NUTRIENT REMOVAL IN NATURAL SWIMMING POOLS
A MASS BALANCE ANALYSIS

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by
Margaret C Hoffman

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The dissertation of Margaret C Hoffman was reviewed and approved* by the following:

Robert D Berghage  
Associate Professor of Horticulture  
Dissertation Advisor  
Chair of Committee

Dan T Stearns  
J Franklin Styers Professor

Rick S Bates  
Associate Professor of Ornamental Horticulture

Robert Shannon  
Associate Professor of Agricultural and Biological Engineering; Coordinator,  
Environmental Resource Management Program

Rick Marini  
Head of the Department of Plant Science  
Professor of Horticulture

*Signatures are on file in the Graduate School
ABSTRACT

Natural swimming pools, (NSPs) are constructed bodies of water, with an impermeable liner between the soil and water, a designed, intentional hydraulic and skimmer system and a complex ecological community as a filter. This technology relies on biological filtration, the interaction of a balanced system including bog vegetation, bacteria and substrate, to reduce nitrogen and phosphorus levels in the NSPs water to less than 30 mg/l NO₃ and 0.01 mg/l P.

Water from three NSPs in central PA were sampled through a 5 month period and inputs and outputs estimated. Aboveground plant biomass of bog vegetation was harvested and analyzed for N and P concentrations. Results indicated the need for additional research conducted in a controlled setting to improve estimation of inputs and output compartments and allow measurement of the substrate compartment. None of the NSPs sampled could explain all nutrient removal through direct uptake and storage by biomass.

Subsequently, research was conducted estimating phosphorus and nitrogen inputs, outputs and storage in two greenhouse studies. Outputs consisted of nutrients contained in harvested biomass while nutrient storage compartments were defined as plant biomass and substrate. Plant direct uptake proved insufficient to remove all nutrient inputs, and substrate type was significant in nutrient storage.

In an experiment testing four substrates planted with Saururus cernuus and Iris versicolor, haydite/clay was the only substrate able to lower phosphorus levels of the mesocosm water to 0.01mg/l. All substrate treatments reduced mesocosm water NO₃ to target levels of 30 mg/l. Iris produced significantly higher biomass than the lizards tail, and biomass was positively correlated with NO₃ removal.

A subsequent experiment testing the removal capabilities of treatments consisting of Canna x generalis and Iris versicolor planted in a substrate consisting of calcined clay and
activated alumina, showed vegetated treatments removed more NO$_3$ and P than the unvegetated control. *Canna x generalis* produced higher biomass and removed more NO$_3$ and P than *Iris versicolor*. Again, biomass production and NO$_3$ removal were highly correlated. The substrate of calcined clay/activated alumina proved very effective at removing P from the mesocosm water, PO$_4$ levels were below 0.01 mg/l when measured using the Murphy-Riley method for both the *Canna x generalis, Iris versicolor* and unvegetated treatments.
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Chapter 1

Introduction

Water is increasingly recognized as an at risk resource. Water quantity and quality are high priorities in most communities, and as a society we recognize this priority with regulations and standards designed to protect drinking water and waterways. The development of Best Management Practices (BMP) and Low Impact Development (LID) practices are strategies used to mimic original hydrology and reduce stormwater volume and pollutant concentration entering receiving waters. Existing technologies, such as constructed wetlands and bioretention, reduce human impact on water resources, while minimizing the use of chemicals and energy resources. Natural swimming pools are a technology developed in the 1970’s in Europe as a response to increasing concerns over loss of wetland habitat, ecological niches and the health effects of chemical filtration in swimming pools. Natural swimming pools employ the same technologies as constructed wetlands and bioretention facilities mimicking natural wetlands’ physiochemical processes and biocenoses, purifying water without the use of chlorine.

Natural swimming pools, (NSPs) are constructed bodies of water, with an impermeable liner between the soil and water, a designed, intentional hydraulic and skimmer system and a complex ecological community as a filter. NSPs use the established technologies developed and tested for CWs and BTs. NSPs provide an alternative to chemically managed swimming pool systems by providing a complex ecosystem that effectively removes excess nutrients and filters harmful bacteria, establishing not a sterile environment, but a safe, clean swimming landscape amenity where a regeneration area eliminates impurities. Using this complex living system, these
pools adjust to different contaminant levels and inputs, utilizing the living organisms and water chemistry inherent in the system to transform and remove undesirable pollutants.

This ability to filter a swimming pool without the addition of harmful chemicals provides an alternative to chlorine. Among the deleterious effects of chlorine are asthma related issues and irritation of skin and eyes. (Bernard et al. 2009) The elimination of chlorine or other harsh chemicals establishes a NSP as a more sustainable landscape amenity than a chemical pool. In fact, in this time of increasing concern over ecosystem services, NSPs provide a unique approach to filling recreational, ecological and aesthetic niches. Additionally, unlike a traditional swimming pool which provides one service to the user, that of recreation, for a limited period annually, a NSP provides a multitude of services, throughout the year. NSPs increase habitats for pollinators and wildlife, maintain biodiversity, and purify and conserve water while providing recreational and aesthetic opportunities. It is this difference in intent, to provide year round aesthetics and ecological services, which is at the heart of NSP design.

The Sustainable Sites Initiative, published in 2009, outlines a framework for landscape design embracing the philosophy of stewardship. It encourages design professionals to look past traditional philosophies and voluntarily meet national guidelines and performance benchmarks for sustainable landscape design. Recognizing that all landscapes have the potential to contribute ecological services to the community, locally and globally (Sustainable Sites Initiative. 2009), the SSI establishes the following ‘guiding principles’: Precautionary: examine all alternatives and ensure that the design does not create risk.
Design with nature and culture in mind

Preserve, conserve and regenerate

Provide regenerative systems for future generations

Support living processes: use an ecosystem approach

Be ethical

Foster environmental stewardship (Sustainable Sites Initiative. 2009).

How do NSPs fit into the guidelines established by the Sustainable Sites Initiative of 2009? NSPs use biological filtration to clarify and purify the water in the system. Biological filtration combines plants, beneficial bacteria and microorganisms to remove nutrients and harmful materials from the water, enhancing clarity and purification. A water feature that uses chemical filtration cannot meet the criteria of a sustainable site design because it does not meet the “do no harm” criterion nor is it a regenerative technology. Traditional pool technology aims for a sterile environment, not a purified environment. Chemical filtration forces an artificial condition on the water system, and pushes the water towards a biological vacuum while biological filtration supports a great diversity of life and therefore draws other life forms to the water feature. This increased diversity of life in the water coupled with a mixed plant population in the bog filter forms a healthy ecological community. Landscape designers are able to use native wetland plants and materials, such as mountain stone and river gravel, to reflect local culture and ecosystems. Families are able to foster a sense of environmental stewardship, employing the NSP as both an educational tool and outdoor lab, while enjoying the increased invertebrate, vertebrate and vegetative community that the NSP
supports. As societies’ children become increasingly isolated from the natural world, NSPs provide an opportunity to reconnect and renew (Louv 2005).

NSPs are a technology still in infancy. Though first introduced in Austria in the 1970’s, North America is only now becoming aware of the technology and verification of performance requires a body of reviewed research. While constructed wetlands and bioretention technologies have an established body of research quantifying removal processes, none exist for natural swimming pools. This research will provide the landscape industry in North America requisite validation for the NSP system or, alternatively, show that the system’s gravel filtration with emergent macrophytes is not sufficient to meet standards for nitrogen, phosphorus and clarity. My research will develop a mass balance equation that estimates system nutrient inputs, nutrient removal compartments and outputs. This approach quantifies system performance and allows the development of simple predictive models (Breen 1990). Mass balances provide a good measure of nutrient removal compartments because evapotranspiration is considered (Tanner 2005) and this analysis is widely used in constructed wetland nutrient research (Johengren 1993, Breen 1990, Borin and Salvato 2012).

Why is it necessary to conduct research on NSPs if they are similar to constructed wetlands and stormwater biofiltration? As mentioned earlier, constructed wetlands (CWs) are engineered systems designed to closely mimic the hydrology, physiochemical and biological functioning of natural wetlands, that as a complex ecosystem together remove nutrients and pollutants from water before releasing the water to the environment (Kadlec and Wallace 2009; Mitsch and Gosselink 2000). Inputs into CWs are typically domestic, agricultural, industrial and mining wastewaters, and as such may contain high
levels of nitrogen, phosphorus and heavy metals. With a few exceptions, treatment water levels are typically constant. Stormwater bioRetention removes suspended solids, nutrients, metals, hydrocarbons, and bacteria from runoff and reduces flow volumes before they enter receiving waters. Stormwater biofiltration must effectively function through periods of inundation and drought. Natural swimming pools kept in a very low nutrient state, present a different set of challenges for the biological filtration function.

While both CWs and BTs are open systems, discharging water to receiving bodies, NSPs are closed systems. Except for precipitation events, once the system is started only small additions of water occur in the form of fill water. No water is discharged from the system unless water escapes through overflow in a high precipitation storm event.

Neither CW or BTs are considered landscape amenities, except in the case of some LID technologies, i.e. rain gardens and bioswales. As an ecosystem services management tool, aesthetics are an important aspect of NSP function. Many CWs are monocultures, pure stands of *Phragmites spp* or *Typha spp*, neither of which are considered ornamental plants by the landscape industry. An NSP bog is designed as a plant community, not a monoculture, therefore research is needed to determine if ornamental aquatic plants are effective in the planted filter.

Treatment wetlands and BTs are assessed by their ability to meet EPA and municipal discharge standards. If they can meet these standards, the amount of algae in the system is of little concern. However, in a NSP, algae is detrimental to the system in two ways. The algae creates a safety hazard if it interferes with water clarity and visibility. Some guidelines specify a Secchi depth reading of 3 meters for clarity (FLL guidelines). Algae also competes with plants for essential nutrients in the NSPs already
low nutrient environment. The presence of algae inhibits the establishment of emergent macrophytes and healthy populations of aquatic organisms though nutrient competition, stimulating harmful pH swings and changes in dissolved oxygen levels. Beyond these water chemistry factors, the most homeowners equates algae with impurity, which would discourage use of the pool while decreasing its aesthetic value. Control of algae involves reduction of NO₃ and P levels to <30 mg/l and 0.01 mg/l respectively, lower target P levels than for either CWs or BTs.

The process of adsorption is one of the important P removal mechanisms in wetlands. While both constructed wetland and bioretention technologies target P for removal, neither is completely effective. Substrate chemical composition and water pH determines substrate ability to adsorb P. More research is needed to determine if gravel substrate is able to reduce P to desired NSP levels, or if gravel substrate should be enhanced by the addition of P adsorbing materials or precipitants.

In conclusion, this research will provide designers, pool owners, and public officials with the knowledge they need to design, build, operate and evaluate a natural swimming pool. Development of a mass balance model allows the formation and adoption of standards for North American NSPs for the Green industry, establishing the legitimacy of NSP technology.

Goals

The goals of this research are:

1. Measure inputs, outputs and storage of nitrogen and phosphorus in plant tissue, substrate and the NSP system water.
2. Comparison of systems with ornamental bog plants, *Iris versicolor*, *Canna x generalis*, *Sarurus cernuus* for nutrient removal of N and P.

3. Assessment of the ability of different substrates to remove and store N and P from the swimming pool water

4. Explore possible media alternatives and additives by quantifying phosphorus removal from the swimming pool water for each substrate treatment.

5. Analyze the contribution of above ground plant harvesting to nutrient removal.

The following section will discuss current level of technology and effectiveness of biofilters in constructed wetlands (CWs) and bioinfiltration technologies (BTs). The two main components of a biofilter, media and macrophytes, will be discussed in detail and related to natural swimming pools. Issues specific to these technologies and NSPs, are the reduction of N and P and control of algal populations. Following the literature review, I will present methods, analysis and discussion of studies involving macrophytes and various substrates, planted in greenhouse mesocosms, and tested for N and P removal abilities. These studies will contain mass balance calculations and removal efficiencies as well as relationship between macrophytes tested and removal efficiencies, biomass and substrate, and substrate and removal efficiencies.
Chapter 2

Literature Review

In the following chapter, I review and discuss two types of technologies, constructed wetlands and stormwater bioretention, which utilize biological filtration to remove nutrients and other pollutants from water. Constructed wetlands and stormwater bioretention have different removal parameters and their designs vary accordingly, but both utilize the same basic nutrient removal processes. Specifically, I summarize current research concerning the effect macrophytes and media have on N and P removal and the processes involved. Research has identified several key contributions of media and macrophytes in biological filtration and these contributions can be applied to natural swimming pools. I then discuss the design of a natural swimming pool’s biological filtration, comparing and contrasting the similarities and differences between the constructed wetlands, stormwater bioretention and natural swimming pools. These processes include volatilization, sorption, precipitation, ammonification, nitrification, denitrification, mineralization and direct uptake.

Nitrogen and phosphorus are the two key nutrients targeted for removal in constructed wetlands, bioretention and NSP systems. Excess nitrogen and phosphorus cause eutrophication if they are above recommended levels in aquatic systems. Eutrophication occurs when high levels of N and P are introduced to an aquatic ecosystem, usually through human activity, and stimulate excess plant growth. A few sources of eutrophication are agricultural and fertilizer runoff and leaching, sewage, wastewater and industrial waste. This enrichment of the system leads to anoxia (no oxygen), hypoxia (low oxygen), and moves the system towards plant monocultures such
as algae and duckweed. Phosphorus is the major limiting nutrient for algae production in freshwater systems, although nitrogen is also strongly related to primary production (Correll 1999, Currie and Kalff 1984, Powers et al 1972). P is the nutrient most associated with eutrophication and occurs as a mix of particulates and dissolved P, usually orthophosphates (PO$_4^{3-}$) (Reed et al 1988, Correll 1999). The association of P and N with eutrophication is why CWs and BTs target N and P contained in influent before release into receiving waters.

All systems with biological filtration use chemical, physical and biological processes to remove nutrients and pollutants from water. Chemical processes of particular interest include the volatilization of ammonia (NH$_3$), and the precipitation and sorption of P. Physical processes include mechanical filtration of large pieces of organic matter and sediment, usually achieved with a skimmer and/or filter. Ammonification, nitrification and denitrification are biological processes that remove nitrogen. Other biological processes include microbial uptake and plant uptake which have significance for both N and P removal, although are of greater importance for N removal.

**Chemical processes**

Volatilization of ammonia (NH$_3$) occurs when ammonia (aqueous) is transforms to ammonia (gas) and releases into the atmosphere. This transformation is pH dependent, requiring pH above 8 for significant volatilization. In wetlands, pH can be driven above 9 when photosynthesis from algal population blooms is high. Volatilization only occurs with nitrogen, while precipitation and sorption are significant removal processes for phosphorus. Phosphorus precipitates when calcium or iron is present in the soil substrate.
and PO$_4$ is relatively high. Sorption, is a physical-chemical reaction where a compound in solution is attracted to the surface of a solid. If the compound concentrates on the solid it adsorbs and if it concentrates in the surface it is absorbed. Organic nitrogen and phosphorus can both be sorbed. Sorption is also dependent on contact time of the solution with the solid, called retention time, sorption increasing with both concentration and contact time. Solution pH, cation exchange capacity and organic matter have a positive correlation with P sorption while pH has a negative correlation with P sorption (Vymazal 2011). As a result, media with high Al, Ca and Fe have high sorption potential (Richardson 1985). In constructed wetlands media selection, any chemical additions, external filters and manipulation of pH influence the chemical processes, flow rates and retention times (Kadlec and Wallace 2009).

**Biological Processes**

**Ammonification and mineralization**

Ammonification is the process which transforms organic nitrogen to NH$_3$. When NH$_3$ becomes ionized it forms ammonium, NH$_4^+$. The conversion of organic nitrogen to inorganic N is also called mineralization. This process is important because ammonium in the soil is one of the nitrogen forms available to plants. The optimum pH for ammonification is 6.5 - 8.5 and the reaction improves with the correct C/N ratio, nutrients and temperature.

**Nitrification and denitrification**

Understanding the nitrogen cycle is crucial in the control of nitrogen in natural
and constructed wetlands. The nitrogen cycle exists in every body of water and is responsible for converting ammonia to nitrates and other less harmful nitrogen compounds. Some ammonia can be removed via plant root uptake but the majority needs to be converted. This is a living cycle, (Figure 2-1) dependent on bacteria to transform ammonia to nitrites and nitrites to nitrates. This process makes nitrogen available to plant roots. The nitrogen cycle needs both heterotrophic and autotrophic bacteria to function efficiently. Heterotrophic bacteria derive energy from outside sources, breaking down complex organic compounds, and act as sludge degraders. They function with or without oxygen and prefer to obtain nitrogen from organic sources. Autotrophic bacteria use nitrogen from inorganic sources, namely ammonia and nitrites. These bacteria are aerobic and fix inorganic carbon to make their own food sources. Two of the most important of these autotrophic bacteria are the Nitrosomonas and the Nitrobacters. Nitrosomonas convert ammonia to nitrites and nitrobacters convert nitrites to nitrates. Nitrates and ammonium are the forms of nitrogen available to plant roots. Autotrophic bacteria are slow growing and double in population every 15-24 hours, while the heterotrophs will double in population in as little as 15 minutes to one hour. Denitrification occurs when facultative, heterotrophic bacteria use nitrates as an alternative to oxygen in their respiration and reduce nitrates to nitrogen gas, releasing the nitrogen gas to the atmosphere.
Figure 2-1: The nitrification cycle (Hilleary 2006)

Available oxygen and carbon are important drivers of the nitrogen cycle. Oxygen is necessary for the first part of the cycle, nitrification, while the second part, denitrification, proceed when conditions are anaerobic. The process of denitrification requires carbon as an energy source for denitrifying bacteria. Kadlec and Knight calculate that 2.5 grams of carbon is needed to convert 1 g of N. Carbon also encourages respiration of aerobic heterotrophs, a process that consumes O$_2$ thereby encouraging the anaerobic conditions needed by denitrifying bacteria.

The following discussion focuses on media and direct uptake of nutrients by
plants, while discussing other key contributions of plants to the system. The processes just discussed are key to all nutrient removal processes in the sections that follow.

**Plant uptake**

N and P are essential nutrients for life. N is present in amino acids, proteins, DNA, RNA and chlorophyll and is 1 to 6% of plants dry weight. P is a component of DNA and RNA, and provides the cell energy in the form of ATP and NADPH for plant metabolism (photosynthesis and respiration) and comprises .02% to 1% of most plants dry weight. Plants preferentially uptake inorganic N, in the form of NO$_3^-$ and NH$_4^+$; and inorganic P, in the form of PO$_4^{3-}$. (McClain and Trisca 1998) Accumulation of N and P in plant tissues is species dependent, and many obligate wetland plants store more P than upland plants. (Dunne et al 2005, Richardson et al 1978, Richardson and Marshall, 1986) Plant direct uptake can range from .05-1.8 % for P and 1-5.8 % for N of total biomass (Greenway 2004, Corbridge 2000). Brix (1997) found that some aquatic macrophytes are capable of nitrogen uptake rates as high as 685 mg N m$^{-2}$d$^{-1}$. In general, plants can only take up a given amount of nutrient (Vymazal, 2005) except in the case of luxury consumption. Luxury consumption occurs when more nutrients are available than optimum and a plant’s uptake is in excess of their needs. The amount of nutrients removed in a constructed wetland attributable to plant uptake depends largely on loading rate (nutrients available). In wetlands with lower loading rates, plants often represent a larger portion of the total nutrient removal amount (Kadlec and Wallace 2009). Other factors such as climate, plant growth cycles and site characteristics all play a role in plant uptake and storage of N and P (Vymazal 2005, Tanner 2001).
As well as contributing to nutrient removal through direct uptake and storage of nutrients in biomass, plants moderate pH and oxygen exchange in the aquatic environment, while providing increased surface area for microbial populations (Reddy and DeBusk, 1987, Brix 1997, Iamchaturapatr and Rhee 2007, Tanner 2001, Kadlec and Wallace 2009, Vymazal 2007). Richardson (1985) and Braskerud (2002) found that most P uptake is in early spring during the period of rapid expansion of plant tissues, and is stored in above ground tissues through the summer till it is translocated to the rhizomes for winter storage. Above ground tissues lose 30% of the nutrients to decomposition within a couple of days of senescence. Death and decomposition of aboveground tissues release P and N to the water column, while decomposition of belowground parts release nutrients to the soil (Vymazal, 2007). In the case of P decomposition, 35-75% of the P in plant tissues is released into the nutrient pool. (Richardson 1985)

Macrophytes compete with algae and bacteria for available N and P in biological filters. The rate of uptake by microorganisms is considerably faster than plants allowing bacteria to outcompete plants initially for nutrients (Vymazal 1994). However, in systems with retention times measured in days not hours, the bacteria go through several cycles of nutrient acquisition and release as cells are disrupted and die. As a result they are not part of the longterm storage of nutrients for a wetland. Over time, plant roots are able to outcompete bacteria and acquire the released N and P.

Plants do not always increase the removal of nutrients, especially P (Iamchaturapatr 2007), but still play an important role as what Tanner (2001) calls ‘ecosystem engineers’. Plants influence wetlands when they entrap sediments and reduce turbulence while roots provide an increase in surface area for microbial growth and
oxygenate the rhizosphere. Dissolved oxygen (DO) levels in the wetland water column fluctuate with bacterial oxidation of NH$_4$ to NO$_3$ and the decay of organic matter. If DO is not high enough to meet the biological demands of the system, aquatic health declines. Plants contribute to N removal by increasing microbial populations for denitrification and microbial transformation (Gersberg et al. 1986). Aerenchyma in aquatic plant tissues transfers atmospheric oxygen to the rhizosphere providing additional oxygen for both nitrification and reducing transformations (Kadlec and Wallace 2009, Mitsch and Gosselink 2000).

The decomposition process of plant vegetative matter supplies carbon to fuel the denitrification process and buffers nutrient release (Tanner, 2001; Kadlec and Knight, 1996, Mitsch and Gosselink, 2000). There is evidence that denitrification rates are 6-9 times higher around the root zones of aquatic plants than in unplanted flooded soil (Reddy et al. 2009). Plants also create new stable residuals through increased organic matter deposition, which accrete in the wetland providing long-term storage of phosphorus (Richardson 1985, Nguyen 2000). Having established the contributions of media and vegetation to biological filtration, the next discussion focuses on research quantifying these processes in constructed wetlands and bioretention technologies.

**Media**

Media in both CWs and BTs have traditionally been coarse (2-20mm) river gravel or sand, (Kadlec and Wallace 2009) and serve several purposes. Media acts as a physical support for plant roots, physically traps particulates and sediments, provides increased
surface area for bacterial populations, a surface for adsorption and absorption and helps
distribute water to all wetland areas for treatment (USEPA 2000).

The significance of both vegetation and media in biological filtration has been
established, and leads this discussion to their specific roles in constructed wetlands and
bioretention technologies. Constructed wetlands and bioretention technologies will be
defined and the research involving vegetation and media for each summarized in the next
sections.

**Constructed wetlands and bioretention technologies.**

**Constructed wetlands**

Constructed wetlands (CWs) are engineered systems designed to closely mimic
the hydrology, physiochemical and biological functioning of natural wetlands that as a
complex ecosystem together remove nutrients and pollutants from water before releasing
the water to the environment (Kadlec and Wallace 2009, Mitsch and Gosselink 2000).

**Constructed wetlands have:**

- A liner or impermeable basin to control water exchange between the constructed
  wetland other water sources
- Substrate for plants and sorption processes
- Plants for direct uptake and support of microbially mediated processes
- A designed nutrient loading rate and hydraulic retention time to manage nutrient
  removal and storage.
There are two types of CWs discussed in this section, free water surface (FWS) wetlands and subsurface flow (SSF) wetlands.

**FWS wetlands**

FWS wetlands are characterized by water level of varying depths over the wetland substrate. (Figure 2-2) Typically, FWS substrate is coarse river gravel, providing good hydraulic conductivity and some physical support for emergent plants. Because FWS wetlands have open water they are able to support communities of free floating plants (water hyacinth), submerged plants (potamegeton) and rooted emergent macrophytes (cattails), or some combination of all three, and have the appearance of natural marshes. Usually a FWS wetland only contains one wetland macrophyte community type, but could be designed to support different communities, thereby increasing ecological services. Ancillary benefits of FWS CWs include increased habitat for invertebrates and other wildlife, ecological diversity and some recreational opportunities (bird watching etc) for the community (Knight et al. 2001). FWS wetlands are often utilized for stormwater treatment because the system is able to adjust with changing water levels and the operating costs and technological expertise required are low. FWS wetlands are typically a very sustainable technology because of low operational energy requirements (Kadlec and Wallace 2009).

In these wetlands, plant uptake, subsequent harvesting and soil accretion are the major mechanisms for removing TP, while nitrification-denitrification, volatilization and plant uptake remove N (Vymazal 2007). P&N are stored in plant tissue and P adsorbed onto media, both saturable processes, and once these compartments are saturated,
nutrients are released back into the system. Soil accretion is the only phosphorus storage compartment that is not saturable (Tanner et al 1999, Richardson 1985) as wetlands continue to accrete new sediments throughout its lifetime. FWS wetlands have high TSS, BOD, NH$_4$, TP, and TN removal efficiencies (Kadlec and Wallace 2009).

![Figure 2-2: Free water surface wetlands](image)

Because FWS wetlands have open water areas, algae can often become a nuisance organism. The presence of algae indicates that P and N exist at higher than optimum levels (Cronk and Fennessy 2001). When algae are a problem, cyclical periods of population bloom and crash occur. A subsequent decrease in dissolved oxygen levels
from algal decomposition adversely affects wetland organisms. High algal populations cause pH swings and changes in water chemistry. As photosynthesis occurs, CO$_2$ is removed from the water, increasing the pH to as much as 9.5 by increasing the level of hydroxides. Respiration in the evening produces CO$_2$, lowering hydroxides and decreasing pH. The large swings in pH are harmful to aquatic organisms, including daphnia, bacteria, macrophytes, invertebrates, fish, and affect plant nutrition (Mitsch and Gosselink 2007). The changes in pH also catalyze changes in the form of N available, increasing the amount of NH$_3$. Plants are not able to utilize NH$_3$ at the same levels they preferentially uptake NH$_4$ (Vymazal 1995). Aside from the aquatic system health issues, large algal mats can cause undesirable odors, water discoloration and in the case of a natural swimming pool, become a safety hazard, reducing clarity. This has the consequence of reducing a NSPs utility as a backyard amenity.

In North America, another plant that can be a problem in a FWS wetland is duckweed, a small plant capable of reproducing quickly. Similar to algae, it often occupies open water areas in eutrophic conditions. While there are biofilters designed to exploit the ability of duckweed and algae to rapidly reproduce and remove nutrients through direct uptake and subsequent harvest (Zimmo 2004, Kadlec 2005), this paper will not examine these systems.

**Subsurface flow wetlands**

Subsurface flow (SSF) (Figure 2-3) constructed wetlands are often employed for treating effluent from single households and small communities, or for use in a treatment train where sediments and solids are removed before entering the SSF wetland.
SSFs provide medium to high efficiency treatments for tertiary BOD, TSS, COD, NO3 and pathogen reduction (Kadlec and Wallace 2009). SSF wetlands have a bed depth close to .6 m and the water surface is below the top of the substrate surface. The substrate in a SSF is most often gravel or local sands which serve as support for macrophytes and increase the surface area available for microbes and bacteria. Plants are established in a layer of soil or peat at the top of the treatment media. When SSF wetlands use a substrate that contains Ca, Fe or Al, they have major potential for P removal. The most appropriate media for a SSF constructed wetland is a media that does not raise the pH of the wetland, allows for good hydraulic conductivity, can support plant life and is cost effective.

Because the water level stays below the surface of the substrate, SSF wetlands have the added advantage of eliminating algae and mosquito populations and decreasing
interactions with pest animals (Vymazal 2007). Additionally, because the water is below the substrate surface when used as wastewater treatment, there are no objectionable odors or pathogens. SSF constructed wetlands operate under lower flow rates than FWS (Table 2-1), usually higher loadings, and frequently require less space. However, they can have issues with clogging of media, especially at the inlet, and do not provide the ancillary benefits of a FWS (Vymazal 2007). SSF wetlands are capable of operation in colder conditions than FWS wetlands because of the obvious insulation values of the media and vegetation. Because the temperatures are moderated, nitrification and denitrification occur through the winter, even if reduced because of the colder temperatures.

In reality, very little performance effectiveness separates the two types of constructed wetlands, FWS and SSF (Kadlec 2010). Free water surface wetlands have been traditional in the United States, probably to mimic a natural wetland’s wildlife and ecological values. In Europe there exists a general perception that SSF wetlands require a smaller footprint (Solano et al 2004, Kaseva 2004) and are more cost effective for small communities and single homes. Kadlec (2010) states that many of these preconceptions about SSF vs FWS do not hold up under scrutiny with the information now available to researchers and designers. Areal coverage, expense, nutrient removal effectiveness and operational costs are approximately the same, leading to his recommendation that other considerations (for instance ancillary benefits) should drive the decision making when deciding which system is installed (Kadlec 2010). Even the accretion of sediments exists in both types of CW with *Phragmites* leaf litter accounting for 40% of P removal in one study of SSF reed beds (Headley et al 2004) and *Melaleuca* litter a significant contributor to the permanent sediment compartment in an Australian FWS study (Bolton and
Greenway 1997). Since the nutrient removal compartments are basically the same for both types of constructed wetland and bioretention technologies (BTs), I will discuss the effects of vegetation and media on N and P removal after introducing BTs in the next section.

**Stormwater Bioretention systems**

The USEPA identifies urban stormwater runoff as a major nonpoint pollution source (USEPA 2000) and Daniel et al, (1998), has shown agricultural runoff as a major contributor in watershed degradation. Many organizations and regulatory commissions recommend the use of low impact development tools (LIDs) and best management practices (BMPs) to decrease peak flows, runoff volumes and pollution. Excess fertilizer, livestock waste, TSS, hydrocarbons and heavy metals can be reduced through the use of many BMP’s including raingardens, greenroofs, bioswales and detention basins (Carpenter et al 1998, Davis and Shokouhian 2001). All of these technologies utilize vegetation and media and rely on the same principles of nitrogen and phosphorus removal as the previously discussed CW’s. Bioretention technologies (BTs), therefore, utilize planting media with good drainage characteristics, (usually gravel or sandy loam) covered with a mulch layer. They are vegetated with a wide range of plants, to remove sediments, nutrients and decrease the volume and flow rate of stormwater runoff entering receiving waters. Underneath the soil filter a drain is often placed to transport water away from the bioinfiltration area to receiving waters or a reservoir. The primary mechanisms to reduce stormflow are evapotranspiration, infiltration and retention, while reductions of sediments in the stormwater occur through adsorption. Microbially
mediated transformations and plant uptake reduce P and N. Bioretention systems differ from constructed wetlands in the following ways: bioretention systems are designed to minimize pooling time eliminating potential mosquito and safety issues, bioretention require a smaller footprint than constructed wetlands, and bioretention systems require a smaller fraction of the surface of the catchment to achieve the same water quality objectives (Fletcher et al 2003). (Table 2-1) The source, type and quantity of the pollutants; retention time and variable hydrologic environment plants must survive; drive design differences for these technologies. Most BT’s are designed to remove particulate matter and sediments from stormwater runoff, whereas a constructed wetland removes nutrients from wastewater. Heavy metals and nutrients are often associated with sediments although they can also be part of the dissolved fraction and therefore removed through direct uptake and through adsorption (Davis et al 2001, Vaze 2002). Usually, the detention time, 9.3days (FWS), 2.9 (SSF) and 12 hours (BT), and the total loading rate for pollutants are greater in a treatment wetland. In addition, the hydraulic regimen differs profoundly. In a BT, the collected precipitation is detained and allowed to slowly percolate directly into groundwater or is released slowly into lakes, rivers and streams. This means an important design consideration is rate and volume of release into the surrounding ecosystem. Vegetation choices are defined by the hydraulic regime and plants must be able to tolerate inundation, periodic drought and low nutrient loads. In contrast, in a CW, water is introduced in a controlled fashion from a specific source with given nutrient and water input levels while for a bioretention technology (BT), water depth and moisture levels are dependent on precipitation events, temperatures and soil hydrology. Because BT’s often target heavy metals such as copper, lead and zinc rather
than nutrients (Davis et al. 2001) plants must be able to thrive in high heavy metal conditions.

In general, research indicates that stormwater bioinfiltration technologies are effective in nutrient, heavy metal and sediment reduction. Removals of 70-85% for P, 60-80% of ammonium and TKN and over 90% of Cu, Pb and Zn have been recorded (Davis et al. 2001, Davis et al. 2005, Bratieres et al. 2008, Hsieh and Davis 2005, Hsieh et al. 2007, Hatt et al. 2009, Read et al. 2007). Occasionally a BT will act as a source, not a sink for P and NO$_3$ (Hunt et al. 2006) leaching P once the saturation capacity is reached in the media, also releasing NO$_3$ (Dietz and Clausen 2006, Davis et al. 2006). The following sections examine two important components of nutrient removal, media and vegetation, and the role of each in bioretention technologies.
<table>
<thead>
<tr>
<th></th>
<th>NSP</th>
<th>FWS</th>
<th>SSF</th>
<th>BT</th>
</tr>
</thead>
<tbody>
<tr>
<td>nutrient removal</td>
<td>TP, TN</td>
<td>P, BOD, NH4, TN, TSS</td>
<td>tertiary BOD, TSS, NO3, pathogens</td>
<td>NH4, TSS, metals</td>
</tr>
<tr>
<td>water fraction</td>
<td>bog area 40%</td>
<td>40%</td>
<td>40%</td>
<td>varies</td>
</tr>
<tr>
<td>Media</td>
<td>river gravel 6mm</td>
<td>gravel and sands, 3/8-3/4&quot;</td>
<td>gravel layer of topsoil or mulch</td>
<td>sandy loam, sand</td>
</tr>
<tr>
<td>Operating depth</td>
<td>Bog - gravel .7M and water .3M above</td>
<td>30 cm</td>
<td>60 cm</td>
<td>75-120 cm</td>
</tr>
<tr>
<td>vegetation type</td>
<td>EAV, SAV, FAV</td>
<td>EAV, SAV, FAV</td>
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<td>EAV</td>
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<tr>
<td>water fraction</td>
<td>40%</td>
<td>40%</td>
<td>varies</td>
<td></td>
</tr>
<tr>
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<td>7 cm/day</td>
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<td></td>
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<td>energy consumption</td>
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<td>low</td>
<td>depends on configuration</td>
<td>low</td>
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<td>bed depth</td>
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<td>.2-.8m</td>
<td>.6m</td>
<td></td>
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<tr>
<td>ancillary</td>
<td>increase habitat, increase</td>
<td>support different plant communities,</td>
<td>mosquito, algae and odor control,</td>
<td>wildlife habitat, species diversity,</td>
</tr>
<tr>
<td>benefits</td>
<td>diversity of plants, pollinator, aesthetics, water capture, promote stewardship</td>
<td>increase habitat, aesthetics, inexpensive</td>
<td>aesthetics</td>
<td>aesthetics, low engineering and energy requirements</td>
</tr>
<tr>
<td>---</td>
<td>---</td>
<td>---</td>
<td>---</td>
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<tr>
<td>retention time</td>
<td>12 hrs</td>
<td>9.4 days</td>
<td>2.9 days</td>
<td>8 hrs</td>
</tr>
</tbody>
</table>

Table 2-1: Comparison of NSP, CWs and BTs (Kadlec and Wallace 2009, FLL Guidelines 2005, Vymazal 2005, Davis et al 2006)
Media in CW

While plant uptake and harvesting provides good nutrient removal for nitrogen and improves phosphorus removal, media is an essential component of nutrient removal and storage for CWs and BTs. The ideal media acts as a substrate for biological activity, is able to support rapid establishment and growth of plant material and has the necessary characteristics for the chemical processes of adsorption and precipitation (Arias and Brix 2001). Other characteristics of media essential for optimal function are hydraulic conductivity, specific gravity and pH. Hydraulic conductivity and specific gravity affect the hydraulic retention time and distribution of water through the system and pH affects the health of aquatic organisms and sorption precipitation reactions. Substrates with Al, Fe and Ca, have a high affinity for P but also have pH values either much lower or higher than neutral and can contribute to an environment not conducive to the health of aquatic organisms.

Many studies have explored the P removal capabilities and physio-chemical properties of alternative media. Some have found that certain media, such as Damolin, while excellent for P sorption, clog easily and are therefore not suitable in a CW (Arias and Brix 2001). Studies with shale (Drizo et al 1997, Forbes et al 2005) have demonstrated high removal efficiencies due to shale’s excellent hydraulic efficiency and high metal content (Fe and Al), providing it with high P binding capabilities. Arias and Brix (2001) tested the properties of various sands used as SSF media for P removal properties and found the most important characteristic for P binding was the presence of calcium in the media. The same study also tested various other media; expanded clay,
crushed marble, diatomaceous earth and calcite, and found that the addition of crushed marble or calcite to the filter media increased its P removal efficiency. However, once saturation occurs, the media lose their effectiveness and release P back into the system. Vohla et al. (2005), in a study exploring the removal capacities of different media, found that SSF planted filters, constructed with local sands, become saturated over a 5-6 year period. As the filter ages, its P removing efficiency initially improves, then levels off, finally decreasing and becoming a source for P. In the same study, batch experiments showed a high P removal capability (98.9%) for oil shale fly ash. Local gravels that were high in Ca, Mg, AL and Fe performed better than sands, regardless of particle size, demonstrating the importance of chemical content for improved sorption. Further research by the same author on filter media, showed a high correlation of P retention to CaO and Ca content but a weak correlation to pH (Vohla et al. 2011). Cui et al. (2008) had similar results; the researchers found that P sorption for 8 tested materials was highly correlated to Fe, exchangeable Al, CEC and organic matter; while it was negatively correlated to media pH and specific gravity. The study ranked the media in order of decreasing P sorption as follows: turf, blast furnace artificial slag, blast furnace slag, coal burn slag, top soil coal burn artificial slag, midsized artificial sand, gravel and midsized sand. These results indicate that traditional media substrates, sand and gravel, are not appropriate if P removal is a treatment goal.

Browning and Greenway (2003) also explored the effect of media on water treatment in addition to particle size and P removal capabilities. In both this study and Vohla et al. (2011) gravel size did not seem to be related to P removal, although other
studies have established a relationship between particle size and sorption capacity. Age of treatment cell in Browning and Greenway (2003) was directly related to performance, with the newest cell in the study the most effective. The media in this cell was new and consequently did not have all the chemically receptive sites saturated, whereas the media in the older cells (one was 5 yrs old) was already saturated.

Because of the difficulties in removing P from wetlands, three strategies have been suggested for enhanced P removal and storage. A wetland could have chemically enhanced media throughout, a prefilter that chemically precipitates P or a separate filter chamber enhanced with specific media (Arias et al. 2003). The cost of enhancing a whole treatment wetland with special media, such as Filtralite-P would be prohibitive and the effective life span of the filter material would still only be around 5 years. During those 5 years the filter material would decline in function and possibly experience episodic release of P. Use of special media throughout the wetlands creates the additional problem of disposal and removal when the media is saturated. Special P filter material could be used as a polishing filter under low loadings if the rest of the P could be removed using chemical precipitation. Rentz et al. (2009) found that iron oxides enhanced P removal. These results were supported by Del Bubba et al. (2005) and Zeng et al. (2004). Several studies have shown electric arc furnace (EAF) slag to have one of the highest affinities for P at both high and low concentrations, 40-400 mg/l and 5 - 10 mg/l respectively (Drizo 2003, Naylor 2003, Johansson 1999), but the EAF significantly raises pH. Naylor (2003), found that the pH of water treated with EAF slag was detrimental to Phragmites and Typha (over 10), reducing standing stock, thereby
affecting nutrient removal efficiency. One planting medium in this study incorporated peat moss with the EAF slag at 50% weight, decreasing the pH, which while increasing biomass, negatively impacted the substrates ability to remove P. This led Naylor to recommend the installation of a 2 stage treatment where fish farm effluent entered a planted gravel filter to remove N, then flowed to an unplanted filter with EAF slag and limestone removing P. In the design, the high pH experienced in the P removal filter would not be detrimental to plants and their nutrient removal performance since the two are completely separated.

**Media and BTs**

Originally, many BTs were constructed with sand or gravel for optimum infiltration. Problems associated with these media include low nutrient levels, CEC capacities and poor water storage and holding capacity, which decrease a plants ability to withstand periods of drought and thrive. On the other hand, these soils have increased infiltration capacities so will filter greater amounts of storm runoff (Hutchinson et al 2011). In DeBusk et al. (2007) the study tested various media including peat, wollastonite, limerock and sand concluding that wollastonite was an effective filter additive to remove P. The presence of calcium and ferrous ions in the wollastonite contributed to P removal through precipitation and adsorption. Other research has tested the addition of different forms of organic matter to the filter and measured the effect on N removal. One study tested the nitrogen removal ability of various organic filter additions, newspaper, sawdust, leaf litter, wood chips and straw. All the media had acceptable N
removal rates, with newspaper supporting the highest rate of denitrification at all nitrogen concentrations and flow rates, results verified by pilot tests providing N removal rates of 75-80% (Kim et al 2012). Because denitrification requires not only anoxic conditions but also an electron donor and carbon source to fuel the transformation, Kim et al. (2012) combined the media additions with a submerged zone that created the anoxic conditions necessary for denitrification. Excellent performance results were attained and further investigation indicated that while the organic additions would have to be replaced when NO$_3$ removal started to decrease, performance should be satisfactory for an extended time. The need to eventually replace the media is similar to media enhancements designed to increase P removal. Replacement and disposal costs and disposal locations must be considered before implementation and have clear implications for sustainable and low-impact technologies.

Another recent trend is the design of BTs using soil-based media instead of sands, with sediment sizes that closely relate to the expected sediments entering the system. Hatt et al. (2007) desired to increase the initial growth of vegetation and improve particulate matter capture in a column study of soil based media. The researchers found not only the media provided significant treatment removal differences, but also the presence of vegetation significantly contributed to removal capabilities. Soil based filters removed greater than 80% and 90% of sediments and metals (lead, copper, zinc), respectively, but unvegetated soil based filters were net exporters of nitrogen.

Li and Davis (2009), in a study testing media for removal capabilities over a wide range of contaminants, found that for 2 drainage cells with sandy loam and sandy clay
loam, nitrogen and phosphorus concentrations were increased slightly in discharge water due to the additional nutrients contained in the flora and fauna of the mulch (organic) layer. Additionally, Bratieres et al. (2008), found that sandy-loam mixed with vermiculite/perlite had a PO$_4$ and TP removal of greater than 83 and 89% respectively with the addition of 10% compost or a layer of mulch, the system became a net producer of PO$_4$. Both Dietz and Clausen (2006), and Hunt et al. (2006), found similar increases in discharge N and P levels. Clearly, studies quantifying and defining removal mechanisms in bioretention systems suffer from the same ambiguity and inconsistency of results as CW research.

Erickson (2007) explored enhanced sand filtration to improve P removal from stormwater runoff. In sand filters, stormwater runoff delivered to a sand filtration system is physically sieved, with any particulates larger than the filter pore size removed. In this study, sand filters were amended with seven different additives, including three different BOF slag amendments, steel wool, limestone, aluminum oxide and calcareous sand. Two of the amendments improved P removal, calcareous sand and limestone, but clogged the sand filters. A third, steel wool, improved P removal without decreasing the hydraulic function. Between 34 to 81% of dissolved P was removed by steel wool in the 6 units tested, with a minimal increase in cost. In an additional study, Erickson found including a mix of 5% iron shavings in an existing sand filter, enhanced P removal and captured 80% of dissolved phosphorus. This result supports the conclusions of Cui et al (2008) and Rosenquist et al (2010) concerning iron oxides and P removal. Other media studied for improved nutrient removal are calcined clay (White et al 2011), modified zeolite (Gibbs
2009), alum and Phoslock (Gibbs et al 2010). In a study measuring the efficacy of modified zeolite, alum and Phoslock on P binding in lakes, all three additives were effective at removing P, but had negative affects on nitrification, decreasing NO$_3$ removal by 35%. In Gibbs (2009), Z2G1, a modified zeolite, acts as a carrier for aluminum. It effectively bound phosphorus in a New Zealand lake, also absorbing NH$_4$. It is the only additive of the three previously mentioned that effectively reduces both P and N.

The associated issue with filter additives that precipitate or adsorb P is when the P saturation is reached the filter media must be replaced or it starts to release P back into the environment, similar to the issue with N and media enhancements. Therefore, further research needs to be done establishing the active life cycle of a filter and disposal issues should be considered. For instance, some studies have tested saturated treatment filters for use as planting media in nurseries after the P saturation, allowing the removed P to then become a resource. (White et al 2011).

**The role of vegetation in constructed wetlands**

In the 1970’s, research was conducted with Eichornia crassipes as a harvestable plant treatment for excess N and P in the Florida Everglades. Water hyacinth is capable of removing 126 gPm$^2$yr$^{-1}$ (Vymazal 1995) but is also capable of aggressively spreading throughout wetlands and ecosystems and escaping to adjoining waterways. As time went on, research and practice shifted from water hyacinth monocultures to other mixes of plants species and plant types (Kadlec 2005), although hyacinth monocultures are still used in the tropics. The majority of wastewater treatment CW research concerning plants
has concentrated on measuring the removal effectiveness of monoculture plant filters by measuring the N and P nutrient uptake and storage in the plant tissues (Drizo et al, Chung et al. 2008, Tanner 2001). Plants often planted in monoculture in treatment wetlands are *Phragmites, Typha, Phalaris, Schoenplectis* and *Carex* spp. (Vymazal 2011, Headley et al. 2003, Kadlec and Wallace 2009, Meuleman and Logtestijn 2003). Later studies explored mixed plantings for improved nutrient removal and improved ecosystem services. (Read et al. 2008, Liang et al. 2011). Although most CW research studies monocultures it is logical to consider the benefits of a mixed species community in addition to nutrient removal. Diverse ecosystems have improved disease and pest avoidance, are able to respond to changing environmental conditions while maintaining function, have improved aesthetics and provide greater ecosystem services. These species are adapted to a range of growing conditions and can tolerate low soil oxygen and water fluctuations. *Typha* and *Phalaris* tolerate a wide pH range, 3-8.5 and 3.7-8 respectively, although biomass production suffers at the extremes of the range (USEPA 2000). A few studies have looked at the ability of ornamental plants and native wetland plants to remove N and P, (Maschinski et al 1994, Zurita et al 2009, Polomski et al 2007, Taylor et al 2006) and found canna, iris and pickerelweed to be effective components of biofilters indicating that aesthetics can be considered when designing a treatment wetland without sacrificing function (DeBusk et al 1995). This is encouraging for NSP research, as aesthetics is an important function of a backyard amenity.

There is much discussion in the literature over the degree plants increase wetland nutrient removal. Some studies indicate that there is substantial N and P uptake and
storage in the biomass of vegetated filters while others indicate that the main contribution of plants is in areas other than direct uptake (Tanner 2001). (See Table 2-2)

<table>
<thead>
<tr>
<th>Plants in FWS</th>
<th>Effects on all CW</th>
</tr>
</thead>
<tbody>
<tr>
<td>Increase sedimentation by reducing wind-induced mixing and resuspension</td>
<td>Added carbon for denitrification, nitrification</td>
</tr>
<tr>
<td>Add surface area in H2O volume; increasing biofilms and soluble pollution uptake</td>
<td>Increased O2 diffusion to water column from plant roots</td>
</tr>
<tr>
<td>Increased surface area for particle interception</td>
<td>Chemical exudates used by plants detoxify root environment</td>
</tr>
<tr>
<td>Increase shade; moderating water temperatures and algae population</td>
<td>Additional fungi species introduced by roots, increases N uptake</td>
</tr>
<tr>
<td>Increased flocculation of smaller colloidal particles into large settleable particles</td>
<td>Symbiotic bacteria introduced by plant roots</td>
</tr>
<tr>
<td>Surface insulation</td>
<td>Increase habitat for invertebrates</td>
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<td></td>
<td>Nutrient uptake</td>
</tr>
<tr>
<td></td>
<td>Aesthetics</td>
</tr>
<tr>
<td></td>
<td>stabilize bed surface</td>
</tr>
</tbody>
</table>

Many studies quantify how planted filters have enhanced N (Borin and Salvato 2012) and P removal performance (Headley et al. 2003) but do not agree on the actual amount of nutrients the plants remove (Greenway and Wooley 2001, Tanner 1994, Browning and Greenway 2003, Zhang et al. 2007, Bratieres et al. 2008, Read et al. 2008, Brix 1997, Chung et al. 2008, Gottschall et al. 2007) As mentioned earlier, many factors affect nutrient uptake and storage, namely species, climate, nutrient loading, type of root system, seasonality and (Tanner 2007, Vymazal 2005, Kadlec 2005) contributing to the wide variation of results. These factors also increase the difficulty applying these results to different wetlands.

Several researchers have conducted studies demonstrating that nutrient uptake and storage in plants can vary between species in the same wetland and between wetlands (Browning and Greenway 2003, Tanner 2001, Vymazal 2005). In one CW study, Gagnon et al. (2012) found higher removal efficiencies for Phragmites in the removal of TKN, NH$_4$, TP and PO$_4$ than Typha and Scirpus. The researchers postulated it could be attributable to increased carbon exudates from the phragmites root system. A study of a SSF wetland planted with phragmitess in a shale based medium, measured substantial increases in N removal, with NH$_4$ completely removed in planted treatments, compared with 40-75% removal in unplanted treatments (Drizo et al. 1997). Additionally, NO$_3$ removal increased from 45-75% in unplanted treatments to 95% in planted. Huett et al. (2005) also studied Phragmites nutrient removal ability for N and P in beds treated with nursery runoff. Nutrient removal improved from less than 16% and 45% of TN and TP removed respectively in unplanted treatment, to more than 96% of TN and TP removed.
in planted treatments. It seems clear that vegetation does contribute to nutrient removal in both FWS and SSF constructed wetlands, but it is unclear if researchers can assume a species will perform at consistent levels in different wetlands.

As mentioned earlier, FWS wetlands are designed with a mix of vegetation types, rooted emergent macrophytes, free floating macrophytes and underwater macrophytes. SSF constructed wetlands and BTs contain emergent macrophytes only. There is evidence that the type of plant community, (emergent, free-floating, submerged), affects treatment performance, but the plant species contained in the plant community does not (Kadlec 2005, Iamchaturapatr 2007). The reason for this difference is related to the substantial differences in uptake and storage between emergent, free-floating, and submerged macrophyte species. After harvest of biomass from each type of system the emergent contained 30-150kgP/ha.yr and 200-2500 kg N/ha.yr, floating 350kg P/ha.yr and 2000 kg N/ha.yr, and submerged <100kg P/ha.yr and 700 kg N/ha.yr (Kadlec and Wallace 2009). Harvesting and removal of above ground plant biomass can remove 10-20g P m⁻² yr⁻¹ when nutrient loadings are low (Vymazal 2007).

Research has also examined the potential of monocultures versus mixed stands to remove N and P. Recent research by Liang et al. (2011) indicates that there is no significant difference in nutrient removal between monocultures and mixed plantings, a conclusion supported by Fraser et al. (2004). Fraser tested four wetland species in monoculture and one treatment of all four plants and found that while the presence of plants had a significant positive effect on nutrient removal there was no difference between monoculture and mixed planting treatments.
Tanner (1996) used tissue nutrient levels as only one measure of plant performance when rating species performance, also considering stand biomass, harvestable production, seasonality and root zone aeration. Using this rating system *Zizania, Glyceria* and *Phragmites* were the highest scoring plants over *Schoenoplectus, Baumea, Cyperus*, and *Juncus* (Tanner 1996). This study presents results for growth and performance based on a warm temperate climate and biomass production, seasonality and nutrient removal could be substantially different in a cool season temperate climate. However, the fact remains that it makes sense to consider other factors then tissue nutrient levels when rating plant performance.

Rate of uptake seems to be proportional to net productivity and concentration of the nutrient in plant tissues (Borin and Salvato 2009, Richardson 1985, Polomski et al 2007) indicating that plant selection for species with rapid growth, large storage capacity and high standing crop is advised (Reddy and DeBusk 1987). Further supporting this conclusion is the evidence from a plant biomass study of *Typha, Schoenoplectis* and *Eleocharis*, indicating that as plant biomass increased so did nutrient uptake (Greenway and Woolley 2001). Amount of nutrient available positively correlates to biomass production, as does ratio of nutrients between aboveground and belowground plant tissue for many plants (Zhang et al 2008, Polomski et al 2007, Polomski et al 2008). In Polomski et al( 2007, 2008) different commercial ornamental plant species were tested for ability to grow and remove nutrients in SSF CW. Five different loading rates (mg/L) of nitrogen and phosphorus, from 0.39-36.81 and .07-6.77 were applied every 2 days for 8 weeks. At the end of this period, biomass was harvested, weighed, and then nutrient
content for each species, divided into above and belowground portions were analyzed. *Pontedaria cordata* (pickerelweed) and *Iris x louisiana* (Louisiana hybrids) had the highest percent shoot nutrient concentrations with pickerelweed shoots containing greater than 61-76% of total N in the plant and Iris greater than 90%. At all N loading rates *Canna, Colacasia, Peltandara, Oenanthe* and *Thalia* had greater N aboveground concentrations, while at loading rates above 10.4 and 21.6 mg/L N, pickerelweed and iris had higher concentrations of N in shoots than roots. *Typha minimus* roots were the dominant sink at the low N loading concentration and shoots were the dominant sink at the two higher loading rates. In contrast, P was highest in shoots for *Canna, Pontedaria, Oenanathe* and *Thalia* at every loading rate.

Research considering shoot to root ratios of nutrient partitioning are helpful if plant harvesting and removal of biomass as a nutrient removal tool is being considered. Harvesting of aboveground material has been explored by Browning and Greenway (2003), Headley et al (2003), Kim and Geary (2001), Vacca (2011) and Polomski et al (2007), Meuleman and Logtestijn (2003). Problems encountered range from inability to recover growth momentum after harvesting (Browning and Greenway 2003, Tanner 2001, Granelli et al 1992), and variable removal efficiencies. Researchers have noted that removal of aboveground biomass when nutrients have not yet translocated to rhizomes decreases the vigor of the plant and can reduce nutrient uptake as a result (Granelli et al. 1992, Asaeda et al. 2008). Additionally, Kim and Geary (2001) indicated percent of nutrients removed and stored by the plant biomass is not substantial enough to make the resource expenditure of harvesting cost effective. More research examining
timing of harvests could help designers to optimize nutrient removal as demonstrated by Vymazal (2007) where maximum P uptake occurred in spring before maximum growth rate and in autumn P moved from the aboveground portion of the plant to rhizomes. Amount of storage in aboveground portions of plants varies from study to study, depending on species, biomass, length of time to reach harvestable standing stock and different nutrient uptake rates.

The cell with the highest biomass and removal rate in a study by Browning and Greenway (2003) of vegetated wetland SSF cells, still only accounted for 11% of N and 3% of P removed. In another study of changes in plant biomass and nutrient removal over three years, growth rate was examined after cropping. Typha and Schoenoplectis would be able to reach harvestable biomass in 6 months after harvest while Eleocharis would be ready in 3 months (Greenway and Wooley 2001). In this case, harvesting increased productivity and the percent of nutrients removed by plants of the total nutrients removed from the wetland. Also supporting the potential benefits of harvesting, an examination of N removal capabilities of wetland plant found that 53-75% of nitrogen load was absorbed by the plant material, and of this 51-83% was allocated to aboveground portions (Borin and Salvato 2012). This supposes a substantial portion of nutrients partitioned in plant tissues.

However, not all research agrees that biomass stores significant amounts of nutrients. In Kim and Geary’s (2001) study, nutrient partitioning in plant tissue was small. Harvesting temporarily increased P uptake but there was no statistical difference in P removal between harvested and unharvested treatments. Overall 90% of P in the
system was compartmentalized in the substrate with less than 10% in biomass. More than 50% of the 10% found in plant tissue was held in underground plant biomass suggesting that, in this case, harvesting doesn’t really affect nutrient removal. Drizo et al (2008) had similar results, below ground biomass of _Schoenoplectis_ had significantly higher P than the aboveground portions and so harvesting would not contribute to nutrient removal. On the other hand, Headley et al. (2003), in a three year study suggests that plant uptake was the most important compartment for P removal, accounting for over 75% of P removed. Media accounted for only 12% of the P removed the first year and increased that number to 30% the second year. While the phragmites beds consistently removed greater than 96% of P and reduced P from .5 mg/l to .005 mg/l, the amount of P held in above and below ground portions of the plant was greatly influenced by seasonal stages. In spring, the phragmites experienced a period of rapid expansion and fueled growth partially by the P stored in the plant rhizomes. This period of rapid expansion was followed by a summer period of nutrient uptake that was controlled by the influent P loading rate. During this period, the amount of P in the roots was stable. At the onset of senescence P translocated from aboveground portions to belowground portions when the underscoring the importance of timing of harvest.

Another study of eight emergent macrophytes found that enrichment increased the shoot to root ratio, doubling it in most of the plant species, which has obvious implications for plant harvesting (DeBusk et al 1995). It is clear that if recommending a nutrient management strategy involving harvesting, knowledge of the effect of different loadings on nutrient partitioning is necessary. For a low nutrient environment
encountered in a NSP, harvesting as a tool for nutrient management would have limited effectiveness, except as a fall cleanup of standing stock.

**Vegetation and Stormwater Bioretention systems**

Henderson et al. (2007), in a bioinfiltration mesocosm study comparing the nutrient removal abilities of three different media and vegetated and unvegetated treatments, found that unvegetated treatments released nutrients back into the system regardless of media type. In this study, vegetated sandy loam and sand media treatments were more effective than gravel vegetated and all unvegetated treatments, achieving overall N and P removals of 63-77% and 85-94% compared to 10% in unvegetated sand for N and 75% of P in unvegetated loam. Further, the sandy loam media supported greater plant growth than the other two medias, with plants in gravel performing poorly (Henderson et al 2007). As discussed earlier, water holding capacity of media and nutrient availability can impact the ability of vegetation to withstand variable environmental conditions. However, since of the three media types, sandy loam had the poorest hydraulic conductivity, supporting a productive plant community would have to be weighed against the need for the BT to drain in a reasonable amount of time.

A stormwater design optimization study by Bratieres et al. (2008) recorded similar results. Sandy loam soil was found to be the best media for plant growth, consistently removing 70% of N and 85% of P although the addition of organic matter to the media reduced P removal. The study tested five plant species and three different media types for nutrient removal capabilities, finding that vegetation had a significant
effect on both TN and NOx removal improving performance over unvegetated. Plant species strongly correlated to TN and NOx removal. Of the five species tested, *Carex spp.*, with its dense root system, performed best, although *Melaleuca* also performed well. *Melaleuca* supports arbuscular mycorrhizal fungi in its roots, which increase the absorptive surface area of the roots. *Melaleuca* exhibited an increased removal of N over time unlike three other tested species that showed a net production of NOx and declining removal performance over time. Read et al. (2008) who conducted research on 20 plant species and their effectiveness at removing nutrients from stormwater found significant differences between species and related these differences to plant morphology and root architecture. It seems it is not enough to simply vegetate BT’s, but to utilize plants with the greatest chance of removing nutrients, especially N, since plant uptake removal is more significant for percent N removal than P.

Lucas and Greenway (2009), explored the role of vegetation in nutrient removal by dosing vegetated and unvegetated experimental columns planted in three different types of media (pea gravel, sand and loam) with synthetic stormwater, at the same levels as Henderson (2008). The presence of vegetation significantly increased the removal of both TN and TP in unsaturated media even though plant uptake accounted for a fraction of total nutrient reduction. The experiment showed significant increase in P removal in the initial dosing of stormwater by vegetated treatments as compared to unvegetated for all media, but no improvement in the final dosing runs where P removal sites were already saturated. In gravel media the improved retention far exceeded plant uptake projections, but the increased retention in vegetated loam and sand treatments was less
than projected plant uptake. Improvement in retention of TP was attributed to the presence of an aerated rhizosphere, which oxidized ferrous iron to ferric thereby increasing P attraction, and to the presence of arbuscular mycorrhizae. Vegetation had a significant effect on TN retention in all media. This change was attributed to plant uptake because the change between vegetated and unvegetated equaled projected plant uptake (Lucas and Greenway 2009). In addition to direct uptake, vegetation provides increased infiltration, crucial to reduction of storm runoff. In a BT, vegetation not only improves nutrient removal as mentioned for CWs but also improves soil structure increasing infiltration and reducing runoff (Henderson 2011).

**Differences between NSPs, CW and BT**

NSPs differ from treatment wetlands in several key areas: treatment goals, operating parameters, aesthetics and maintenance. Treatment wetlands and bioretention systems are open systems, and one of the primary design goals for a BT is reducing stormwater runoff volume as well as reducing specific pollutants. Both systems have inputs from outside the system (precipitation, wastewater, fertilizer, sediment etc.) and after treatment release the effluent from the system. A NSP is a closed system. (See Figure 2-5) Pool fill water introduces the nutrients to the pool and the water simply recirculates through the biofilter during the swimming season without discharge. Regulating agencies often set treatment levels for CW and BT’s, but the standard for a NSP is simply clear water.
Because of the difference in treatment goals, operating procedures vary accordingly. For constructed wetlands and bioretention systems optimum retention time to reach treatment goals is measured in days and hours, respectively. Retention time has a different context in a NSP, because water continually circulates through the filter, without discharge, although volume running through the filter at any one time is low, \(1 \text{m}^3/\text{m}^2/\text{day}\). Most of the nutrient additions are from user impurities, pollen, insects, organic matter (leaves, etc) and fertilizer runoff. See Figure 2-4 for NSP filter diagrams.

Figure 2-4: Construction sideview of NSP bog or regeneration area
If the pool is designed correctly precipitation events add volume to the system, but there should be very little runoff or sedimentation introduced. However, certain areas of the country (usually in agricultural areas) have high levels of P in municipal water causing the introduction of substantial levels of P with the fill water. EPA guidelines advise that N and P be lower than 10 mg/l and 1.5 mg/l respectively, well below the 30 mg/l and above the 0.01mg/l recommended by FLL (2003, 2006) guidelines for N and P. If animals and fish are excluded from the system, the nutrient load introduced to the pool should be relatively low. See Figures 2-5 and 2-6 for NSP and CW inputs and outputs.

Figure 2-5: Natural swimming pool system inputs and outputs
When considering plants for a NSP the designer must consider several important criteria, aesthetics, the need for rapid establishment and consistently low nutrient levels.

As a landscape amenity, NSP & BT plantings should follow garden design principles of order, unity and repetition and therefore contain at least two species and contain a good mix of bog plants. Mixed plantings have three advantages. Aesthetically, from a landowner’s perspective, mixed plantings have more interest and a longer bloom time. More importantly, mixed plantings have biological diversity and with diversity comes the ability to fill different niches both in habitat and nutrient removal. Research indicates monocultures are not as effective in modulating seasonal fluctuations as systems planted with a variety of species (Picard C.R. et al 2005). Unfortunately, plants that form monocultures have many desirable qualities from a nutrient removal perspective. *Typha, Scirpus and Phragmites* have vigorous root systems, high biomass accumulation rates, rapid establishment, and can accommodate different nutrient environments. Despite those advantages, *Typha, Baumea* and *Phragmites* all have sharp,
pointed rhizome tips, very capable of penetrating a pool liner, and should be avoided in natural swimming pools.

NSP regeneration areas need plants that are quick to establish, most pool owners want to minimize downtime. Bog plants must also be easy to locate for purchase, tolerate a low nutrient environment and play nicely with others, in other words, less likely to form a monoculture. Since nutrient removal capabilities vary between types of aquatic vegetation, (submerged, floating and emergent), and NSP bog filter design incorporates the ability to vary depths accommodating different aquatic vegetation, it seems prudent to incorporate all three types into the bog filter. A combination of species with different prime uptake periods would be optimum, for instance planting *Iris versicolor* and *Hibiscus moscheutos*. *Iris versicolor* is an early riser, one of the first bog plants to break dormancy, while hibiscus starts late and stores P primarily in the fall during flowering (Billings 2011). A NSP would not be likely to use either *Typha* or *Phragmites* in the regeneration area, for reasons stated previously, rather would use more ornamental and less ecologically damaging species such as *Pontedaria cordata* and *Saururus cernuus*. Both are native plants with attractive qualities and good growth habits, and planted in some constructed wetlands (Kadlec and Wallace 2009).

NSPs, CWs and BTs experience different nutrient loading rates and water depths. In treatment wetlands, plant stress occurs from the high levels of nutrients and heavy metals that can cause toxicity for some plant species, especially at higher loading rates and longer retention times (Chung et al. 2008). A NSP has, hopefully, persistent low, nutrient levels. Its nutrient regime may change after a weekend with many users, or after
a three inch rainfall, but still doesn’t fluctuate widely. Nutrient levels stay lower than you would typically find in a waste treatment wetland and would be more consistent with the levels found in storm water biofilters after a rain event. Plants respond to nutrient shortages by altering resource allocation, increasing root biomass to increase surface area for uptake (Hermans et al 2006). NSP and FWS wetlands have consistent water depths; vegetated filters in BTs have the challenge of adjusting to periods of inundation and drought. Also, CWs and NSP use gravel for planting substrate, which is more difficult, mechanically for root penetration, affecting establishment time. So the stresses on plants in a NSP are different from that of a constructed wetland and consist primarily of low nutrient levels, the undesirability of monocultures, and the challenge of low nutrient loading, and plant root establishment in a gravel filter.

In treatment wetlands and storm water biofilters there is a gradual buildup of organic matter from plant senescence and decay. While decay releases nutrients into the system, the process where these plant parts break down and become part of the soil is crucial to the functioning of the wetland. It is in these sediments that much of the phosphorus is held until the P sorbing sites are all saturated. This compartment is not available in a NSP as any sediment that formed is removed regularly to maintain aesthetic appearance, control accumulation of organic matter and protect the system from clarity issues from suspended solids. However it would be reasonable to speculate that a seasonal soil accretion rate would develop, compartmentalizing a certain amount of P each growing season, allowing the P to be removed with the sediments at the end of the growing season. If removed at a regular rate the amount of sediment to remove will
never be overwhelming or present a disposal issue. These sediments could be used locally as garden amendments or topdressing.

Maintenance and upkeep are both considerations when managing these technologies. A crucial maintenance element for natural swimming pools is harvesting emergent aboveground biomass and free floating biomass. Traditionally harvest has occurred in the fall, in which case it should be scheduled before nutrients start to translocate to the root systems, but for some aquatic species it may be possible to intermittently harvest throughout the growing season. Free floating vegetation should only be used in areas where the species does not present an ecological risk. As landscape amenities, periodic inspection of both NSPs and BTs for insect and disease issues is necessary. Use of fertilizer, pesticides and fungicides is not advisable in or around natural swimming pools because of the negative consequences to the biocenoses.

Treatment wetlands with free water surface and macrophyte populations are assessed by their ability to meet EPA discharge standards. If they can meet these standards, the amount of algae in the system is of little concern. However, in a NSP, algae are detrimental to the system in two ways. The algae create a safety hazard if it interferes with water clarity and visibility. FLL guidelines specify a secchi depth reading of three meters for clarity. Algae also competes with plants for the nutrients in an already low nutrient environment. The presence of algae inhibits the establishment of emergent macrophytes and healthy populations of aquatic organisms though nutrient competition, by stimulating harmful pH swings and DO levels. Beyond these two factors, the general
population equates algae with impurity, which would discourage use of the pool while decreasing its aesthetic value.

To summarize, quality research on constructed treatment wetlands and stormwater biofilters exists, but the differences between these and NSPs make it difficult to apply results from CW and BT research to NSPs. The inconsistent nutrient removal performance of existing studies, demonstrates the need for a concentration model for NSP biofiltration, contrasting vegetated and nonvegetated systems and assessing the ability of different substrates to remove P. In this study ornamental native, emergent aquatic plants were chosen, to vegetate each filter type. At the end of each experiment plant tissue concentrations were obtained, and substrate and water NO₃ and P measured for the purpose of developing a mass balance model for the biofiltration of a NSP.

**Research Design and Objectives**

Low nutrient environments create a challenging environment for establishment of plants in NSPs. No data is available on the nutrient removal and survivability of plants in NSPs. Nutrient removal pathways of NSPs must be quantified to understand the significance of both compartments, substrate and macrophytes, allowing the development of a mass balance model for natural swimming pools. Each study presents mass balance assessments estimating nutrients added to the system, nutrients stored in plant biomass, removed by harvest, and stored in substrate.
FLL guidelines suggest design guidelines to obtain recommended NO₃ and P levels. These design guidelines must be tested to assess the ability of a NSP to effectively remove NO₃ and P to 30 mg/l and 0.01 mg/l respectively.

Substrate characteristics have significance for the adsorption of phosphorus and removal from pool water. This research tests five substrates for N and P removal abilities and their effect on plant establishment. Rapid plant establishment is essential for effective system function, especially in N removal. Growth indexes are developed to measure plant health for correlation assessments with nutrient removal.

**Assumptions**

1. FLL guidelines for measurable output levels
   a. total phosphorus after treatment ≤ 0.01mg/l P
   b. NO₃ level after treatment ≤ 30 mg/l
   c. ammonia ≤ .5 mg/l
   d. pH 6.0-9.0
   e. retention time .05 to 1.5 m3/m2xd
   f. .5-1 inch river gravel in regeneration area 1ft deep

2. Assume no sediment accretion if pool properly maintained and designed

3. Assume P is algae limiting. This impacts model measurements. P will be measured as TP (measurement of all forms of P dissolved or particulate) and SRP (measure of orthophosphate, the filterable soluble inorganic fraction of phosphorus, the form directly
taken up by plants.) Particulate P has less impact on environment, as it is less available to plants and algae.

4. Assume algae are undesirable in these systems. Even though algae have filtering abilities for nutrients its affect on pool aesthetics and safety are negative.

5. Assume no animals are allowed in the natural swimming pool. They add too many nutrients to the system.

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Chapter 3

Nutrient Balance of Three Natural Swimming Pools

Introduction

The Sustainable Sites Initiative, published in 2009, provides a framework for landscape design that embraces the philosophy of stewardship. It encourages design professionals to look past traditional philosophies and design limits while recognizing that all landscapes have the potential to be ecosystems that contribute ecological services to the community, locally and globally. Its this attitude of seeing beyond the site, realizing that even small contributions contribute to the whole, that fosters a sense of responsibility to community, client and the future. After all, sustainability at its core is about legacy, bequeathing a future for subsequent generations, what the Sustainable Sites Initiative (SSI) calls ‘intergenerational equity’.

How do natural swimming pools (NSPs) fit into a discussion about the SSI? NSPs are a sustainable alternative to chemical pools. Chlorine pools contain levels of chlorine harmful to aquatic organisms. Recent research indicates a relationship between respiratory problems and high exposure to swimming pool chlorine, especially adolescents and children (EPA 1994, Bernard et al. 2009). In contrast, the ecosystem functions that a NSP offers include water quality enhancement and conservation, increased personal health and well-being, and a sense of connection to nature. Research into personal connectiveness with nature and emotional health underscores the...
importance of experiencing small wild spaces in day to day living (Manuel 2003, Louv 2008).

The SSI stipulates that the design, implementation and maintenance of a sustainable site must be supportable through its life cycle without incurring economic hardship or unrealistic maintenance levels to sustain its function. Chlorine pools require daily pH, chlorine checks, skimmer cleanouts and shocking. Natural swimming pools require the occasional removal of leaves and debris from the skimmer, harvest of plant material in the fall and seasonal water quality testing (Littlewood 2005, von Berger 2010). None of the maintenance requirements present a danger to the environment or the user. No studies have been conducted on the maintenance time requirements as compared to a chlorine pool.

This research assesses the design and implementation of NSPs for nutrient removal. NSPs must be able to reduce NO$_3$ and TP to FLL recommended concentrations, <30 mg/l and < 0.01 mg/l respectively (FLL 2003). FLL standards are formulated by the German Landscape Research, Development and Construction Society and were published in 2003 and 2006. Maintenance of pool water at or below these levels eliminates algae, creating a safe, attractive and healthy pool system. Recommendations for operation and maintenance of a NSP include fall harvest of plant standing stock, removal of sediment accumulation and prohibition of pets and fish (FLL 2003, Littlewood 2005). It is also recommended that outdoor showers be installed with NSPs. Showering before entering the pool decreases nutrient (dirt, sweat, sunscreen) introduction.
This field study followed three NSPs from June 5 through Nov 1, 2009. NO$_3$, NH$_4$, TP, pH, total alkalinity and hardness (CaCO$_3$) were recorded at the start and end of the experimental period. Total phosphorus (TP) is the measure of all forms of P, dissolved or particulate. Total alkalinity (CaCO$_3$) is the measure of acid neutralizing capacity or more simply, how easily pH is shifted in a pool. In November, plant biomass was harvested, weight recorded and analyzed for %N and %P. This research determined if the NSP operated and functioned within approved parameters. If not, and if excessive maintenance is required for adequate nutrient removal and water clarity, then the system as designed, is neither sustainable or useful as a landscape amenity.

Planted filters usually outperform unplanted in removing nutrients (Hunter et al. 2001, Fraser et al. 2004). Plants increase removal of both nitrogen and phosphorus from the system with peak removal periods from June to October (Picard C.R. et al. 2005). There are seasonal variations in effectiveness of biological filtration, but less variance in phosphorus removal because it is dominated by sediment adsorption as compared to biological processes (Spieles and Mitsch 2000, Richardson 1985). Planted filters are less subject to monthly fluctuations in nutrient removal as the plants act to mediate temperature changes (Hunter et al. 2001). Some studies suggest planted monocultures aren’t as effective as mixed species in modulating seasonal fluctuations (Picard C.R. et al. 2005). Other research determined no difference between nutrient removals in monocultures vs mixed species plantings. Because NSPs are year round landscape amenities, water nutrient levels and the resulting clarity must be maintained in every season.
Unplanted regeneration areas are still able to remove nutrients indicating microbial processes as a major pathway of nutrient removal. Plants contribute to microbial processes by supplementing nutrient removal and increasing the diversity of the microbial population (Ottova et al. 1997). Plants also supply carbon to microbes allowing them to be more successful in surviving and removing nutrients (Lin et al. 2002).

Vymazal (2007) states that nutrient removal through plant harvest is low in all constructed wetland systems, but contributes more to nutrient reduction in low load systems. In low load systems, the nutrients in plant biomass through uptake represent a higher portion of the total load. A NSP is a low load system. The majority of phosphorus is introduced through pollen, fill water, precipitation and user impurities. Precipitation and user impurities are the primary sources of nitrogen.

It is the hypothesis of this section that a critical plant biomass level exists to remove nutrients that algae need to grow, and if this critical level is not achieved then the pool will experience algal blooms. Level of plant coverage needed could be measured by the point where nitrogen and phosphorus levels in the system, including N & P stored in aboveground plant biomass, at the end of the season, are less than the levels in spring. When input of nitrogen and phosphorus equals output of same as measured by nutrients present in plant biomass the natural swimming pool is in balance. Measured inputs were atmospheric nitrogen and phosphorus measured from data collected from the National Atmospheric Deposition data station in University Park, PA. This amount was added to the nutrients added by fill water volume. Base level data would include surface area,
volume, NO$_3$, NH$_4$, TP, pH, total alkalinity and hardness (CaCO$_3$). If plant biomass has accumulated more nutrients than atmospheric input, we know on-site nutrient contributions exist. If plant biomass nutrient levels are less than inputs, especially in the case of P, then we can assume that at some critical level of nutrient accumulation the pool will not be in balance and will experience algal blooms.

This study estimated inputs of N and P, measured final NO$_3$ and P water concentrations and determined biomass storage. The three pools in the study were constructed between June 2006 and the summer of 2008. Because of the difference in age there will be a corresponding difference in plant coverage of the bog area, providing an opportunity to learn more about the time period involved in achieving optimal plant densities for maximum nutrient removal.

Natural swimming pools differ from natural wetlands in that they are sealed against interaction with local hydrology. EPDM liner, or some waterproof material, prevents water intrusion or seepage into groundwater systems. No exchange of water occurs between adjoining streams, lakes or wetlands and a NSP. The NSP represents a closed system with nutrients and water introduced to the system, remaining within the system with the exception of ET.

Natural swimming pools also attempt to limit nutrient introduction. FLL guidelines recommend placing a shower next to the pool entrance to eliminate nutrient additions through user skin, sunscreen etc. Guidelines also recommend the pool stays free of fish, eliminating possible nutrients from food and waste products. Mechanical components, (skimmers), remove particulate matter before accumulation in the nutrient
pool (FLL 2003, Littlewood 2005, von Berger 2010). All of these elements separate a NSP from a natural wetland.

Elements in common with natural wetlands are vegetation types, appearance of the bog and biocenoses, or the community of organisms that contribute to wetland function. Biocenoses include bacteria, periphyton, zooplankton, and emergent, submergent and floating macrophytes. Precipitation is an inflow for both wetlands and NSPs and evaporation and splash is the major water removal process in NSPs.

Natural wetlands have a water budget that can be described by equation 5.1

\[
\Delta V/\Delta t = P_i + S_i + G_i - ET - S_o - G_o
\]

(5.1)

where

\[ \Delta V/\Delta t = \text{change of volume per unit time} \quad S_o = \text{surface outflows} \]
\[ P_i = \text{precipitation in} \quad G_o = \text{groundwater outflows} \]
\[ S_i = \text{surface inflows} \]
\[ G_i = \text{groundwater inflows} \]
\[ ET = \text{evapotranspiration} \]

The water budget of a natural swimming pool is better illustrated by equation 5.2

\[
\Delta V/\Delta t = P_i + F_i - ET - O
\]

(5.2)

where

\[ \Delta V/\Delta t = \text{change of volume per unit time} \]

66
Precipitation contributes to both wetlands and NSPs, but the amount of precipitation that adds to water volume (throughfall) varies by interception from surrounding vegetation. A forested area may intercept 8 to 35% of precipitation before it reaches the wetland, and may divert an additional 3-5% by stemflow (Mitsch and Gosselink 2007). A NSP in an urban environment, with the typical open backyard will have very little precipitation lost to a protective canopy.

EPDM liners or traditional swimming pool shells seal NSPs against groundwater intrusion. Properly designed, minimal surface runoff should enter the NSP. Surface runoff is undesirable because it is an unpredictable source of nutrients and includes lawn fertilizers, pesticides and organic matter. NSPs do receive fill water throughout operation, supplied by tap or well water. Water from both sources is often a source of phosphorus and nitrogen (NO₃⁻). For instance, a Penn State tap water sample analyzed for P in Dec. 2011 contained .7 mg/l, substantially higher than the target level of 0.01 mg/l of P for a NSP. The design for a NSP could include a reservoir to capture stormwater for use when needed, but for this discussion, I assume a local water source, either tap water or well water.
Evapotranspiration, water lost from the system through evaporation from soil and water surfaces and moisture passed through plant tissues to the atmosphere, is seasonal and peaks for both wetlands and NSPs in summer. Many factors affect evapotranspiration losses, including wind, air temperature, anoxic conditions and humidity (Cronk and Fennessey 2001). One method of estimating evaporation volume losses from water bodies is by utilizing data collected from NOAA evaporation pan technical reports. Researchers have concluded that substantial differences do not exist between open water and vegetated wetland evapotranspiration, nor between different species type (Mitsch and Gosselink 2007). For this research, evapotranspiration will be estimated by Class A evaporation pan data. Other volume losses can occur due to overflow, but only in the event of a major precipitation event, and therefore is not really considered a loss, but a shunting of excess precipitation.

In this study, I examine three existing NSPs in the same geographic area for water budget and vegetative nutrient removal. All three utilize 2b (1.5 “) local river gravel as bog substrate. However, the pools vary in important design considerations, namely flow rate, proportion of bog surface area to swimming surface area, volume of water and age of system. Because of these considerations, direct comparisons are not possible between pools. This study does provide the opportunity to increase understanding of NSP operation and function, and the role of vegetation in nutrient removal.
Methods

Three pools used as NSPs were examined for water budget and vegetative nutrient removal. Pool 1 and pool 2 are located in Lewisburg, Union County, Pennsylvania, 40°56’44”N, 76°54’47”W, and pool 3 located in Northumberland Co, Pa, 41°2’29”N, 76°49’26”W. Water samples were drawn June 5, 2009 and Nov. 1, 2009 and sent to the Penn State Agricultural Analytical Services Laboratory, University Park, PA, for analysis. Water samples were analyzed for NO$_3$, NH$_4$, P, alkalinity, hardness, and pH. Samples of water used for fill were collected for each pool and tested for the above parameters. National Atmospheric Deposition Program (NADP) data for the period June 1 through Nov1 2009 was obtained, and the amount of precipitation and NO$_3$, NH$_4$, and pH in precipitation recorded. Data was collected and recorded at the Penn State NADP (site id PA15) station. Evaporation pan data from the NOAA Technical Report NWS 34, 1982 was used to estimate evaporative losses from the pools. Bog plants were harvested on Oct 30 and air dried in a greenhouse until constant moisture levels were reached. All plants were pooled, for pool and specie, and then weighed. A sample of the pooled biomass was oven dried at 65.6 C (150 F) for one week then weighed and sent to the Penn State Ag Analytical Lab for %P and %N analysis. Plant tissue P % was determined using the dry ash method in Miller (1998), and %N using the combustion methods in Horneck and Miller (1998). N and P content were calculated by multiplying plant dry weight by nutrient concentration.

Pool volume was estimated by using formula 5.3
Pool volume = length x width x ave. depth (5.3)

Where pool shape was not symmetrical the pools were divided into sections and volumes estimated for each section. Bog volumes were measured separately, and adjusted for gravel displacement.

Precipitation volumes for each pool were estimated by taking the precipitation amounts recorded by the NADP and applying the following formula:

\[
\text{Precipitation} \times \text{pool surface area} = \text{total precipitation volume} \quad (5.4)
\]

Monthly evaporation rates obtained from NOAA Technical Report NWS 34, (1982) were added, and the total multiplied by the Class A pan correction factor (0.75) to find the total corrected evaporation amount. This value was multiplied by pool and bog surface area for total water losses by evaporation. This method provides an estimate only, as site conditions and the amount of stream and waterfall surface area affect evaporation. Higher waterfall surface area leads to greater loss through splash and evaporation. Still, this method gives a legitimate standard of comparison. Fill water was assumed as the difference between evaporation and precipitation volumes.

Nutrient concentrations (NO₃ and P mg/l) were obtained for rain, fill and pool water through NADP data and Penn State Agricultural Analytical Lab analysis. (ICP EPA 6010 1986) Nutrient content (g) was determined by multiplying concentrations (mg/l) by pool volumes.
Analysis Pool 1

Water Analysis

Pool 1 is a large NSP, \((\text{vol} = 237.16 \text{ m}^3)\), operating since Aug of 2008. The bog was planted Aug 15, 2008. Bog surface area is approximately 50% of the pool surface area. Water samples drawn June 1 and Nov 1, 2009 are summarized in Table 3-1.

The pool received .59 m of rain between June 5 and Nov 1 for a total volume of 153.38 m³ for the pool and bog. Rain volume introduced 151.84 g, 782 g, 16.10 g of NH₄, NO₃, and P. (See Table 3-2). Evaporation losses for June 1 though Nov 1, 2009 were estimated at .9 m for a total pool volume loss of 177.42 m³. Pool fill volume was estimated as evaporative loss – precipitation addition, or 24.05 m³. Fill water for Pool 1 was Lewisburg, PA municipal tap water. NH₄ levels were below detection for fill water, so there was no additional mass of NH₄ introduced. However fill water contributed 17.48 and 1.21 g of NO₃ and P. (Figure 3-1)

<table>
<thead>
<tr>
<th>Date</th>
<th>NO₃-N</th>
<th>P</th>
<th>Amm NH₄-N</th>
<th>pH</th>
<th>Total alkalinity CaCO₃</th>
<th>Hardness/ CaCO₃</th>
</tr>
</thead>
<tbody>
<tr>
<td>Jun 5, 2009</td>
<td>.59 mg/l</td>
<td>.09 mg/l</td>
<td>&lt;1 mg/l</td>
<td>7.8</td>
<td>50.2 mg/l</td>
<td>76.3 mg/l</td>
</tr>
<tr>
<td>Nov 1, 2009</td>
<td>1.33 mg/L</td>
<td>.13 mg/L</td>
<td>&lt;1 mg/l</td>
<td>7.8</td>
<td>75.1 mg/L</td>
<td>102 mg/L</td>
</tr>
</tbody>
</table>

Table 3-1: Water Analysis Pool 1 for June 1 and Nov 1, 2009
Vegetation Nutrient Analysis

Vegetation in Pool 1 was only in its 14th month, completing one growth cycle. Vegetation density in the bog reflects the early stage of development for the NSP so surface area coverage was poor. Plant harvest yields by species are contained in Table 3-2.

Total grams of N and P removed by all biomass equaled 167.5 and 6.96 g respectively.
Canna produced the highest biomass at 2806.6 g with *Juncus/Acorus* producing the least, 247.4 g. The algae/leaf litter portion exhibits the highest N and P concentration, 2.5 and .24%. This portion was composed of filamentous algae and leaf litter removed from the bog during harvest. The .24% P is substantially higher than the next highest plant species, an unidentified plant volunteer which contained .04 % P.

Table 3-2: Pool 1 Plant Biomass by Species, Nutrient Percent and Nutrient Content

<table>
<thead>
<tr>
<th>Pond</th>
<th>Plant</th>
<th>N%</th>
<th>P%</th>
<th>dry weight*</th>
<th>N(g)</th>
<th>P (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pond 1</td>
<td>algae/leaf litter</td>
<td>2.5</td>
<td>0.24</td>
<td>1902.2</td>
<td>47.6</td>
<td>4.57</td>
</tr>
<tr>
<td></td>
<td><em>Juncus effusus/Acorus</em></td>
<td>0.8</td>
<td>0.01</td>
<td>247.4</td>
<td>2.0</td>
<td>0.02</td>
</tr>
<tr>
<td></td>
<td><em>Hibiscus moscheutos</em></td>
<td>1.4</td>
<td>0.02</td>
<td>918.3</td>
<td>12.9</td>
<td>0.18</td>
</tr>
<tr>
<td></td>
<td><em>Iris versicolor</em></td>
<td>1.6</td>
<td>0.03</td>
<td>317.1</td>
<td>5.0</td>
<td>0.10</td>
</tr>
<tr>
<td></td>
<td><em>Eichornia</em></td>
<td>2.1</td>
<td>0.04</td>
<td>750.1</td>
<td>15.4</td>
<td>0.30</td>
</tr>
<tr>
<td></td>
<td><em>Myosotis scorpioides</em></td>
<td>2.0</td>
<td>0.04</td>
<td>969.2</td>
<td>19.8</td>
<td>0.39</td>
</tr>
<tr>
<td></td>
<td><em>Pontedaria cordata</em></td>
<td>1.3</td>
<td>0.02</td>
<td>**</td>
<td>**</td>
<td>**</td>
</tr>
<tr>
<td></td>
<td><em>unidentified</em></td>
<td>2.4</td>
<td>0.04</td>
<td>641.5</td>
<td>15.1</td>
<td>0.26</td>
</tr>
<tr>
<td></td>
<td><em>Lysimachia nummularia</em></td>
<td>1.8</td>
<td>0.03</td>
<td>1031.3</td>
<td>18.0</td>
<td>0.31</td>
</tr>
<tr>
<td></td>
<td><em>Canna xgeneralis</em></td>
<td>1.1</td>
<td>0.03</td>
<td>2806.6</td>
<td>31.7</td>
<td>0.84</td>
</tr>
</tbody>
</table>

*aboveground biomass only, harvested Nov 13, 2009
**missing data

Total grams of N and P removed by all biomass were 167.5 and 6.96 g respectively.

Canna produced the highest biomass at 2806.6 g with *Juncus/Acorus* producing the least, 247.4 g. The algae/leaf litter portion exhibits the highest N and P concentration, 2.5 and .24%. The .24% P is substantially higher than the next highest plant species, an unidentified plant volunteer. Examination of the plant nutrient analysis demonstrates the need for fall cleanup, the algae/leaf litter contained substantial amounts of P.
The most efficient species for nutrient reduction was *Canna x generalis*, (canna) for both N and P. Because of high biomass production, canna exhibited both the highest nutrient concentrations (%), and content (g). The five top species for both N and P removal were canna, water hyacinth, water forget-me-not, creeping jenny and an unidentified plant.

**Mass Balance Model**

Figure 3-2 describes the nutrient removal/storage compartments of a mass balance model for Pool 1. It is clear that key components for both nutrient deposition and storage are not accounted for, harvested biomass and Nov. pool water account for 69 and 83% of introduced N and P. Harvested biomass accounted for 48% of N added to the pool (fill and precipitation) and June water N. Unaccounted N is 92.8 g N. NO₃ increased from .59 mg/l in June to 1.33 mg/l in November. Final NO₃ concentration is well within the treatment goal of <30 mg/l.

P is much higher than the FLL treatment goal of P < 0.01 mg/l, and increased from .09 in June to 13 mg/l in Nov. 2009. Harvested biomass accounted for 15% of P added to the pool (fill and precipitation) + June P water content. (Table 3-1) Harvested biomass and November water P(g) account for 83% of total P load leaving .25 g P unaccounted. A high percentage of the accounted inputs are in the November water, 68%. Normally we would expect P to be adsorbed to substrate, and the substrate storage compartment to be the largest in the wetland. The low adsorption of P indicates that the river gravel does not contain enough Al, Fe or Ca to adsorb P.
Mass Balance for Pool 1

Figure 3-2: Mass Balance Model for Pool 1

Analysis Pool 2

Water Analysis

Pool 2 was a small NSP (37.19 m²) in operation since June of 2006 with a bog surface area of approximately 33% of the pool surface area. Water samples drawn June 1 and Nov 1, 2009 are summarized in Table 3-3.
The pool NO₃ and P concentrations increased from the start to end of testing period, although the NO₃ concentrations in November measured lower than the NSP goal of <30 mg/l NO₃. P concentrations are considerably higher than the FLL goal of P < 0.01 mg/l, and increased till the end of the experiment.

<table>
<thead>
<tr>
<th>Date</th>
<th>NO₃-N mg/l</th>
<th>P mg/l</th>
<th>Amm NH₄-N mg/l</th>
<th>pH</th>
<th>total alkalinity CaCO₃</th>
<th>hardness/ CaCO₃</th>
</tr>
</thead>
<tbody>
<tr>
<td>6/1/09</td>
<td>.5</td>
<td>.1</td>
<td>&lt;1</td>
<td>8.2</td>
<td>126 mg/l</td>
<td>124.5 mg/l</td>
</tr>
<tr>
<td>11/1/09</td>
<td>1.47</td>
<td>.15</td>
<td>&lt;1.00</td>
<td>8.3</td>
<td>146.5 mg/l</td>
<td>187.1 mg/L</td>
</tr>
</tbody>
</table>

Table 3-3: Water Analysis Pool 2 for June 1 and Nov 1 2009

Precipitation added .59 m of rain between June 5 and Nov 1 for a total bog/pool volume of 28.3 m³. Rain volume introduced 21.72, 53.1 g, 2.81 g of NH₄, NO₃, and P. (See Figure 3-3). Evaporation losses for June 1 though Nov 1 2009 were estimated at .9 m³ for a total loss of 45.1 m³. This means estimated fill volume, evaporative loss – precipitation addition, equals 23.1 m³. Well water was the fill water for Pool 2. NH₄ levels were below detection for fill water, so there is no way of calculating additional mass of NH₄. However, fill water contributed 11.56 and .9 g of NO₃ and P.
Vegetation Nutrient Analysis

Bog plants had grown 2.5 growing seasons at the time of the study. Plant coverage is good but was not yet complete. Species that produced the highest biomass (harvestable) were *Oenanthe javanica/Myosotis scorpioides* and *Juncus/Acorus* with 1267.6 g and 2067.6 g respectively, dry weight, followed by *Saururus cernuus* 1213.8g, *Hosta spp.* 678.9 g, and *I. versicolor* 522.4 g. Although the *Juncus/Acorus* mix has a higher biomass dry weight it was substantially lower in N and P removal than the *Oenanthe/Myosotis* combination, especially for P removal. The *Oenanthe/Myosotis* stored in aboveground tissue, 3 times the P content than the *Juncus/Acorus* component. See Table 3-4.
Table 3-4: Pool 2 Plant Biomass by Species, Nutrient Percent and Nutrient Content

<table>
<thead>
<tr>
<th>Pond</th>
<th>Plant</th>
<th>N%</th>
<th>P%</th>
<th>dry weight*</th>
<th>N(g)</th>
<th>P (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pond 2</td>
<td>Iris versicolor</td>
<td>1.4</td>
<td>0.03</td>
<td>522.4</td>
<td>7.5</td>
<td>0.16</td>
</tr>
<tr>
<td></td>
<td>Saururus</td>
<td>0.8</td>
<td>0.01</td>
<td>1213.8</td>
<td>10.0</td>
<td>0.12</td>
</tr>
<tr>
<td></td>
<td>Hosta sp</td>
<td>1.3</td>
<td>0.03</td>
<td>678.9</td>
<td>8.9</td>
<td>0.20</td>
</tr>
<tr>
<td></td>
<td>Oenanthe and l</td>
<td>1.7</td>
<td>0.05</td>
<td>1267.9</td>
<td>21.8</td>
<td>0.63</td>
</tr>
<tr>
<td></td>
<td>Juncus effusus</td>
<td>0.9</td>
<td>0.01</td>
<td>2067.6</td>
<td>18.8</td>
<td>0.21</td>
</tr>
</tbody>
</table>

Mass Balance Model

Figure 3-4 illustrates the measured mass balance compartments in this NSP.

Mass Balance for Pool 2

![Mass Balance Diagram for Pool 2](image_url)

Italics represent unmeasured compartments

Figure 3-4: Mass Balance Diagram for Pool 2
In this mass balance model the NSP water experiences a .74 mg/l (27.42 g) gain in NO$_3$ and a .04 mg/l (1.41 g) gain in P. Of the 34.71 g of N added to the system, biomass harvest removed 206%. Harvesting also removed 20% of the added P. Fill water, rain and initial water concentration contained 34.71 g of N. Of this, 76.39 g N was recovered in plant tissue and pool water, or a total of 234% recovery. Inputs totaling 43.8 g N were introduced to the system from some unmeasured compartment or combination of compartments. A total of 5.56 g P was accounted for by biomass and pool water in Nov., out of 6.54 g P in precipitation, fill and June water content, a removal of 85%. Final water nutrient concentration of 1.47 mg/l for NO$_3$ met treatment FLL goals, but final P water nutrient concentration failed to meet P values < 0.01 mg/l. In fact, water concentration levels for both N and P increased from the experiment start date to the end of the experiment. See Table 3-5.

<table>
<thead>
<tr>
<th>Total added Nutrient</th>
<th>Pool 2 Nutrients at Start and End</th>
<th>Total Nutrient Accounted</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>NH$_4$ (g)</td>
<td>N (g)</td>
</tr>
<tr>
<td>fill</td>
<td>11.56</td>
<td>2.61</td>
</tr>
<tr>
<td>prec.</td>
<td>21.72</td>
<td>16.9</td>
</tr>
<tr>
<td>June H2O</td>
<td>14.13</td>
<td>3.19</td>
</tr>
<tr>
<td>Nov H2O</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Harvested biomass</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>21.72</td>
<td>16.9</td>
</tr>
</tbody>
</table>

Table 3-5: Nitrogen and phosphorus content of fill water, precipitation, pool water and biomass for pool 2 in June and November 2009
Analysis Pool 3

Water Analysis

Pool 3 is a large NSP (209 m$^2$) in operation since Aug of 2008. Bog surface area is approximately 25% (53.5m$^2$) of the pool surface area. Water analysis of samples drawn June 1 and Nov 1, 2009 are summarized in Table 3-6. Water volume for Pool 3 is 295.1 m$^3$ including bog and pool.

<table>
<thead>
<tr>
<th>Date</th>
<th>NO3-N</th>
<th>P</th>
<th>Amm NH4-N</th>
<th>pH</th>
<th>total alkalinity CaCO3</th>
</tr>
</thead>
<tbody>
<tr>
<td>6/1/09</td>
<td>&lt;.5mg/l</td>
<td>0.04 mg/l</td>
<td>&lt;1 mg/l</td>
<td>7.6</td>
<td>83.3 mg/l</td>
</tr>
<tr>
<td>11/1/09</td>
<td>1.77 mg/L</td>
<td>.03 mg/L</td>
<td>&lt;1.00mg/L</td>
<td>7.8</td>
<td>137.5 mg/L</td>
</tr>
</tbody>
</table>

Table 3-6: Water Analysis Pool 3 for June 1 and Nov 1 2009

Pool 3 met FLL guidelines for NO3 concentration at 1.77 mg/l, but did not meet the FLL guidelines for P concentrations, measuring .03 mg/l. Nitrate concentrations more than doubles between June and November while P decreased from .04 to .03 mg/l. Precipitation added .59 m of rain between June 5 and Nov 1 adding a total of 154.9m$^3$ and introducing a total of 153.53, 374.86, 16.26 g of NH$_4$, NO$_3$, and P. Evaporation losses for June 1 though Nov 1 2009 are estimated at .9 m for a total loss of 179.2 m$^3$. This means estimated fill volume, evaporative loss – precipitation addition, equals 24.3 m$^3$. Pool 3 used well water as fill water. NH$_4$ levels were below detection for fill water,
so there is no additional mass of NH₄ introduced. However, fill water contributed 17.48 and 1.21 g of NO₃ and P. (Figure 3-5)

Figure 3-5: Water budget for pool 3

**Vegetation Nutrient Analysis**

Perennial bog plants were planted in Aug of 2008. To increase vegetation biomass *Eichornia crassipes*, water hyacinth, was added to the bog areas. Water hyacinth spread vigorously, achieving complete coverage of the bogs. *Juncus* and *I. versicolor* grew along the margin of the bog and pool. See Figure 3-6.
Nutrient content is a product of high biomass production and tissue nutrient concentration. As a result, water hyacinth removed approximately 9 times more N and P than either the iris or rush/sweet flag. Nutrient concentrations were also higher for the water hyacinth than the other species, 2% N and .04% P.

**Mass Balance Model**

Figure 3-6 illustrates the measured and unmeasured mass balance compartments in this NSP. The water of pool 3 experienced a 1.27 mg/l (50.17 g) gain in NO₃ and a 0.01 mg/l (2.96 g) decrease in P between June and November. Of the 137.04 g N in the system from the NH₄ and NO₃ contained in rain, fill water and June pool concentrations, biomass harvest removed 152.5%. Combined N (g) for November water N and biomass harvest was 238% of the original N amount. This leads to the conclusion there were substantial N inputs that were unmeasured.

Biomass harvesting accounted for 11% of the P present in rain, fill and June pool water. P in Nov pool water and biomass represented 33.8% of the total of P in June water, fill and rain. P storage was largely unaccounted. Other possible compartments for P are algal and bacterial uptake. Water hyacinth was
able to remove the entire N measured inputs, but did not remove enough P to balance the pool. The water still contained 8.85 g P, so P was available for plant uptake.

![Mass balance diagram for pool 3](image)

*italics denote unmeasured compartments*

**Figure 3-6:** Mass balance diagram for pool 3

**Conclusion**

All pools met NO$_3$ FLL guidelines but none met FLL guidelines for P, namely P < 0.01 mg/l. Pool 1 was a young system, plants had not had much chance to become established and bog coverage was poor. It is possible that as the NSP matures, function would improve. This possibility is supported by research that shows a relationship
between maturity of a wetland and plant N storage. Harvestable biomass in the two older NSPs in this research exhibited high N storage, removing over 150% of the measured N inputs. This compares to the NSP that had only been in operation for 9 months where harvestable N accounted for 48% of N. The difficulty of predicting the amount of nutrients plants remove through direct uptake is compounded by factors affecting uptake, climate, age of plant, plant health, growth stage, and substrate, loading rate of nutrients.

Most likely, plants have limited function as P storage compartments, even with plant harvesting. Research has shown that while plants account for much of the N storage in a wetland, they represent a small fraction of the P removal (Lucas and Greenway 2009). Kadlec and Wallace (2009) find that plant filters are most important in low load systems, such as NSPs, and state that most research records removals for P of less than 10% through harvesting. For the pools in this study, plant harvesting was not as efficient for P removal as N removal, but still removed between 11% and 20% of P inputs. (Table 3-7) It is clear that plant harvesting is important to N and P removal in a NSP, but the amount removed by harvest is not consistent between pools or plant species. It is likely that vegetative filters alone are not capable of removing P to levels \( \leq 0.01 \) mg/l.

<p>| Nutrient content (g) of harvested biomass and % of inputs removed by harvest |
|---------------------------------|----------------|----------------|-----|-----|</p>
<table>
<thead>
<tr>
<th>Pond</th>
<th>Total N harvested</th>
<th>Total P harvested</th>
<th>%N</th>
<th>%P</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>167</td>
<td>6.96</td>
<td>48</td>
<td>15</td>
</tr>
<tr>
<td>2</td>
<td>67</td>
<td>1.32</td>
<td>206</td>
<td>20</td>
</tr>
<tr>
<td>3</td>
<td>209</td>
<td>4.26</td>
<td>152</td>
<td>11</td>
</tr>
</tbody>
</table>
Since a NSP serves the dual purposes of a backyard amenity and nutrient reduction technology, one of the design requirements is effective, timely nutrient control. So the question is, how can we improve early function of this technology, so that it not only looks good but also provides a quality recreational experience?

The primary role of vegetation in NSPs may not be in nutrient uptake but in other wetland ecology functions (Tanner 2001, Kim and Geary 2001). Since P removal is especially important to a NSP system because of its effects on primary production (algae), substrate is considered a key component of system filtration and nutrient reduction. Substrates other than river gravel may provide improved P removal. Some of these potential substrates include, Norlite, iron, EAF slag, lightweight expanded clay (LECA), and calcined clay (Vohla et al. 2005, Vohla et al. 2007, Hill et al 2008, Erikson et al 2008). Substrates other than 2b river gravel may also decrease establishment time for plants.

This field study provided a window into the function of three NSPs of various vegetation types, size and maturity. While all the NSPs were capable of meeting FLL guidelines for N removal, none achieved P target levels. Removal efficiencies were also highly variable. Because there existed large unaccounted compartments of N inputs and smaller unaccounted P and N output compartments, it is clear that this model does not fit the system and should be expanded. Expanding the model to include substrate and increasing control of nutrient inputs and outputs should help to identify some of the

Table 3- 7: Nutrient content (g) of harvested biomass and % inputs removed by harvest
complex processes critical to nutrient removal in a NSP. Further greenhouse studies are required to monitor inputs and outputs, assess plant vigor and control the operating environment.

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Chapter 4

The Relationship of Four Substrates to N and P Removal and Biomass Establishment in a Greenhouse Natural Swimming Pool Experiment

Introduction

Natural swimming pools (NSPs) are recent technological introductions to North America, designed to provide an attractive landscape amenity that also serves as a chemical free swimming pool. Based on constructed wetland technology, natural swimming pools use biological filtration, a designed, intentional hydraulic system, and an impermeable liner to separate the system from the surrounding natural hydrologic cycle. Biological filtration involves the use of plants and substrate to closely replicate natural wetland environments. Biological filtration creates conditions that encourage the establishment of organisms, plants and bacteria to filter excess nutrients and harmful bacteria from aquatic systems, without the use of harmful chemicals. These systems offer a variety of ecosystem services, supplying wildlife habitat and four season aesthetics, fostering stewardship, eliminating ecologically deleterious chemicals and supporting a diverse vegetative community.

Nutrients of particular concern for natural swimming pools are nitrogen and phosphorus, because of their association with eutrophication and subsequent high algae levels. In natural freshwater bodies, the USEPA mandates that NO$_3$-N levels be $\leq 10$ mg/l and recommends TP levels less than .1 mg/l (US EPA 1986). However, P levels as low as .02 mg/l are implicated in nuisance algal blooms (Daniel et al. 1998). In Europe,
recent standards developed for NSP operation specify levels for N and P of 30 mg/l (NO$_3$) and 0.01 mg/l P respectively (FLL 2003).

Many constructed wetland studies indicate an increase in nutrient removal with vegetated biofilters compared to unvegetated, in wastewater treatment (DeBusk et al. 1995), nursery runoff treatment (Berghage 1999; Chen & Bracy, 2009; Huett 2005; Taylor et al. 2006), and stormwater filtration ((Bratieres et al. 2008; Browning & Greenway, 2003; Greenway & Jenkins 2004; Hatt, Deletic, & Fletcher 2007; Henderson, Greenway, & Phillips, 2007; Johengen & LaRock, 1993; Moortel et al. 2010; Read et al. 2008; Roy-Poirier et al. 2010; Zhang et al. 2011). No studies, however, have examined the nutrient removal capabilities of the NSP biofilter. NSP systems differ from other nutrient reduction systems because they are closed systems, sealed to prevent infiltration and exchange with the surrounding hydrology, are recreational and aesthetic landscape amenities and nutrient loadings are low. Because NSPs are landscape amenities, the biofilter vegetation must fill the dual purposes of efficient nutrient reduction and ornamental enhancement. In addition, values such as fostering stewardship, and environmental assets including plant diversity and wildlife habitat suggest that plant monocultures, usually found in wastewater treatment, are not the optimum choice for NSP systems.

Ornamental obligate and facultative wetland species have been studied for nutrient removal in bioretention systems, (Chen and Bracy 2009, Maschinski et al. 1999), and constructed wetlands (DeBusk et al. 1995; Read et al. 2008, Polomski et al. 2007),
but no research has quantified the nutrient removal and establishment of ornamental, native obligate wetland species in natural swimming pools.

This study quantifies the contribution of two native, obligate wetland plants, *Iris versicolor* (blue flag) and *Saururus cernuus* (lizards-tail). *I. versicolor*, is an obligate, native wetland plant commonly found on lakeshores and in wetlands, often growing in shallow water. In many wetlands, the aggressive, non-native *Iris pseudacorus* (yellow flag iris) has replaced native blue flag iris, forming dense colonies and reducing plant community diversity. Although these attributes make *I. pseudacorus* well suited for nutrient removal, (Chen and Bracy, 2009, Dunne et al. 2005, Calheiros et al. 2007, Berghage et al. 1995) they are less well suited for a landscape amenity providing diverse ecosystem services. Other species in the iris genera studied for nutrient removal include *Iris ensata* (Lamchaturapatr et al. 2007) and *Iris* ‘Louisiana Hybrids’ (Polomski et al. 2009). *I. versicolor* was only incidental to research conducted by Drizo et al. (2008), Keppler and Martin (2008), and Kohler et al. (2004), and iris biomass nutrient removal was not quantified. This research quantifies the performance of *I. versicolor* in a NSP biofilter.

Lizards-tail is an obligate, native wetland plant often found along river shores and the margins of marshes and streams. Lizards-tail has fragrant, white inflorescences and can grow 3-4’ tall, frequently producing high standing biomass (Mitsch and Gosselink 2007). Mitsch and Gosselink (2007) recommend lizards-tail for constructed wetlands used to treat stormwater and agricultural wastewater.
The high above ground production and attractive appearance of both blue flag and lizards-tail make each a good candidate for a NSP biofilter, which requires a high aesthetic standard and ability to remove nutrients through high uptake and standing stock harvest. Since this study is also interested in ecosystem services, plants chosen for inclusion are native to the Northeastern US. A third species, *Pontedaria cordata* (pickerel weed) was originally included in the study, but replaced by *I. versicolor* when the plants failed to become established. Because most studies that investigate species effect on nutrient removal efficiency are unable to determine if the effect is species or health/growth related (Brisson and Chazarenc 2009) this study measures plant health using both biomass weight and growth index to assess removal efficiency relationships. Plant growth index provides NSP designers with the ability to specify plants tolerant of the specific nutrient levels, pH and operating conditions found in NSPs. Rapid establishment is unlikely for a plant with low growth index measures, making it a poor candidate for a NSP.

The two types of constructed wetlands most often used for nutrient remediation, free water surface (FWS) and subsurface flow (SSF) are effective for NSP biofilters. FWS wetlands, characterized by a water level of varying depths over the wetland substrate, typically contain emergent vegetation, commonly *Typha spp.*, *Scirpus spp.* or *Phragmites spp.* Traditionally FWS substrate is coarse river gravel, providing good hydraulic conductivity and some physical support for emergent plants. Although normally the FWS wetland vegetation consists of emergent plant communities, submergent and free-floating plant communities may also populate these wetlands.
(Kadlec and Wallace 2009). In contrast, the water level of a SSF wetland is below the surface of the substrate and emergent plants established in a shallow mulch or soil layer on top of the substrate (Kadlec and Wallace 2009). Nutrient removal capabilities are similar for both, but they differ in surface area requirements, aesthetics, control of algae and other nuisance organisms and accompanying ancillary values (Tanner 1996, Kadlec 2009, Knight et al. 2001). In the case of a NSP, both FWS and SSF systems have potential advantages and disadvantages. In consideration of these competing, but equally valid values, both types of wetlands are included in this research.

While many studies agree that vegetated filters increase nutrient removal, nutrients are released back into the system, 30% within the first few days of senescence, if plant above ground portions are not harvested (Vymazal 1995). FLL guidelines for NSP operation recommend removal of aboveground tissue immediately after fall dormancy to prevent leaching of nutrients back into the system.

Constructed wetlands are often efficient at removing nitrogen, but P can be problematic. NSPs, with P removal targets of < 0.01mg/l (FLL 2003) are especially challenging (FLL 2003). Evaluation of alternative media for P removal shows removal improvements with the incorporation of a clay-based material (White et al. 2011, Brix et al. 2001, Chang et al. 2010), materials high in Al, Fe or Ca (Brix et al. 2001, DeBusk et al. 1997, Hsieh and Davis 2006) and lightweight expanded clays (LECA) and expanded shales (Vohla et al. 2011). Despite excellent P removal performance, some alternative media create secondary issues, such as high pH, poor hydraulic conductivity and rapid P saturation. These constraints limit the usefulness of the media. Electric arc furnace slag
(EAF), high in CA, AL and Fe show removal rates of 78% to 98% for P (Naylor et al. 2003, Cameron et al. 2003). However Naylor et al. (2003) found EAF increased pH above levels able to sustain a healthy aquatic system. Most natural wetlands operate from pH 7-8, except for acidic peat based wetlands, while bacteria needed for nitrification and denitrification prefer a pH of 6.5 to 7.5 (Kadlec and Wallace 2009). Phosphorus reactions, specifically precipitation and adsorption, are very pH dependent as are decomposition and nutrient availability to plants. Therefore, ideal substrate choices for NSPs have high hydraulic conductivity, high P adsorption capacities, are able to maintain adequate plant growth, have long term P saturation potential, do not clog, maintain a reasonable pH and are easily available and cost effective.

In this NSP experiment, media targets were P removal and biomass accumulation. The four media in this research were selected for their nutrient removal abilities and support for biomass establishment and growth, cost effectiveness and availability. Each is currently used for nutrient remediation in greenroofs, aquaculture or constructed wetlands, and has potential for use in natural swimming pools. The control is local river gravel, commonly used in natural swimming pools, constructed wetlands and bioretention. The other three substrates are growstone, haydite and norlrite. Growstone, a recycled glass product having a high surface area to volume ratio, is first finely ground followed by the application of a natural foaming agent, 1.0 – 5.0 % of calcium carbonate. Growstone’s high surface area ratio, calcium carbonate, and extremely lightweight nature make it an attractive candidate for phosphorus removal. Haydite is a lightweight expanded shale amended for this study with calcined clay. Haydite’s chemical content
includes 9.6% ferric oxide, 19.4% alumina and 3.4% calcium oxide, important for P removal. Norlite (ESCE), often used in greenroofs, bioswales and wastewater treatments, is a lightweight coarse aggregate of fired shale, clay and slate. All are easily obtainable and cost effective. All four substrates are summarized in Table 4-1.

<table>
<thead>
<tr>
<th>Substrate</th>
<th>Composition</th>
</tr>
</thead>
<tbody>
<tr>
<td>River Gravel</td>
<td>neutral, depends on local source</td>
</tr>
<tr>
<td>Growstone</td>
<td>recycled glass 95-99+%, Calcium carbonate 0.5 - 5%</td>
</tr>
<tr>
<td>Haydite/clay</td>
<td>fired shale,(9.6% ferric oxide, 19.4% alumina, 3.4% calcium oxide, magnesium oxide 5.6%, silica 57.6%)</td>
</tr>
<tr>
<td>Norlite</td>
<td>fired shale, clay, slate (Silica (SiO2) 64.20%, Alumina (Al2O3) 20.24%, Iron Oxide (Fe2O3) 4.86%, Titanium Oxide (TiO2) .70%, Calcium Oxide (CaO) 2.00%, Magnesium Oxide (MgO) 3.62%, Alkalies 3.16%, Sulphur Trioxide (So3) .69%)</td>
</tr>
</tbody>
</table>

Table 4-1: Composition of 4 media used as substrate in study of the effect of substrate on nitrogen and phosphorus removal in a natural swimming pool.

A mass balance approach estimates system nutrient inputs and nutrient removal and storage through various compartments. This approach quantifies system performance and the contribution of various nutrient removal compartments and allows the development of simple predictive models (Breen 1990). Mass balance provides a good measure of nutrient removal because it considers evapotranspiration. Removals are calculated using actual nutrient amounts, not concentrations. As a tool, it is widely used
in constructed wetland and bioretention research (Johengren 1993, Breen 1990, Borin and Salvato 2012).

**Materials and Methods**

This greenhouse study was conducted from Feb 2012 to June 2012 at The Pennsylvania University Department of Horticulture greenhouse complex at University Park, Pennsylvania (lat. 40°8019'N, long 77°8627'W). Plant plugs obtained from Maryland Aquatics Nursery on Sept 29, 2011 were placed in tubs and maintained in an actively growing condition until the experiment started. Plants selected were *I. versicolor* (blue flag iris), *P. cordata* (pickerelweed), and *S. cernuus* (lizard’s tail). Plants were fertilized with Peters 15-15-15 monthly until replanted in experimental mesocosms. All plants were weighed Nov 9, 2011 and planted in the mesocosms to allow plants time to acclimatize. All units in this experiment received one plant each of lizard’s tail, pickerelweed and blue flag. The pickerelweed failed in all units and each was replaced by one blue flag, on Feb 2, 2012, resulting in a final mix of 2 Iris and one lizard’s tail. Baseline weights were obtained for and N and P content measured. The initial measurements were subtracted from final weight and nutrient content for mass balance calculations.

The experiment is a complete randomized design with two conditions. Figure 4-1 shows the experimental design. Four substrates were randomly assigned to eight experimental units, four of the eight units assigned to subsurface flow conditions, and
four to free water surface flow conditions. Units were randomly assigned their location
on two benches.

A hole drilled above the media in the bog container allowed the water to return
via gravity flow to the pool unit. Each treatment had eight bog units, four free water
(FWS) and four subsurface (SSF) flow. The FWS flow return water hole was located 2”
above the bog surface, while the SSF drainage hole was located at the top of the substrate
and under 2” of 3/8 to ½” pea gravel. Bogs were elevated 1’ above the pool units. Both
pool and bog units were rubbermaid containers (69L, 61 cm x 47 cm x 40 cm) filled with
45.6 L DI water or the bog materials indicated by each treatment. Figure 4-1 shows
design setup and Figure 4-2 illustrates greenhouse layout.

Water pumped at a rate of 22.5 L per 12 hrs, resulted in total turnover once every
24 hrs. Artificial lights controlled by a timer were on from 5:00 pm to 10:00 pm daily to
augmented natural lighting. Greenhouse temperature was kept at 82 F during the day and
76 F at night. Water levels were adjusted to a consistent level, checked weekly and DI
water added as necessary, with the volume of each addition recorded. On February 13,
2012 each container was fertilized with 6 g of 20-10-20 granular fertilizer. The same
amount was applied on March 2, 2012, April 23, 2012 and May 25, 2012.
Figure 4-1: Experimental unit design for natural swimming pool and bogs in the experiment ‘The Relationship of Four Substrates to N, P Removal and Biomass Establishment in a Greenhouse Natural Swimming Pool Experiment’
Fifty ml water samples were drawn weekly from the pool units and measured for relative algae levels on a Bausch and Lomb Spec 20 colorimeter. The water samples were also measured for nitrate, pH, EC and water temperature using Forstan LabNavigator, (Synaptic Sensors, LLC, 401 Mathews St., Fort Collins, CO) probes. Probe calibration occurred before each sampling event and measurement accuracy verified using spiked samples. Assessments of plant vigor, included using fan length and numbers for iris and plant height and number of shoots for lizards-tail. These measurements develop a method of assessing plant health and its correlation to a plant’s nutrient removal ability. Chung (2008) used the same measures when assessing plant health for Typha spp.

Additional water samples were drawn every 2 weeks, (50 ml) and filtered through a 45μm filter and P was determined by the colorimetric molybdenum blue method (Murphy and Riley, 1962). Prepared samples were measured on a Perkins Elmer Lambda 25 spectrophotometer with UV WINLAB V 2.85 software, Tyson Building, University Park, PA. All samples were filtered immediately, but if samples could not be tested immediately after filtering, they were stored in a freezer until testing could be done.

At the end of the experiment, a final 50 ml water sample from each of the 32 pool units was analyzed on a Bausch and Lomb spec 20, Dept of Plant Science, HH2 Lab, University Park, PA, for chlorophyll a absorbance followed by P analysis. NO₃, P, pH, water temperature and EC were measured for each pool unit. Water from the bog and pool for each unit was weighed on a field scale to determine final volume for mass
balance purposes.

The collected 50 ml substrate samples sent to the Penn State Ag Analytical Lab were analyzed using the combustion method from Bremer 1996 TN and P was analyzed using the Mehlich 3 extraction method (Recommended Soil Testing Procedures for the Northeastern United States 2009).

Plants were harvested on June 5, 2012 and rinsed with DI water so the roots were free of substrate. Final plant vigor measurements were obtained, measuring shoot length and shoot number for lizard’s tail and number and height of fans for iris. These growth index measurements were averaged and reported by unit. The plants were pooled for each unit and weighed. Plants placed in a dryer for a week, were then weighed, and a sample extracted from the pooled samples and weighed again. These samples were sent to the Penn State Ag Analytical Lab and analyzed for %P and %N. Plant tissue P was determined using dry ash method in Miller (1998), and N using the combustion methods in Horneck and Miller (1998). N and P content were calculated by multiplying plant dry weight by nutrient concentration.

Aphid infestation was problematic throughout the experiment. Control strategies ranged from wiping down each plant with 2% insecticidal soap to releasing predators in the greenhouse. Use of biologicals failed to control aphids, possibly impacting plant growth. Iris exhibited symptoms of iron deficiency, namely interveinal chlorosis, resulting in application of iron chelate, Sprint 330 (10% iron), as a foliar spray on Apr 11 and Apr 23, 2012. Lemna was introduced to the system from the roots of the plants and became established in a few units. Lemna was regularly removed and analyzed for TN
and TP at the end of the experiment. It is reported separately and not included in the mass balance calculations.

**Statistical analysis**

Data from nutrient removal compartments, plant biomass, substrate and pool water, was analyzed with a univariate analysis of variance (ANOVA) using SPSS (version 20 for MAC, IBM, Armonk, New York). A two-way fixed effect ANOVA model was used to determine interaction effects between treatment and water flow and a further multiple comparison analysis of substrate effect using Tukey’s method. Repeated measure analysis of variance was conducted to determine the effect of time on nutrient removal. Substrate was considered a between-subject effect and dates a within-subject factor. Q-Q charts, residual plots and Levene’s Test assessed model fit to ensure necessary assumptions of normality and constant error variance were met. Bivariate correlation and Pearson’s correlation determined the relationship of biomass and nutrient removal. When assumptions of normality were not met, two nonparametric correlation tests, Kendall’s Tau and Spearman’s rho were employed. The significance level (α) used to reject the null hypotheses for all tests is 0.05.

Nutrient removal efficiencies were calculated by using the ratio of the mass removed (nutrient in final biomass plus nutrient in substrate at end) to the mass added by fertilization. Total mass removed was calculated by subtracting the nutrient mass present in the plant biomass and substrate from the mass added to the system. Biomass accumulation was contrasted with nutrient uptake efficiency. Calculation of biomass
nutrient content was accomplished by multiplying the plant dry weight by nutrient percent.

**Results and discussion**

**Biomass Accumulation**

Treatment exhibited a highly significant effect on biomass accumulation (harvested biomass dry weight – initial biomass dry weight) with a p < .001. Treatment 2, Growstone, was the least productive treatment with a dry biomass mean of 142 g compared to 313 g for Haydite/clay. Control (river gravel) and haydite/clay were very similar statistically although there was less biomass variability in the haydite/clay treatment. Biomass for iris was significantly greater than lizard’s tail for all units. Because of poor growth, lizard’s tail contributed minimally to total biomass per unit. The lizard’s tail exhibited extremely slow establishment, and did not start growing until quite late in the experiment. Lizard’s tail shows a slow dormancy break and responds to dormancy cues quite strongly in the fall, (Billings 2012). Iris responded quickly to growing conditions and experienced rapid biomass accumulation after planting. Plant health measures show vigorous growth in Iris, both height and fan number, and slow growth in the lizards tail.

Although flow condition, (FWS vs. SSF) did not have a significant treatment effect on biomass, SSF biomass was greater than FWS biomass for every treatment. (Figure 4-3) This was consistent with findings in Arnold et al. 1999, where growth of *I. pseudacorus* was greater in SSF treatments than FWS treatments, with iris heights an average of 143cm for SSF versus 131 cm in FWS. In this study, blue flag weights were
24 g, 137g and 48g heavier for FWS flow conditions in control, Growstone and Norlite, but 164g greater in the SSF condition for Haydite/clay. Iris and lizards tail heights at the end of the experiment are listed in Table 4-2.

Figure 4-3: Mean total biomass in dry wt (g) by substrate and flow condition at end of study ‘The Relationship of Four Substrates to N, P Removal and Biomass Establishment in a Greenhouse Natural Swimming Pool Experiment’. 
Although flow condition, (FWS vs SSF) did not have a significant treatment effect on biomass, SSF biomass was greater than FWS biomass for every treatment. (Figure 4-3) This was consistent with findings in Arnold et al. 1999, where growth of *I. pseudacorus* was greater in SSF treatments than FWS treatments, with iris heights an average of 143 cm for SSF versus 131 cm in FWS. In this study, blue flag weights were 24 g, 137 g and 48 g heavier for FWS flow conditions in control, growstone and norlite, but 164 g greater in the SSF condition for haydite/clay. Iris and lizards tail heights at the end of the experiment are listed in Table 4-2.

Table 4-2: Mean and SD of total biomass, height and growth index for treatments at end of study ‘The Relationship of Four Substrates to N, P Removal and Biomass Establishment in a Greenhouse Natural Swimming Pool Experiment’

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Biomass (g)</th>
<th>ave iris ht(cm)</th>
<th>lizards tail height(cm)</th>
<th>Growth Index*±SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>river gravel</td>
<td>301.4+-118.2</td>
<td>160.7</td>
<td>17.5</td>
<td>771±280</td>
</tr>
<tr>
<td>growstone</td>
<td>142 +/- 115</td>
<td>111.2</td>
<td>32.5</td>
<td>331±203</td>
</tr>
<tr>
<td>haydite/clay</td>
<td>312.75+/-.86</td>
<td>192</td>
<td>32.5</td>
<td>694±258</td>
</tr>
<tr>
<td>Norlite</td>
<td>235 +/- 46.2</td>
<td>194.8</td>
<td>16.3</td>
<td>593±135</td>
</tr>
</tbody>
</table>

*i*iris Gl= ht*# fan
lizards tail Gl= ht* # of shoots
Treatment averages for iris height ranged from 111 cm to 195 cm. Lizard’s tail measurements were more modest with treatment height averages of 16 cm to 36 cm. It is quite interesting that the river gravel treatment produced the highest growth index, 771, followed by the haydite/clay with 694. Both were substantially higher than the other treatments, especially growstone with a growth index of 331. Biomass weights and growth index were correlated in this study, (Pearson’s Correlation Coefficient = .028) and all nonparametric correlations were highly significant (p<.001). The high correlation indicates that growth index is a reasonable means of comparing plant health.

The following figures, 4-4 and 4-5 show the relationship of iris and lizards tail growth index to treatment and date. Julian dates correspond to the following dates: 4/12/12, 4/18/12, 5/5/12, 5/24/12. Lizards tail in particular exhibited different growth index by treatment, growth index in river gravel outperforming all other treatments.
Figure 4-4: Iris growth index by treatment for Julian dates 12103 (Apr 12, 2012), 12109 (Apr 18, 2012), 12126 (May 5, 2012), 12145 (May 25, 2012)
Figure 4-5: Lizard’s tail growth index by treatment for Julian dates 12103 (Apr 12, 2012), 12109 (Apr 18, 2012), 12126 (May 5, 2012), 12145 (May 25, 2012)

N and P Recovery

Lizards tail nutrient concentration was greater than iris in every treatment, although because iris biomass production is higher in every treatment, the actual nutrient removal was greater with iris. Previous studies measuring nutrient reduction in vegetated filters have discovered lizards tail to have a limited contribution to biofilters performance. In a study measuring nutrient mitigation in stormwater ditches, Saururus removed the lowest percentage of NO$_3$ for five species and third of five for P removal (Moore and Kroger 2010). DeBusk et al. (1995), measured similar P concentrations for Saururus, .29% for unenriched and .44% for enriched conditions, compared to .50% for this study. In an earlier field natural swimming pool (NSP) study where autumn
harvested NSP bog plant biomass was analyzed for nutrient content, lizards-tail and iris had N and P concentrations of .82 and 0.01 percent and 1.37 and .03 respectively. These concentrations reflect plants harvested in the fall from three NSP biofilters. These biofilters varied in size, but were all harvested after a full years establishment. This preliminary data seems to indicate that plants grown in the relatively low nutrient environment of a NSP have lower tissue nutrient concentrations and that a difference in nutrient uptake probably exists between greenhouse and field plants. One possible explanation for this is the different allocation ratios between roots and shoots in high nutrient and low nutrient environments. Iris tissue concentrations of N ranged from 1.63% to 1.93% and for P,.3% to .41%. There were no treatment differences for N tissue concentration but a significant difference exists between the Haydite/clay treatment and all other treatments for iris P tissue concentration. Polomski et al. (2007) reported similar N tissue concentration levels for a Louisiana iris hybrid, although the P tissue concentrations reported in that study were well below those of this study, ranging from .14% to .29% instead of the .3% to .41% in this research. However, both studies report concentrations in the range of expected P tissue values of .05 to 1%. Treatment averages for N and P concentrations for total unit biomass are shown in Figures 4-8, 4-9. Polomski (2007) iris N concentrations were .91% and .7% for a treatment with an input of .39mg/l contrasting with the concentration means in this study of .4, .41, .30 and .41% for Treatments 1 through 4 respectively. In Polomski (2007), the .39mg/l was batch loaded at the start of the study and added every 2 days throughout the 8 weeks of the study to keep water levels consistent. In this study, the nutrient additions were
introduced four times during the 15 weeks of the experiment. Biomass concentration differences for iris tissue could be attributable to loading regimen, species difference and stage of growth. Species difference could also affect nutrient uptake. Polomski (2007), used *I. pseudacorus*, a vigorous invasive iris found commonly in wetlands, while the iris in this study was *I. versicolor*, a native iris with less vigorous growth habit. Plants in early rapid growth experience a different rate of uptake than an established plant. Allocation of P and N is also different between root and shoot depending on nutrient regimen and stage of establishment.

Figure 4-6: P mean % biomass concentration at experiment end by treatment and flow condition (free water and sub surface flow)

Percent of nutrients recovered by biomass of total nutrients removed for each treatment, ranged from 46% for growstone and 92% for haydite/clay and norlite for P,
and 28% to 72% for Growstone and haydite/clay respectively for N. The control treatment of river gravel had an assumed percent recovery of 100% for N and P by biomass, because of the low surface area ratio of the gravel and its lack of chemical attraction for P. This is reasonable for N also, as other studies have found that N in gravel is a small component of the mass balance in vegetated wetland mesocosms (Borin and Salvato 2012). To verify this assumption, the uptake efficiency was calculated for N and P across all treatments. The uptake efficiency, calculated by dividing the total P in biomass by total P applied to the unit is compared to the optimal uptake rate, 100% of applied nutrients. (Table 4-3) Uptake efficiency for the control treatment was 101% and 49% for N and P respectively.
This indicates while N is totally removed by plant uptake in this treatment, much of P is removed through other mechanisms than plant uptake. Uptake efficiency for the other treatments ranged from a low of 48 and 22 percent in growstone, 87 and 37 in norlite and 104 and 38 in haydite/clay for N and P. Uptake efficiency for iris exceeded that of lizards tail. Uptake efficiency was based on total plant biomass, not just shoots. Iris grown in N concentrations between 10.4 to 21.6 mg/l had greater concentrations of N in shoots than roots in Polomski et al. (2007). The iris in that study was the native
Louisiana iris, while in this study, it is blue flag, but results appear to follow the same trend.

Table 4-3: Nitrogen and phosphorus uptake efficiency by treatment

<table>
<thead>
<tr>
<th>Treatment</th>
<th>N uptake efficiency</th>
<th>P uptake efficiency</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>101.42</td>
<td>48.75</td>
</tr>
<tr>
<td>Growstone</td>
<td>47.53</td>
<td>21.75</td>
</tr>
<tr>
<td>Haydite/clay</td>
<td>104.21</td>
<td>37.95</td>
</tr>
<tr>
<td>Norlile</td>
<td>92.93</td>
<td>38.78</td>
</tr>
</tbody>
</table>

In terms of harvestable biomass, the iris had considerably more biomass production in all treatments than lizards tail. While the lizards tail had a higher percent N and P than iris, it was not as good a choice for nutrient reduction because it doesn’t produce enough biomass to remove significant amounts of nutrients, or to improve the removal rate through harvesting.

Given the low biomass production in the growstone units, it is reasonable that the growstone treatment has the lowest nutrient removal percentage for N. (Table 4-4) growstone has a porous surface due to its manufacturing, with a resultant high surface area. The high surface area to volume ratio increases adsorption potential and this treatment removed 99.7% P from the pool water, but plants in the growstone treatment were probably unable to access the P held by the substrate. The haydite/clay treatment had the best nutrient removal percentage from solution for both P and N at 99.95 and
94.46 respectively. All treatments had high final P removal percentages by concentration, over 99%, but ranged from 50% to 95% for N removal (Table 4-5). P removal results are high compared to literature values and hard to explain considering P removal is highly dependent on physio/chemical processes. Because P removal mechanisms are largely dependent on pH, surface area and metal content, determining the ability of the substrate to adsorb and precipitate P, it is logical to expect some P removal differences between treatments based on chemical characteristics of the substrates. Other studies measuring P removal from vegetated mesocosms have ranged from less than 50%, (Braskerud 2002, Fink and Mitsch 2007) to greater than 75% (Tanner 1996). Henderson, 2008 reported removal efficiencies for P of 90 – 100% for four vegetated treatments of different substrates, loam, sand, gravel and gravel-sand. Taylor et al. (2006), reported that P removal varied widely in a 38 month study of a FWS wetland treating nursery runoff, from an export of 39.8% to assimilation of 30.4%. The P effluent concentration measured in Taylor (2006) averaged 1.41 mg/l PO4, nowhere near the levels needed for optimum NSP operation. FLL standards recommend N and P concentrations of < 30mg/l and < 0.01 mg/l. It is possible that this NSP study would experience less efficient P removal if conducted in the field and over a longer time period

<table>
<thead>
<tr>
<th>Treatment</th>
<th>P recovered %</th>
<th>N recovered %</th>
</tr>
</thead>
<tbody>
<tr>
<td>river gravel</td>
<td>100</td>
<td>91.7 +/- 23.4</td>
</tr>
<tr>
<td>growstone</td>
<td>46.2 +/- 21.7</td>
<td>55.6 +/- 13.1</td>
</tr>
<tr>
<td>haydite/clay</td>
<td>91.7 +/- 4.3</td>
<td>79.7 +/- 4.2</td>
</tr>
<tr>
<td>Norlite</td>
<td>92.1 +/- 4.7</td>
<td>71.3 +/- 17.3</td>
</tr>
</tbody>
</table>

Table 4-4: Percent recovered P and N by biomass of total N and P removed
Final P (mg/l) in the pool water was highly significantly different by treatment with a p < .001. Although all removals were high, only the haydite/clay treatment reduced P in the water to the target level of 0.01 mg/l. All other treatments were above the recommended FLL level of 0.01 mg/l.

All final water NO₃ levels, except for growstone, reached target levels of NO₃, (NO₃ < 30 mg/l), as set by FLL guidelines. Haydite/clay reduced NO₃ to 8.5 mg/l while growstone levels were reduced to 53 mg/l, well over FLL recommendations. Nutrient recovery rate for N was calculated by dividing the total outputs (N in substrate and final biomass) by total inputs (N in substrate and biomass at the start and N added to the system).

Biomass production is important for several reasons. Since plant coverage is important to biofilter function, the quicker coverage is obtained the better the filtration. From a user standpoint, most clients do not want to wait a season to use their new pool and desire it to become established with a minimum of downtime.

Statistical analysis shows that substrate had a significant effect on both N and P removal. This was not unexpected and is supported by results from Chung et al. (2008), Browning and Greenway (2003), Brix et al. (2001), just a few of the studies that have explored substrates and nutrient removal in constructed wetlands and bioretention technologies. N removal is probably correlated with substrates because of substrate support or inhibition of biomass production. Treatment effect was significant for nitrogen removal at p < .008 and data demonstrated a strong correlation between nitrogen
removal and biomass production.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>P removal %</th>
<th>N removal %</th>
</tr>
</thead>
<tbody>
<tr>
<td>river gravel</td>
<td>99.75+/-.11</td>
<td>73.16+/-.11</td>
</tr>
<tr>
<td>growstone</td>
<td>99.68+/-.16</td>
<td>59.66+/-.16</td>
</tr>
<tr>
<td>haydite/clay</td>
<td>99.95+/-.06</td>
<td>95.56+/-.65</td>
</tr>
<tr>
<td>Norlite</td>
<td>99.91+/-.08</td>
<td>80.21+/-.14</td>
</tr>
</tbody>
</table>

Table 4-5: Final mean and standard deviation of the percent treatment removal

Figure 4-10 illustrates P removal rates after application of 6 grams of 20-10-20 fertilizer to the swimming pool units. Applications on the dates of Feb 13, March 2, April 23 and May 25 2012 resulted, in most cases, with a spike of P in the pool water followed by a rapid modulation of P concentrations. The exception is the April 23rd addition, which raised the P level considerably and took almost 2 weeks to remove from solution. Possible explanations include the addition of insects and tap water to the systems when the plants were sprayed with tap water to knock down a dual infestation of spider mites and aphids. Two applications of Sprint 330 iron chelate were sprayed on all units to ameliorate symptoms from low nutrient levels in the pool water. Spray was applied both April 11 and April 23, 2012 and ladybugs released to control aphids. Unfortunately, a high proportion of the ladybugs died and ended up in the pool water, also contributing to nutrient addition. However, once the system adjusted, P removal reached rates of 98% or above for all systems. It seems clear that as the systems mature, P removal occurs at a faster rate after addition of P.

In this study, P removal is not correlated with water pH or temperature. Water pH
is kept stable in this study since only DI water is used as fill. Though biofilms established on the surface of the containers and substrate, the chlorophyll a measurements with the Spec 20 showed planktonic algae was not an issue in most units. Since pH is modulated by algal respiration and photosynthesis, the low algae levels contributed to steady pH levels (Vymazal 1995, Mitsch and Gosselink 2007).

N removal rates also improved over time, (Figure 4-11) but became most efficient in the haydite/clay treatment, with the haydite/clay treatment ultimately removing 95% of NO$_3$ in the water. Not only is the removal efficiency high but it also appears that the system become able to buffer the water from swings in nutrient levels as it matures. The last nutrient addition at Julian 12144 (5/24/12) barely caused an increase in NO$_3$ levels for the following sampling date. Though haydite clay removes the most N, river gravel and norlite treatments exhibit similar responses to N additions. Growstone displays the largest spike in NO$_3$ water concentration after the application of fertilizer and total N removal percent is only 50% of NO$_3$ added. Table 4-5
Figure 4-8: P concentration (mg/l) in pool water by treatment and date

Figure 4-9: N concentration (mg/l) in pool water by treatment and date

N removal in this study is comparable to levels of nitrogen removal by vegetated
mesocosms in Borin and Salvato (2012), and Iamchatturapatr et al. (2007), with removals of 78% and 89-90% respectively. The N removal by Haydite/clay at 95% is very close to the average remediation efficiency of 94.7% experienced during summer months in Taylor et al. (2006). N removal is significantly correlated to water pH. NO$_3$ water concentrations decreasing with rising pH until the pH reaches 8.5. Statistical analysis found no correlation between water temperature and NO$_3$ pool water concentrations. Water temperatures are within optimum removal ranges for biological activity, only varying between 17°C and 27°C.

**Biomass removal**

This study measured similar biomass removal efficiencies to Henderson (2008), with biomass assimilating 71%, 80% and 92% of N removed by the systems in the norlite, haydite/clay and river gravel treatments. The Henderson study measured between 78 and 71% removal of N by aboveground vegetation in loamy-sand and sand treatments. Biomass in the growstone treatment only removed 56% of the total N removed, which is still significantly greater than N removed by biomass in Chung et al. (2008), where plant uptake accounted for 2.6-3.1% of N removed and Browning and Greenway (2003), who reported biomass removals of between 1-11% N. There is no significant difference between biomass uptake in FWS and SSF conditions.

For P, biomass is responsible for 45%, 9%, 35%, 35% of total P removed in the river gravel, growstone, haydite and norlite treatments. These numbers are much higher than reported in Browning and Greenway (2003), 1-3%, and Tanner et al. (1995), 1.9-5.3% for biomass removal of total P removed. Influent levels were higher for both of
these studies then the levels used in this study, and Kadlec and Wallace (2009) state that biomass removal is of greatest significance for low loading rates.

**Mass Balance**

Mass balance is used to partition N and P between substrate, pool water and plant biomass. Differences in N and P concentrations (mg/l) in pool water between the beginning and end of the experiment are calculated, as are the mass changes (g). Differences between the N and P (g) at the beginning and end of the experiment for the substrates and biomass are also measured. Mass balance is calculated using the following formula:

Total mass balance = inputs – outputs.

The inputs equal the nutrient present in the substrate before it is added to the treatment + nutrient present in the biomass at the start of the experiment + grams of nutrient in fertilizer additions throughout the experiment. (Figure 4-10) Differences in this mass balance analysis and many for wastewater or stormwater treatment, is that this mass balance treats the system as a closed loop, no water leaves the system as effluent, but continually recirculates. Final N and P in the water is not considered an output, but a treatment goal that needs to be satisfied. No nutrients were added with fill water, as greenhouse DI water is used to add water to the units. Both substrate and biomass nutrient contents before the experiment are determined from representative samples sent to the Penn State Analytical Lab for analysis, where percent nutrient x dry weight = mass of nutrient contained in the sample. Outputs are considered the nutrients in the total
biomass and substrate and are calculated by unit. Since there are no significant
differences in nutrient removal or biomass for flow conditions, results were pooled for
each treatment for a total of eight units per treatment. Unmeasured compartments for
nutrient inputs and outputs are insects, algae cycling, bacterial transformation and loss of
water through splash.

Duckweed, *(Lemna sp)* introduced from the plant roots when obtained from the
nursery, became a measurable component of plant uptake in some of the units.
Duckweed totals are obtained through weekly harvests from units where needed with
subsequent drying. The identical procedures are followed for obtaining nutrient levels
for duckweed as are used for iris and lizards’-tail.

Statistical analysis shows a significant relationship between treatment and mass
balance (p< .001), but no significant relationship between flow condition and mass
balance. N and P mass balances are correlated (.008). Analysis indicates recovery rates
of N and P are significantly different between treatments, p < .03 and 0.01 respectively.
The growstone treatment exhibits the lowest N and P recovery rate for all treatments.
Recovery rates for N are quite high, ranging from 80 to 113%. If the amount of N and P in the water is added to the recovered portion in the output, (biomass + substrate) then nutrients in the water, sediment and biomass are accounted for. For the growstone treatment, 80% of N is recovered, indicating that N is bound to some compartment in unrecoverable form or was exported from the system. Possible export mechanisms would be denitrification, or biomass loss. River gravel and norlite have over 100% recovery. Three of the treatments, river gravel, haydite/clay and norlite have recovery.

Table 4-6: Nitrogen and phosphorus mass balance

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Input N (g)</th>
<th>Output N (g)</th>
<th>Final N in Water (g)</th>
<th>Mass Balance</th>
<th>Recovery Rate of N</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 (River Gravel)</td>
<td>5.46</td>
<td>4.87 +/- 1.75</td>
<td>1.29 +/- .68</td>
<td>.59 +/- 1.75</td>
<td>113</td>
</tr>
<tr>
<td>2 (Growstone)</td>
<td>8.10</td>
<td>4.01 +/- 1.19</td>
<td>2.42 +/- .95</td>
<td>4.09 +/- 1.19</td>
<td>80</td>
</tr>
<tr>
<td>3 (Haydite/clay)</td>
<td>6.86</td>
<td>6.25 +/- 1.52</td>
<td>.27 +/- .39</td>
<td>.65 +/- 1.52</td>
<td>95</td>
</tr>
<tr>
<td>4 (Norlite)</td>
<td>7.24</td>
<td>6.81 +/- 2.88</td>
<td>1.89 +/- .88</td>
<td>.43 +/- 2.88</td>
<td>120</td>
</tr>
</tbody>
</table>

*Input = N(g) added by fertilizer and N(g) in starting biomass +N(g)in start and substrate
*Output = g(N) in biomass +g(N) in substrate

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Input P (g)</th>
<th>Output P (g)</th>
<th>Final P in Water (g)</th>
<th>Mass Balance</th>
<th>Recovery Rate of P</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 (River Gravel)</td>
<td>2.59</td>
<td>1.17 +/- .37</td>
<td>.003 +/- .001</td>
<td>1.42 +/- .37</td>
<td>45</td>
</tr>
<tr>
<td>2 (Growstone)</td>
<td>5.83</td>
<td>1.36 +/- .95</td>
<td>.003 +/- .002</td>
<td>4.47 +/- .95</td>
<td>23</td>
</tr>
<tr>
<td>3 (Haydite/clay)</td>
<td>2.65</td>
<td>.99 +/- .16</td>
<td>.0005 +/- .0006</td>
<td>1.67 +/- .16</td>
<td>37</td>
</tr>
<tr>
<td>4 (Norlite)</td>
<td>2.62</td>
<td>1.01 +/- .08</td>
<td>.0009 +/- .0009</td>
<td>1.61 +/- .08</td>
<td>39</td>
</tr>
</tbody>
</table>

*Input = P(g) added by fertilizer and P(g) in starting biomass +P(g)in start and substrate
*Output = g(P) in biomass +g(P) in substrate
rates close enough to 100% that we can say with confidence that this research is accounting for all the sinks and losses for N in the mesocosms.

Recovery of P is much less in all four treatments. Growstone only recovered 23% of the input P. River gravel had the highest recovery at 45%. It seems likely that P was tightly bound to the growstone, haydite and norlite treatments and the Mehlich 3 was unable to remove the P for measurement. The river gravel, with less reactive sites, would release more P. Still, even the river gravel has a large portion of P unaccounted. It is possible that the unmeasured contribution of sediments that accumulated in each unit could have contributed to P removal, as well as duckweed and algae uptake. Analysis of P in water samples was conducted after filtering through a 45µm filter, so would not measure the P lost to algal sequestration. Recovery rates for both N and P are highest for river gravel, most likely because it has the lowest surface area ratio and least reactive sites for adsorption.
Mass Balance for River Gravel Substrate
Treatment 1

Harvestable Biomass

Biomass storage
4.87g N
1.17g P

Other
0g N
1.42g P

Pool Water
1.29 g N
.003 g P

Unaccounted Inputs
.76 g N
0 g P

Substrate storage
0g N
0g P

Inputs
5.4g N
2.59g P

Mass Balance for Growstone Substrate
Treatment 2

Harvestable Biomass?

Biomass storage
2.28g N
.52g P

Other
1.67g N
4.47g P

Pool Water
2.42g N
.003g P

Substrate storage
1.73g N
.83g P

Inputs
8.1g N
5.8g P
Figure 4-10: Inputs, outputs and storage by substrate type for a natural swimming pool greenhouse study
Conclusion

Iris appears to be a good candidate for a NSP biofilter with acceptable vigor and biomass accumulation. While lizard’s tail contains a higher tissue nutrient concentration, its uncertain establishment and low growth index indicates it may not be a good candidate for NSP biofilters. Nonetheless, many researchers agree that measuring plant contributions toward nutrient removal should not be based only on harvestable (aboveground) biomass (Tanner 2005). Other plant-mediated pathways beneficial to nutrient removal include plant effects on microbial processes, species effect on denitrification by producing carbon in the rhizosphere and decomposition of plant material for sediment formation. In addition, though lizard’s tail did not have high biomass accumulation in this study, it would be interesting to compare these results to biomass production by lizards tail in a field scale NSP under a longer study period.

Two substrates demonstrate potential as substrate for bogs in natural swimming pools. Norlite and haydite/clay reduced both nutrients to 260.01 and 5.83 mg/l for NO₃, and .02 and 0.01 mg/l for P respectively. (Table 4-7) Norlite reduced P levels in the water to .02, very close to the 0.01-mg/l target. The haydite used in treatment 4 mixed with calcined clay, achieved the target level of 0.01 mg/l. It seems probable that the addition of calcined clay to norlite might also improve P removal to targeted levels (White et al. 2011).

River gravel and growstone, reduced NO₃ and P levels in the pool water to 28.24 and .06 (NO₃) mg/l and 53.04 and .07 mg/l (P) respectively. To improve the performance of the river gravel, smaller diameter river gravel could be used since adsorption is the
main removal mechanisms for P and surface area is a key characteristic. River gravel decreased to 16mm has been found to increase P adsorption (Akritos and Tsihrintzis 2007) although the source of river gravel and chemical characteristics would determine exchange capacity.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>NO3 mg/l</th>
<th>P mg/l</th>
</tr>
</thead>
<tbody>
<tr>
<td>control</td>
<td>28.24</td>
<td>0.06</td>
</tr>
<tr>
<td>growstone</td>
<td>53.04</td>
<td>0.07</td>
</tr>
<tr>
<td>Haydite/clay</td>
<td>5.83</td>
<td>0.01</td>
</tr>
<tr>
<td>Norilite</td>
<td>26.01</td>
<td>0.02</td>
</tr>
</tbody>
</table>

Table 4-7: Mean water concentrations of NO₃ and P (mg/l) at end of experiment by treatment

Mass balance calculations for this study indicate that biomass removed a significant portion of N and P. When investigating pathways for N removal it is important to remember that NO₃ does not sorb well, and therefore removal is generally through either denitrification or plant uptake. Literature is divided when reporting N mass balance amounts retained by plants. Many report that N removal by plants is a minor fraction of TN removal with denitrification representing the largest removal process (Maine et al 2007). However, we could expect plant N uptake to be more significant in this study since water is consistently circulating, oxygenating the substrate and reducing optimal denitrification conditions. Also, with the low levels of N introduced to each mesocosm, plants are forced to mine the substrate for nutrients to support active growth. Stormwater nutrient removal research indicates that in most cases plants improve NO₃ removal (Read et al. 2008, Fletcher et al. 2007), especially in low
load systems (Kadlec 2005). N removal in the haydite/clay treatment probably is reflected by the improved biomass production over any chemical substrate characteristics, as is the poor performance in the growstone treatment with the lowest biomass production of all treatments. Kadlec and Wallace, 2009, estimate N uptake of 51g-N.m2/y is possible for plants. If Table 4-7 is examined, it is clear that in the case of a NSP, where N loads are much less than 51 g/m2/yr, biomass potentially could remove a large portion of total added N.

While biomass is frequently cited as a significant compartment for N removal, many studies report a smaller portion of removed P stored in the standing stock. For instance, Drizo et al. (2002) found that while N was highly correlated to the presence of macrophytes, P was not. In Chung et al. (2008), planted treatments significantly increased P removal over unplanted, but in the mass balance, substrate was the main removal pathway with less than 1% of P removed by plant uptake. In conclusion, if properly designed and harvested as recommended, a substrate of Haydite and calcined clay will support biomass production sufficient for plant assimilation of greater than 90% of N and will also remove TP to target levels of 0.01 mg/l.

Probably the most important aspect of managing P in NSP pool water is minimizing the use of fill water. Since P levels are not of particular concern for drinking water, significant amounts of P can be introduced through the addition of tap and well water. Early experiments at the Penn State greenhouse measured .26 mg/l, .7mg/l and .3 mg /l of P in tap water. In an earlier field study, tap and well water P inputs were measured at .15 mg/l well above the 0.01 mg/l target level. The FLL (2003, 2006)
guidelines recommend that all fill water contains < 0.01 mg/l of P, a recommendation that clearly would be almost impossible to follow.

N is mandated by the EPA to be less than 10 mg/l in drinking water, but is still present in small amounts in tap water and additionally may share the same input sources as P. Typical stormwater runoff input values from 0.019 to 0.6 mg/l of P (Dietz and Clausen 2005, Davis 2007, Hunt et al, 2006, Passeport et al. 2009) and 2.6 mg/l N (Hatt et al.) have been reported in stormwater management nutrient reduction research. Table 4-8 lists other sources of N and P, such as sunscreen, pollen, leaf litter and tree seeds.

Table 4-7: Nitrogen and Phosphorus Sources for NSP Systems

<table>
<thead>
<tr>
<th>Deposition source</th>
<th>Nitrogen</th>
<th>Phosphorus</th>
</tr>
</thead>
<tbody>
<tr>
<td>Atmospheric deposition</td>
<td>.25 mg/l (1)</td>
<td>2 – 30 µL/yr (3)</td>
</tr>
<tr>
<td>Surface runoff in urban area</td>
<td>2.6 mg/l</td>
<td>.019-.6 mg/l</td>
</tr>
<tr>
<td>Fill water</td>
<td>2.2 mg/l (6)</td>
<td>.26 mg/l(6)</td>
</tr>
<tr>
<td>User/sunscreen</td>
<td>unknown</td>
<td>.46 mg/l(7)</td>
</tr>
<tr>
<td>pollen</td>
<td>.12 mg/l (2)</td>
<td>15 mg P/m2/yr (2)</td>
</tr>
<tr>
<td>Plant litter</td>
<td></td>
<td>.97 ppm/yr</td>
</tr>
</tbody>
</table>

1= data from Waller 1977; 2= Nicholls and Cox 1978; 3 = Reinfelder et al 2004
4= Roy-Poirier 2010; 5= Davis 2004; 6= Penn State tap water; 7= p.c. BioNova Inc

Table 4-8: Potential nitrogen and phosphorus inputs for a NSP
As stated in Hatt et al 2009, selection of a media with low P content is essential for effective P removal. This is reflected in the removal data for P in the growstone treatment, the substrate with the highest original P concentration and the lowest P removal in this study. Final growstone treatment water P concentrations could have been a result of leaching from the substrate, since it had a higher amount of P inherent in the substrate before system operation.

Additionally river gravel and Norlite P removal capacities may be improved if they are mixed with calcined clay, as is the Haydite substrate. The addition of calcined clay would be a cost effective alternative if mixed with river gravel, but more research would be needed to determine effectiveness. P retention is very high in the greenhouse mesocosms, however, it is reasonable to assume that as the system ages, the substrate will approach saturation and removal efficiencies will decrease.

Evidence for this assumption is contained is observations from Hsieh et al (2007), White et al. (2011), Davis et al. (2006) and Arias et al. (2001) where declines in P adsorption were measured when the systems approached P saturation. NSP loading is much lower than that of most treatment wetlands and stormwater technologies. Logically, media saturation would occur much later than that of a wastewater treatment wetland, possibly functioning beyond the expected lifetime of the other NSP components, such as EPDM liner, qualifying the biofiltration as a sustainable technology with much lower carbon costs than a treatment wetland.

While this study did not measure sediment formation, new sediment deposition occurred in all mesocosms and possibly was the source of a portion of the unaccounted P
outputs. Therefore it seems likely that the bog filter in a NSP would form sediments as it ages, increasing P removal due to sediment accretion. Removal of sediments periodically would permanently remove adsorbed P, while the sediments could easily be treated as soil amendments and added to compost piles or gardens.

The 4 treatments examined in this research exhibits a good fit to the input/output model presented, for N cycling. Evidence suggests P would also fit the model well if a different method was used to measure bound P in the substrate compartment. Including measures of algae and biofilms nutrient uptake would also improve model fit.

Further research is needed to determine NSP biofilter performance in field conditions. Previous greenhouse mesocosm research has illustrated concerns about artificial inflation of removal efficiencies because of increased vegetative densities/surface area. In addition, NSPs need to operate efficiently through the year, with peak performance during spring and summer when owners’ pool use is high and additional nutrient loading stress exists, such as pollen deposition and spring storm runoff. Therefore, further research is needed to assess seasonal performance and the effect of plant senescence and cold temperatures on nutrient removal capabilities.
Literature Cited


FLL, 2006. Recommendations for the planning, construction and maintenance of private swimming and natural pools, Bonn.


Chapter 5


Introduction

Natural swimming pools (NSPs) are recent technological introductions to North America, designed to provide an attractive landscape amenity that also serves as a chemical free swimming pool. Based on constructed wetland technology, natural swimming pools use biological filtration, a designed, intentional hydraulic system, and an impermeable liner to separate the system from the surrounding natural hydrologic cycle. Biological filtration is the use of plants and substrate to closely replicate natural wetland environments, filtering excess nutrients and harmful bacteria from the pool system without the use of harmful chemicals. These systems offer a variety of ecosystem services, supplying wildlife habitat and four season aesthetics, fostering stewardship, eliminating ecologically deleterious chemicals and supporting a diverse vegetative community.

Nutrients of particular concern for natural swimming pools are nitrogen and phosphorus, because of their association with eutrophication and subsequent high algae levels. In natural freshwater bodies the USEPA mandates that nitrate levels be \( \leq 10 \text{ mg/l} \) and recommends TP levels less than .1 mg/l (US EPA 1986). P levels as low as .02 mg/l
are implicated in nuisance algal blooms (Daniel et al. 1998). In Europe, recent standards developed for NSP design specify levels for N and P of 30 mg/l (NO₃) and 0.01 mg/l (TP) respectively (FLL 2003).

Many constructed wetland studies indicate an increase in nutrient reduction abilities with vegetated biofilters in wastewater treatment (DeBusk et al 1995), nursery runoff treatment (Berghage 1999; Chen & Bracy, 2009; Huett 2005; Taylor et al 2006), and stormwater filtration ((Bratieres et al. 2008; Browning & Greenway, 2003; Hatt, Deletic, & Fletcher, 2007; Henderson, Greenway, & Phillips, 2007; Johengen & LaRock, 1993; Moortel et al. 2010; Read et al., 2008; Roy-Poirier et al. 2010; Zhang et al. 2011). No studies, however, have examined the nutrient removal capabilities of the NSP biofilter. NSP systems differ from other nutrient reduction systems because they are closed systems, sealed to prevent infiltration and exchange with the surrounding hydrology. In addition, NSPs are recreational and aesthetic landscape amenities and nutrient loadings are low. Because NSPs are landscape amenities, the biofilter vegetation must fill the dual purposes of efficient nutrient reduction and ornamental enhancement. In addition, values such as fostering stewardship, and environmental assets including plant diversity and wildlife habitat suggest that plant monocultures, usually found in wastewater treatment, are not the optimum choice for NSP systems.

Many traditional constructed wetland plants often are not suitable for natural swimming pools. Sharp, pointed rhizomes typical of some commonly used obligate wetland plants; prohibit their use in NSPs that utilize an EPDM liner. Other common constructed wetland species are simply not ornamental. A few ornamental obligate and
facultative wetland species have been studied for nutrient removal in bioretention systems, (Chen and Bracy 2009, Maschinski et al. 1999), and constructed wetlands (DeBusk et al. 1995; Read et al. 2008, Polomski et al. 2007), but not in the conditions specific to a natural swimming pool.

Relatively low nutrient loads and certain substrate characteristics in NSPs can delay plant establishment. Although plants are often an essential polishing removal mechanism (Mitsch and Gosselink 2007, Vymazal 1995, Kadlec and Wallace 2009), substrate is the major P storage compartment. Ideal substrate characteristics for NSP systems are the ability to promote rapid plant establishment, support high standing stock, high hydraulic conductivity and contain the characteristics necessary for high P adsorption. These characteristics include surface area, clay content, or the presence of Al, Fe and Ca (ion exchange capacity) (Kadlec 2005, Vacca 2011). Studies of media and nutrient removal conclude that for highest P removal capabilities prefilters, special media or media enhancements are necessary (Brooks et al. 2000, Hunt et al. 2006). However, some media enhancements create environments limiting to plant establishment and growth (Vohla et al. 2011). EAF slag and limestone increase pool water pH to levels harmful to many aquatic organisms even though P removal is high (Naylor et al. 2001). Other drawbacks to alternative media are cost, availability and saturation timeframes.

This study explores the use of calcined clay, an easily obtained and inexpensive product, enhanced with alumina. Calcined clay possesses high P adsorption potential, (White et al. 2011; Arias et al. 2003; Brix et al. 2001) high hydraulic conductivity and
provides an excellent root media for most emergent wetland plants. Alumina attracts P and binds it, but also has been shown to increase the availability of P to plant roots.

**Substrate Storage of Phosphorus**

Activated alumina is both effective in wastewater treatment (Vohla 2011) and as a P adsorbant recommended for traditional outdoor swimming pools (Douglas et al. 2004). Phosphorus removal on activated alumina is mainly ion exchange promoting adsorption and precipitation (Narkus and Meiri 1981). Activated alumina manufacturing creates a very porous material with high surface area, which increases P exposure to adsorbent surfaces.

Because activated alumina does not influence pH, has no ecological toxicity and binds P to very low residual levels it is a recommended material to adsorb P in swimming pools (Douglas et al. 2004). Originally, activated alumina was examined to remove P from low loaded natural water bodies, such as lakes, where influent P levels were .1mg/l (Hano et al. 1997), suggesting it might also be valuable in the low nutrient NSP system. Additional research suggests activated alumina when combined with planting media also correlates to an increase in seedling growth without a corresponding increase of P in leachate (Elliot 1989, Oguto 2008).

The substrate in this study was a mix of expanded clay, sand and activated alumina. Arias et al. (2003) identified a calcite material (kitty litter) that possessed high P binding capability and hydraulic conductivity. The Arias et al. (2003) study tested the calcined clay for P removal in a design with high levels of P influent and short contact
times, only 28 to 99 minutes. The calcined clay exhibited high P removal and decreased P concentration by 75%. Removal declined however as the material became saturated, with some filters becoming saturated at 7 weeks after startup. Once saturation occurred, P removal decreased and P released into the system from the filter. One of the goals of this study is to explore if combining the calcined clay with activated alumina would increase the adsorptive period of the media and inhibit release of P into the pool water.

**Ornamental Plants and Nutrient Recovery**

Two ornamental wetland plants, *Iris versicolor* and *Canna x generalis*, were tested for their ability to produce aboveground biomass and nutrient removal capabilities. *I. versicolor*, blue flag iris, is an obligate, native wetland plant commonly found on lakeshores and in wetlands, often growing in shallow water. Although blue flag was studied incidentally in Drizo et al. (2008), Keppler and Martin (2008), and Kohler et al. (2004), its direct contribution to nutrient removal was not quantified. These studies conducted in constructed treatment wetlands operating in high nutrient levels, are not reflective of the low nutrient environment of a natural swimming pool.

*Canna x generalis* (canna spp.) is a common ornamental and can be grown either in wetland environments or in garden conditions. Canna is a vigorous grower reaching 6’ and hardy to USDA zone 7. Canna grown north of zone 7 must be dug after frost and overwintered indoors. Canna is often studied in constructed wetlands (Liang et al. 2011, Zhang et al. 2008, Polomski et al. 2008, Calheiros 2007), stormwater (Zhang et al. 2011)
and aquaculture (Konnerup 2011) research, but has not been examined as part of a NSP biofilter.

Canna production of standing stock was greater than *I. pseudacorus* ‘Golden Fleece’ in a greenhouse study by Chen et al. (2009) and showed high nutrient removal ability. Despite lower biomass production, Golden Fleece iris had higher N tissue concentration than canna, and P concentration was very similar. Tissue N & P concentrations in shoots were higher than roots for both canna and iris, suggesting aboveground tissue harvests could remove substantial nutrients.

While research disagrees on the importance of harvesting plant material for nutrient removal, Chen et al. (2009) indicates that over half of the nutrients contained in biomass are removed through harvesting. Supporting evidence for higher nutrient concentrations in shoots than roots is found in a study of nutrient recovery for five ornamental plants by Polomski et al. (2008). Canna had higher N & P tissue concentrations in shoots at every treatment level except at low loadings of .39 mg-N/l and .07 mg-P/l. Canna showed increasing P shoot/root concentration ratios as P loading increased, although even when loadings were < 1.86 mg/l, 64 – 74% of tissue P was contained in shoots. Iris showed similar results, with 90% of tissue P & N contained in shoots, regardless of loading level (Polomski et al. 2008). The low nutrient treatments in this study adversely affected the canna, as they exhibited visual deficiency symptoms, but had little effect on the iris.

The goal of this study was to determine nutrient removal capabilities for *I. versicolor* and *Canna x generalis*. This study included a mass balance determination of
substrate storage, biomass removal, water retention of N and P and unaccounted removal compartments. Plants used in NSPs should have the following characteristics;

- Not present an ecological danger to local plant communities
- Tolerance for local weather conditions, pests and diseases
- Tolerance of low nutrient conditions
- Rapid establishment
- Ability to contribute significantly to N and P removal through direct uptake or support of microbially mediated removal pathways (modified from Cronk and Fennessy 2001).

This study also determines if canna and iris, plants recommended for high nutrient environments (Cronk and Fennessey 2001), will tolerate the lower nutrient conditions in a NSP.

Appropriateness of the substrate materials, calcined clay, sand and alumina was assessed for plant establishment and nutrient removal. Water pH and algae levels and possible negative interactions with the system were analyzed.

**Methods and Analysis Experiment Three**

This greenhouse study was conducted from April 4, 2012 to June 5, 2012 at The Pennsylvania State University Department of Horticulture greenhouse complex at University Park, Pennsylvania (lat. 40°8019′N, long 77°8627′W). Iris obtained from Maryland Aquatics Nursery as plugs on Sept 29, 2011, were placed in tubs to maintain growth until the experiment started. *Canna x generalis* was obtained from a local grower
in October 2011 and stored in the greenhouse coolers until planted. Peters 15-15-15 was applied to plants monthly until replanted in experimental mesocosms. All plants were weighed before planting then planted in the mesocosms to allow plants time to acclimatize. Baseline weights were obtained for each plant species, dry weight was determined, and N and P content measured. The initial measurements were subtracted from final weight and nutrient content for mass balance calculations. The Penn State Agriculture Analytical Lab conducted analysis of samples for N and P percent for later mass balance calculations using the dry ash method and the Vario elementar analyzer.

The experiment included 24 units and 3 treatments (Iris virginica, Canna x generalis, unvegetated), with 8 units each. All treatments are divided into 2 conditions, free water surface (FWS) and subsurface flow (SSF), with 4 units in each condition. The experimental design was a complete randomized design. (See Figure 5-1 for experimental layout).

Each bog (17.9L Rubbermaid container, 42.9cm x24.9cm x24.7cm) was constructed with 1.5 cm of free space on the bottom by placing a seed starting tray on the bottom of the unit, followed by a layer of geotextile and 6” of substrate. See Figure 5-2 for pool/bog design. Water was pumped from the ‘pool’ unit, and introduced to the free water space at the bog bottom via a PVC pipe, where it diffuses through the bottom and percolates through the substrate. A hole drilled above the media in the bog container allowed the water to return via gravity flow to the pool unit. Each treatment had 8 units, four each free water and subsurface flow. The free water flow return water hole was located 2” above the bog surface, while the subsurface flow drainage hole was located at
the top of the substrate and under 2” of pea gravel. Bogs were elevated 1’ above the swimming pool units. Swimming pool units were also 69L Rubbermaid containers, (60.7 x 40.4 x 30.9 cm). Pools were initially filled with 45.6 L DI water. Bog units were filled with 6100 g substrate, composed of 3700 g calcined clay, 2390 g sand, and 66 g activated alumina. See Figure 5-1 for experimental unit setup

Water pumped at a rate of 22.5 L per 12 hrs, resulted in total turnover once every 24 hrs. Artificial lights controlled by a timer that turned on the lights from 5:00 pm to 10:00 pm daily augmented natural lighting. Greenhouse temperature was kept at 82 F during the day and 76 F at night. Water levels were adjusted to a consistent level, checked weekly and DI water added as necessary. The volume of each water addition was recorded. Each container was fertilized with 4 g of 20-10-20 granular fertilizer on 4/12/2012, 4/23/2012, and 5/25/2012.

Figure 5-1: Experimental layout of treatments and flow condition for an experiment determining the effect of Iris versicolor, Canna x generalis and unvegetated treatments* in free water and subsurface flow** conditions using calcined clay and activated alumina substrate on nitrogen and phosphorus removal

*Top number refers to treatment (plant species)  **Bottom number refers to flow condition
1= Iris versicolor  0=free water surface
2= Canna x generalis  1=subsurface flow
3= control (no vegetation)
Figure 5-2: Experimental unit setup for pool and bogs in an experiment measuring the effect of *Iris versicolor*, *Canna x generalis* and unvegetated treatments in free water and subsurface flow conditions using calcined clay and activated alumina substrate on nitrogen and phosphorus removal

Fifty ml water samples were drawn weekly from the pool units and measured for relative algae levels on a spec 20 and then measured for nitrate, pH, EC and water temperature using Forstan LabNavigator probes, Synaptic Sensors, LLC, 401 Mathews St., Fort Collins, CO. Probe calibration occurred before each sampling event and measurement accuracy verified using spiked samples.

Additional 50 ml water samples, drawn every 2 weeks, were filtered through a 45μm filter and P (PO4) determined by the colorimetric molybdenum blue method (Murphy and Riley, 1962) and measured on a Perkins Elmer Lambda 25 spectrophotometer with UV WINLAB V 2.85 software. Samples were filtered immediately, and either tested immediately or stored at 4°C.

At the end of the experiment, final 50 ml water samples were drawn from each of
the 32 swimming units and analyzed on a Bausch and Lomb Spec 20 spectrophotometer for chlorophyll a absorbance, followed by filtering for P analysis. The Forstan LabNavigator was used to measure NO₃, pH, water temperature and EC for each pool unit. Final 50 ml samples were analyzed for P concentration using the colorimetric molybdenum blue method (Murphy and Riley, 1962) and measured on a Perkins Elmer Lambda 25 spectrophotometer with UV WINLAB V 2.85 software. A field scale weighed the water in bog and pool units for volume determination.

For each bog unit, 50 ml substrate samples were collected and sent to the Penn State Ag Analytical Lab. TN analysis conducted using the combustion method from Bremer (1996), and P analyzed using the Mehlich 3 extraction method (Recommended Soil Testing Procedures for the Northeastern United States 2009).

Plants were harvested on June 5, 2012, rinsed with DI water to remove substrate and oven dried at 65.6 C (150 F) for one week. These samples were sent to the Penn State Ag Analytical Lab and analyzed for percent P and percent N. Plant tissue P % was determined using dry ash method in Miller (1998), and N% using the combustion methods in Horneck and Miller (1998). N and P content was calculated by multiplying plant dry weight by nutrient concentration.

Aphid infestation was problematic throughout the experiment. Control strategies ranged from wiping down each plant with 2% insecticidal soap to releasing predators in the greenhouse. Use of biologicals failed to control aphids, possibly impacting plant growth. Iris exhibited signs of iron deficiency resulting in application of iron chelate as a foliar spray on Apr 11 and Apr 23, 2012. Lemna was introduced to the system from the
roots of the plants and became established in a few units. It did not prove problematic in this experiment so is not included in the mass balance.

**Statistical analysis**

Data from nutrient removal compartments, plant biomass, substrate and pool water, were analyzed with a univariate analysis of variance (ANOVA) including post hoc Tukey’s using SPSS (version 20 for MAC, IBM, Armonk, New York). A two-way fixed effect ANOVA model was used to determine interaction effects between treatment and water flow. Repeated measure analysis of variance was used to evaluate the effect of time on nutrient removal, treating plant species as the between-subject effect and date a within-subject factor. Q-Q charts, residual plots and Levene’s Test were used to ensure necessary assumptions of normality and constant error variance were met. The relationship of biomass and nutrient removal was determined through bivariate correlation and Pearson’s correlation. In the case the assumptions of normality were not met, two nonparametric correlation tests, Kendall’s Tau and Spearman’s rho were also used. The significance level ($\alpha$) used to reject the null hypotheses for all tests is 0.05.

Determinations of nutrient removal efficiencies were calculated using the ratio of the mass removed (nutrient in final biomass plus nutrient in substrate at end) to the mass added by fertilization. Total mass removed was calculated by subtracting the nutrient mass present in the plant biomass and substrate from the mass added to the system. Biomass accumulation was contrasted with nutrient uptake efficiency and biomass nutrient content calculated by multiplying the dry weight by nutrient percent.
Discussion and Analysis

Nutrient Levels in the Pool Water

The unvegetated treatment did not remove N as efficiently as the vegetated filters. There was no interaction effect between flow condition and plant type, and flow condition alone had no significant effect on either N or P removal. Because flow had no effect, results were pooled within each treatment. Treatment effect for plant species and final NO$_3$ levels in the pool water were highly significant with a p value < .001. It was clear that plants did have significant capabilities to improve removal of NO$_3$ compared to the unvegetated treatment. Canna removed larger amounts of NO$_3$ than iris, probably due to greater biomass production. The correlation of biomass and NO$_3$ removal was highly significant, p<.001, using Pearson’s correlation. Time had an effect on NO$_3$ removal, but Figure 5-3, demonstrates it is a very complicated relationship and hard to define. Spikes in the graph coincide with nutrient additions. It is clear that nutrient removal improves over time only in the 2 vegetated treatments.

Presence of canna or iris had no significant effect on removal of P from pool water (p = .241). Plant biomass was highly correlated with final P in the water (P < .004 using Pearson’s correlation, and using Kendall’s and Spearman’s nonparametric correlation test,  p<.001). Even with this correlation, all treatments did achieve high removal efficiencies and obtained the P removal goal of TP< 0.01mg/l. Except for the nutrient addition before Julian 12117 (4/26/12), additional P (mg/l) in the water from fertilizer applications was removed by the next sampling date (See Figure 5-3).
There is a large unexplained spike in water NO3 and P at Julian date 12117 (4/26/12). This spike occurred in both this experiment and a concurrent experiment in the same greenhouse.

Figure 5-3: Mean NO3 concentrations for sampling dates by treatment in an experiment measuring NO3 removal in natural swimming pool mesocosms.
Figure 5-4: Mean P concentrations as measured by Murphy- Riley method, for sampling dates by treatment in an experiment measuring P removal in natural swimming pool mesocosms.

Canna produced over twice as much biomass as iris in the 10 week experimental period and contained almost twice the N and P in plant tissues as the iris. However, iris tissue nutrient concentration was higher for both N and P. (See Table 5-1 )

Canna nutrient concentration was 1.81% and .21% for N and P respectively. Canna harvestable biomass concentration is 2.2% and .23% for N and P respectively. There was higher N allocation to shoots and P to roots (Table 5-2). The N concentration was higher than canna N shoot concentrations in Polomski et al. (2009) but the P concentration was comparable. ‘Yellow King Humbert’ in that study contained 1.6%N and .23 %P respectively. Chen et al. (2009) reported canna shoot concentrations of
1.51% and .51% for N and P. In this NSP study, canna harvestable biomass accounted for 52% and 43% of total biomass N and P. (See Table 5-2)

<table>
<thead>
<tr>
<th>Table 5-1: Mean Unit Biomass and Tissue Concentration</th>
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<tbody>
<tr>
<td><strong>Plant Biomass Weight, Nutrient % and Nutrient Content (g)</strong></td>
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<tr>
<td></td>
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<tr>
<td>Treatment</td>
</tr>
<tr>
<td>Iris</td>
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<tr>
<td>Canna</td>
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</table>

nutrient in biomass = nutrient % x biomass dry wt

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<thead>
<tr>
<th>Table 5-2: Final total shoot biomass, nutrient content and percent nitrogen and phosphorus contained in harvestable biomass.</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Shoot Nutrient Percent, Nutrient Content and Percent of Fertilizer input Removed by Harvest of Shoot Biomass</strong></td>
</tr>
<tr>
<td>Treatment</td>
</tr>
<tr>
<td>Iris</td>
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<tr>
<td>Canna</td>
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</table>

% removed by harvest = g of nutrient in shoot (input g) of N and P from fertilizer

Iris tissue concentrations were slightly lower than reported in Chen et al. (2009), which measured *Iris pseudacorus* productivity over a ten week period. The USDA Plant database describes *I. pseudacorus* as a vigorous and aggressive wetland species, with the tendency to outcompete other wetland plants. Given its aggressive growth habit, *I. pseudacorus* should produce greater biomass than *I. versicolor*, but in Chen’s study, mean *I. versicolor* dry weight was 30g while the *I. pseudacorus* dry weight was only 13.23. Since the differences in biomass accumulation cannot be attributed to growth habit, specific growing conditions, such as media, most likely account for the difference.
Direct comparisons across studies are difficult because of variations in nutrient levels, flow rates, retention times and plant species.

Whole plant iris nutrient concentrations were 2.03% and .22% for N and P respectively, while Chen et al. (2009) reported N iris concentrations of 2.72, 1.7 and 1.23 for shoots, rhizomes and roots, and P concentrations of .39, .13 and .16 percent respectively. For comparison, this study divided representative random samples of iris units between roots, shoots, and determined nutrient concentrations. Analysis of iris shoots for N and P show shoot concentrations greater than roots for N but not P; the separated shoot samples contain 2.22% and .22% N and P, still less than levels reported by Chen et al. (2009).

Polomski et al. (2009) reported that iris shoots contained greater than 90% of plant tissue N, although actual differences in N concentration levels were slight. Iris biomass production is primarily in aboveground standing stock, and N is allocated to shoots in the period of rapid expansion. P concentration levels in Polomski et al. (2009) were also higher in shoots than roots, but only at very low and high nutrient inputs. However, iris shoots contained greater than 90% of tissue P content. In this study, iris harvestable biomass accounts for 83% of N and 90% of total P in plant tissues, very similar to Polomski’s values. However, in Chen et al. (2009), iris harvestable biomass was only 48% of total plant biomass. This difference could be attributable to the difference in growing conditions. Polomski (2007) and this study grew plants in substrate, but a simulated floating system was used in Chen et al. (2009).
Higher iris shoot N and P concentrations than canna may be due to differences in root architecture. Iris has a very fibrous root system, while canna has thick fleshy rhizomes. The iris fibrous root system with increased surface area could increase acquisition ability while the thick canna rhizome might store more nutrients. Bratieres et al. (2008) postulated Carex spp. removed the most P because of its extensive root system, with microscopic root hairs, which enhanced phosphorus acquisition. Further, Gahoonia and Nielson (1997) showed root hairs do increase P acquisition in their study of radio labeled P uptake from soils via root hairs.

Harvestable Biomass

Gersburg et al. (1986) estimated harvesting of plants could only account for 12-16% of total N removal in a bulrush monoculture. Browning and Greenway (2003) and Chung et al. (2008) reported plant uptake accounted for 11% and <3.2% N and 3% and <1% of P removed, respectively. These findings are partially supported by work demonstrating that plants can only assimilate and store a given predetermined amount of nutrients, and once this storage potential is reached plant uptake becomes minimal (Howard –Williams 1985). Subsequent research by Breen (1990) reported higher N and P removals due to plant uptake, of 51% and 67% of total influent, while Zhang et al. (2008) reported 19-42% and 4-39% removals for N and P. Other research that reported similar high levels of nutrient removal due to plant uptake include Borin and Salvato (2012), Fraser et al. (2004), Tanner (1996).
Although in this study harvestable biomass does not represent as high a proportion of nutrient removal as other studies, it is still high enough to support end of season harvest. Harvesting the aboveground biomass would remove 25 and 21% of N inputs for canna and iris, and 21 and 12% of total inputs of P, amounts which justify the energy and time expenditure of harvest. Variations in removal due to uptake between research is attributable to differences in plant species, temperatures, growing conditions, nutrient load and water contact time with roots.

**Mass Balance Model**

Figures 5-5, 5-6, 5-7, illustrated the mass balance model for all treatments. The models include nutrient addition, substrate storage, biomass storage, biomass removal through harvest and nutrients present in the pool water at the end of the experiment. Nutrient inputs for each treatment differ by a small amount reflecting the difference of nutrients present in the vegetation at start.

**Substrate**

It was expected that the substrate would be the primary storage compartment for P. Original mass balance calculations showed a large unaccounted P portion, with a small amount of P bound to the substrate. This is a highly unlikely scenario as P is a recalcitrant mineral, it does not have a gaseous phase, and the major phosphorus removal mechanism is adsorption and precipitation. Clay and activated alumina have strong P removal capabilities, and in low load systems substrate becomes an even larger P removal
compartment (Kadlec and Wallace 2009). While plant uptake is a major compartment for nitrogen storage (Huett et al. 2005), it is a smaller compartment for phosphorus. Plants assimilate greater amounts of nitrogen than phosphorus and plant tissues contain 2-5% N and only .05 to 2% P. In this study, substrates were tested for TP using the Mehlich 3 extractant. Total phosphorus (TP) is a measure of all the forms of phosphorus, dissolved or particulate, found in a sample. Soluble reactive phosphorus (SRP) is a measure of orthophosphate, the filterable inorganic fraction of phosphorus, the form directly taken up by plant cells.

Since phosphorus does not have a gaseous phase and is not commonly involved in microbial cycles, a microbially mediated process is not likely to account for the unmeasured P. Plant uptake amounts are comparable to other nutrient removal mass balance studies and sampling includes all plant tissues, it is unlikely any of the unaccounted portion is in the plant biomass.
Figure 5-5: Inputs, outputs and storage for nitrogen and phosphorus in iris treatment in the experiment measuring nitrogen and phosphorus removal in natural swimming pool mesocosms.
Figure 5-6: Inputs, outputs and storage for nitrogen and phosphorus in canna treatment in the experiment measuring nitrogen and phosphorus removal in natural swimming pool mesocosms.
Figure 5-7: Inputs, outputs and storage for nitrogen and phosphorus in unvegetated treatment in the experiment measuring nitrogen and phosphorus removal in natural swimming pool mesocosms.

<table>
<thead>
<tr>
<th>Table : Mass Balance for N and P (g)</th>
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<tr>
<td>Plant spp.</td>
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<tr>
<td>Iris</td>
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<tr>
<td>Canna</td>
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<td>unvegetated</td>
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Table 5-3: Mass balance for nitrogen and phosphorus in canna, iris and unvegetated Treatments in the experiment measuring nitrogen and phosphorus removal in natural swimming pool mesocosms.
Table 5-4: Nutrient recovery rate for nitrogen and phosphorus in canna, iris and unvegetated treatments in the experiment measuring nitrogen and phosphorus removal in natural swimming pool mesocosms.

<table>
<thead>
<tr>
<th>Table: Treatment Recovery Rate</th>
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<tr>
<td><strong>Plant spp.</strong></td>
</tr>
<tr>
<td>Iris</td>
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<tr>
<td>Canna</td>
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<tr>
<td>unvegetated</td>
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Recovery rate = nutrient recovered in the pool water divided by the total nutrient input x 100

Table 5-5: Percent N and P by storage and uptake by treatment and analysis of adjusted P substrate storage.

Algae is not a possible storage compartment either, as spec 20 readings indicated low chlorophyll a levels. It seems most likely that the P held by the alumina in the substrate was bound so tightly that the acid used in the Mehlich 3 analysis was unable to strip the P from the alumina binding sites, resulting in the large unaccounted portion.
Thus for the purpose of this analysis the unaccounted for P was applied to substrate storage compartments. Differences in percent removal are presented in Table 5-5.

If we accept that the unaccounted for P is tightly bound to the alumina and not solubilized with the Mehlich 3 then substrate is the highest removal compartment for both N and P for all treatments. N storage in the substrate experiences very little variation between the three treatments, ranging from 46-49% of total N inputs. Substrate storage of P accounted for 88, 65 and 100% of total P inputs for iris, canna and unvegetated treatments. The substrate is capable of adsorbing sufficient P to meet FLL standards for P without vegetation while supplying plants with sufficient P to establish quickly and support biomass production. Since the unvegetated treatment is not able to achieve FLL standards for NO₃ in water, plants must play an important role in increasing NO₃ removal in a NSP.

**Biomass**

Plant uptake accounts for a small amount of P removed from input P, 14 and 34% for iris and canna respectively. N uptake by plants is a higher fraction of the total N input, representing 25 and 51% of total N removed from inputs. Differences in nutrient content are explained by biomass productivity differences not tissue concentration. Biomass is significantly correlated to nutrient removal, p< .001, and iris and canna mean biomass equals 121g and 268g.
**Water Fraction**

The unvegetated treatment met the treatment goal for phosphorus of P < 0.01 mg/l, as did both vegetated treatments. However, the unvegetated treatment failed to achieve the removal goal for NO$_3$ (NO$_3$ < 30 mg/l.) (Table 5-6) The canna treatment achieved the highest removal for both nutrients, 99% for NO$_3$ and almost 100% for P, followed by iris with 96% and 99% N and P removal from the swimming pool. (Table 5-4) The unvegetated treatment final pool water NO$_3$ concentration was 75 mg/l, for a total recovery rate of 18%, while P recovery rate was 99%. Recovery rate refers to the nutrient present in the pool water at the end of the experiment divided by the total nutrient added. The numbers in Tables 5-4 and 5-5 illustrate the essential role of plant uptake for N removal and substrate for P removal.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>NO$_3$ (ave/SD)</th>
<th>P ave/SD</th>
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<tbody>
<tr>
<td>iris</td>
<td>3.6+-5.4</td>
<td>.001+-0</td>
</tr>
<tr>
<td>canna</td>
<td>1.13+-0.87</td>
<td>0+-0</td>
</tr>
<tr>
<td>unvegetated</td>
<td>74.8+-8.6</td>
<td>.01+-0</td>
</tr>
</tbody>
</table>

Table 5-6: Final average and standard deviation of NO$_3$ and P concentrations(mg/l) of pool water in iris, canna and unvegetated treatments in the experiment measuring nitrogen and phosphorus removal in natural swimming pool mesocosms.

**Unaccounted Fraction**

Not all N inputs are accounted for in the mass balance model. In the iris and unvegetated treatments, 27 and 23% of N inputs remain unaccounted. Biofiltration nutrient removal research assumes unaccounted N is transformed though denitrification.
Wetlands contain both epiphytic biofilms and bacteria on the substrate. Potential denitrification by epiphytic biofilms can be as high as denitrification in sediments (Bourgues and Hart 2007). It is possible that the increased surface area of the fibrous iris root system augmented biofilm establishment, transforming much of the input N. Oxygen levels in the unvegetated media are probably lower than vegetated, lacking an oxygenated rhizosphere, also potentially supporting denitrification.

Conclusions

The excellent P removal experienced in all three treatments supports the hypothesis that activated alumina combined with calcined clay is an effective media for adsorption of P in a NSP system. All treatments reached treatment goals for P removal and experienced > 99% removal of influent P. Activated alumina contained in the media strongly binds P while simultaneously releasing a low concentration of P to roots, supporting acceptable plant growth. However, while the media is also effective when combined with plants to remove more than 96% of influent N, the media did not meet the NO3 treatment standard of < 30mg/l NO3 in the unvegetated treatment.

Because activated alumina removes P to an equilibrium level without releasing it back into the system, it is a satisfactory media enhancement for NSP systems. Studies show that activated alumina in low nutrient systems reaches saturation within 500 days, this period is highly dependent on many system factors. (Hano et al. 1998) Since biomass does not account for all P inputs, removing 14 % and 34 % of P in this experiment for the iris and canna treatments, P would eventually saturate the activated
alumina. Using the activated alumina in a prefilter could solve this problem, providing high removal efficiencies, and a low-tech filtration option for pool owners. When the prefilter reaches saturation it could be removed and used to fertilize gardens, compost piles or even released into the bog to support plant growth. Traditional gravel substrate has low P adsorption capabilities compared to other substrate materials (Cui et al. 2008), however is used because it is normally an inexpensive as well as locally obtainable material. A prefilter of calcined clay and activated alumina combined with local gravels as substrate could improve the river gravel substrate P removal to FLL standards. Future research is needed on saturation time in a NSP system and the cost effectiveness of prefilters or river gravel enhancements needs to be evaluated. It is clear from this research that vegetated filters improve nutrient removal and are crucial for attainment of N removal standards. It is not clear that species type makes a difference. While treatment differences were significant, both iris and canna treatments removed N and P well below testing standards. Further research could assess whether mixed plantings would perform better than monocultures of these two species. Both iris and canna performed well and produced acceptable biomass levels, though canna exhibited some deficiency symptoms. Harvesting biomass in the fall before aboveground tissues release nutrients into the system would reduce significant nutrient inputs. This research demonstrates that certain ornamental wetland plants, canna and blue flag iris, perform well in the low nutrient environment of a NSP and substantially improve biological filtration.
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FLL, 2006. Recommendations for the planning, construction and maintenance of private swimming and natural pools, Bonn.


VITA (for Ph.D. only)

Margaret C Hoffman

Education:
Ph.D., Horticulture, The Pennsylvania State University, Aug 10, 2013
  Dissertation: Development of a Mass Balance Model for Biological Filtration of a Natural Swimming Pool.
  Coursework included Residential Landscape Design, Plant Design, Greenroofs, Riparian Ecological Restoration, Extension Education, Public Administration
B.S., Forestry, minor in Urban Forestry, University of Minnesota, College of Forestry. 1982

Professional Experience

Director, Landscape Division, Country Farm and Home Garden Center  March, 2013 to present
  Manage department, setting goals and strategic planning
  1 Design, estimate and bid all design jobs.
  2 Maintain client contacts developing longterm relationships.
  3 Installed hardscapes including flagstone patios, mountain stone walls, patios, water features
  4 Responsible for scheduling, budget management and sales

  Hands on owner, I expanded company from landscape design company to design/build firm. Pioneered the design and installation of natural swimming pools in central Pennsylvania. Under my management the company experienced a steady increase in sales and profit.
  5 Managed 1 to 2 crews, 3 to 5 employees
  6 Estimated and bid all jobs enabling company to post a 20% profit.
  7 Managed projects consisting of $100,000 plus installations, integrating teams of subcontractors to improve scheduling and production.
  8 Installed hardscapes including flagstone patios, mountain stone walls and patios, water features
  9 Responsible for budgeting and sales
  10 Publish quarterly newsletters
  11 Contribute regular gardening column to Susquehanna Life Magazine

Forester 1983-1984
  Pennsylvania Department of Environmental Resources, Bald Eagle District Mifflinburg, PA
Urban forester, City of St Paul, MN

171
Summers 1978-1982

Assistant Extension Forester, Univ. of Minnesota, St Paul, MN
1978 to 1982

1. Diagnosed disease and insect problems and identified plant material for clients
2. Exercised public speaking and written communication skills responding to clients inquiries concerning urban forestry, sugarbush management and IPM
3. Published bulletin on developing home nurseries
4. Job required strong written and verbal communication skills