus how grazing might affect nutrient export from the wetland. An isolated, created wetland at USDA's Beltsville Agricultural Research Center (BARC) in Beltsville, MD was divided into four grazed and four ungrazed plots. Two rounds of grazing significantly reduced *Phragmites* height, stem count, and biomass and increased some measures of plant diversity. Grazing significantly elevated soil water total nitrogen and total phosphorus levels and reduced soil watersoluble phosphorus levels. The nutrient pool analysis indicates that grazing reduced the fertility of the system. The results of this study will inform the development of an alternative, sustainable approach to controlling *Phragmites* that integrates the local agricultural community while benefitting the local ecology.

GRAZING AS A MANAGEMENT TOOL FOR CONTROLLING PHRAGMITES AUSTRALIS AND RESTORING NATIVE PLANT BIODIVERSITY IN WETLANDS

By

Jennifer Brundage

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Advisory Committee: Dr. Andrew Baldwin Dr. Joshua McGrath Dr. David Tilley © Copyright by Jennifer Brundage 2010

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Chapter 1: Introduction

Phragmites australis [(Cav.) Trin. Ex. Steud] is an invasive, cosmopolitan reed forming monocultures in U.S. wetlands that rapidly replace native vegetation (Chambers et al., 1999; Meyerson et al., 2000; Warren et al., 2001). Phragmites began invading Mid-Atlantic wetlands of the U.S. approximately 100 to 150 years ago (Rice et al., 2000). It is especially invasive in inland freshwater emergent wetlands, and in disturbed, urbanized, and constructed wetlands (Havens et al., 2003). It is also invasive in coastal, brackish, and freshwater tidal wetlands (Meyerson et al., 2000). It forms monotypic stands that exclude native vegetation (Meyerson et al., 2000; Warren et al., 2001) and expands linearly at rates of 1-3\% annually (Warren et al., 2001) which often equates to rates of several meters per year (Farnsworth and Meyerson, 1999). While *Phragmites* is generally treated as an invasive species in the U.S., it appears from peat cores and fossil records to have been present for thousands of years in low abundance in mixed stands (Kraft, 1971; Orson, 1999). The invasive *Phragmites* appears to be of a different genotype, more akin to the European strain than the native type (Saltonstall, 2002, 2003 a,b). It can grow to 6 m tall (typically 2-4 m) (Meyerson et al., 2000) and reproduces primarily via rhizomes, though it also produces thousands of seeds of low viability annually (Harris and Marshall, 1960; Haslam, 1972; Greenwood and MacFarlane, 2006).

Its efficient growth under high nutrient conditions allows *Phragmites* to outcompete native vegetation for light, growing space, and other resources (Onimaru and Yabe, 1996; Burdick and Konisky, 2003). Levine et al. (1998) hypothesized that light is limiting where nutrients are abundant, therefore *Phragmites* may succeed because of its superior competitive

ability for light. In addition to its tall growth during the growing season, its tall culms remain standing throughout the winter and it produces abundant litter that blocks light from reaching the ground (Meyerson et al., 2000; Burdick and Konisky, 2003). In the U.S., Meyerson et al. (2000) reviewed the literature on *Phragmites* ecology. They found that *Phragmites*-dominated sites had lower species richness than non-invaded marshes, and that eradication of *Phragmites* led to increased species richness *in all reviewed cases*.

Because of its perceived negative effects on diversity and the local ecology (Gusewell and Klotzli (1998), *Phragmites* is often treated by U.S. land managers with herbicide, burning, mowing, or, rarely, flooding with saltwater. Herbicide treatment is often the most cost-effective of these methods to reduce *Phragmites* biomass and restore plant biodiversity (Farnsworth and Meyerson, 1999; Warren et al., 2001). However herbicide can have negative impacts on non-target plants and other organisms. Herbicide use to control *Phragmites* has also been shown to cause a net increase in porewater ammonium concentrations for up to two years following treatment, which suggests that herbicide use increases the risk of nitrogen export from the wetland (Findlay et al. 2003).

Grazing is a potential alternative management tool for controlling *Phragmites* that has been little studied in the U.S. Studies in Europe have shown that using livestock to graze *Phragmites* communities successfully increases plant species richness (Ausden et al 2005; Vulink et al., 2000) and decreases *Phragmites* density (Bassett, 1980; van Deursen and Drost, 1990; Ausden et al., 2005). Vulink et al. (2000) wrote that *Phragmites* is particularly sensitive to grazing because it has apical meristems well within reach of grazing herbivores. Bassett (1980) and Burnside (2007) demonstrated that excluding grazers from previously-grazed plots containing *Phragmites* caused a dramatic increase in *Phragmites* abundance.

Grazing has been widely used in Europe as a management tool for restoring biodiversity in wetlands over the last few decades (Van Deursen and Drost, 1990; Vulink et al., 2000). Grazing is believed to increase plant diversity by increasing habitat heterogeneity (Rook et al., 2004) and by reducing shading by tall, competitive species (Onimaru and Yabe, 1996). Grazing is thought to re-set the successional clock in nutrient-rich systems to prevent succession to a "climax community" of tall, monotypic stands of reed and has been shown to restore habitat for birds (Milsom et al., 2000) and invertebrates (Davis and Bidwell, 2001).

Grazed marshes can also provide free-range grazing habitat for the local agricultural community. Animal products from free-range ruminants grazed in wetlands in Europe are often considered premium items for which consumers are willing to pay a high price (Gordon et al., 1990, Rook et al., 2004). Charging farmers for use of the wetland as pasture can also raise funds for management of nature reserves (Gordon et al., 1990).

In contrast, grazing is only rarely used as a management tool in the eastern U.S., and it is almost never used in wetlands. Grazing is widely perceived in the U.S. as detrimental to biodiversity in both uplands and wetlands, since studies have shown that high-intensity grazing and grazing in riparian areas can reduce biodiversity (Popotnick and Giuliano, 2000; Jansen and Robertson, 2001).

Tesauro and Ehrenfeld (2007) was the only study to my knowledge to examine the use of grazing as a management tool to control *Phragmites* in the U.S. They demonstrated that grazing can effectively reduce the abundance of *Phragmites* and several other invasive species and increase plant biodiversity in high-nutrient, abandoned pastures in New Jersey and New York. Marty (2005) similarly demonstrated that grazing in vernal wetlands of

California increased diversity of both plants and aquatic invertebrates and reduced the abundance of exotic plants. *Phragmites* was not present in Marty (2005)'s study, however.

A key concern of wetland managers in the Chesapeake Bay region is minimizing nutrient inputs to the Bay. While a handful of studies have shown that grazing increases plant biodiversity in wetlands, none have examined the effect of grazing *Phragmites* on nutrient cycling in wetlands. The impacts of livestock on flooded soils are not well understood (Bohlen and Gathumbi, 2007). It would be imprudent to recommend grazing as an environmentally safe management tool for controlling *Phragmites* in wetlands without an understanding of how this might affect nutrient levels and potential nutrient export to surface or groundwater.

Herbivores can have complex, non-linear effects on nutrient cycling (Pastor and Thompson Hobbs, 2006). They can affect ecosystem nutrient cycles through foraging, trampling, urination, and deposition of feces (Thompson Hobbs, 2006). Foraging directly removes nutrients. Herbivore digestion degrades complex carbohydrates such as lignin and cellulose into simple sugars and nitrogen compounds. Digestion breaks indigestible plant material into smaller particles with a lower C:N ratio than plant litter, which increases both the surface area available for microbial attack and the nutritional quality of the material for microbes (Pastor et al., 2006). Together, urination and deposition of feces help to accelerate decomposition and thus nutrient cycling.

Grazing can increase, decrease, or have mixed effects on nutrient cycling rates in an ecosystem (Bardgett and Wardle, 2003; Pastor et al., 2006). Grazers can affect nutrient pools and cycling rates through multiple pathways, including selection of forage species, uneven deposition of excreta, and effects on the quantity and quality of plant litter (Dubeux et al.,

2007). Plant responses to grazing also affect nutrient pools, such as for example by increasing energy allocation to either aboveground or belowground parts, or by changing their growth form. Grazing has often been studied in grasslands (Dubeux et al., 2007), but anaerobic conditions in wetlands make their nutrient cycling and pools differ from those of upland soils in a number of ways (Mitsch and Gosselink, 2007). There has been no study to my knowledge examining how grazing affects nutrient pools either in wetlands or in *Phragmites*-dominated systems.

Phragmites has been shown to store significant quantities of nutrients in its biomass, and much of this biomass is refractory and slow to decompose (Meyerson et al. 2000). 80-90% of the nitrogen and 50-75% of the phosphorus consumed by grazers is released as excreta (Davidson, 1964; Dahlin et al., 2005; Morse, 1992: Meschy, 2002). Grazing could therefore make the large store of nutrients trapped in *Phragmites* biomass available in the more labile forms found in excreta. Grazers also remove some of the nutrients they consume as biomass, theoretically reducing the fertility of their pasture.

Trampling by herbivores can also significantly accelerate litter breakdown and nutrient release. Zacheis et al., (2002) found in Alaskan salt marshes grazed by snow geese that trampling of the previous year's litter accelerated incorporation of litter into soils, which in turn accelerated nitrogen mineralization. Fecal additions had little effect relative to those of trampling.

Most of the studies done on nutrient releases from grazing systems are conducted in upland grassland ecosystems, and there is little information on nutrient releases from grazing systems set up specifically for biodiversity restoration. Milchunas and Lauenroth (1993) reviewed the literature on grazing in upland ecosystems and found no consistent effects on

soil nutrient levels. Some studies reported increases, and some reported decreases. One study on grazing to enhance plant diversity, Bakker and Heerdt (2005) found that extensive grazing (grazing at a low stocking density) decreased the occurrence of plants indicative of high soil nitrogen levels, suggesting that grazing lowered soil nitrogen levels.

Furthermore, much of the literature on nutrient releases from grazing systems is restricted to nitrate leaching. Two studies, Owens and Bonata 2004 and Anger et al., 2002, both found that extensive grazing in grassland could reduce nitrate concentrations in shallow groundwater. Other studies have found that grazing increases nitrate leaching from upland pasture (Afzael and Adams, 1992; Hack-ten-Broeke and van der Putten 1997; Decau et al., 2004).

There is also a limited amount of information available on nutrient releases from grazing operations in wetlands. One study on grazing in wet pasture, Sigua et al. (2006), found that wetlands that had been converted to pasture 63 years ago had lower total soil nitrogen and phosphorus and equivalent soil water-extractable phosphorus levels to reference wetlands. In their study the effects of grazing were confounded with the effects of wetland drainage; nevertheless it can be concluded that 63 years of grazing did not increase nutrient levels in these pastures compared to their previous ungrazed state. Sigua et al. (2010) similarly found that grazing in an upland pasture contributed negligible amounts of phosphorus to groundwater.

Overall there is a paucity of data on the effects of grazing on nutrient pools in *Phragmites*-dominated in wetlands, and it is unclear how grazing might affect wetland soil and water quality.

Objectives

The overall objectives of this study were to determine whether grazing can restore a diverse native wetland plant community, and whether this can be done without negatively impacting soil or water quality. Specifically, my objectives were to:

- 1) Determine whether goat grazing can effectively reduce *Phragmites* stem density and height, and whether it can increase native plant diversity, in terms of both species richness and Shannon-Weiner diversity indices
- 2) Determine whether grazing elevates ambient soil water levels of nitrogen and phosphorus in a *Phragmites*-dominated wetland,
- 3) Determine whether grazing elevates soil nitrogen and phosphorus levels, and
- 4) Evaluate the effects of grazing on nutrient pools of different ecosystem compartments.

The objectives listed above were addressed using a field grazing experiment as described in the next chapter. Chapter 3 describes the results of the experiment. Chapter 4 analyzes the results, compares them to the current scientific literature, and provides recommendations and limitations for the use of grazing to control *P. australis* in wetlands. Finally, chapter 5 sets out the main conclusions of the study and suggests directions for future research on this topic.

Chapter 2: Methods

This study examined the effects of goat grazing on plant diversity, soil water nutrients, and soil nutrients at a mitigation wetland at the Beltsville Agricultural Research Center in Beltsville, MD. Three rounds of grazing were carried out between September 2008 and September 2009. Four grazed and four ungrazed plots were monitored for *Phragmites* stem height, count, and biomass, plant diversity, soil water nitrogen and phosphorus levels, and soil nitrogen and phosphorus levels. The data was analyzed using Analysis of Variance (ANOVA) and repeated-measures ANOVA.

Study Site Description

This research was conducted at the Beltsville Agricultural Research Center (BARC) in Beltsville, MD. A suitable *Phragmites* patch was identified in a wetland that was constructed approximately 15 years ago as mitigation for wetland destruction nearby (39.01.18.33 N, 76.52.36.27 W) (Figure 1). The wetland is approximately 80 m long by 40 m wide (Figure 2). The wetland is underlain with 7.5-15 cm of bentonite clay (depth varies across the site), which isolates its groundwater hydrology from that of the surrounding area. The clay begins at an average depth of approximately 30 cm below the soil surface, but this varies by several inches across the site (personal observation). The wetland has no stream channel inflows or outflows. The hydrology of this wetland therefore does not reflect "natural" wetland hydrologic conditions, however it reflects conditions common to some constructed mitigation wetlands.

During site visits in early 2008 the wetland was observed to contain 0.3-0.6 m of standing water. The wetland dried out as early as May and stayed completely dry until

October, when it began ponding in some areas following precipitation events. In 2009 a few inches of standing water were visible through July, then the wetland stayed dry through the rest of the study.

Before the wetland was restored a soil survey was completed, and the soil type in the existing field on the site was mapped as a mixture of Elkton Fine Sandy Loam (ElB), Galestown Loamy Sand (GdB), and Sunnyside Fine Sandy Loam (StC2) (Wallace, Roberts & Todd, 1991). During the course of my study no attempt was made to characterize the profile of this soil because it was assumed to be unnatural and highly disturbed. Eight samples from the top 30 cm of soil were textured by the University of Maryland's Pedology Laboratory in August 2009. The soil texture ranged from sandy loam to loamy sand with a clay content ranging from 9-12%.

The wetland was originally planted with *Cephalanthus occidentalis* (L.) (buttonbush), *Taxodium distichum* (L.) Rich. (bald cypress), and other trees, herbs, and shrubs (Wallace Roberts & Todd, 1991). I undertook a vegetation survey of the wetland in late April of 2008. The wetland surrounding the *Phragmites* patch contained a diverse mix of wetland plants, including *Juncus effusus* (L.) (soft rush), *Echinochloa* sp. (P.) Beauv., (barnyard grass), *Panicum virgatum* (L.) (switch grass), and *Typha latifolia* (L.) (cattail). See **Appendix 1: Plant Survey Report March, 2008**for the survey report and species lists. The majority of the interior of the wetland was dominated by a dense monoculture of *Phragmites*, with scattered *C. occidentalis* (buttonbush) and *T. distichum* (bald cypress) emerging above the *Phragmites* canopy. (All plant nomenclature is according to the USDA Plants database, http://plants.usda.gov, accessed 02/09/10.)

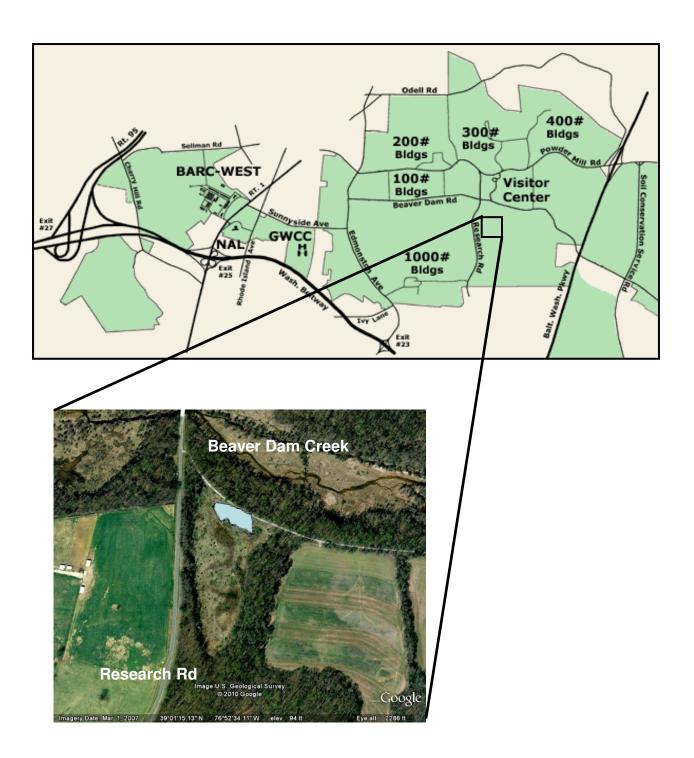


Figure 1: Research site location
The study patch of *Phragmites* is highlighted in blue in the enlarged panel; the aerial photo is a google earth image, map is from BARC website.

Study Design

In August and September of 2008 eight 340m² plots were delimited within the wetland. Each pair of adjacent plots was treated as a block to factor out potential differences in soils, hydrology, and other environmental variables across the site. Within each block, treatment (grazed or ungrazed) was assigned randomly. Each plot was 8.5 m wide by 40 m long and included 21 m² of upland area in both the front and the back to allow the grazing animals a refuge from wetland conditions. Plots were spaced 1 m apart (Figure 2).

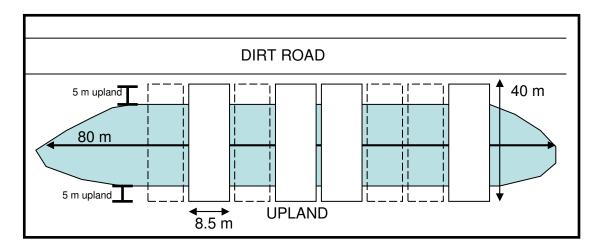


Figure 2: Layout of goat grazing plots
Dotted lines represent ungrazed plots; solid lines represent grazed plots. The blue area represents the *Phragmites* monoculture.

Grazed plots were enclosed by a 1.3-m high combination panel fence erected in September 2008. A combination panel fence contains smaller openings at the bottom and openings that gradually get larger towards the top. The fence was surrounded by a single strand of electric wire as a deterrent to unwanted human and animal visitors. In order to access the interior of the wetland for fence installation, in July 2008 and again in September a 2-m wide tractor mower was run through the wetland to demarcate each plot. Fence construction was completed by personnel from USDA BARC's Resource Support Services (RSS) team, with support from UMD personnel. In the front upland section of each grazed

plot a 4x8 ft (0.8 x 1.84 m) plank of pressure-treated plywood was laid down, on top of which was placed a 1.52 x 2.29 m hutch (Big Foot Calf Nursery, PolyDome) as shelter for the animals.

Two goats were placed in each grazed plot. The goats were mixed-breed domestic caprine species. This number of goats was used because, on the one hand, goats are social animals and it would be detrimental to their health and well-being to separate them into one goat per plot. On the other hand, based on our project veterinarian's advice more than two goats per plot would represent unsustainably high grazing pressure for my relatively small plots (Dr. Bill Hare, personal communication). This equates to a stocking density of 58.82 goats/hectare or 5.88 livestock units/hectare (Food and Agriculture Organization, 2005—one livestock unit is equivalent to one adult dairy cow).

The animals were obtained from the resident BARC herd of 25 goats. Goats were used because of their renowned readiness to eat and ability to digest tough, lignified plant matter (MacKenzie, 1993), because they require less land than cattle, and because they have been shown to effectively reduce *Phragmites* biomass (Tesauro, 2001). Ilius et al. (1999) found that goats choose their forage based on whatever they can ingest the most of the fastest rather than on forage quality. The fact that *Phragmites* has high lignin content relative to other herbaceous wetland plants and grows in monocultures in high abundance suggested that goats would be likely to consume *Phragmites*.

Three rounds of grazing were carried out. The first grazing treatment began on September 17, 2008 and ran until October 9, 2008, lasting 23 days. After this round of grazing the *Phragmites* did not re-sprout (presumably because it was beginning to senesce for the year), so no further grazing was carried out in 2008. The second grazing treatment

ran for a total of 33 days from May 21 to June 23, 2009. The third round of grazing ran for 14 days from August 25 to September 8, 2009.

During all grazing treatments the animals were removed when *Phragmites* was nearly completely grazed down in one plot, i.e. when at least one pair of animals had run out of food. The same individual goats were used in both 2008 and 2009, however individuals were randomly re-assigned to plots for each round of grazing. Both the second and third rounds of grazing began once the *Phragmites* had grown to about 1.5 meters in height. This ensured that there would be a source of food for the animals for several weeks.

A BARC animal care and use permit was approved for the preliminary work carried out in 2008 and was re-approved for 2009. The campus Institutional Animal Care and Use Committee (IACUC) informed me in writing that no animal care and use permit was required for this research since no UMD personnel were handling the animals. All animal care and handling was done by BARC staff.

Sampling Methods

Vegetation Sampling

Research Question 1: Does goat grazing reduce *Phragmites* cover? Does goat grazing increase plant species richness and/or Shannon-Weiner diversity?

Hypothesis: Goat grazing will reduce *Phragmites* abundance and height and will increase plant species richness and Shannon-Weiner diversity.

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Nested vegetation quadrats were established in each plot in a modified form of the method proposed by Peet et al. (1998). In each plot a permanent 100-m² (5x20 m) quadrat was established, and within this large quadrat four permanent 1-m² quadrats were established. The large quadrat was delimited by 10-ft (3.28 m) high PVC poles with string running lengthwise. The smaller quadrats were located systematically at 4, 8, 12, and 16 meters lengthwise and 2.5-m width-wise within the larger quadrat (Figure 3). The number of *Phragmites* stems was counted and height of the five tallest stems was measured in each 1-m² quadrat.

Baseline vegetation measurements were made in July 2008. The impact of the first round of grazing on vegetation was measured in October 2008. Vegetation was again sampled in May 2009, prior to the second round of grazing, and was assessed again in July, August, and September 2009. In August only diversity in the 100-m² plots was assessed and the 1-m² plots were not surveyed.

Aboveground Biomass

On August 12, 2009 live stems of *Phragmites* were harvested from two 1-m² quadrats in each plot. The quadrats were systematically located adjacent to the first and third fixed 1-m² vegetation sampling quadrats in each plot. After harvest the biomass was dried to a constant mass at 105°C and weighed to the nearest 0.01g.

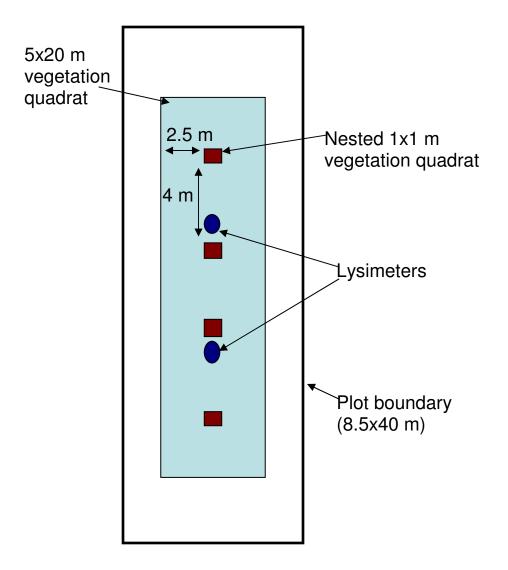


Figure 3: Vegetation sampling plot and lysimeter locations (not to scale)

Groundwater Sampling

Research Question 2: Does goat grazing of *Phragmites* in wetlands affect shallow groundwater nutrient levels?

Hypothesis: There will be no difference in soil water nutrient levels between grazed and ungrazed plots.

Soil water was sampled at a depth of 30 cm below the soil surface with porous cup lysimeters, Model 1920 from SoilMoisture Equipment Corporation (Santa Barbara, CA). Where bentonite clay was shallower than 30 cm, soil water was sampled at the deepest possible point above the clay layer. Two lysimeters were systematically located in each plot at 6.6 m and 13.3 m lengthwise across each 20 m long quadrat (Figure 3). The lysimeters were installed in September of 2008. Each lysimeter was surrounded by a 15-cm diameter PVC pipe with a screw top to protect it from damage by the goats. The pipes were driven about 15 cm into the ground, and a deeper hole was dug into the soil in the center of each pipe for the lysimeter. The hole was then backfilled with a slurry of the soil that had been extracted and tap water. In the grazed plots the pipes were stabilized with stakes.

Vacuum pressure was applied to each lysimeter 1-5 days prior to sampling. Prior to setting vacuum, pressure was applied to each lysimeter to force out previously accumulated soil water. Samples collected were immediately placed in a cooler to minimize microbiologically-mediated transformations of nitrogen and phosphorus. Samples were immediately transported to the University of Maryland Soil Fertility laboratory, where they were filtered to 0.45 microns and analyzed within 24 hours for pH and ammonium, nitrate, and ortho-phosphate using a QuikChem 8500 automated ion analyzer (2006 Latchat Instruments, Loveland, CO). Total phosphorus and total nitrogen were also measured on unfiltered samples following persulfate digestion. Approved Environmental Protection Agency (EPA) procedures for the analysis of Water and Wastewater were used for each analysis (Appendix 2: Methods of Soil Water Analysis). Briefly, ammonium was analyzed via the indophenol method, nitrate and total nitrogen via cadmium reduction, and ortho-phosphate and total phosphorus via the ascorbic acid method.

Soil water pH was determined with room-temperature solutions by immersing pH probe into a sample and waiting for the reading to stabilize. pH was read to the nearest 0.01 pH unit.

In 2008, soil water was sampled the day before treatments began, three weeks into the grazing treatments, and two weeks after grazing ended. In July 2009 soil water was sampled three weeks after grazing ceased, and in September 2009 soil water was sampled one week after grazing ceased.

Soil Sampling

Research Question 3: Does goat grazing of *Phragmites* in wetlands impact soil nutrients?

Hypothesis: There will be no significant difference in soil nutrients between grazed and ungrazed plots.

Soil was sampled at depths of 0-15 and 15-30 cm in each plot using an Oakfield soil sampler. Separating measurements into two depths allowed evaluation of downward movement of nutrients through the soil. 15 subsamples were haphazardly collected from each plot and combined into one composite sample per plot for each depth as per the methods described in Pierzynski (2000).

Baseline soil samples were collected in September 2008 within a week after the grazing treatments began. In 2009 soil samples were collected three weeks after the second round of grazing ceased. Soil samples were not collected after the third round of grazing due

to cost and time constraints. Although goats were present during the baseline sampling, I am assuming that the timeframe was too short for the goats to have affected soil nutrient pools, so that these samples can be considered to reflect pre-treatment nutrient levels.

Soil samples were air-dried, ground, and analyzed for pH and water-extractable phosphorus as well as Mehlich 3 phosphorus, iron, aluminum, and calcium by the University of Maryland Soil Fertility laboratory. Total nitrogen and percent carbon were also measured.

For determination of soil pH, 10 g of air-dried soil was sieved to 2 mm and mixed with 10 mL of deionized water. The mixture rested for 15 minutes, was stirred, and rested again for 15 minutes. The pH probe was then gently swirled in the soil slurry until a stable pH reading was obtained. Soil pH was read to the nearest 0.01 pH unit.

Measuring water-extractable phosphorus theoretically simulates the amount of phosphorus dissolved during rainfall or re-wetting of the soil after the dry season. It has also been shown to be well correlated to concentrations of dissolved reactive phosphorus in runoff (Pote et al., 1996; Maguire and Sims, 2002). For determination of water-soluble phosphorus 2 g of soil was air-dried, weighed, sieved to 2mm, and centrifuged. Samples were then shaken for 1 hr and centrifuged again. The supernatant was immediately filtered. Orthophosphate concentration from the filtered supernatant was measured within 24 hours using a QuikChem 8500 automated ion analyzer (2006 Latchat Instruments, Loveland, CO).

Mehlich 3 phosphorus is a widely used method for quantifying plant-available phosphorus. Thus, measuring Mehlich 3 phosphorus could allow easier comparison of our measured phosphorus levels to those measured in other studies. In most non-basic soils in Maryland soil phosphorus is primarily found bound to iron and aluminum. Dividing Mehlich

3 phosphorus by Mehlich 3 aluminum + iron can be used to calculate a soil saturation ratio, an estimate of the amount of a soil's phosphorus binding sites that are saturated with phosphorus. This ratio indicates the amount of phosphorus that is present in the soil relative to the capacity of the soil to bind phosphorus. Mehlich 3 saturation ratio is also correlated to the likelihood of phosphorus leaching from a soil (Maguire and Sims, 2002).

Soil Mehlich 3 phosphorus, iron, and aluminum were determined by adding 25 mL of Mehlich 3 extractant to 2.5 g of air-dried soil sieved to 2 mm. Samples were then shaken for 5 minutes, filtered, and analyzed using a QuikChem 8500 automated ion analyzer (2006 Latchat Instruments, Loveland, CO)

Total nitrogen and carbon were measured on soil samples that were ground and sieved to 2 mm. A subsample was taken from each sample, ground by mortar and pestle to pass a 1 mm sieve, then dried for 1 hour at 105°C. Nitrogen and carbon were subsequently determined using a Carbon/Hydrogen/Nitrogen Determinator (CHN-2000 Elemental Analyzer, Leco Corporation, St. Joseph, MI). The CHN-2000 combusted the soil, quantified percent soil nitrogen via thermal conductivity, and quantified percent soil carbon by infrared detection.

Soil Bulk Density

Soil bulk density was measured in order to estimate the size of the soil nutrient pool, as well as to determine whether grazing might impact soil bulk density. Samples were collected on March 9, 2010. At the time of sampling there were several centimeters of standing water in the wetland. Litter was brushed away from the soil surface, then a 173.5 cm³ cylindrical aluminum corer was gently twisted into the soil until full. Three subsamples were collected per plot. The bulk density of the three sub-samples was averaged to obtain

average bulk density per plot. Samples were weighed wet, dried at 105°C to a constant weight, then re-weighed to the nearest gram.

Soil bulk density was calculated as

Bulk density $(g/cm^3) = Mass\ of\ dry\ soil\ (g) / volume\ of\ sample\ (cm^3)$

Phragmites tissue nutrient analysis

A random subsample of the *Phragmites* biomass harvested from each plot (see **Aboveground Biomass** above) was ground to <0.02 mesh size and analyzed for tissue nitrogen and phosphorus concentrations by the Pennsylvania State Agricultural Analytical Laboratory. The samples were digested with a hot block digestion (EPA Method 3050b, **Appendix 2: Methods of Soil Water Analysis**) and subsequently analyzed by inductively coupled plasma mass spectrometry. For nitrogen analysis samples were combusted at 1,000°C and N₂ gas was measured using an Elementar Vario MAX Macro Nitrogen Analyzer.

Data Analysis

Vegetation

Analyses of Variance (ANOVA) were run on all baseline data to check for preexisting differences between grazed and ungrazed treatments.

Repeated-measures ANOVA was run on vegetation data to determine the effect of grazing on measured vegetation variables over time (Marty, 2005). For all of the vegetation data there was a significant interaction between time and treatment (grazed/ungrazed), meaning that the effect of the treatment differed significantly on different sampling dates.

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Therefore a separate ANOVA was run for each sampling date for each vegetation variable, and repeated-measures analysis could not be used.

ANOVA was used to determine whether there was a significant difference in average *Phragmites* stem heights and counts between grazed and ungrazed plots on each sampling date. Data collected on species richness and cover from the 100 m² quadrats were similarly analyzed using ANOVA. These data were used to derive a Shannon-Weiner diversity index and evenness index for each plot, and ANOVA was run on the calculated indices.

Shannon-Weiner index was calculated as:

$$H'=-\sum[p_i \ln(p_i)]$$

H'=Shannon-Weiner diversity index

 P_i =the proportion of individuals found of the i^{th} species

For the calculation of the Shannon-Weiner index and species evenness, cover of each individual and total cover were substituted for number of individuals of each species and total number of individuals, as per (Biring et al., 2003). First, the cover classes assigned to each species were converted to the midpoint in % cover of each class, as per Peet et al. (1998). Then cover of each species was divided by the total plant cover in each plot in order to relativize cover values to 1. Next cover*In(relativecover) was calculated for each species. The negative of the sum of these values for all the species in each plot was the Shannon-Weiner index for that plot.

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In order to derive Shannon-Weiner evenness values for each plot, the Shannon-Weiner diversity index was divided by the natural log of the number of species in each plot:

J = H'/ln(S)

J=Shannon evenness index

H'=Shannon-Weiner diversity index

S=*number of species in the plot*

ANOVA was also run to compare the change in number of species, Shannon-Weiner index, and evenness from July 2008 to July 2009 between grazed and ungrazed plots.

All data sets were checked for assumptions of homogeneity of variances and normality and were log-transformed where normality was violated. All analyses were performed using SAS 9.2 (SAS Institute, Cary, NC). See **Appendix 3: Sample SAS Code** for a sample SAS code.

Water Quality

Repeated-measures ANOVA was used to test for differences in soil water nutrients between grazed and ungrazed plots over time. Ammonium and phosphate data were log-transformed and multiplied by 100 before statistical analysis to correct deviations from normality.

Soil Nutrients

ANOVA was also used to compare post-grazing soil nutrient levels and soil pH between grazed and ungrazed treatments. For most soil data there was a significant

interaction of treatment*depth. Therefore most soil data were analyzed separately at the two sampling depths (0-15 cm and 15-30 cm).

A Mehlich 3 molar saturation ratio (M3SR) was calculated for each soil sample as:

M3SR=molar Mehlich 3 P/(molar Mehlich 3 Al +molar Mehlich 3 Fe)
(Mukherjee et al., 2009)

ANOVA was used to compare Mehlich 3 molar saturation ratios between grazed and ungrazed plots.

Nutrient Pool Analysis

The average total amount of nutrients (nitrogen and phosphorus) stored in *Phragmites* biomass in grazed and ungrazed plots was calculated as:

biomass (g/m^2) * % tissue nutrient concentration = total g nutrient in Phragmites biomass/ m^2

Biomass=average live standing biomass in grazed or ungrazed plots, as calculated from the biomass harvest in August 2009

From these data a nutrient pool model was constructed for nitrogen and phosphorus in both grazed and ungrazed plots for August 12, 2009, the date of the *Phragmites* biomass harvest. Goats were not present in the plots on this date, but the effects of the first round of grazing seven weeks previously were taken into account in constructing the model.

In order to estimate goat consumption of *Phragmites* it was assumed that the amount of *Phragmites* available for the goats to eat during their first grazing period was equal to standing live biomass in the ungrazed plots measured in August 2009 minus standing

biomass in the grazed plots. It was assumed that goats consumed 70% of this biomass, based on a visual estimate, and that they returned 30% directly to the litter pool by killing it but not consuming it. It was assumed that the goats retained 15% of the nitrogen and 65% of the phosphorus they consumed as live weight gain (Davidson, 1985; Morse, 1992; Meschy, 2002; Dahlin, 2005. It was also assumed that the amount of non-*Phragmites* plants the goats consumed was negligible since these made up a fraction of the total biomass in each plot.

The sizes of the nitrogen and phosphorus nutrient pools were calculated as:

Soil nutrient (g/m^2) = concentration (mg/L) x measured bulk density (g/cm3) x soil volume per plot (L) x 50% soil solids / plot area (m^2)

For phosphorus, measured Mehlich 3 concentration was used and for nitrogen measured total soil nitrogen concentration was used. To calculate soil volume it was assumed that the bentonite clay sealed the plot at a depth of 30 cm. It was also assumed that 50% of the total soil volume was composed of soil solids and the remaining 50% was pore space (Rabenhorst, 2009).

The size of the soil *water* nutrient pool was calculated as:

Soil water nutrient (g/m^2) = measured concentration (mg/L) x soil volume of plot (L) x 20% / area of plot (m^2) / 1,000

It was assumed that in August 2009 the soil was at saturated at field capacity and therefore 20% of the total soil volume was composed of water-filled pore space (i.e. 50% of the soil volume was solids, 20% was water, and 30% was air) (Rabenhorst, 2009).

Chapter 3: Results

Vegetation

Phragmites Stem Counts

There was a significant interaction between treatment and time in the repeated-measures ANOVA on *Phragmites* stem counts, therefore a separate ANOVA was run for each sampling date. After the first round of grazing in 2008 there was no significant difference in *Phragmites* stem counts between grazed and ungrazed treatments. This was due to uneven grazing intensity in the four grazed plots: the two middle plots were heavily grazed, while the two outer plots were lightly grazed. This was clear from visual inspection of the plots (Figure 4). Notice in Figure 4b, the lightly grazed plot, the stand of virtually untouched *Phragmites* remaining at the very back of the plot. This stand was approximately 5 m deep by 8.5 m wide. In consequence of this uneven grazing intensity, ANOVA were run separately on the heavily grazed and lightly grazed blocks. These still revealed no significant differences between grazed and ungrazed plots, although for the heavily grazed plots the p-value was marginal (p=0.11).

In May, 2009, between grazing applications, there was no significant difference in *Phragmites* stem count between grazed and ungrazed treatments (p=0.2819). Regrowth of *Phragmites* was similar in both previously grazed and ungrazed plots.

After the second round of grazing in May-June 2009, there were twice as many standing live *P.australis* stems in ungrazed than grazed plots (p=0.0181, Figure 5a) on July 1. This difference was maintained after the third round of grazing (p=0.0385). Grazing was

more even during the second round between all four grazed plots, but the two middle plots were still visibly more heavily grazed than the outer two grazed plots.



Figure 4: a=photo of a heavily grazed plot; b= photo of a lightly grazed plot Both photos were taken on July 1, 2009, 10 days after the second round of grazing (first round of grazing in 2009) ended.

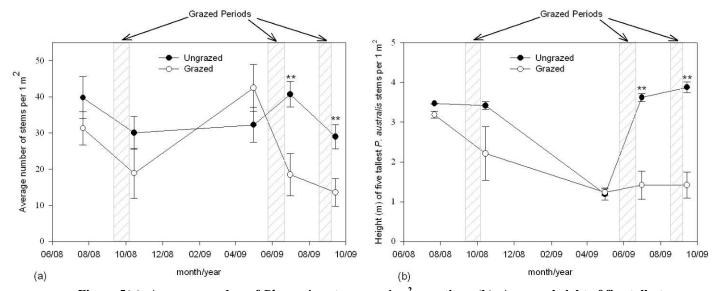


Figure 5(a): Average number of *Phragmites* stems per 1 m² over time; (b): Average height of five tallest *Phragmites* stems per 1 m² over time Error bars represent standard error of the mean.

Phragmites Stem Heights

There was a significant interaction between treatment and time in the repeatedmeasures ANOVA, therefore a separate ANOVA was run for each sampling date. The

^{**}indicates a significant difference (p<0.05) between grazed and ungrazed plots on this sampling date

results for *Phragmites* stem height were similar to those for stem counts (Figure 5b). After the first round of grazing in 2008 there was no significant difference in height of the five tallest *Phragmites* stems between grazed and ungrazed treatments. This was likely due to the uneven grazing intensity in the four grazed plots (Figure 4). In consequence, ANOVA's were run separately on the heavily grazed and lightly grazed blocks. There was a significant difference in *Phragmites* stem heights between heavily grazed plots (0.0277) and their ungrazed counterparts, but there was no significant difference between the lightly grazed plots and ungrazed plots (p=0.5888).

In May, 2009, between the two grazing applications, there was no significant difference in average *Phragmites* stem height between grazed and ungrazed treatments (p=0.6018).

After the second round of grazing, in 2009, *Phragmites* stems in ungrazed plots were two-and-a-half times taller than in grazed plots (p=0.0010). This difference was maintained after the third round of grazing ($F_{1,6}$ p=0.0004).

Phragmites aboveground biomass

Grazed plots had seven times less aboveground *Phragmites* biomass than ungrazed plots in August 2009, after one round of grazing in 2009 and one round in 2008 (p=0.0001, Figure 6).

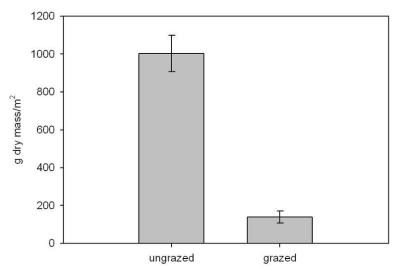


Figure 6: Dry weight of *Phragmites* per 1 m² after two rounds of grazing Error bars represent standard error of the mean.

Plant Diversity Measures

There was a significant interaction between treatment and time in the repeated-measures ANOVA for each measure of diversity, therefore a separate ANOVA was run for each sampling date. Species richness was 38% higher in grazed plots compared to ungrazed plots in August 2009 (p=0.0267), after two rounds of grazing and two months since the last round of grazing ended (Figure 7a). In September 2009, one week after the third round of grazing ended, species richness was over twice as high in grazed as in ungrazed plots, although this difference was not significant (p=0.1299).

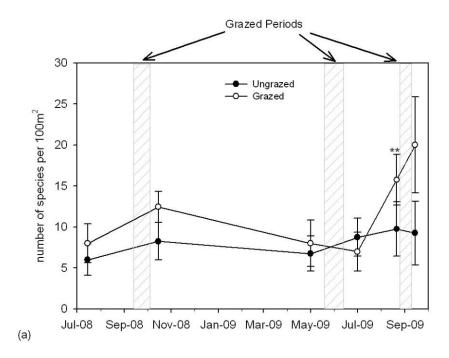
The patterns in the diversity data were similar for species richness, Shannon-Weiner diversity index, and Shannon-Weiner evenness (Figure 7). Shannon-Weiner diversity was twice as high in grazed as in ungrazed plots in August 2009, after the second round of grazing (Figure 7b), and this difference was significant at the 0.1 α level (p=0.0869).

Shannon-Weiner diversity in grazed plots was 58% higher than in ungrazed plots in September 2009, and this difference was again significant at the 0.1 α level (p=0.0887).

Between July 2008 and July 2009 Shannon-Weiner evenness increased in grazed plots by 0.218 units and decreased in ungrazed plots by 0.016 units, and this difference was significant at the 0.05 α level (p=0.0407). Shannon-Weiner diversity indices increased between July 2008 and July 2009 for both grazed and ungrazed plots. However grazed plots gained over three times more Shannon-Weiner diversity than ungrazed plots, and this difference in rate of increase was significant at the 0.1 α level (p=0.0619, grazed=0.412, ungrazed=0.124). Ungrazed plots gained an average of 2.75 species between July 2008 and July 2009 while grazed plots lost an average of 1 species, and this difference was also significant at the 0.1 α level (p=0.0576).

Lightly grazed plots had diversity indices comparable to those of ungrazed plots. The diversity indices of heavily grazed plots were much higher on average, although the experimental design did not allow for enough degrees of freedom to test the significance of this difference. For example, in July 2009, after the second round of grazing, the Shannon-Weiner diversity index of the ungrazed plots was 0.46, of lightly grazed plots was 0.42, and of heavily grazed plots was 1.42. Shannon-Weiner diversity in the heavily grazed plots was 70% higher than in the lightly grazed plots on this sampling date (Figure 8).

A list of the plant species encountered in the study plots can be found in Appendix 4.



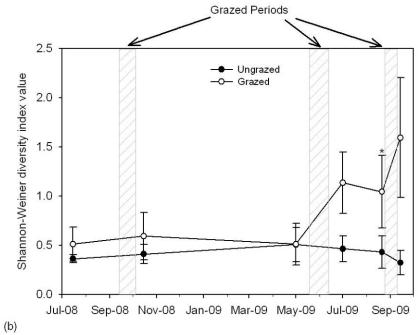


Figure 7: (a) Species richness and (b) Shannon-Weiner diversity index through time Error bars represent standard error of the mean.

^{*} indicates a significant difference at the 0.1 α level between grazed and ungrazed plots on this sampling date

^{**} indicates a significant difference at the 0.05 α level between grazed and ungrazed plots on this sampling date

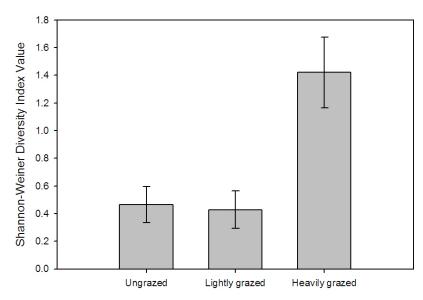


Figure 8: Shannon-Weiner diversity indices in ungrazed, lightly grazed, and heavily grazed plots Error bars represent standard error of the mean.

Soil Water

Soil water pH was lower in grazed than ungrazed plots, and this difference was significant at the 0.1 α level (p=0.0646, Figure 9, 6.88 grazed vs. 7.03 ungrazed). Figure 10 shows that levels of labile soil water nutrients in grazed and ungrazed plots tracked each other closely throughout the study. There were no significant differences in labile soil water nutrient levels between grazed and ungrazed plots. Nitrate, ammonium, and phosphate levels differed significantly between sampling dates (p=0.0261, p=0.0022, p=0.0030, respectively).

Grazing significantly affected total soil water nutrient levels (Figure 11). Grazed plots had higher levels of total nitrogen (3.54 vs.2.78 mg/L, p=0.0483) and total phosphorus (0.166 vs. 0.094 mg/L, p=0.0795) in soil water than ungrazed plots.

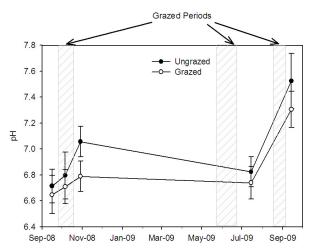
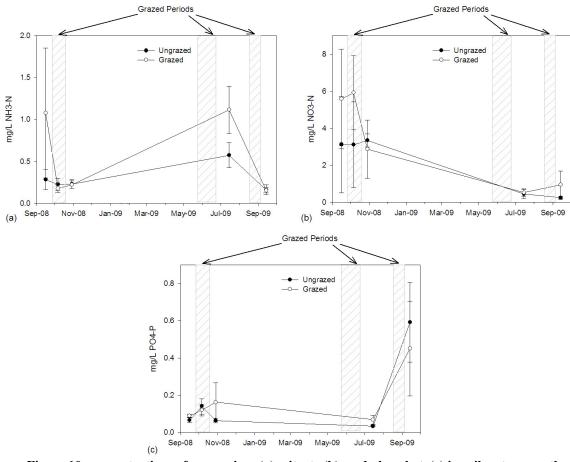
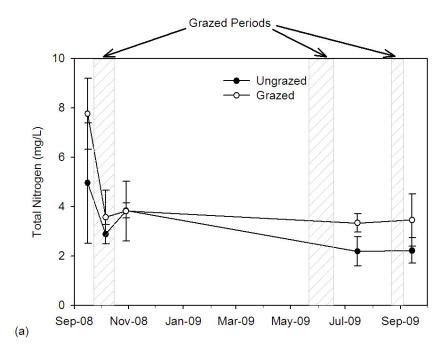


Figure 9: pH of shallow groundwater over time Error bars represent standard error of the mean.



 $Figure\ 10:\ concentrations\ of\ ammonium\ (a),\ nitrate\ (b),\ and\ phosphate(c)\ in\ soil\ water\ over\ the\ course\ of\ the\ experiment$

There were no significant differences between treatments in any soil water nutrient concentrations. Error bars represent standard error of the mean.



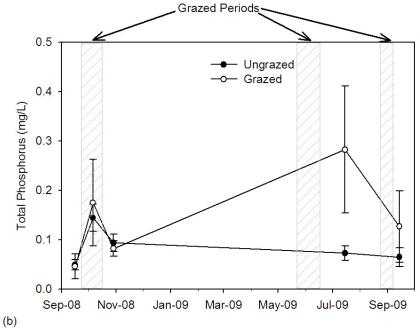


Figure 11: levels of total nitrogen (a) and total phosphorus(b) in soil water over time Error bars represent standard error of the mean.

The Mehlich 3 phosphorus saturation ratio of grazed plots at a depth of 15-30 cm was 3% higher than in ungrazed plots (p=0.0053, Figure 12). Across all soil samples the average Mehlich 3 molar phosphorus saturation ratio was significantly higher after grazing than the baseline level (p=0.0200).

Ungrazed plots had a significantly higher average water-extractable phosphorus level of $1.63~(\pm 0.082)$ compared to $1.24~(\pm 0.155)$ in grazed plots (p=0.0455). There was no significant difference in soil water-extractable phosphorus content between depths.

Baseline total soil nitrogen levels were significantly higher than post-grazing soil nitrogen levels across treatments and depths (p=0.0025, 150±19 vs. 91±14 g/kg), but there were no significant differences between grazed and ungrazed plots.

Soil total carbon levels were not significantly different between grazed and ungrazed plots and ranged between 1.7-2.7% in the top 15 cm of soil and 0.7-1% at a depth of 15-30 cm. The C:N ratio of the soil was approximately 16:1.

There were no significant differences in soil pH between treatments or depths. The average soil pH for all plots, sampling dates, and depths was 6.35 ± 0.04 .

Table 1 shows the average concentrations of important elements in the soil with standard deviations.

	Al	K	Ca	Fe	P	Molar P	Molar Fe	Molar Al
Average	649.4164	34.8602	1065.6453	205.0293	147.9925	0.0048	0.0116	0.0241
Standard								
Deviation	83.2615	4.5173	93.8839	22.2162	19.4918	0.0006	0.0015	0.0031

Table 1: Average Mehlich 3 concentrations and standard deviations of soil nutrients Molar values are the measured amounts converted to molar amounts. There were no significant differences in any of these nutrient concentrations between plots, so averages are presented pooling all plots.

Soil bulk density was higher in grazed plots (0.97±0.066 g/cm³) than in ungrazed plots (0.83±0.087 g/cm³), but the difference was not significant (p=0.24).

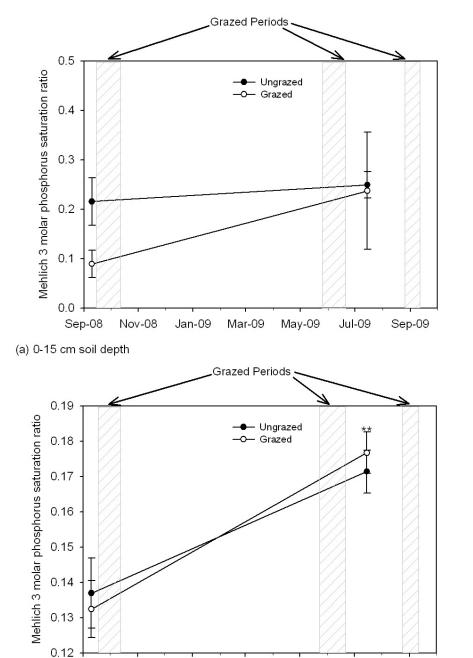


Figure 12: Mehlich 3 molar phosphorus saturation ratios from soil at a depth of 0-15 cm (a) and 15-30 cm(b) $\,$

May-09

Sep-09

Jul-09

Error bars represent standard error of the mean.

Nov-08

Jan-09

Sep-08

(b) 15-30cm soil depth

Mar-09

^{**} indicates a significant difference between grazed and ungrazed plots on that sampling date

Nutrient Pool Analysis

Phragmites in grazed plots had significantly (p=0.0013) higher tissue nitrogen concentrations than in ungrazed plots in August 2009, after two rounds of grazing (2.39%±0.09 N and 1.13%±0.10 N, respectively). Phragmites shoots in grazed plots also had significantly (p=0.0004) higher tissue phosphorus concentrations than ungrazed plots (0.21%±0.010 P and 0.11%±0.008 P, respectively). However, because they had higher biomass, the ungrazed plots contained more than three times the amount of nitrogen (p=0.0001) and almost four times the amount of phosphorus (p=0.0004) as grazed plots (Table 2).

	Ungrazed	Grazed
N	11.11±0.85 / 112.8±8.56	3.33±0.71 / 33.28±7.09
P	1.08±0.07 / 11.03±0.69	0.28±0.07 / 2.85±0.60

Table 2: Calculated amounts (g/m^2) / (kg/ha) of nitrogen and phosphorus stored in *Phragmites* biomass in grazed and ungrazed plots in August 2009 \pm standard error of the mean

Figure 13 shows a nutrient pool analysis for nitrogen and Figure 14 shows a nutrient pool analysis for phosphorus for this experiment for August 12, 2009, the date when *Phragmites* biomass was harvested and samples were collected for *Phragmites* tissue nutrient analysis.

The nitrogen pool analysis (Figure 13) shows that approximately three times as much nitrogen was cycled through *Phragmites* and goat grazing as was stored in the soil. Grazed plots had less nitrogen stored in the soil and more in the soil water compartment than ungrazed plots. Grazing therefore shifted nitrogen out of the *Phragmites* and soil pools and into the soil water, excreta, and goat biomass pools. Storage in goat biomass, denitrification, and volatilization, all provided opportunities for nitrogen loss from the system. The size of

the excreta pool is overestimated, because some, if not most, of the nitrogen from excreta would have been released into the measured soil and soil water compartments or transformed via volatilization and/or denitrification during the time between excretion and August. Table 3 shows that the nitrogen fertility of the system was reduced by approximately 63% after two rounds of grazing.

The phosphorus pool analysis (Figure 14) shows that less than a third of the phosphorus in the system was cycled through the *Phragmites* and grazing pathway; the rest was stored in the soil. Grazing moved phosphorus from the *Phragmites* live standing biomass pool to the Mehlich 3 and soil water pools. The estimate of phosphorus stored in the excreta pool is an overestimate, since some of this phosphorus is likely to have been transferred to the measured soil and soil water pools. There was a potential for permanent removal of phosphorus from the system in the form of goat biomass. Table 4 shows that grazing reduced the phosphorus fertility of the system by 8%. Using the estimates in Figure 14 to extrapolate the return of phosphorus as excreta over both grazing periods in 2009, I estimate that 0.264 total grams of phosphorus were excreted per m² during 2009.

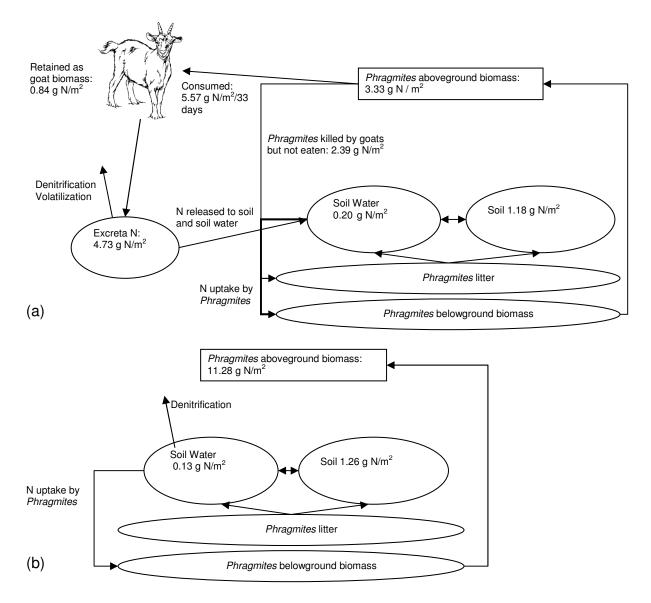
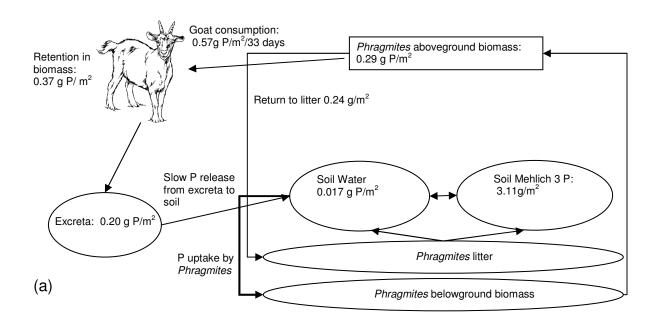


Figure 13: Nitrogen pool analysis for a) grazed and b) ungrazed plots in August 2009 Goats were not present in the plots in August 2009, but they have been included in the diagram to show the effect of their past presence during May-June 2009. Assumptions: 1) goat consumption was 70% of the difference in *Phragmites* standing stock in August between grazed and ungrazed plots (i.e. total growth – growth since the end of the last round of grazing), and 30% was returned as litter. 2) Goats excreted 85% of the nitrogen they consumed and retained 15% as live weight gain (Davidson, 1985; Dahlin, 2005).

	Ungrazed	Grazed
Soil	1.26	1.18
Soil Water	0.13	0.20
Phragmites aboveground	11.28	3.33
biomass		
Sum Total	12.67 g N/m ²	4.71 g N/m ²

Table 3: Measured g N/m² in soil, soil water, and *Phragmites* aboveground biomass



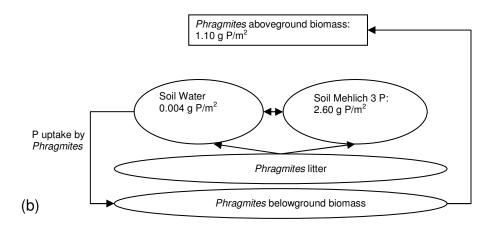


Figure 14: Phosphorus pool analysis for (a) grazed and (b) ungrazed plots in August 2009 Goats were not present in the plots in August 2009, but they have been included in the diagram to show the effect of their past presence during May-June 2009. Assumptions: 1) goat consumption was 70% of the difference in *Phragmites* standing stock between grazed and ungrazed plots (i.e. total growth – growth since the end of the last round of grazing), and 30% was returned as litter. 2) Goats excreted 35% of the phosphorus they consumed and retained 65% as live weight gain (Morse, 1992; Meschy, 2002).

	Ungrazed	Grazed
Soil	2.60	3.11
Soil Water	0.004	0.017
Phragmites aboveground	1.10	0.29
biomass		
Sum Total	3.70 g P/m ²	3.42 g N/m ²

Table 4: Measured g P/m² in soil, soil water, and *Phragmites* aboveground biomass

Chapter 4: Discussion

Vegetation

Effects of Grazing on Phragmites

The results support my hypothesis that goats can effectively control *Phragmites*. The goats consumed *Phragmites* and reduced its biomass, although their consumption rates varied between plots. Initially grazing was more intense in some of the plots than others, but grazing became more even with each subsequent grazing event.

Neophobia (a fear of new things or of the unknown) and personality differences between the animals probably played a role in the observed uneven grazing intensity (Michelena et al., 2008). My study represented a major shift in diet, setting, and herd size for the animals, and some of them apparently took to the change better than others. Studies have shown that sheep are reluctant to eat novel foods, especially when they are pastured in a new location (Chapple and Lynch, 1986; Burritt and Provenza, 1997). In my study the combination of the novel situation and novel food probably reduced the animals' food intake, at least for the first few days. Presumably the animals in the middle two grazed plots watched each other consume *Phragmites* and quickly overcame their aversion to the novel food source. The animals in the further two plots, however, may have taken longer to overcome their aversion since they had less social support. Boissy and Le Neindre, (1990) showed that the social support provided by the presence of peers has positive consequences for individual ruminants exposed to a novel situation.

Another possible reason for uneven grazing intensity could be the spacing of the plots causing more stress for the more isolated goats. The heavily grazed plots were adjacent to each other, while the lightly grazed plots were spaced further away (Figure 15). The tall *Phragmites* in ungrazed plots separated the animals in plots 2 and 8 visually from goats in other plots. Goats are gregarious, and it is likely that the individuals in the edge plots felt anxiety at being reduced from part of a herd of many (their group size before this experiment) to a pair. The anxiety caused by this social isolation could have reduced their appetite. Mills and Faure (1989) similarly found that domestic hens that were prevented from feeding as a group reduced time spent feeding and increased frustration behavior. Kanitz et al. (2009) found that separation of piglets from their herd caused behavioral and physiological signs of stress.

The goats in the outer plots may have also perceived themselves to be running a higher risk of predation, and this could have increased their anxiety levels and thus decreased foraging. Berger (1991) found that when running a predation risk wild bighorn sheep in smaller groups grazed less efficiently than individuals in larger groups. There has been evidence of coyote activity at the BARC facility over the last few years (W. Hare, personal communication) and the goats may have also sensed coyote presence. I personally observed that during the first round of grazing the goats in plot 2, one of the edge plots, ran to the back of the plot and hid whenever I or any vehicles came by. Their increased anxiety and vigilance behavior probably reduced the amount of time they spent foraging.

In contrast, the animals in the middle plots may have felt as if they were effectively a herd of four since they were separated by only three feet of fencing through which they could clearly see each other. In addition to reducing their fear of predation, studies have

shown that this would have increased their feeding rate relative to the more isolated goats. Shrader et al. (2007) found that goats feed faster in the presence of competitors and that domestic goats glean information from watching other goats feed that directly affects their foraging behavior. Chapple and Lynch (1986) similarly showed that sheep can learn to eat novel foods within 15-30 minutes from sheep that have prior experience with the food. The animals in the middle plots may have watched each other forage on *Phragmites* and more quickly determined it was a suitable food source. They may have subsequently been driven by competitive interactions to maximize their intake rate.

It is likely that all of the above factors contributed to producing the uneven grazing intensity observed across my study plots during the first round of grazing. Overall this suggests that the most efficient control of *Phragmites* using ruminants will be achieved when using herds of at least four animals and when having introduced the animals to eating *Phragmites* before moving them to a new pasture containing *Phragmites*.

Grazing rates across plots were more even in subsequent rounds of grazing, presumably because the goats had overcome their neophobia. My results are in accordance with Olson et al (1996)'s recommendation that animals used for biocontrol should have prior experience grazing the weed to be controlled. Popay and Field (1996) also found that when using lambs to control leafy spurge (*Euphorbia esula*) and mountain mahogany (*Cercocarpus montanus*) lambs with previous experience of eating the plants were more likely to be effective control agents. This suggests that more consistent success in controlling *Phragmites* with goats could be achieved by using animals conditioned to consume the plant, either through having grazed on invaded pasture previously, or by having been introduced to the plant as forage in a familiar environment.

The timing of grazing may also be an important factor in maximizing *Phragmites* control and minimizing environmental damage. Based on a study of the seasonal changes in rhizome energy reserves, Karuntanatne et al. (2004) suggested that June was the optimal time to harvest *Phragmites* aboveground biomass if the aim is to weaken the stand. Their results suggested that harvest only in July or August would maintain a healthy stand of *Phragmites* in subsequent years. Future studies on grazing of *Phragmites* should focus on determining the optimum timing, frequency, and intensity of grazing to maximize *Phragmites* control and plant diversity.

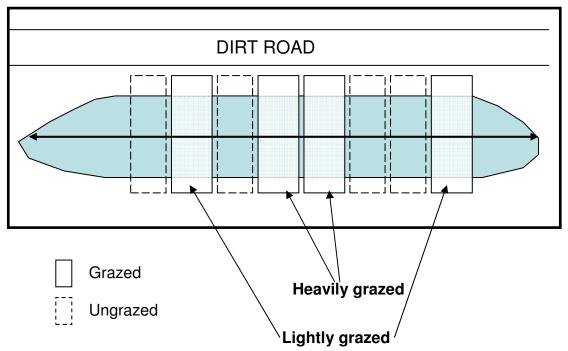


Figure 15: Layout of lightly and heavily grazed plots

Effects of Grazing on Plant Diversity

My study showed that increases in plant diversity are detectable after only one year and two rounds of intensive grazing of a *Phragmites*-dominated wetland. Grazed plots had significantly higher plant species richness and Shannon-Weiner diversity than ungrazed plots

after the second round of grazing. Species evenness remained higher in grazed plots after the third round of grazing. The trends (Figure 7) in diversity data also suggested that after the second round of grazing diversity was increasing in grazed plots but remaining stagnant in ungrazed plots.

Many studies, including Van Wieren et al. (1998), Vulink et al. (2000), Ausden et al. (2005), Tesauro and Ehrenfeld (2007), and others have found that grazing of *Phragmites* leads to increased plant diversity. These and other studies of changes in plant communities following restoration typically examine time scales of at least five to ten years, if not longer. For example, Vilem et al. (2007) used grazing as a tool for restoring plant diversity in upland grassland and found that it took four years for the plant community to show a detectable response. Vulink et al. (2000) found that grazing of *Phragmites* replaced it with a short grass community within four years. My study is the first to my knowledge to document significant increases in plant diversity after only one year of grazing *Phragmites*. Marty (2005) documented that grazed vernal pools had significantly higher cover of native plants and lower cover of exotic grasses than ungrazed plots in the first year of grazing, however *Phragmites* was not a factor in his study. Future studies should continue over longer timescales to better understand the response of wetland plant communities to grazing.

My results suggest that it takes at least two rounds of grazing on *Phragmites* monocultures for diversity effects to begin to appear. I believe that if my study were to continue for another year, grazing intensity would become more even across all grazed plots. This is supported by the decrease in variability of vegetation measurements I observed in subsequent rounds of grazing. This should lead to unambiguous increases in plant diversity in grazed plots compared to ungrazed plots over time.

My data suggest that there was an increase in diversity from grazing, but that the effect was difficult to detect due to large variation in response between plots. Uneven grazing intensity caused large residual error in diversity data in grazed plots, as is suggested by the large error bars for grazed plots in Figure 7 as compared to the error in ungrazed plots. Figure 8 also shows that diversity in lightly grazed plots was very similar to diversity in ungrazed plots. This variability made it difficult to detect a significant difference between grazed and ungrazed plots.

The detection of significant differences in plant diversity between grazed and ungrazed plots was also hampered by the fact that block 1 was an anomaly: the ungrazed plot in block 1 consistently had higher plant diversity than the grazed plot, even in the baseline survey. Graphically block 1 showed a reversed trend compared to the other blocks. This could be attributable to edge effects and that fact that it was one of the lightly grazed plots. However removing block 1 from the analysis did not lead to any significant differences, probably due to the resulting loss of statistical power.

It is notable that two months after the second round of grazing ended grazed plots had significantly higher species richness than ungrazed plots. However a week after both the second and third rounds of grazing there were no significant differences in species richness or Shannon-Weiner diversity between grazed and ungrazed plots. This suggests that it takes between a week and two months after a *Phragmites* monoculture has been grazed for plant diversity to increase. The goats apparently ate every plant in the plots, including plants other than *Phragmites*. Therefore intensive grazing had the immediate effect of decimating the plant community. It apparently took time for plants other than *Phragmites* to be able to grow and take advantage of the newly available resources.

Plants can colonize a site via seed or clonally, and it appeared that both mechanisms were factors in the increased plant diversity observed in my study. Many of the newly colonizing plants were seedlings, such as *Acer rubrum* (red maple) and *Quercus rubra* (red oak), as well as several seedlings that were too small to be identifiable (Appendix 4). Some of the newly colonizing plants may have already been present in the seed bank, but it took time for their seeds to germinate and grow. Although habitat became available once *Phragmites* was cleared, these seeds required suitable germination conditions, e.g. adequate moisture and temperature, and these conditions can vary temporally and spatially. Seeds from other plants may not have been present in the seed bank and would have had to disperse to the site via wind or animal vectors. It is possible that the goats brought seeds of some plants to the site. Other plants colonized clonally, by growing into adjacent areas. Clonal plants that expanded into my grazed plots included *Polygonum perfoliatum* (mile-a-minute weed), *Toxicodendron radicans* (poison ivy), *Vitis sp.*, (grape), *Lonicera japonica* (Japanese honeysuckle), and *Lonicera tatarica* (bush honeysuckle).

I believe that if grazing were used as a management tool for successive years, once the *Phragmites* regrowth has been substantially reduced (probably after two or three years), grazing pressure (number of animals/unit area) should be reduced. European studies have generally shown that extensive or low-intensity grazing maximizes biodiversity benefits while restricting clonal invasives (Van Wieren et al. 1998; De Cauwer and Reheul 2009), as opposed to intensive grazing, which has been shown to be detrimental to biodiversity (Jansen and Robertson, 2001). Marty (2005) showed that a stocking density of one animal unit (cowcalf pair) per 2.4 ha improved plant and aquatic invertebrate diversity compared to ungrazed

wetlands. Future studies on grazing *Phragmites* should attempt to determine an appropriate stocking density to maximize plant biodiversity.

My study did not determine the mechanism by which grazing of *Phragmites* increases plant diversity. Bakker (1985) observed that grazing in salt marshes improved plant diversity and attributed this effect to litter removal and exposure of bare soil by grazers. This is one possible mechanism by which grazing could have increased plant diversity in my study. Another possible mechanism is the removal of tall *Phragmites* culms that prevented sunlight from reaching the soil, potentially inhibiting seed germination. Van Wieren et al. (1998) and Levine (1998) both suggested that light is the limiting resource where nutrients are abundant, as was the case in my study. Besides its tall, dense growth during the spring and summer, *Phragmites* leaves many standing dead culms throughout the winter that block sunlight from reaching the ground in early spring. This has been suggested as a possible mechanism by which *Phragmites* outcompetes other plants (Onimaru and Yabe, 1996; Gusewell and Klotzli, 1998). After the first round of grazing in my study grazed plots had fewer standing dead culms throughout the winter and also less litter. This may have altered the competitive balance against *Phragmites* in favor of native plants.

Phragmites belowground parts have been shown to secrete an allelopathic chemical that causes acute rhizotoxicity in other plants (Rudrappa et al., 2007). By reducing Phragmites aboveground biomass, grazing may also reduce the amount of energy Phragmites roots allocate to production of allelopathic compounds and thus reduce its competitive advantage over other plants.

Soil water

Nitrate, Ammonium, and Phosphate

The results of this study support my hypothesis that grazing would not significantly increase levels of soluble soil water nutrients, and thus risk of pollution. Levels of nitrate, ammonium, and phosphate in grazed and ungrazed plots tracked each other closely throughout the study, but there were no discernible patterns (Figure 10). Nitrate levels were far below the EPA drinking water standard of 10 mg/L (US EPA, 2009). Soil water phosphate levels in both grazed and ungrazed plots were above the EPA standard of 0.1 mg/L for rivers and streams, however.

In contrast, Findlay et al. (2003) found that treating *Phragmites* with herbicide increased porewater ammonium concentrations for at least two years following treatment. Bart and Hartman (2000) similarly found that clipping *Phragmites* led to a 3- to 4-fold increase in porewater ammonium concentrations. In my study grazing did not increase porewater ammonium (or nitrate) concentrations, presumably because the nitrogen was consumed by the goats rather than released from the dying plants in situ. Any mineralized nitrogen could also have been removed via denitrification or volatilization. My results may therefore suggest that grazing *Phragmites* causes less risk of nitrogen export from a wetland than herbicide application or mowing.

The significant effect of time on nitrate, ammonium, and phosphate content of soil water suggests that, despite my high stocking rate, any elevation in labile soil water nutrients attributable to the presence of herbivores was negligible compared with the seasonal fluctuations driven by changes in vegetation, temperature, and soil moisture (Jamieson et al., 1999; Hook and Burke, 2000). Bouman et al. (2008) similarly found significant seasonal

effects in nitrate concentration of soil water. This also suggests that the best way to examine effects of grazers on labile soil nutrients would be to remove the effect of seasonality by comparing samples taken at the same time of year. There are substantial differences in measurements, however, even when comparing the baseline levels in September 2008 to the measured levels after the third round of grazing in September 2009. PO4-P levels were much higher in 2009 than 2008 in both grazed and ungrazed plots, nitrate levels were lower, and ammonium levels were comparable between both years. Both inter-annual and seasonal variations therefore appear to overwhelm the effects of grazing on soil water labile nutrient levels.

On the other hand, it is possible that grazing did increase soil and soil water nutrient levels but that there is a lag time between nutrient application and nutrient release by the wetland, as some authors have shown (Owens et al., 2008; Meals et al., 2010). However these authors measured response of nearby streams and groundwater seeps rather than nutrient levels directly in soil water. Afzal and Adams (1992) found that soil nitrate concentrations under cattle dung and simulated cattle urine patches increased substantially within days of application and peaked within a month, then very gradually fell off over a period of weeks. Based on the rapid response of soil water nitrate levels to urine addition and the gradual decline these authors observed, I believe that my measurements captured treatment effects.

There was high variability in soil water nutrient levels within treatments. In some cases there was a ten-fold difference in readings from the two lysimeters in one plot.

Spatial variability in soil chemical and physical characteristics and readings from suction cup lysimeters have often been reported to be high and underestimated by researchers (Grossman

and Udluft, 1991; Lord and Shepherd, 1993). Grazing has also been reported to increase spatial variability of soil nutrient levels (Afzal and Adams, 1992; Lord and Shepherd, 1993; Augustine and Frank, 2001). These factors suggest that more than two lysimeters should have been installed in each of my study plots. Bouman et al. (2008) used a density of lysimeters four times higher than mine to measure soil water nutrients under extensively grazed pasture. Lord and Shepherd (1993) suggested that in arable rotations 20-25 replicates were required to detect differences of 25%, 5-7 replicates to detect differences of 50%, and more under grazed fields. Based on the variability in my results and the work of these authors, I would recommend that future studies use at least eight lysimeters per 100m² to estimate mean soil water nutrient levels. Future studies should also use higher replication of grazed/ungrazed treatments to gain more statistical power. Despite high variability, I believe that my results show that grazing will not increase ambient soil water labile nutrient levels in the short-term.

The state of the wetland (flooded vs. non-flooded) also probably had a significant effect on measured soil water nutrient levels. For example, ammonium levels were elevated in July 2009 compared with September and October 2008. This was probably because the soil was flooded at the time of sampling in July 2009 but not in the fall of 2008, and under flooded conditions most of the nitrogen in soil water will exist in the form of ammonium. Nitrate levels were lower in July 2009 than in the fall of 2008, which agrees with this hypothesis.

Soil and Soil Water pH

The decrease in soil water pH observed in grazed plots relative to ungrazed plots is difficult to explain. This difference was more pronounced after the first round of grazing and

became less pronounced through 2009. The difference between grazed and ungrazed plots was 0.15 pH units. There was also no significant difference in soil pH at either depth between grazed and ungrazed plots. So, although this difference is statistically significant, it may not be biologically meaningful.

One possible explanation for this effect may be that grazers trampled the detritus, opening up the canopy, increasing aeration and decomposition, and thus increasing the formation of organic acids relative to ungrazed plots. Zacheis et al. (2002) similarly found that grazing by graylag geese in an Alaskan salt marsh decreased soil pH slightly but significantly, but they did not attempt to explain the effect.

The veterinarian on this project, Dr. Hare, also suggested that the sugar-rich *Phragmites* may make the goats' urine more acidic (Bill Hare, unpublished data). Milchunas and Lauenroth (1993) found that the literature on grazing documented no consistent trend in soil pH. Some of the studies they reviewed documented a slight decrease or increase in soil pH with grazing similar to mine, but none attempted to explain this result.

Total Nitrogen (TN) and Total Phosphorus (TP)

I hypothesized that grazing would not affect total soil water nutrient levels, but my results do not support this hypothesis. Grazing increased levels of total soil water nitrogen by 21% and phosphorus by 43% relative to ungrazed plots. Since ammonium, nitrate, and phosphate levels were not similarly elevated, and assuming that nitrite levels were negligible, the observed increase in total nutrient levels must have been caused by elevated dissolved organic nitrogen and phosphorus in soil water.

Relatively few studies on grazing have measured levels of total dissolved nutrients or dissolved organic nutrients in soil water. One study, Foote & Hornung (2005), found that

cattle grazing in prairie pothole wetlands did not increase total nitrogen or phosphorus levels in surface waters. Campbell et al. (2009) found, however, that grazing in riparian areas increased total nutrient levels in adjacent water bodies. Campbell et al. (2009) hypothesized that both excretion and physical disturbance caused the increase they observed.

There are several potential mechanisms by which grazing could have increased dissolved organic nutrient levels in my grazed study plots. It is possible that the observed increase in total nutrient levels in soil water was made up of direct inputs of organic nitrogen and phosphorus from goat excreta. Although these nutrients were deposited on the soil surface, they could have rapidly moved down into the soil water via macropore flow (Grossman and Udluft, 1991; Pakrou & Dillon, 1995; McGechan, 2003).

Trampling of the vegetation may also have increased organic nutrient levels in grazed plots relative to ungrazed plots. The physical abrasion of litter from trampling could have made more surface area available for microbial attack. This could have accelerated decomposition and the release of organic acids as well as increasing litter incorporation into the soil. As discussed in the nutrient pool analysis below, the goats in my study also killed some *Phragmites* without eating it, increasing returns to the litter pool in the middle of the growing season. The added organic nutrients in soil water could therefore have been derived from decomposing plant matter. Zacheis et al. (2002) similarly found that trampling by geese in an Alaskan salt marsh stimulated nitrogen mineralization and attributed this effect to greater litter incorporation into the soil.

In my study grazing also maintained a higher percentage of nitrogen and phosphorus in *Phragmites* tissue, and this could have made *Phragmites* litter in grazed plots more easily decomposable (Sirotnak and Huntley, 2000).

Trampling, higher plant tissue nutrient levels, and input of dung could all have contributed to accelerating decomposition and could therefore have stimulated microbial activity in grazed plots. The increased organic nutrient concentrations in soil water could therefore have been at least partially derived from microbial biomass. Ruess and McNaughton (1987) similarly found that grazing in Serengeti grassland stimulated the microbial community.

Dissolved organic nitrogen has recently come to be regarded as an overlooked pathway of nitrogen loss from agricultural soils and a pollution source to receiving waters (Oleman et al., 2007; van Kessel et al., 2009). Colloidal transport of phosphorus through soils via macropore flow has also recently been documented, and manure is believed to be an important source of colloids for phosphorus transport (Nash and Halliwell, 2000; McGechan and Lewis, 2002). The elevated total nutrient levels in my study therefore raise concerns that there is a potential for grazing in wetlands to cause nutrient pollution via export of dissolved organic nutrients.

Wetlands, however, are efficient at removal of total nitrogen and phosphorus from wastewater (Kadlec, 2003; Tanner et al., 2004; Kadlec, 2009). As long as the retention time of water in a grazed wetland is long enough to process these nutrients, I believe it is unlikely that increased levels of TN and TP in the soil water would translate to increased export of TN and TP from the wetland. The excess organic nitrogen in the soil water could eventually be stored in the soil, taken up by plants, or removed via volatilization or denitrification. The excess phosphorus could be stored in the soil as well, although the soil in my study wetland may already be nearing phosphorus saturation, as discussed in the "Soil" section below.

Nevertheless, I would recommend that future use of goats to control *Phragmites* minimize the risk of nutrient pollution by reducing the stocking density. The stocking density I used was 20% higher than even intensive grazing systems in grassland (e.g. Dormaar and Willms, 1998; de Klein and Ledgard, 2001), and was much higher than the extensive grazing systems used in Europe to control *Phragmites* and other clonal dominants (Bakker, 1985; Gusewell et al., 2007; de Cauwer and Reheul, 2009). Any *Phragmites* control effort would need to balance a desire for quick *Phragmites* eradication, requiring a higher stocking density, with the need to minimize the risk of nutrient pollution, which would require a stocking density as low as possible. Studies of extensive grazing systems in grasslands have consistently shown that extensive grazing does not increase nutrient pollution and can in fact be used as a tool to decrease nitrate leaching (Anger et al., 2002; Owens and Bonata, 2004; Bouman et al., 2008). Future studies on grazing to control *Phragmites* should determine the optimum stocking density for minimizing pollution risk while maximizing *Phragmites* control and increases in plant diversity.

Restricting grazing late in the growing season could also help to minimize the risk of pollution. When grazing early in the growing season nutrients are released at a time when plants need them and can rapidly assimilate them. However when grazing late in the growing season nutrients are released at a time when plants are no longer taking them up as quickly, therefore they are likely to remain in the soil or soil water, available for export (Stout et al, 1997).

Other studies (e.g. Afzal and Adams, 1992) have found that most of the nitrogen excreted from grazers is mineralized within a few days. In my study the excreted nitrogen was apparently not rapidly mineralized, but remained as organic nitrogen for at least six

weeks. This could have occurred because the waterlogged soil conditions in my study could have prevented urea mineralization, thus elevating organic nitrogen levels instead of mineral nitrogen levels. The soil in my study became waterlogged briefly towards the end of the first grazing period, in the fall of 2008, and was waterlogged during most of the first grazing period in May-June 2009. The soil was relatively dry, however, during the last grazing period, yet no increases in mineral nitrogen levels were measured. It is also possible that waterlogged soil conditions could have caused denitrification of some or most of the nitrogen that was mineralized, and that under dry conditions some nitrogen could have volatilized.

It is also possible that during my study some nutrients were translocated from the wetland to the adjacent upland area. It is unlikely that the upland area on which the goat hutches were placed was underlain with the bentonite clay that lined the wetland. There was a high concentration of animal excreta visible around these resting areas. Nutrients may have been translocated from the wetland to the upland area and then leached from the soil. The ground in the upland area was very hard, however, having probably been modified when the adjacent dirt road was constructed. Therefore it is possible that much of the water flow on the area would have been surface runoff rather than percolation through the soil, and since the area sloped towards the wetland, these nutrients could have been returned to the wetland.

Soil

This study found that grazing increased soil molar phosphorus saturation ratio (MPSR) slightly at depth and decreased soil water-soluble phosphorus levels. Total soil nitrogen and carbon levels were not affected by grazing. These findings agree with those of Augustine and Frank (2001), who found that grazing of grassland by wild ungulates in

Yellowstone National Park did not increase total soil nitrogen, mineral extractable nitrogen, total soil carbon, or nitrogen mineralization potential of the soil.

Soil Water-Soluble Phosphorus

Soil water-soluble phosphorus levels were 24% lower in grazed plots than in ungrazed plots. This suggests that in grazed plots more water-extractable phosphorus was being removed from the soil, presumably by either plants or microbes. Grazing could have accelerated *Phragmites* phosphorus uptake, since grazed *Phragmites* grew new shoots that had twice the tissue phosphorus levels of *Phragmites* in ungrazed plots. Grazing could also have stimulated the microbial community (Harrison and Bardget, 2008) and thus increased the amount of soil phosphorus from the water-soluble pool incorporated into microbial biomass.

It is also possible that, assuming grazing reduced the overall amount of decaying plant litter in plots, grazing reduced water-soluble phosphorus release from decaying plants. Decaying plants have been found to release significant amounts of water-soluble phosphorus (Nash and Halliwell, 2000). It is reasonable to suppose that the goats consumed much of the aboveground biomass that would have eventually become litter, senesced and decayed, releasing phosphorus.

Trampling the soil also presumably increased mixing and contact of aluminum and iron soil minerals with water-soluble phosphorus, and thus may have increased phosphorus bound in the top layer of soil. This would have reduced water-soluble phosphorus and increased Mehlich 3 phosphorus in shallow soil.

Baseline soil Mehlich 3 phosphorus levels were well above the levels at which Sims et al. (2002) recommends environmental action should be taken for *agricultural* soils and Mehlich 3 phosphorus saturation ratios were well above those predicted by Mukherjee et al. (2009) to cause phosphorus release from wetland soils to water. The high baseline nutrient levels are probably attributable to the fact that the area was used for agriculture for decades before wetland creation/restoration. The soil was undoubtedly heavily fertilized while it was under cultivation. Wetlands with high baseline phosphorus fertility could pose a greater risk of phosphorus export from the wetland, but they also present a greater opportunity for phosphorus removal from the system.

In my study the soil molar phosphorus saturation ratio (MPSR) was significantly higher in grazed than ungrazed plots at a depth of 15-30 cm after the second round of grazing. The difference between grazed and ungrazed plots was 3%, which is so small as to be of questionable importance. Mean MPSR in grazed plots also increased at the 0-15 cm depth relative to ungrazed plots, but its measure was so variable that no significant difference could be detected.

Activities at the surface of a soil, such as grazing, very rarely affect Mehlich 3 phosphorus saturation ratio at depth. It is therefore difficult to explain the increase I observed as a treatment effect. It may have been caused by hydrologic factors, such as phosphorus release from minerals in the bentonite clay.

The soil MPSR increased by about a third for both grazed and ungrazed plots between September 2008 and July 2009. This is probably attributable to the fact that the soil was saturated during soil sampling in 2009 but not in 2008. The Mehlich 3 test releases most

iron- and aluminum-bound phosphorus, therefore dissolution of these bonds via reduction probably did not contribute much to the observed increase. Flooding releases phosphorus through cell lysis and other mechanisms (Wright et al., 2001).

Nutrient Pool Analysis

Although my results for soil water above suggest that heavy grazing of a *Phragmites*-dominated wetland can increase nutrient availability, the nutrient pool analysis suggests that grazing can reduce the fertility of a *Phragmites*-dominated wetland. Nitrogen can be removed via grazer biomass, denitrification, and volatilization, and my results suggested that 62% of the nitrogen in the system was removed after two rounds of grazing. Phosphorus can be removed in the form of grazer biomass, and my results suggest that grazing reduced the phosphorus fertility of the wetland by 8%. Reducing fertility could be a key to long-term *Phragmites* control, since *Phragmites* is adapted to high-nutrient environments. By permanently reducing nutrient availability, grazing could eventually make the habitat unsuitable for *Phragmites* re-colonization.

Figure 13 shows that nitrogen excreted from the goats can be removed from the system via denitrification or volatilization. Grazing makes a large amount of nitrogen rapidly available in the soil and soil water, and this typically overwhelms plant demand for nitrogen (Stout et al., 1997). This provides an opportunity for denitrification or volatilization losses (Stout et al., 1997; Rotz et al., 2005). This is also the point in the cycle at which there is a potential for nitrate to leach or be exported from the system in surface water.

The rates of denitrification and volatilization will vary depending on climatic conditions and were not measured in this study. Wetter pastures should experience higher

denitrification rates and lower volatilization losses, and the opposite should be true at drier times of year. Frazier et al. (1994) reported that 28% of the nitrogen applied by grazing animals in a wet pasture was lost to denitrification. Zhou et al. (2009) reported that 30-40% of nitrogen applied to a rice paddy as both fertilizer and liquid cattle waste was lost via denitrification and 2.5-4% was lost via ammonia volatilization. Treading by the goats may also enhance denitrification rates. Menneer et al. (2005) found that treading by cattle increased denitrification rates by up to 22 times the baseline rate for a period of 28 days following the event.

In addition to denitrification and volatilization losses, nitrogen can be lost from the *Phragmites* pasture through goat uptake. Approximately 15% of the nitrogen goats consume will be removed in the form of goat biomass (Davidson, 1985; Dahlin, 2005), assuming the animal does not die and decompose on site.

Grazing can similarly remove phosphorus from the ecosystem in the form of biomass. Goats retain 65-75% of the phosphorus they ingest as live weight gain (Morse, 1992; Meschy, 2002). Thus only a small proportion of the phosphorus goats ingest will be returned to the system. Almost all of the phosphorus excreted by a ruminant is returned in the form of feces, and this is relatively slowly decomposed. The slow decomposition rate means that phosphorus can be released at a rate at which plants can use it, as opposed to nitrogen in urine, which is typically released so rapidly that it overwhelms plant demand.

Reducing the phosphorus fertility of the soil could be a long process, however, since the nutrient pool analysis shows that grazing reduced phosphorus fertility by only 8%.

Slightly less than 1/3 of the Mehlich 3 phosphorus in the system was being cycled through

Phragmites and goat grazing. It could take many years to draw out the soil store of phosphorus and remove it in the form of animal biomass.

Although as discussed above heavy grazing may increase the risk of nutrient export from a wetland, heavy grazing may nevertheless promote reductions in soil fertility. Li et al. (2008) found that historic *light* and *moderate* sheep grazing on the Mongolian steppe did not affect soil total nitrogen or phosphorus levels, but *heavy* grazing decreased soil total nitrogen by 11.4% and total phosphorus by 7.6%.

Richardson (1999) suggested that over a range of wetlands, including both mineral and peat soils, phosphorus additions of more than 1 g/m²/year will generally be too high for the wetland to absorb and will lead to phosphorus export from the wetland. I estimated that the goats in my study returned approximately 0.26 g/m² phosphorus to the wetland in the form of excreta in 2009, well below the 1 g/m²/yr threshold. This suggests that grazing is unlikely to cause phosphorus export from the wetland. Rather, the wetland should be able to assimilate this phosphorus.

Grazing increased the concentrations of N and P in *Phragmites* biomass. This probably made the *Phragmites* litter in grazed plots more easily decomposable and thus sped up nutrient cycling. Many other studies have similarly found that grazing increased plant tissue nutrient concentrations (e.g. Hik and Jefferies, 1990; Fox et al., 1998; Sirotnak and Huntley, 2000). As discussed above, this could have accelerated decomposition in grazed plots.

Grazing also affected the timing of *Phragmites* additions to the litter pool in a way that probably accelerated decomposition. Since the goats consumed only an estimated 70% of the *Phragmites* in their plots, grazing released approximately 30% of the live standing

stock of *Phragmites* to the litter pool in the middle of the growing season, when soil temperatures and trampling would have facilitated decomposition. In contrast, in the ungrazed plots *Phragmites* senesced in the fall, and it quickly became too cold for soil organisms to decompose the fresh litter.

It is possible that there were nutrient inputs to the wetland system from nutrient pools in the litter layer and belowground biomass of *Phragmites*. The size and fluxes to and from these pools were not measured or estimated in my study. It is likely that grazing moves nutrients out of these two compartments and into the goat-excreta-soil water pathway. I expected grazing to deplete *Phragmites*' rhizome store of energy and nutrients, and the nutrient pool analysis suggests that this may have occurred. The rhizome must have had to release energy and nutrients to replace its grazed aboveground parts. Alternately, some belowground parts of the *Phragmites* clone could have senesced and released their nutrients directly to the soil and soil water.

Nutrients could also have been released from the litter layer more quickly in the grazed plots than in the ungrazed plots. The previous year's litter could have been more quickly decomposed in the grazed plots by trampling and stimulation of the microbial community, as discussed earlier (Zacheis et al., 2002). The litter layer can contain a large proportion of nitrogen in an ecosystem system. For example, Robertson et al. (1993) found that the litter layer in a grassland contained 30-50% of the nitrogen in the system.

It is unlikely that the goats imported nutrients to the site, since they were not given any supplemental feed while they were in the *Phragmites* enclosures. It is also possible that soil bacteria fixed some nitrogen, but this was not measured, and there is no reason to suppose nitrogen fixation would have been favored in this system. There were few to no

nitrogen-fixing plants present and no cyanobacteria visible. Since there are no surface water inflows or outflows to the wetland and there is little to no groundwater connection, it is unlikely that nutrients entered through hydrologic pathways. Atmospheric deposition could have also contributed nitrogen to the system (Rotz et al., 2005), but this was not measured, and it would have been equal across grazed and ungrazed plots.

Overall the nutrient pool analysis suggests that heavy grazing shifts nutrients between ecosystem compartments, ultimately making them more available to biota and decreasing the fertility of the system. Grazing has the potential to reduce the fertility of a *Phragmites*-dominated wetland through removal of nutrients as goat biomass and loss of nitrogen through denitrification and volatilization. Future studies should measure nutrient pools and fluxes from the litter layer and belowground biomass in order to assess the magnitude of these fluxes.

Recommendations for Phragmites Control in Wetlands

This study showed that grazing by goats can effectively control *Phragmites*. My recommendations for maximizing control efficiency are:

- 1) Use herds of at least four animals
- 2) Use goats with previous experience eating *Phragmites*, or with experience of the location, or both, if possible. If this is not possible, the goats should be introduced to eating *Phragmites* before they are moved onto the *Phragmites* patch, and/or animals without prior experience eating *Phragmites* should be pastured on the patch with animals that are experienced so they can quickly learn from each other.

- 3) Repeated grazing will be required over at least two growing seasons before reductions in *Phragmites* biomass and increases in plant diversity are measureable. It is likely that grazing will need to continue for an extended period of time in order to continue to suppress *Phragmites*. Although Vulink et al. (2000) found that it took only four years of grazing to replace a *Phragmites* community, Bassett (1980) and Burnside (2007) have shown that in areas where historic grazing ceases *Phragmites* re-invades. A wetland with high nutrient levels will remain favorable habitat for *Phragmites*. Grazing over many years may reduce nutrient levels enough to make the habitat no longer favorable for *Phragmites*, but actions in the wetland's watershed could also have a major impact on nutrient levels.
- 4) Grazing in June and July will probably weaken the clone most efficiently (Karunaratne et al., 2004) and maximize nutrient uptake by biota in order to minimize nutrient export from the wetland. Grazing in the fall increases the risk of nutrient export from the system because plants do not have enough time to assimilate the nutrients (Stout et al., 1997).
- 5) Extensive grazing (grazing at a low stocking density) should be employed after a year or two of intensive grazing. Extensive grazing has been shown to maximize biodiversity benefits while effectively controlling invasive clonal species.

Limitations of Grazing as a Management Tool to control *Phragmites*

It is likely that grazing could only control rather than completely eradicate *Phragmites*, and that cessation of grazing will cause *Phragmites* to re-invade (Bassett, 1980; Burnside, 2007). I would expect this to be especially true in areas such as our study site

where grazed areas are adjacent to ungrazed *Phragmites*-dominated areas. In such cases *Phragmites* will almost certainly maintain rhizomes running underneath the grazed area and continue to send up new shoots there each year, and the habitat will probably remain favorable for many years before nutrient levels could be depleted.

In my study grazing caused no immediate environmental damage. However, I believe that in certain situations grazing could cause unacceptable environmental damage and should not be used as a management tool. For example, on peat soils grazing animals could cause poaching of the soil, destroying the soil structure. Poaching occurs when livestock, typically at high density, trample wet soils. This reduces soil porosity and damages plant meristems. The result is a muddy slurry rather than a healthy ecosystem. I believe that grazing would probably also be damaging in low marsh, where the plants are already subjected to severe environmental stress via frequent or permanent inundation. Van Wieren et al. (1998), showed that grazing promoted diversity in high salt marshes but not in low or middle salt marshes, where the ratio of species affected negatively to affected positively by grazing was about 1:1.

I would also not recommend grazing in riparian wetlands, where nutrients could be released into the water column by grazers and directly transported to surface waters without a chance to be processed by the wetland. Finally, I would caution against grazing in recharge wetlands where nutrients released by grazers could be leached to the water table. I suggest that further study is required to determine the impact of grazing on recharge wetlands.

Before allowing grazers into a wetland I would recommend a survey to ensure that it does not contain rare or threatened plants or other organisms that may be negatively impacted by grazing.

Grazing by large herbivores is probably not a natural disturbance in Mid-Atlantic wetlands of North America (Middleton et al., 2006). Therefore although it can control *Phragmites*, grazing may not be able to return the plant community to its pre-*Phragmites* invasion state. Changes in physio-chemical wetland characteristics that made the wetland susceptible to *Phragmites* invasion in the first place, such as increased nutrient levels, remain altered. Grazing may reduce nutrient levels in the wetland over time, or it may not affect them due to the overwhelming runoff of nutrients into the wetland from the watershed. But I believe that carefully managed grazing would create an ecosystem with higher biodiversity and value for wildlife than a *Phragmites* monoculture. Grazing may therefore be an unnatural disturbance to achieve a biodiversity objective rather than a tool to restore the system to a previous state.

Veterinary Study on Goat Health

In conjunction with our study on the effects of grazing on wetland health, BARC personnel and Dr. Reginald Harrell collaborated on a study of the effects of grazing in wetlands on goat health. Samples of urine, blood, and stool were regularly taken during the study. According to the veterinarian, Dr. Hare, there were no detrimental health effects on the goats of grazing on *Phragmites*. All animals gained weight during the study period. The goats had low levels of magnesium after the 2009 grazing treatment, but these were not low enough to produce any negative health effects.

Chapter 5: Conclusions

Grazing as a Management Tool for Phragmites Control

Grazing can successfully control *Phragmites* within two growing seasons, but grazing effectiveness can vary by an order of magnitude depending on the size of the herd and the animals' familiarity with the food and location.

Grazing as a Management Tool for Increasing Plant Diversity

Heavy grazing, which completely eliminates *Phragmites* in each round of grazing, can increase plant diversity. *Phragmites* should be completely grazed for this strategy to be successful, and it takes at least two rounds of grazing or two growing seasons of grazing for significant effects to be detectable.

Effects of Grazing on Wetland Nutrient Levels

In the short-term grazing is not detrimental to soil or soil water nutrient levels, although it can increase total nutrient levels in the soil water. Further study is required to determine whether these increases would translate into nutrient export from the wetland as well as to determine an appropriate stocking density to minimize increases in total nutrient levels. Longer-term study is needed to determine whether grazing can be recommended in the Chesapeake Bay watershed as an effective means of controlling *Phragmites* and reducing nutrient levels.

Direction for Future Research

Future research should:

- 1) Determine the correct stocking density to maximize *Phragmites* control and native plant diversity while minimizing environmental damage;
- 2) Investigate the long-term effects of a *Phragmites* diet on ruminant health;
- 3) Test the use of other animals and cattle in particular, which European studies have shown are the most effective grazers for control of *Phragmites*;
- 4) Be of longer duration; many other studies of grazing for restoration purposes track ecosystem recovery over periods of years, e.g. Lindborg and Eriksson (2004) and Krahulec et al. (2001) 6 yrs, Andresen et al. (1990)—9 years; an adequate timescale will be one at the end of which the pattern of change in plant diversity and nutrient levels is clear, or these measures are no longer changing with time;
- 5) Study the effects of grazing on different types of wetlands (e.g. recharge, riparian, discharge, peat-based);
- Monitor nutrient levels long-term; there could be a delayed response of nutrient levels to grazing;
- 7) Investigate the effects of grazing on animal communities, such as invertebrates and birds;
- 8) Investigate mechanisms by which grazing increases plant diversity, such as reduction in litter, increased light penetration, and altered soil nutrient availability or spatial patterns;

MEMO

To: Bill Hare

From: Jennifer Brundage, University of Maryland

Subject: Threatened and Endangered Plant Survey USDA Beltsville Agricultural Research

Center

Date: April 29, 2008

A proposal has been made by researchers at the University of Maryland to fence and graze a patch of *Phragmites australis* (Cav. Trin. ex Steud.) growing at the USDA Beltsville Agricultural Research Center. Following concerns raised by the local ecology committee, a survey was conducted to determine whether there are any plant species of concern growing in the proposed project area. Notes were also made on animal species observed, though no specific animal surveys were undertaken.

The survey was conducted on Thursday, April 24, 2008, between 9:30 and 11:30 am. The weather was sunny, with temperatures around 70°F. The survey was conducted by graduate students Jennifer Brundage and Peter Sharpe of the University of Maryland with assistance from Rose Johnson, undergraduate laboratory assistant in UMD's Wetland Ecology Lab. Specimens of species that could not be identified in the field were transported to the lab and identified using stereo zoom dissecting microscope and taxonomic references by Jennifer Brundage and Dr. Andrew Baldwin.

An aerial photo of the survey site is shown in Figure 1 below, and Figures 2 and 3 show photos of the site. Plants observed in all areas that could potentially be fenced and subjected to grazing pressure were noted. This included inside the *Phragmites* patch as well as the grassy north and south margins. The east and west margins of the patch are probably too wet to permit grazing, however species within about 5 feet of the *Phragmites* on theses margins were recorded.

Plants noted in the survey are listed in tables 1 and 2 below. No species of concern were observed. Observed fauna within the *Phragmites* patch were red-winged blackbirds (*Agelaius phoeniceus*) and green frogs (*Rana clamitans melanota*). Most of the plant species observed were growing in the margins around the *Phragmites* patch. Very few plants (besides *Phragmites*) were growing inside the patch, apart from a few bald cypress

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(*Taxodium distichum*) which had apparently been planted prior to *Phragmites* invasion, a few maple saplings, and some poison ivy.

It is of note that water levels in and around the *Phragmites* patch were several inches lower on this visit than on two previous visits during the winter. It is possible that water levels will continue to drop throughout the summer, making the area increasingly suitable for small ruminants.

Overall no species of concern were observed, and it is not anticipated that fencing and grazing of this *Phragmites* patch would have any negative impacts on the local ecology.

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Contributors

Jennifer Brundage

Professional Experience: 4 years

Education: B.S., Environmental Biology

M.S., Candidate MEES Program

Peter J. Sharpe

Professional Wetland Scientist, PWS No. 1404

Professional Experience: 11 years

Education: B.S., Biology

M.S., Environmental Pollution Control

Ph.D. Candidate MEES Program

Dr. Andrew Baldwin

Associate Professor and Director of the Wetland Ecology and Engineering Laboratory University of Maryland

Plant List

Table 1: Herbs, Shrubs and Vines

Species and Naming	species and Naming Common Name		Rare/
Authority			Threatened
Apocynum sp. (L.)	Dogbane	v.	No
Asclepias sp. (L.)	Milkweed	v.	No
Aster puniceus (L.)	Purple-stemmed aster	V.	No
Carex vulpinoidea (Michx.)	Fox sedge	v. fl.	No
Cirsium arvense (L. Scop.)	Canada thistle	V.	No
Coronilla varia (L. Lassen)	Crown vetch	v. fl.	No
Festuca sp.(L.)	n/a	v. fl.	No
Galium sp. (L.)	Bedstraw	v.	No
Juncus effusus (L.)	Soft rush	v.	No
Lamium purpureum (L.)	Red dead-nettle	v. fl.	No
Lolium perenne (L.)	Ryegrass	v. fl.	No
Lonicera japonica (Thunb.)	Japanese honeysuckle	v.	No
Lonicera tartarica (L.)	Tartarian	v.	No
	honeysuckle		
Lychnis alba (Mill.)	White campion	v. fl.	No
Oxalis sp. (L.)	Wood sorrell	v.	No
Parthenocissus quinquefolia	Virginia creeper	v.	No
(L. Planch.)			
Phragmites australis (Cav.	Phragmites	v.	No
Trin. ex Steud.)			
Plantago sp. (L.)	Plantain	v.	No
Potentilla norvegica (L.)	Rough cinquefoil	v. fl.	No
Ranunculus recurvatus (Poir.)	Hooked crowfoot	v. fl.	No
Rhus radicans (L.)	Poison ivy	v.	No
Rosa multiflora (Thunb.)	Multiflora rose	v.	No
Rumex crispus (L.)	Curly dock	v.	No
Sedum sp (Michx.)	Wild live-forever	v. fl.	No
Stellaria sp.	Chickweed	v.	No
Taraxacum officionale (F.H.	Dandelion	v. fl.	No
Wigg.)			
Typha latifolia (L.)	Broad-leaved cattail	v.	No
Veronica persica (Poir.)	Persian speedwell	v. fl.	No

Table 2: Trees

Species	Common Name	Rare/Threatened
Acer negundo_(L.)	Boxelder	No
Acer rubrum (L.)	Red maple	No
Acer saccharinum (L.)	Silver maple	No
Celtis occidentalis (Hackberry	No
Cephalanthus occidentalis	Buttonbush	No
(L.)		
Fraxinus pennsylvanica	Green ash	No
(Marsh.)		
Liquidambar styraciflua	Sycamore	No
(L.)		
Morus rubra (L.)	Red mulberry	No
Prunus sp.(L.)	Various	No
Quercus alba (L.)	White oak	No
Taxodium distichum (L.)	Bald cypress	No
Rich.		

v. = vegetative

Contact Information

The University of Maryland 1423 Animal Sciences Building College Park, MD 20742-2315

Jennifer Brundage Cell: 703-582-8931

Dr. Andrew Baldwin Office: 301-405-7855

fl. = flowering

v. fl. = vegetative and flowering

Appendix 2: Methods of Soil Water Analysis

Analyte	Lachat	EPA .	Summary of Method
	standard procedure	Equivalent Method	
Ammonia	10-107-06-2	350.1	"The sample is buffered at a pH of 9.5 with a borate buffer in order to decrease hydrolysis of cyanates and organic nitrogen compounds, and is distilled into a solution of boric acid. Alkaline phenol and hypochlorite react with ammonia to form indophenol blue that is proportional to the ammonia concentration. The blue color formed is intensified with sodium nitroprusside and measured colorimetrically."*
Nitrate	10-107-04-1	353.2	"Nitrate is quantitatively reduced to nitrite by passage of the sample through a copperized cadmium column. The nitrite (reduced nitrate plus original nitrite) is then determined by diazotizing with sulfanilamide followed by coupling with N-(1-naphthyl)ethylenediamine dihydrochloride."**
Total nitrogen	10-107-04-1	353.2	Persulfate digestion, then as for nitrate above
Ortho- phosphate	10-115-01-1	365.1	"ammonium molybdate and antimony potassium tartrate react in an acid medium with dilute solutions of phosphorus to form an antimony-phospho-molybdate complex. This complex is reduced to an intensely blue-colored complex by ascorbic acid. The color is proportional to the phosphorus concentration."***
Total phosphorus	10-115-01-4 I	365.3	Persulfate digestion, then same as for orthophosphate above
Plant tissue digestion	n/a	3050b	A representative 1-2 gram (wet weight) or 1 gram (dry weight) sample is digested with repeated additions of nitric acid (HNO ₃) and hydrogen peroxide (H ₂ O ₂). The resultant digestate is reduced in volume while heating and then diluted to a final volume of 100 mL.

*O'Dell, J.W., 1993. Method 350.1: The determination of ammonia nitrogen by semi-automated colorimetry. Available at http://www.epa.gov/waterscience/methods/method/files/350_1.pdf Accessed 02/11/10

**Wendt, Karin, 2000. QuikChem Method 10-107-04-1-A: DETERMINATION OF NITRATE/NITRITE IN SURFACE AND WASTEWATERS BY FLOW INJECTION ANALYSIS (LOW FLOW METHOD). Available at http://swroc.cfans.umn.edu/soilandwater/lab/sop/nitrate_water.pdf Accessed 02/11/10

*** Extract from manual:

U. S. Environmental Protection Agency. 1983.

Phosphorus, All Forms. Method 365.1 (Colorimetric, Automated, Ascorbic Acid). pp.365-1.1 -- 365-1.7. In *Methods for Chemical Analysis of Water and Wastes*, EPA-600/4-79-020. U.S.E.P.A., Cincinnati, Ohio, USA.

Available at: http://www.uga.edu/sisbl/epa-po4.html Accessed 02/11/10

Appendix 3: Sample SAS Code

6

7

8

3

4

4

20.25

24.75 3.43

26

3.32

3.715 U

U

G

```
title 'Jennifer Brundage';
title2 'Goat veg data repeated measures ANOVA grazed v ungrazed August 10, 2009';
DM 'log; clear; out; clear;';
options ls=75 ps=60 pageno=1;
data veg;
input plot block count height trt$ date;
datalines;
                     3.28
              41.5
1
       1
                           U
                                   20081015
2
       1
              35.5
                     3.32
                           G
                                   20081015
3
      2
              32.5
                     3.355 U
                                   20081015
4
      2
              10.25 1.16
                           G
                                   20081015
      3
5
                    0.925 G
              5
                                   20081015
6
       3
              20.25 3.32
                           U
                                   20081015
7
      4
              26
                     3.715 U
                                   20081015
8
      4
              24.75 3.43
                           G
                                   20081015
       1
1
              45
                     3.41
                           U
                                   20090701
2
       1
              35.5
                     2.29
                           G
                                   20090701
3
      2
              48.25 3.5
                           U
                                   20090701
4
      2
              16
                    0.91
                           G
                                   20090701
5
      3
              13
                    0.765 G
                                   20090701
6
      3
              36
                     3.7
                           U
                                   20090701
      4
7
              33.5
                     3.87
                           U
                                   20090701
8
      4
              9.5
                     1.6933 G
                                   20090701
1
       1
              20.75 0.795 U
                                   20090501
2
       1
              53.25 0.99
                                   20090501
                           G
3
      2
              42.75 1.14
                           U
                                   20090501
      2
4
              48.5
                     1.28
                           G
                                   20090501
5
      3
              44.25 1.28
                           G
                                   20090501
6
      3
              37.25 1.37
                           U
                                   20090501
7
      4
              28.25 1.465
                           U
                                   20090501
8
      4
              24
                     1.39
                           G
                                   20090501
data october08;
input plot block count height trt$ date;
datalines;
1
       1
              41.5
                     3.28
                           U
                                   20081015
2
       1
                     3.32
                           G
              35.5
                                   20081015
3
      2
              32.5
                     3.355 U
                                   20081015
4
      2
              10.25 1.16
                           G
                                   20081015
5
       3
              5
                    0.925 G
                                   20081015
```

20081015

20081015

20081015

```
proc sort data=veg;
by plot date;
run;
proc print data=veg;
run;
proc mixed data=veg;
class plot block trt date;
model height=trtldate /ddfm=kr outp=resids;
random block;
repeated date / subject=plot(block trt) type=CS;
lsmeans trt date trt*date/ adjust=tukey;
run;
proc plot data=resids vpercent=50;
plot resid*pred/vref=0;
plot pred*trt;
plot resid*trt;
run;
Proc univariate data=resids plot normal;
var resid:
run;
proc mixed data=veg;
class plot block trt date;
model count=trtldate /ddfm=kr outp=resids;
random block;
repeated date / subject=plot(block trt) type=CS;
lsmeans trt date trt*date/ adjust=tukey;
run;
quit;
/*Tested different covariance structures for each repeated measures ANOVA and chose the
one with the best fit statistics balanced against having not too many covariance parameter
estimates, which decreases power*/
proc plot data=resids vpercent=50;
plot resid*pred/vref=0;
plot pred*trt;
plot resid*trt;
run:
Proc univariate data=resids plot normal;
var resid;
run;
```

```
/*significant interaction between trt*time for both counts and heights, so used a straight ANOVA grazed v. ungrazed at sampling date*/
```

```
proc mixed data=october08;
class block trt;
model count=trt /ddfm=kr outp=resids;
random block;
lsmeans trt;
run;
proc plot data=resids vpercent=50;
plot resid*pred/vref=0;
plot pred*trt;
plot resid*trt;
run;
```

Proc univariate data=resids plot normal; var resid; run;

Appendix 4: List of Plant Species Encountered

Scientific Name	Common Name
Acer rubrum	red maple
Alisma plantago-aquatica	common water-plantain
	American water
Alisma subcordada	plantain
Ascelipias incarnata	swamp milkweed
Aster sp.	
Aster vimineus	small white aster
Baptisia tinctoria	wild indigo
Bidens sp.	beggarticks
Bohemeria cylindrica	false nettle
Carex sp.	sedge sp.
Cephalanthus occidentalis	buttonbush
Cirsium sp.	thislte
Cyperaceae sp.	
Cyperus odoratus	fragrant flatsedge
Eleocharis sp.	spikerush
Epilobium coloratum	purpleleaf willowherb
Epilobium glandulosum	fringed willowherb
Eupatorium sp.	boneset
Fragaria indica	indian strawberry
Fraxinus pennsylvanica	green ash
Galium trifidum	three-petal bedstraw
Gallium sp.	bedstraw
Glechoma sp.	ground ivy
Juncus effusus	soft rush
Lamium purpureum	red deadnettle
Leersia oryzoides	rice cut-grass
Liquidambar styraciflua	sweetgum
Lonicera japonica	Japanese honeysuckle
Lonicera tatarica	bush honeysuckle
Ludwigia palustris	marsh seedbox
Mikania scandens	climbing hempvine
Mimulus ringens	monkeyflower
Morus sp.	mulberry
Murdannia keisek	wartremoving herb
Nastertium microphyllum	onerow yellowcress
Parthenocissus quinquefolia	Virginia creeper
Penthorom sp.	stonecrop
Phragmites australis	common reed
Phytolacca americana	American pokeweed
Pilea pumila	clearweed
Polygonum hydropiperoides	swamp smartweed
Polygonum perfoliatum	mile-a-minute weed
Polygonum punctatum	dotted smartweed

Polygonum persicaria	lady's thumb
Quercus sp.	oak
Ranunculus sceleratus	cursed buttercup
Ranunculus sp.	buttercup
Rorippa islandica	marsh yellow cress
Rosa multiflora	multiflora rose
Rumex sp.	dock
	American black
Sambucus canadensis	elderberry
Solanum chacoense	chaco potato
Sonchus sp.	sow thistle
Tax officionalis	dandelion
Taxodium distichum	bald cypress
Toxicodendron radicans	poison ivy
Typha latifolia	broad-leaved cattail
Verbena hastata	blue vervain
Viburnum dentatum a.k.a.	
recognitum	southern arrowwood
Viburnum sp.	Viburnum
Vitis sp.	grape
Waldsteinia fragarioides	barren strawberry

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