NITROGEN ACCUMULATION IN A CONSTRUCTED WETLAND FOR DAIRY
WASTEWATER TREATMENT

by

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ABSTRACT

Constructed wetlands manifest a significant potential for removing nitrogen from wastewater. The study was conducted at a free water surface constructed wetland receiving partially treated dairy wastewater. A series of four cells was lined with plastic and a parallel series lined with clay. The objectives were to evaluate 1) nitrogen removal from the wastewater stream and 2) nitrogen accumulation in the soil and plant biomass. Above and below ground plant tissues and soil samples were collected from twenty-four locations during one year. Total nitrogen removal from wastewater was about 17 percent. The clay-lined cells accumulated more nitrogen in the soil, and less in plant biomass compared with the plastic-lined cells. Nitrogen accumulated was distributed evenly among the soil, above, and below ground plant biomass. Maximum total nitrogen in the soil was found during the second sampling event (about 1100 mg kg^{-1}). About 90 percent of the accumulated nitrogen was organic.

1. INTRODUCTION

Background: Nitrogen Removal from Constructed Wetlands

In recent years wetlands, an ecotone between terrestrial and aquatic systems, have increasingly been recognized as important ecosystems because of their ability to transform and store organic matter and nutrients. This ability has been exploited in the use of Constructed Wetlands (CWs) which can be a cost effective, energy efficient technique for water quality improvement. In these artificial systems aquatic plants play a key role in wastewater treatment.

Brix (1993) classified CW systems according to macrophytic community as Free Floating-Based Systems, Submerged-Based Systems, or Emergent-Based Systems. Extensive work has been published about Free Floating-Based Systems and, overall, these systems serve as an upgrade for stabilization ponds. The Submerged-Based Systems are still in the experimental phase, even though present knowledge suggests that these systems have potential as an alternative for tertiary treatment of wastewater. The Emergent-Based Systems can be characterized by the flow pattern of the system. An Emergent-Based Systems with the water surface exposed to the atmosphere has been classified as a Free Water Surface (FWS). Subsurface-horizontal Flow (SF) is a system in which the water level is maintained below the surface of typically a gravel media. A third type of Emergent-Based Systems is a Vertical Non Saturated Flow system where water is applied in a batch flow and percolates through the media. Since the following study was conducted with emergent macrophytes this paper focuses mainly on Emergent-Based Systems.

One of the most prevalent water pollutants is nitrogen (N), especially as nitrate (NO_3^-). The main sources of N pollution in the environment are municipal effluent, agricultural waste and storm water runoff, and industrial effluents (usually from food production processing facilities). The N cycle in CW is complex because of the presence of inorganic and organic N forms in gaseous, liquid, and solid phases. Moreover, transformations among various N forms occur through a variety of biochemical, chemical, and physical reactions. Nitrogen is primarily contributed to wastewater streams as either dissolved or particulate organic compounds. Hammer and Knight (1994) summarized 52 CWs in North America and reported that the common N forms are ammonium (NH_4^+), nitrate (NO_3^-), nitrite (NO_2^-) and organic compounds.

Constructed wetlands have been used to remove N from wastewater. Removal rates of total nitrogen (TN) from CW range from 0.2 to 3.3 $\text{g m}^{-2} \text{d}^{-1}$ (Tanner, 1994; van Oostrom and Russell, 1994; Hammer and Knight, 1994). Total N removal from CW usually ranges between 25 and 70 percent of the initial loading. FWS systems appear to have an overall higher N removal rate than SF systems (Brix, 1994b). In stimulated conditions, such as improving aeration and supplying carbon, many studies report N removal of 90 percent and higher (Gersberg *et al.*, 1983, 1984, 1986; Burgoon *et al.*, 1991; Rogers *et al.*, 1991; Gale *et al.*, 1993).

The potential N removal mechanisms in CW are assimilation into plants and other living biomass, accumulation in soil, ammonia volatilisation, nitrate leaching, and denitrification. The major processes reported to effectively remove N from effluent in CW are denitrification (Gersberg *et al.*, 1984; Stengel *et al.*, 1987; van Oostrom and Russell,

1994), plant uptake (Breen, 1990; Rogers *et al.*, 1991; Adcock and Ganf, 1994), and accumulation in soil (Schierup *et al.*, 1990; van Oostrom and Russell, 1994). Constructed wetland systems designed to remove N by denitrification are preferable since these systems should behave as self sustained systems with low maintenance. Designing CW to remove N by plant uptake and soil accumulation can be an inefficient removal approach unless the biomass that accumulated would periodically be removed from the system by harvesting.

A Need for Evaluation of N Accumulation in High Organic Load Constructed Wetlands.

The use of constructed wetlands for treatment of non-point source pollution from Confined Animal Feeding Operations (CAFO) is rather recent and still in the experimental stage (DuBowy and Reaves, 1994). The NH_4^+ concentration reported in most effluent streams from CAFOs commonly exceeds 100 mgN L^{-1} and can range from 300-400 mgN L^{-1} (Cronk, 1996). Ammonia in high concentrations can be toxic to biological activities (Hills *et al.*, 1997), and at high NH_3 concentrations sufficient O_2 is crucial for nitrification and mineralization processes (Bowmer, 1987).

Little data has been published on the performance of systems containing wastewater with high N concentrations. The National Resources Conservation Service (NRCS) recommended that NH_4^+ -N concentrations from livestock operations discharged into surface wetlands should be $<100 \text{ mg L}^{-1}$. There are questions regarding the ability of constructed wetlands to adequately treat more concentrated waste and whether the processes active in low nutrient load systems are also active in high nutrient load systems.

Therefore, the capacity of constructed wetland systems to accumulate N in aquatic plants and soil profiles needs to be studied.

The objectives of this study were to 1) evaluate the removal of organic N, NH_4^+ , and NO_3^- different N species from wastewater in a free water surface constructed wetland charged with partially treated dairy wastewater, and 2) evaluate N accumulation in plant biomass and the wetland soil.

2. LITERATURE REVIEW

This review discusses the biological and physical mechanisms that influence N removal processes in CW and evaluates the performance of high N load CW systems.

Dominant N Cycle Processes in Constructed Wetlands

Mineralization (Ammonification). Mineralization is the microbial breakdown of dissolved and suspended organic material to ammonium ($\text{NH}_4\text{-N}$). This reaction is accelerated in aerobic conditions. In a CW an exponential decline of organic N was observed with increasing length of the flow path (Kadlec, 1995). The amount of this decline never exceeded the background concentration ($0.5\text{-}2.5\text{ mg L}^{-1}$) of organic N, which is equal to the equilibrium between the liquid, the organic sediments, and the biomass decay (Kadlec, 1995). Ammonification is possible in a wide range of redox potentials (Eh) and the optimum pH for ammonification is between 6.5 and 8.5. Mineralization is highly positively correlated with temperature and the mineralization rate fluctuates diurnally. In addition, Bowmer (1987) suggested that observed summer and winter differences in N removal are attributed to the effect of temperature on mineralization rate. The optimal temperature for mineralization is $40\text{-}60^\circ\text{C}$ (Hammer and Knight, 1994). At any temperature above 0°C the mineralization rate increases with temperature (Hershkowitz, 1986). In a SF gravel bed planted with reed the ammonifying bacteria were mostly associated with roots (May *et al.*, 1990; Williams *et al.*, 1994). In this setup the ammonification activity was higher in the rhizome biofilms than in the gravel bed biofilms.

Also, high NH_4^+ concentrations in the water reduced ammonification in the biofilms (Williams *et al.*, 1994).

Nitrogen Fixation. Nitrogen fixation is the process of reducing atmospheric N to ammonia by specialized bacteria and blue green algae. van Oostrom and Russell (1994) claim that this is an insignificant N loading contribution to CW systems receiving wastewater with high N loading. Furthermore, the presence of high concentrations of NH_4^+ inhibit fixation (Kadlec, 1995).

Ammonia Volatilization. Ammonia volatilization is the removal of unionized ammonia (NH_3) from the solution to the atmosphere. The unionized NH_3 fraction in the solution increases with pH and temperature. At pH 7 and $<25^\circ\text{C}$ the fraction of unionized ammonia is less than 1 percent (Kadlec, 1995). Ammonia volatilization can be appreciable at $\text{pH} > 8$ (Freney *et al.*, 1985). In systems with high NH_3 concentration ($> 20 \text{ mg L}^{-1}$), high pH, and high water temperature, ammonia volatilization may be a significant N removal mechanism. Atmospheric parameters above the CW such as wind velocity, air temperature, and NH_3 concentration in the air may significantly impact the rate of the ammonia volatilization process.

Nitrification. Nitrification is the microbial oxidation of NH_4^+ to NO_2^- and NO_3^- .

Nitrification in wetlands is thought to be a first order reaction controlled by the flux of oxygen (Kadlec and Knight, 1995). Nitrification is considered to be a limiting reaction for

N removal in CW systems receiving high organic N and/or $\text{NH}_4\text{-N}$ effluent (Schierup *et al.*, 1990; van Oostrom and Russell, 1994). High organic N increases the oxygen demand and high $\text{NH}_4\text{-N}$ lowers the redox potential, both of which inhibit nitrification (Bowmer, 1987). Based on the stoichiometric relationships for nitrification, 4.6 grams of O_2 are required to oxidize one gram of $\text{NH}_3\text{-N}$. Dissolved oxygen values of 0.3 to 2 mg L^{-1} have been reported as threshold values for nitrification (Nichols, 1983; Reddy and Patrick, 1984; Bowmer, 1987; Hammer and Knight, 1994). Although O_2 is mainly replenished by atmospheric mass-transfer, some O_2 is released from the emergent aquatic plant roots. The role of aquatic plants as oxygen suppliers, however, is difficult to assess. Some studies suggest that the aquatic plant oxygen contribution is insignificant (Brix, 1990; Howard-Williams, 1985). In high temperature conditions, however, with a fully developed root system a macrophyte may transfer enough oxygen to induce detectable nitrification (Brix, 1990; Reed and Brown, 1995; Gersberg *et al.*, 1986). The rate of oxygen release from macrophyte roots ranges from 0 to 28 $\text{g O}_2 \text{ m}^{-2} \text{ d}^{-1}$ (Brix, 1990; Armstrong *et al.*, 1990; Kadlec and Knight, 1995). Some researchers claim that in order for nitrification to occur the effluent has to contact the oxygenated root zone. Common restraints found in existing systems suggest that in many SF systems the root zone is not fully developed and as a result effluent flows below the root system (Reed and Brown, 1995). Likewise, in many FWS the soil hydraulic conductivity is low and therefore restricts the flow from the oxygenated root zone (Schierup *et al.*, 1990). The use of batch

flow in SF systems (versus continuous flow) has been suggested as a method to enhance air flow into the medium and support mineralization and nitrification. Burgoon *et al.* (1995), however, showed that batch flow in a gravel base system did not enhance oxidative processes.

For nitrification to occur in CW the air temperature should be between 10-35°C. The optimum temperature for nitrification is between 20-30°C (Nichols, 1983), and the optimum pH is 7.2-9.0 (Kadlec and Knight, 1995). Nitrification rate decreases significantly below pH 6, and below pH 5 nitrification activity is negligible (Nichols, 1983).

High concentrations of salt, $\text{NH}_4\text{-N}$, sulfide, and BOD inhibit nitrification. High salt concentrations interfere with the nitrifying bacteria (Hammer and Knight, 1994). Ammonium and sulfide cause reducing conditions (Williams *et al.*, 1994; Bowmer, 1987, respectively), and BOD competes for the available dissolved oxygen with the nitrifying bacteria. Williams *et al.* (1994) found that nitrification occurs more rapidly in rhizome biofilms than in gravel bed biofilms, while in both biofilms high concentrations of NO_3^- in the water decreases the nitrification rate.

Denitrification. Denitrification is the stepwise reduction of nitrate ($\text{NO}_3^-_{(\text{aq})}$) to nitrite ($\text{NO}_2^-_{(\text{aq})}$), to nitrous oxide ($\text{N}_2\text{O}_{(\text{g})}$), and nitrogen gas ($\text{N}_{2(\text{g})}$). Stengel *et al.* (1987) reported that 75-90 percent of the nitrate removal in a constructed wetland planted with reed (*Phragmites australis*) resulted from denitrification. Many researchers have reported that

denitrification is the major mechanism for N removal from CW (Gersberg *et al.*, 1984; Hammer and Knight, 1994; Nichols, 1983; Gale *et al.*, 1993; Brix, 1994; van Oostrom and Russell, 1994; De Busk *et al.*, 1983).

Denitrification is an anaerobic process that in CW requires a dissolved O_2 of $<3 \text{ mg L}^{-1}$ (Stengel *et al.*, 1987), although in some CW denitrification has been observed with dissolved $O_2 >3 \text{ mg L}^{-1}$ (van Oostrom and Russell, 1994). This can be explained by a dissolved O_2 gradient from the water surface to the wetland sediments that creates side by side zones with anaerobic and aerobic conditions. Under acidic conditions the denitrification rate is slower compared to neutral or alkaline conditions. At a pH of < 6 , the reduction of N_2O to N_2 is inhibited and below pH 5 chemical rather than biochemical reactions can convert N to gaseous forms (Nichols, 1983).

The primary limiting factor for denitrification is the presence of carbon in sufficient quantities (Gersberg *et al.*, 1984; van Oostrom and Russell, 1994). In CW organic matter serves as a source of C, energy, and H for denitrification. Some 5-9 g of BOD or 2.1 g of carbon at 20°C is required to convert 1 g of $NO_3\text{-N}$ to N_2 (van Oostrom and Russell, 1994). Gersberg *et al.* (1983) demonstrated the importance of organic matter for denitrification. They compared N removal in three FWS CW loaded with municipal secondary wastewater, using different sources of C. They found N removals of >95 percent and 60-86 percent, when methanol and plant mulch were added as supplement sources of C. In treatments that did not receive any C supplement the removal rate was only about 25 percent. Gersberg *et al.* (1983) and van Oostrom and Russell (1994) report

that C biomass produced by aquatic plants in CW is sufficient to sustain optimal denitrification.

Nitrate concentrations are usually not a limiting factor for denitrification. However, the NO_3^- diffusion rate toward the denitrification sites may be a limiting factor (Nichols, 1983). Nitrate tends to diffuse into anaerobic soil layers from the aerobic medium where it is created (Reddy and Patrick, 1984; Kadlec and Knight, 1995). Denitrification can occur in temperatures from 25 to 60°C (Hammer and Knight, 1994; Nichols, 1983). The denitrification rate doubles with each 10°C increase between 11-35°C, and decreases rapidly below 11°C, although it is still detectable at 2°C (Nichols, 1983).

Denitrification rates in rhizome biofilms have been reported to be 10 fold higher than in gravel bed biofilms (Williams *et al.*, 1994). Furthermore, Russell *et al.* (1994) reported that the denitrification potential of nitrified liquid in a floating plant mat was significantly higher than in the liquor and sludge in a Free Floating Based System planted with *Glyceria maxima*. The higher potential for denitrification in these zones is due to available C from plant decay and root exudation.

Nitrogen Assimilation and Release. Nitrogen assimilation refers to a variety of biological processes that convert inorganic N forms (nitrate and ammonia) into organic compounds in living cells. Flora and fauna also release organic N and $\text{NH}_4\text{-N}$ during decomposition. Studies have shown that non-vegetated CW beds consistently remove less N than

vegetated beds (Wolverton *et al.*, 1983; Gersberg *et al.*, 1984, 1986; Burgoon *et al.*, 1991; Breen, 1990). Under most conditions aquatic plants favor the assimilation of NH_4^+ over NO_3^- as a source of N. However, in nitrate rich-water, NO_3^- can become the major N source for plants. Aquatic plants produce enzymes such as nitrate reductase to convert oxidized N to a useable form, although production of nitrate reductase is inhibited in high NH_4^+ concentrations (Kadlec and Knight, 1995).

Some researchers claim that macrophyte uptake can account for only a small portion of overall N removal (Gersberg, *et al.*, 1983). However, plant assimilation has the potential to contribute significantly to the removal of N in CW. For example, the average N content in plants receiving wastewater is 2.3 percent (Kadlec, 1995), and the above ground biomass of cattail (*Typha* spp.) and reed (*Phragmites* spp.) ranges between 4,500-24,000 $\text{g m}^{-2} \text{yr}^{-1}$ (dry weight) in both temperate and tropical climates (Brix, 1994). Therefore, the potential plant N uptake is 0.28 to 1.5 $\text{g m}^{-2} \text{d}^{-1}$ with an annual average of 0.75 $\text{g m}^{-2} \text{d}^{-1}$ (Tanner, 1996). Rogers *et al.* (1991) reported that plants are responsible for more than 90 percent of the N removal in a VNSF type of system. This system apparently improved the root contact with the nutrient which enabled plants to effectively compete with microbes for the available N.

Plant N uptake rate in CW ranges from 0.6-4.4 $\text{g m}^{-2} \text{d}^{-1}$ (Zhu and Sikora, 1994; Burgoon *et al.*, 1991). The N content in plant tissues and N removal due to plant assimilation is correlated to the hydraulic loading rate (Burgoon *et al.*, 1991). In a low N loading system 0.07- 2.7 $\text{g m}^{-2} \text{d}^{-1}$ plant acquisition was assumed to be responsible for 65

percent of the N removal from the wastewater (Roger *et al.*, 1991). Tanner (1996) investigated eight aquatic species in a SF system receiving high $\text{NH}_4\text{-N}$ loading (87 mg L^{-1}). He reported that all plants showed positive growth without toxicity or nutrient deficiency symptoms. However, different plant species acquired N at different rates. The absence of NO_3^- and a C source enhances the NO_3^- uptake by plants, and the addition of C significantly reduces plant assimilation, while increasing the denitrification rate (Gersberg *et al.*, 1984; Zhu and Sikora, 1994). In emergent aquatic plants, the above ground to below ground biomass ratio ranges from 0.35 to 3.35 (Tanner, 1996) and the rhizomes contain about 90 percent of the below ground biomass (Adcock and Ganf, 1994). This above/below ground ratio suggests that above ground harvesting may remove <50 percent of the biomass. The above and below ground concentration of N ranges from 1.5-3.2 percent of the dry weight and maximum plant accumulation is some 135 g N m^{-2} (Tanner, 1996).

Nitrogen removal by plants is considered a temporary storage mechanism, which requires harvesting for permanent removal because N assimilated by the biota is partially released during the decay process. Kadlec (1995) reported a 20 percent weight loss of plant mass from (*Typha*) and a greater loss for soft tissue plants (*Sagittaria* spp.) during death and decay. The N return rate from decaying plant biomass to the system is $0.04\text{-}0.07 \text{ g m}^{-2} \text{ d}^{-1}$. This rate is divided equally between below and above ground biomass (Kadlec and Knight, 1995). This is a calculated daily value based on yearly values that are not spread evenly over the entire year. The life cycle of emergent above ground biomass is shorter (5-9 months) than the below ground biomass (i.e., 1.5-2 yr for *Typha* spp., and

more than 3 yr for *Phragmites* spp.) (Zhu *et al.*, 1994). Assimilation of N by microorganisms and algae have not been studied in CW for wastewater treatment, and the magnitude of these processes are not well understood (Kadlec and Knight, 1995).

The information above suggests that any design of CW with plant assimilation as a major N removal mechanism should consider plants with a high above/below ground ratio and no seasonal senescence.

Nitrogen Accumulation in the Soil. Nitrogen flux as deposition to the soil as organic matter results from settling of solids, death of below ground biota, and litter fall.

Quantitative information on the relative importance of accumulation of N by the soil is scarce, since few experimental CW for wastewater treatment systems have been operated over long periods (Howard-Williams, 1985; Kadlec *et al.*, 1979). In effluent that contains Total Suspended Solids (TSS) $>20 \text{ mg L}^{-1}$, CW have been shown to be efficient in removing TSS (66 - 98 percent). Solids removal is mainly related to sedimentation to the bed media (Kadlec and Knight, 1995). The quantity of N found in suspension is variable and depends mainly upon the organic matter discharged in the wastewater. Some studies have shown that N removal due to sedimentation of suspended solids is a significant process (Schierup *et al.*, 1990). In a typical CW for municipal wastewater treatment $1\text{-}2 \text{ mm yr}^{-1}$ of low bulk density solids (about 0.03 g cm^{-3}) are accumulated. This solid accretion gradually increases toward the CW inlet section (Kadlec *et al.*, 1995). It has been reported that one third of a CW volume was filled with a *Glyceria maxima* mat after

20 months of operation. Sediment accumulation accounted for 40 percent of the solids removed from the liquid (van Oostrom and Russell, 1994).

Residence time is a major factor that influences the sedimentation process. Vegetation density also plays a role in the deposition of organic matter into the bed by decreasing the flow velocity. Conversely, vegetation also can contribute to preferential flow which can decrease sedimentation (Brueske *et al.*, 1994). Other factors that influence residence time are operational parameters such as hydraulic rate load and water depth. Literature is scarce on soil deposition processes that are responsible for N removal in CW and most of the reports are from studies done on natural wetland sediments. In many natural wetland ecosystems, approximately 95 percent of the total N is present in the soil; moreover, the N is proportional to the peat accumulation rate. In these systems the organic-matter microbial decomposition is incomplete and the remaining nutrients are stored as peat (Kadlec *et al.*, 1979). It seems that soils in natural wetlands that have received effluent for some duration would show a N concentration increase. However, in several studies no N accumulation was found (Howard-Williams, 1985). Quantitative information on the relative importance of sediment adsorption processes versus uptake by the various plant and microbial communities in CW is not readily available (Howard-Williams, 1985; Kadlec *et al.*, 1979; and Kadlec and Knight, 1995).

Ammonium Adsorption. Ammonium adsorption is the process of ionized NH_4^+ removal from the solution through cation exchange adsorption to soil minerals or organic matter. In systems that contain $10 \text{ mg L}^{-1} \text{ NH}_4\text{-N}$ the top 20 cm of the wetland substrate may

contain up to 20 g m^{-2} exchangeable $\text{NH}_4\text{-N}$. This storage of N is established quickly even in systems with low N loading (Kadlec, 1995). Sikora (1994), claims that from wastewater containing 50 mg L^{-1} $\text{NH}_4\text{-N}$ sorption onto the gravel exchangeable sites removed some 34 mg L^{-1} . Tilton (1977) reported a removal rate for $\text{NH}_3\text{-N}$ of $3.74 \text{ mg L}^{-1} \text{ d}^{-1}$ over sediment compared with only $0.5 \text{ mg L}^{-1} \text{ d}^{-1}$ in water alone partially due to soil sorption. The decrease of $\text{NH}_4\text{-N}$ in columns with no sediment was due to particulate organic matter adsorption of ammonium ions.

Nitrogen removal from CW with high nitrogen load

Because of the success of CW in treating domestic and municipal wastewater, attempts are being made to use these systems for Confined Animal Feeding Operations (CAFO). Biochemical Oxygen Demand (BOD) concentration in CAFO wastewater is usually at least an order of magnitude higher than in domestic effluent (Cronk, 1996). Most successful systems used to treat CAFO wastewater have involved pretreatment application and/or dilution of the effluent prior to the CW discharges to reduce N concentration in the effluent. Few studies have been conducted on CAFO derived wastewater with high N concentration ($\text{TN} > 80 \text{ mg L}^{-1}$). Systems with high N have resulted in a wide range of N reduction (0- 70 percent) depending on hydraulic parameters, environmental conditions, macrophyte characteristics, and N constituents in the influent (Finlayson and Chick, 1983; Bowmer, 1987; Tanner 1994; van Oostrom and Cooper, 1990).

Finlayson and Chick (1983) used an inflow concentration of about 100 mg L^{-1} TKN. They found a reduction of 14-56 percent in total N while the ammonium fraction was $<40 \text{ mg L}^{-1}$. The inflow of this system was actively aerated and nitrification was observed. Nevertheless, effluent dominated by $\text{NH}_4\text{-N}$ and organic N impaired N reduction, probably because of a lack of mineralization and nitrification (Bowmer, 1987; van Oostrom and Cooper 1990; Tanner, 1994). This has further been supported by van Oostrom and Russell (1994) who used wastewater with high NO_3^- concentration and showed a total N reduction greater than 90 percent. Cathcart *et al.* (1994) reported high N removal from a high $\text{NH}_4\text{-N}$ discharge system receiving wastewater from a swine farm. They designed a FWS that encompasses principals of passive aeration and sufficient residence time for nitrification and denitrification to occur sequentially. Bere *et al.* (1995) also reported successful removal of high NH_4^+ concentrations from dairy wastewater. In their system they incorporated a series of cells of vertical and horizontal flow at different flow rates. This design increased aeration and stimulated nitrification. Because a high NH_3 input concentration is associated with high pH, $\text{NH}_4\text{-N}$ volatilization may be a significant N removal process. Moreover, in high organic N loads suspended solids removal may contribute greatly to the overall N reduction since a high fraction of the organic N is present in the suspended solids. Further research is needed to study the removal processes in high N load systems.

3. THESIS FORMAT

This thesis includes a summary of the findings of my study which are appended as a manuscript. The manuscript (**APPENDIX A**) describes the performance of constructed wetland for N removal from dairy wastewater stream and the ability of the constructed wetland to accumulate N in the soil and plant biomass. The manuscript will be submitted for publication to the Journal of the American Water Resources Association.

This study was conducted as a part of a demonstration project to evaluate the feasibility of using a free water surface constructed wetland to treat dairy wastewater for reuse and/or recharge. The principal investigators are Mr. Robert J. Freitas from the Department of Soil, Water, and Environmental Science and Dr. Martin M. Karpiscak from the Office of Arid Lands Studies, both at the University of Arizona. The wetland was constructed in the summer of 1996 at a large commercial dairy in Glendale, Arizona. More details about the experimental site are given in the manuscript.

The study was planned under the supervision of Dr. Thomas A. Doerge. Water samples were collected as a part of a biweekly sampling routine from the site while the N accumulation study was performed during the 1997/98 season, during three major sampling events. The samples were collected and prepared by the author, Mr. Freitas, Dr. Karpiscak, Mr. Glenn France (a research specialist) and student research assistants. The samples were analyzed by Scott White (a research specialist) in the laboratory of Dr. Thomas L. Thompson, who is my major advisor. I wrote this original manuscript under the supervision of my major advisor and the principal investigators.

4. PRESENT STUDY

The description of the study site, methods, results, discussion and conclusions are presented in the manuscript appended to this thesis. The following is a summary of the most important findings in this study.

Summary of Methods

To quantify soil and plant N biomass accumulation in constructed wetland for dairy wastewater treatment, Perforated Core Liners (PCLs) were inserted 60 cm into the soil media in three locations in each of eight wetland cells (total of twenty four locations) during the final phase of the system construction. The PCLs were designed to contain an intact soil column and plant biomass confined to a known volume and area. Each PCL unit was constructed of schedule 40 PVC pipe, 15 cm in diameter, 70 cm long, perforated with twenty two holes one cm in diameter, and plugged at the bottom. Each PCL contained one plant. One PCL from each sampling location was removed from the cells using a crane on Jun. 1997, Oct. 1997, and Feb. 1998. Above ground plant biomass was harvested from the removed PCLs at the time of removal. Each PCL was cut in to three segments using a saw. The content of each segments was then separated into below ground plant biomass and soil. All plant tissue samples were analyzed for total N using a modified micro Kjeldahl method. Soil samples were analyzed for $\text{NH}_4\text{-N}$, $\text{NO}_3\text{-N}$, and total N. In addition, bi-weekly water samples from the wetland inlet and outlet were also analyzed for all of the above.

Summary of Results

Nitrogen Removal. Data from water samples collected from Mar. 1997 to Feb. 1998 showed average reductions of 17, 13, and 35 percent for total N, organic N, and $\text{NH}_4\text{-N}$, respectively. Concentrations of $\text{NO}_3\text{-N}$ in wastewater were $< 2 \text{ mg L}^{-1}$ during the study period. The final N concentrations observed in the system outflow were higher than originally anticipated (247, 112, and 128 mg L^{-1} for total N, organic N, and $\text{NH}_4\text{-N}$, respectively). The major factors suspected to be responsible for this are insufficient aeration for aerobic processes such as nitrification and mineralization, and short residence time of the wastewater in the wetland cells (about 4 days).

Nitrogen Accumulation. Accumulations of total N and $\text{NH}_4\text{-N}$ in the soil profile were not correlated with distance from the system inlet. The clay-lined cells had significantly higher concentrations of N in the soil during the three sampling events and for the three depth segments. However, accumulation of N in the plant biomass was higher in the plastic-lined cells.

At all three sampling events the soil organic N concentrations decreased with depth both in the plastic and clay lined cells, with the upper 15 cm having substantially more organic N than the deeper layers (15-60 cm). Organic N in the soil at the second sampling event was not significantly different from organic N at the third sampling event in both liner types. In the three sampling events 80-86 percent of the total N accumulated in the soil was organic whereas the remainder was $\text{NH}_4\text{-N}$.

Ammonium accumulation in the soil profile showed different trends for the clay and the plastic liners. In the plastic-lined cells the $\text{NH}_4\text{-N}$ concentration decreased with depth for the three sampling events whereas in the clay-lined cells the differences between depth segments became smaller with time. In the clay-lined cells the concentration increase as a function of time was smaller. This indicated that the system has a limited capacity to accumulate $\text{NH}_4\text{-N}$.

Overall, plant biomass accumulated about 60-70 percent of the total N accumulated within these cells, approximately 3,000-3,500 kg TN/ha. These values were calculated from the first two cells only. The above and below ground biomass ratio was about 0.8. Average N content in the above ground dry tissue was 2 ± 0.15 percent and in the below ground tissues was 1.56 ± 0.2 percent, respectively.

Summary of Conclusions

The use of a constructed wetland to treat dairy wastewater reduced the concentration of the N species in the water. However, only about 17 percent of the total N introduced into the wetland was removed from the wastewater stream. Plant biomass accumulated about 60 percent of the total N accumulated in the wetland system. This accumulation was divided almost equally between the above and below ground tissues. The cell liners (plastic versus clay) performed differently with respect to N accumulation in the soil and plants. Overall, the clay-lined cells had higher accumulation of N in the soil and less accumulation of N in the plant biomass. Organic N and $\text{NH}_4\text{-N}$ concentrations decreased in the lower soil profile. In addition, the concentration in the soil of $\text{NH}_4\text{-N}$ in

the clay-lined cells, and organic N in both liners did not appear to increase from the second to the third sampling event. This suggests that soil bed based FWS constructed wetlands that receive high nutrient concentrations have a limited capacity to accumulate N in soil and plant biomass.

APPENDIX A:
NITROGEN ACCUMULATION IN A CONSTRUCTED WETLAND FOR
DAIRY WASTEWATER TREATMENT

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NITROGEN ACCUMULATION IN A CONSTRUCTED WETLAND FOR DAIRY WASTEWATER TREATMENT

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ABSTRACT: Constructed wetlands have a potential to remove nitrogen (N) from wastewater. A free water surface (FWS) constructed wetland was installed at a dairy in Glendale, Arizona. The system consists of eight cells (70 x 9 x 1.5 m) planted with *Typha domingensis*, *Scirpus validus*, and *Phragmites australis*. A series of four cells was lined with plastic and a parallel series was lined with clay. The cells received approximately 250 m³ d⁻¹ of partially treated effluent. The objectives were to evaluate 1) N removal from the wastewater and 2) N accumulation in soil and plant tissues. Above and below ground plant tissues, and soil samples were collected from twenty-four locations during one year. Total N removal from wastewater was about 17 percent. The clay-lined cells accumulated more N in the soil, and less in plant biomass compared with the plastic-lined cells. Plant biomass was accounted for approximately 60 percent of the total N accumulated in cells that established dense plant community. Maximum total N in the soil (1,100 mg kg⁻¹) was found eight month after establishment. Ninety percent of the accumulated N was organic. The results suggest that the system had a limited capacity to accumulate N.

(KEY TERMS: constructed wetlands; dairy wastewater; nitrogen removal; nitrogen accumulation; CAFO; emergent macrophytes)

INTRODUCTION

Constructed wetlands are an alternative technology for the removal of nitrogen (N) from wastewater. They are being used to treat a variety of wastewater including municipal sewage, industrial wastes, and acid mine drainage (Hammer and Knight, 1994). The most effective and sustainable N removal approach for constructed wetlands is sequential biological nitrification - denitrification (Gersberg *et al.*, 1986). However, studies have identified other significant N removal mechanisms in constructed wetlands such as plant uptake (Rogers *et al.*, 1991), sedimentation (Kadlec and Knight, 1995), and NH_3 volatilization (Skarda *et al.*, 1994). The effectiveness of N removal mechanisms depends on many factors, including season and temperature, (Newman and Clausen, 1997), hydraulic retention time (Kadlec and Knight, 1995), and availability of dissolved O_2 (Brix and Schierup, 1990).

The use of constructed wetlands for treatment of nonpoint source pollution from Confined Animal Feeding Operations (CAFOs) is recent and still experimental (DuBoway and Reaves, 1994). The NH_4 concentrations in most effluent streams from CAFOs commonly exceed 100 mgN L^{-1} and can range from 300-400 mgN L^{-1} (Cronk, 1996). Ammonium at these high concentration can be toxic to biological activities (Hill *et al.*, 1997). At such high $\text{NH}_4\text{-H}$ concentrations, sufficient oxygen is crucial for nitrification and mineralization to proceed (Bowmer, 1987).

Most studies conducted with CAFO wastewater report diluting the wastewater with fresh water to reduce N concentration (Hunt *et al.*, 1994). Thus, little data has been published on the performance of systems containing high NH_4 concentrations. The

National Resource Conservation Service recommended that NH_4 concentrations from livestock operations discharged into surface wetlands should be $<100 \text{ mg L}^{-1}$ (NRCS, 1991). There are questions regarding the ability of constructed wetlands to adequately treat highly concentrated wastewater and whether the active N removal processes in low nutrient load systems remain effective in high nutrient load systems. Moreover, the capacity of constructed wetland systems to accumulate N in the aquatic plants and soil profile needs to be studied.

The present study was conducted at a free water surface (FWS) constructed wetland receiving partially treated dairy wastewater. The objectives of this study were to 1) evaluate the removal of different N species from wastewater and 2) evaluate N accumulation in plant biomass and soil.

MATERIALS AND METHODS

Study site

Eight free water surface (FWS) wetland cells were constructed in July 1996, on a dairy farm in Glendale, Arizona, USA. Each cell measured 70 m long, 9 m wide, and about 1.5 m deep. The combined surface area of the eight cells was about 0.5 ha. Depth of wastewater flow varied between 0.15 and 0.3 m. The constructed wetland was operated as two parallel multi celled systems (Figure 1). Water flowed from cell to cell by gravity due to a slope gradient of 0.3 percent between the cells. One system (cells 1, 3, 5, and 7) was lined with an impermeable 20 mil high-density polypropylene plastic liner

placed on a 0.3 m deep compacted bed of local topsoil (sandy clay loam). The other system (cells 2, 4, 6, and 8) was lined solely with 0.6 m of compacted local topsoil. Both systems were then back filled with an additional 0.6 m of local subsoil (sandy loam) that had been excavated during the wetland construction. The backfilled soil was compacted to a bulk density of about 1.6 g cm^{-3} .

The wastewater entering the wetland system was produced in the milk house and was mixed with any rainfall and runoff from the dairy. The milkhouse wastewater is used for feed lane flushing in one of the central barns from the manure before it enters the treatment system. Before the wastewater enters the constructed wetland it undergoes primary treatment in pair of solid separators, followed by additional treatment in a pair of anaerobic lagoons, and finally a pair of shallow aerobic ponds for atmospheric aeration. For additional details on the overall dairy waste management system configuration and performance see Karpiscak *et al.*, (submitted).

During the first five months of operation (Mar -Jul., 1997) the wetland system received an intermittent (about once per week) batch flow of dairy wastewater. In the summer months (Aug-Sep, 1997) the system received batch flows of irrigation water because of the high salt concentration found in the wastewater. Starting in Sep. 1997 the wetland system received a continuous flow of wastewater at a rate of $150\text{-}300 \text{ L min}^{-1}$ with an approximate hydraulic retention time of 20 hr per cell. During the study period the system received 120 mm of rainfall and the daily average water temperature ranged from 16 - 22°C.

Vegetation

Emergent aquatic plant species growing naturally in Arizona and typically used in constructed wetlands were chosen for this study. In Sep. 1996 cattail (*Typha domingensis*) and soft-stem bulrush (*Scirpus validus*) transplants were placed into cells 1/2 and 3/4, respectively. Soft-stem bulrush seedlings from a different source were placed into cells 5/6 in Oct. 1996. In Nov. 1996 rhizomes of common reed (*Phragmites australis*) were planted in cells 7/8. When the common reed rhizomes showed little growth as of Feb. 1997, cells 7/8 were replanted in Mar. 1997 with transplants. The initial planting density in all the cells was approximately 1 plant per m² (about 600-800 plants per cell). Before the wetland cells began receiving dairy wastewater in Feb. 1997, all cells received a weekly batch flow of irrigation water to foster plant growth. During the study period canopy cover of the plants were fully established in cells 1/2, while within the other wetland cells the plant canopy was only partially established (Karpiscak *et al.*, (submitted)).

Sampling methods

To quantify soil and plant N biomass accumulation several Perforated Core Liners (PCLs) were installed within each wetland cell. The PCLs were used so that an intact soil profile and plant tissue could be collected from a known volume and area. The PCLs were made of 15 cm diameter schedule 40 PVC pipe, and were 70 cm long and perforated with twenty two 1.5 cm diameter holes in order to simulate the flow in the surrounding environment (Figure 2). A 15 cm PVC coupling was placed on top of the PCLs to form a reinforced collar. Holes were drilled through the collar, to enable hooks to be attached for

removal of the PCLs from the cells. A 15 cm inside griper plugs were used to seal the bottoms of the PCLs.

The PCLs were inserted vertically 60 cm into the soil during the final phase of the system construction. A ceramic tile was placed below each unit to protect the plastic liner. The PCLs were then filled with the surrounding soil and a single plant was planted in each unit. Five PCLs were placed about 2 meters apart in each of three locations along the length dimension of each cell (Figure 1). One PCL from each location within each cell was removed by crane on 30 Jun, 1997; 12 Oct, 1997; and 5 Feb, 1998. The two PCLs that remained in the ground in each sampling location will be extracted in the future.

Sample processing and analysis

Above ground plant biomass from the removed PCLs was harvested. Each PCL was cut into three segments (0-15, 15-30, and 30-60 cm) using a mechanical saw. The contents of each segments were separated into below ground plant biomass and soil. Soil and plant samples were oven dried at 60°C for about seven days. The dried plant tissue samples were ground to pass a 40 mesh sieve and the soil samples were ball milled to < 100 mesh. The soil samples were extracted with KCl and $\text{NH}_4\text{-N}$ and $\text{NO}_3\text{-N}$ were determined using steam distillation (Mulvaney, 1996). Plant samples and extracted soil samples were analyzed for total N using a modified micro Kjeldahl method (Bremner, 1996).

Water samples were collected every two weeks from the wetland inlet and outlet and were analyzed for total N and inorganic N. Organic N concentrations were calculated as the difference between total N and the sum of the inorganic N species.

RESULTS AND DISCUSSION

Nitrogen Removal

Chemical properties of wastewater from the constructed wetland inlet and outlet are presented in Table 1. Data are from samples collected from Mar. 1997 to Feb. 1998, excluding the period that the system received irrigation water. We observed reductions of $\text{NH}_4\text{-N}$, total N, and organic N of 35, 17, and 13 percent, respectively.

The NRCS (1991) recommended $<100 \text{ mg NH}_4\text{-N L}^{-1}$ for wetlands treating livestock wastewater, and a total N input rate of $<10 \text{ kg ha}^{-1} \text{ d}^{-1}$. The $\text{NH}_4\text{-N}$ and organic N concentrations that entered this wetland system (about 200 and 130 mg L^{-1} , respectively) and total N ($> 20 \text{ kg ha}^{-1} \text{ d}^{-1}$) all exceeded these recommendations. As a result, the N concentrations in the effluent leaving the wetland were higher than originally anticipated. The N removal was less than desired, the most likely reasons being insufficient aeration for aerobic processes such as nitrification and mineralization (Bowmer, 1987), and short residence time of the wastewater in the wetland cells (about 4 days) (Gale *et al.*, 1983).

Nitrate concentrations in the water were $<2 \text{ mg L}^{-1}$. This may be due to either limited nitrification in the system (van Oostrom *et al.*, 1994) or nitrification followed immediately by denitrification (Sievers, 1997). Constructed wetlands have been reported to denitrify NO_3 in effluent efficiently (up to about $30 \text{ kg ha}^{-1} \text{ d}^{-1}$) due to induced anaerobic conditions and high C availability (Gersberg *et al.*, 1986; and van Oostrom *et al.*, 1994).

High NH_4 concentrations stimulate reducing conditions that decrease rates of oxidative processes. Furthermore, some of the plant species used in our study may be

susceptible to high NH_4 concentrations (Finlayson *et al.*, 1983; Hill *et al.*, 1997). The high concentration of organic N was accompanied by a high concentration of organic C. This would result in a high biochemical oxygen demand, while at the same time the low dissolved O_2 created competition between heterotrophic bacteria and autotrophic nitrifying bacteria.

While processing the PCLs for analysis, we observed small root systems that in most of the samples did not extend below the upper 15 cm. Furthermore, almost no rhizomes developed. Mature root systems in constructed wetlands are reported to be a crucial factor for N removal. However, extensive root systems seldom occur in constructed wetlands (Brix and Schierup, 1990). The factors limiting root growth include: insufficient contact between the roots and the wastewater flow (Kadlec and Knight, 1995), deposition of toxic substances and creation of a black anoxic layer below the root zone (Biddlestone *et al.*, 1991), and by plants acquiring nutritional requirements directly from the wastewater flow (Karpiscak 1998). In our system both low hydraulic conductivity of the bed and abundance of nutrients in the flow are suspected as reasons for inhibited root growth.

Nitrogen Accumulation

Accumulation of total N and $\text{NH}_4\text{-N}$ in the soil was weakly correlated to distance from the system inlet. Most of the regressions indicate that the accumulation of N was not correlated with distance from the system inlet (avg $r = 0.36$ and 0.55 for total N and $\text{NH}_4\text{-N}$, respectively). This weak correlation was probably because 1) during the first operation period (March - June 97) the wetland was operated with an intermittent batch flow, and 2)

during the continuous flow operation period (September 97 - February 98) the wastewater residence time within the wetland cells (about four days) was not sufficient to create a gradient in N concentration along the length of the cells. Therefore, for the purpose of statistical analysis all PCLs within the cells were considered to be replicates.

Concentrations of total N and $\text{NH}_4\text{-N}$ in the soil were higher in the clay-lined cells compared to the plastic-lined cells at each of the three sampling events and the three soil depth segments (Table 2). Ammonium accumulation in the soil had higher differences between the liners compared with total N.

The higher soil N accumulation in the clay-lined cells versus the plastic-lined cells was unexpected. The initial hypothesis was that losses of N due to leaching in the clay-lined cells is negligible, since the surface horizontal flow rate (about 100 m d^{-1}) was 2 to 3 fold faster than the estimated infiltration flow rate. Results of subsurface water content measurements using neutron probes (Frietas and Karpiscak, 1997) indicted that leaching occurred below the clay-lined cell while, but not below the plastic-lined cells. Bias due to experimental error should have been minimized since all the cells were constructed from the same soil to a uniform compaction, received the same wastewater at the same flow rate, and the soil samples were processed and analyzed at the same manner. Therefore, these differences remain unexplained.

Means of total N and organic N accumulations by soil depth are presented in Figures 3 and 4, respectively. For all three sampling events the total N concentrations in the soil decreased with depth. The upper 15 cm accumulated a substantially higher concentration of total and organic N than the middle (15-30 cm) and the deepest (30-60

cm) layers. The upper 15 cm accumulated 42-48 percent and 52-57 percent of the total N accumulated during the first and the later sampling events, respectively. Differences in total N accumulation between the middle and the deepest layers were small. The middle layer accumulated 24 percent and 31 percent of the total N in the soil during June 97 and the later dates, respectively. Thus, the results show that in the soil bed of a surface flow constructed wetland the lower soil profile (15-60 cm) may serve as a considerable sink of nutrients.

The total N accumulated in the three soil depths in October 97 (1,800 and 2,270, mg N kg⁻¹ soil, for the plastic and clay, respectively) were not significantly different from the total N values found at the second sampling event (February 98). This probably indicates soil saturation with respect to total N accumulation by the time of the second sampling event. This assumption might be confirmed in the future if additional sampling of the remaining PCLs is conducted. The same apparent trend of saturation was observed for organic N (Figure 4). Most of the organic N measured in the soil can be attributed to the sludge that accumulated on the soil surface, organic material that penetrated into the lower layers from channels created by roots and cracks, and dissolved organic N transported with the water flow.

Ammonium in the soil (Figure 5) is from both the soil solution and NH₄ adsorbed onto exchange sites on mineral and organic matter. In the plastic-lined cells the NH₄-N concentration decreased with soil depth. The lowest NH₄-N concentration in the soil was found in the second sampling event and the highest was in the third sampling event. This could be explained by the introduction of low NH₄-N concentration irrigation water

(August -September, 97), which may have removed some of the NH_4 from the soil.

In the clay-lined cells the $\text{NH}_4\text{-N}$ concentrations were generally higher than in the plastic-lined cells. However, the differences between the soil depth segments became smaller with time. Moreover, during the second sampling event the middle layer had a higher concentrations than the upper layer. (109 mg kg^{-1} and 94 mg kg^{-1} , respectively). At the last sampling event we observed a continuing increase of NH_4^+ concentrations in all three depths of soil with insignificant ($P < 0.05$) differences among the layers in the clay-lined cells. These trends observed in the clay-lined cells suggest that the system reached saturation for $\text{NH}_4\text{-N}$ accumulation.

The nitrate concentrations for all the soil samples were $< 2 \text{ mg N/kg soil}$, suggesting that little nitrification occurred due to anaerobic conditions in the soil column.

Overall, the soil profile proved to be a major sink for N, accumulating about 30-40 percent of the N accumulated in the system. Little is known on the role of soil as a sink for N species in FWS with soil bed and the effect of liner type on the N accumulation in the soil. Further research to determine the processes contributing to these phenomena should be conducted.

Nitrogen accumulation by plants could be compared only between the first two cells (cells #1 and 2). These cells were planted with cattail (Table 3), and were the only cells that had an established mature dense plant community during the study period (Karpiscak *et al.*, Submitted). Plants grown in the plastic-lined cells produced about 30 percent more dry weight and accumulated about 20 percent more N both in the above and below ground tissues. During the study period the plants in the plastic-lined cells had

more vibrant growth, a denser canopy, and less mortality compared to the clay-lined cells. Since all plants received the same treatment, these variations between the liners were unexpected.

The overall distribution of total N accumulated in the system is presented in Figure 6. This analysis is the mean of the first two cells that were planted with cattail (cells 1 and 2). Overall, the plant biomass accumulated about 60-70 percent of the total N accumulated within these cells, approximately 3,000-3,500 kg N ha⁻¹. The above below ground total N biomass ratio was about 0.8. Based on these results, a harvesting program to remove the above ground plant biomass would remove approximately 10-20 percent of the total N introduced into the wetland. This assumes five harvests per year, complete canopy cover in the cells, and a mean flow rate of 120 L min⁻¹. Although a significant quantity of N can be removed in this manner (about 1,500-2,500 kg N yr⁻¹) significant amounts of N will remain in the below ground biomass and soil. Some studies have reported that plant uptake can account for > 90 percent of the nitrogen removal (Rogers *et al.*, 1991). However, the general consensus is that plant uptake is responsible for 10-20 percent of the N removal (Gersberg *et al.*, 1986; and Reed *et al.*, 1988).

CONCLUSIONS

The use of a constructed wetland to treat dairy wastewater reduced the concentrations of total N and NH₄-N from the stream in 17 and 35 percent, respectively. However, the wetland outflow water quality will require additional treatment because of high loading of NH₄ and organic N. Plant biomass accumulated about 60 percent of the

total N accumulated in the wetland system. This accumulation is approximately equal divided between the above and below ground tissues. About 80-86 percent of the total N accumulated in the soil was organic N and the reminder was $\text{NH}_4\text{-N}$. The clay-lined cells accumulated more N than the plastic-lined cells, although the plant biomass removed more N in the cells that were lined with plastic.

Overall, organic N and $\text{NH}_4\text{-N}$ concentrations decreased with soil depth. In addition, total and organic N concentrations in the soil did not increase from the second to the third sampling event, as well as $\text{NH}_4\text{-N}$ in the clay-lined cells. These suggests that this type of constructed wetland has a limited ability to accumulate N in the soil. In conclusion, more research is required in order to modify the overall system for achieving improved N removal.

Figure 1. Schematic of the wetland cells and the sampling locations

Figure 2. Cross section of a Perforated Core Liner (PCL)

TABLE 1. Average wastewater parameters and percent reduction in the wetland inlet and outlet¹

	Wetland Inlet (mean $\pm$ s.d.)	Wetland Outlet (mean $\pm$ s.d.)	Percent Reduction ²
Biochemical oxygen demand (mg L ⁻¹)	285 $\pm$ 97	277 $\pm$ 101	3
Total suspended solids (mg L ⁻¹)	943 $\pm$ 669	606 $\pm$ 380	36
Total kjeldahl N (mg L ⁻¹)	296 $\pm$ 109	247 $\pm$ 110	17
Organic N (mg L ⁻¹)	129 $\pm$ 104	112 $\pm$ 57	13
NH ₄ -N (mg L ⁻¹)	196 $\pm$ 113	128 $\pm$ 86	35
NO ₃ -N (mg L ⁻¹)	<2	<2	
pH	8.1 $\pm$ 0.3	8.3 $\pm$ 0.45	
Dissolved oxygen (mg L ⁻¹)	<1	<1	

¹Average values for Mar. 97 - Mar. 98 excluding Aug - Sep .97.

²Average percent reduction for Mar. 97 - Mar. 98 excluding Aug - Sep .97.

TABLE 2. Total N and $\text{NH}_4\text{-N}$ soil accumulation mean values in the plastic and the clay liners as functions of depth and time.

		June 97		October 97		February 98	
		Plastic liner (mean)	Clay liner (mean)	Plastic liner (mean)	Clay liner (mean)	Plastic liner (mean)	Clay liner (mean)
0-15 cm	Total N (mg kg^{-1})	661	685	1024	1189	921 ^a	1175
	$\text{NH}_4\text{-N}$ (mg kg^{-1})	116 ^b	156	88	101	151	157
15-30 cm	Total N (mg kg^{-1})	457	505	438	557	449	505
	$\text{NH}_4\text{-N}$ (mg kg^{-1})	92 ^a	151	68 ^a	149	123	157
30-60 cm	Total N (mg kg^{-1})	278 ^a	441	337	526	358 ^a	564
	$\text{NH}_4\text{-N}$ (mg kg^{-1})	28 ^a	105	38 ^a	126	87 ^a	166

^a Mean values for the liners at this date were different at $P < 0.01$

^b Mean values for the liners at this date were different at $P < 0.05$

TABLE 3. Above and below ground plant biomass mean values for cells 1 and 2 (cattail)

		June 97			October 97			February 98		
		Plastic liner (mean ± s.d)	Clay liner (mean ± s.d)		Plastic liner (mean ± s.d)	Clay liner (mean ± s.d)		Plastic liner (mean ± s.d)	Clay liner (mean ± s.d)	
Above ground (cattail)	N (%)	1.6 ± 0.2	1.8 ± 0.2		1.3 ± 0.1	1.3 ± 0.2		1.7 ± 0.3	1.7 ± 0.2	
	Dry weight (g)	209 ± 70	144 ± 22		139 ± 47	111 ± 58		157 ± 9	140 ± 95	
	Accumulated N (g)	3.2 ± 0.7	2.5 ± 0.2		1.8 ± 0.5	1.4 ± 0.6		2.7 ± 0.3	2.5 ± 1.9	
Below ground (cattail)	N (%)	1.6 ± 0.2	2.1 ± 0.2		2.0 ± 0.1	2.1 ± 0.1		2.3 ± 0.2	2.1 ± 0.3	
	Dry weight (g)	178 ± 65	138 ± 11		155 ± 60	120 ± 57		164 ± 82	117 ± 83	
	Accumulated N (g)	2.8 ± 1.0	2.9 ± 0.6		3.0 ± 1.0	2.4 ± 1.1		3.6 ± 1.5	2.4 ± 1.6

Figure 3. Total N (mean $\pm$ s.d.) in the soil for the plastic and clay lined cells as a function of soil depth and time.

Figure 4. Organic N (mean $\pm$ s.d.) in the soil for plastic and clay lined cells as a function of soil depth and time.

Figure 5. $\text{NH}_4^+ \text{-N}$ (mean $\pm$ s.d.) in the soil for plastic and clay lined cells as a function of soil depth and time.

Figure 6. Total Nitrogen Distribution in the Plants and Soil of Cells # 1 and 2

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APPENDIX B:
RAW DATA

TKN concentration in the soil as found in the PCLs expressed in mg N per Kg of soil

PCL Location ¹	June 97			October 97			February 98		
	0-15 cm	15-30 cm	30-60 cm	0-15 cm	15-30 cm	30-60 cm	0-15 cm	15-30 cm	30-60 cm
1N	829.7	367.2	299.2	754.8	1407.6	387.6	1050.0	357.0	371.0
1C	707.3	438.1	374.0	938.4	700.4	421.6	693.0	707.0	469.0
1S	410.0	320.9	188.0	761.6	434.0	537.2	861.0	602.0	441.0
3N	521.0	560.6	443.3	1217.2	326.4	340.0	784.0	343.0	266.0
3C	555.5	291.5	208.7	1366.8	N/A	353.6	602.0	322.0	371.0
3S	741.2	477.8	476.0	768.4	319.6	319.6	777.0	364.0	322.0
5N	1040.0	609.0	236.3	1067.6	380.8	319.6	1673.0	539.0	350.0
5C	640.0	300.2	201.8	791.0	367.2	285.6	728.0	679.0	448.0
5S	374.3	362.2	176.0	1169.6	299.2	224.4	847.0	385.0	280.0
7N	884.9	666.4	263.9	1353.2	374.0	346.8	651.0	364.0	406.0
7C	555.5	603.7	238.0	897.6	265.2	231.2	1169.0	399.0	308.0
7S	672.8	483.0	231.2	1210.4	380.8	285.6	1218.0	329.0	266.0
2N	581.3	564.4	382.9	1244.4	618.8	516.8	882.0	728.0	343.0
2C	872.9	602.0	770.0	1400.8	782.0	408.0	840.0	525.0	511.0
2S	948.7	569.2	201.8	1353.2	782.0	421.6	1253.0	406.0	406.0
4N	539.9	479.6	389.9	958.8	530.4	544.0	1274.0	651.0	784.0
4C	700.4	422.6	414.8	1033.6	401.2	571.2	854.0	0.0	819.0
4S	831.5	523.6	643.4	1292.0	476.0	644.0	1428.0	644.0	483.0
6N	671.0	505.4	441.0	1706.8	510.0	523.6	1547.0	644.0	525.0
6C	591.6	452.0	408.0	856.8	564.4	469.2	812.0	581.0	497.0
6S	719.3	341.6	410.6	1047.2	462.4	598.4	1484.0	532.0	658.0
8N	762.4	465.7	436.4	1366.8	476.0	571.2	1120.0	413.0	623.0
8C	300.2	534.8	429.5	1026.8	625.6	537.2	1568.0	602.0	574.0
8S	698.6	602.0	362.2	986.0	448.8	510.0	1036.0	336.0	546.0

¹ The integers represent the cell number, while the letters represent the location within the cells: N = North, C = Center, S = South, C = Center, S = South.

Organic N concentration in the soil as found in the PCLs expressed in mg N per Kg of soil ¹

PCL Location ²	June 97			October 97			February 98		
	0-15 cm	15-30 cm	30-60 cm	0-15 cm	15-30 cm	30-60 cm	0-15 cm	15-30 cm	30-60 cm
1N	631.0	125.8	258.4	597.7	1275.3	336.2	890.1	159.9	259.9
1C	560.0	290.9	250.7	838.7	644.5	348.9	520.6	453.7	250.4
1S	358.2	282.9	169.2	597.3	319.3	426.5	782.3	418.9	318.0
3N	330.0	258.6	420.0	1117.8	258.6	315.8	303.8	243.8	214.4
3C	402.9	253.3	199.2	1238.0	N/A	333.0	438.2	154.9	326.7
3S	614.8	387.3	446.0	653.1	261.8	294.8	617.4	202.5	271.8
5N	984.1	583.9	221.4	1033.8	351.3	302.8	1549.8	521.3	285.1
5C	517.4	230.9	184.7	694.9	301.3	263.0	593.3	567.9	375.0
5S	262.7	306.0	164.6	1096.5	138.1	196.6	729.8	336.2	185.3
7N	816.6	635.4	248.1	1339.6	351.0	323.0	589.8	237.1	302.6
7C	489.8	579.4	226.4	865.8	231.5	198.7	1111.9	362.8	269.0
7S	569.2	437.7	213.0	1169.5	349.0	260.9	1112.9	253.9	199.3
2N	348.1	424.5	191.9	1089.5	534.0	431.1	798.4	610.9	294.5
2C	609.4	405.7	559.1	1286.5	516.4	275.1	590.8	231.9	341.4
2S	739.0	377.5	104.4	1184.0	625.1	311.8	1013.3	92.6	146.5
4N	399.6	270.7	211.9	818.3	278.5	472.6	1079.0	382.4	537.1
4C	553.6	200.3	322.7	939.3	176.6	438.4	707.0	-160.8	574.9
4S	748.6	314.9	587.2	1154.5	329.3	452.4	1233.8	495.8	272.1
6N	521.0	396.5	416.8	1654.8	366.9	305.2	1415.8	583.6	371.2
6C	466.1	246.1	373.8	790.5	404.7	369.1	692.7	441.2	322.2
6S	560.8	291.6	288.1	915.4	372.5	466.8	1347.5	439.8	524.8
8N	659.3	420.3	401.1	1341.5	401.2	474.3	988.5	284.0	531.1
8C	190.7	449.4	371.5	981.9	512.1	458.1	1413.0	516.1	438.4
8S	546.1	458.8	208.0	909.4	367.3	351.1	929.5	258.5	418.4

¹ The organic N is calculated from the difference between the TKN and NH₄-N.² The integers represent the cell number, while the letters represent the location within the cells: N = North, C = Center, S = South.

NH₄⁺-N concentration in the soil as found in the PCIs, expressed as a mg N per kg of soil

PCI location ¹	June 97			October 97			February 98		
	0-15 cm	15-30 cm	30-60 cm	0-15 cm	15-30 cm	30-60 cm	0-15 cm	15-30 cm	30-60 cm
1N	198.7	241.4	40.9	157.2	132.3	51.4	150.0	197.1	111.1
1C	147.2	147.3	123.3	99.8	55.9	72.7	172.4	253.3	218.6
1S	51.8	37.9	18.9	164.3	114.7	110.7	78.8	183.1	123.0
3N	191.	302.0	23.6	99.4	67.8	24.2	480.2	99.2	51.6
3C	152.5	38.3	9.5	128.8	46.8	20.6	163.8	167.1	44.3
3S	126.4	90.6	30.0	115.3	57.8	24.9	159.6	161.5	50.2
5N	55.9	25.3	15.0	33.8	29.5	16.8	123.2	17.7	64.9
5C	122.6	69.2	17.1	96.1	65.9	22.6	134.8	111.1	73.0
5S	111.6	56.2	11.3	73.2	161.1	27.8	117.3	48.8	94.7
7N	68.4	31.1	15.9	13.7	23.0	23.8	61.2	126.9	103.4
7C	65.7	24.4	11.6	31.8	33.7	32.6	57.2	36.2	39.0
7S	103.6	45.3	18.2	40.9	31.8	24.7	105.1	75.1	66.7
2N	233.2	139.9	191.0	154.9	84.8	85.7	83.6	117.1	48.5
2C	263.5	196.3	210.9	114.3	265.6	132.9	249.2	293.1	169.6
2S	209.8	191.7	97.4	169.2	156.9	109.8	239.8	313.4	259.5
4N	140.3	208.9	177.9	140.5	251.9	71.4	195.0	268.6	246.9
4C	146.8	222.4	92.1	94.3	224.6	132.8	147.0	160.8	244.1
4S	82.8	208.7	56.2	137.6	146.7	191.6	194.3	148.2	210.9
6N	150.1	109.0	24.2	52.0	143.1	218.4	131.3	60.4	153.8
6C	125.5	205.9	34.2	66.3	159.7	100.1	119.4	139.8	174.8
6S	158.5	50.0	122.4	131.8	89.9	131.6	136.5	92.2	133.2
8N	103.2	45.4	35.3	25.3	74.8	97.0	131.5	129.0	91.9
8C	109.4	85.3	58.0	44.9	113.5	79.1	155.0	85.9	135.6
8S	152.5	143.3	154.3	76.6	81.5	158.9	106.5	77.5	127.6

¹ The integers represent the cell number, while the letters represent the location within the cells: N = North, C = Center, S = South.

NO₃-N concentration in the soil as found in the PCLs expressed in mg N per Kg of soil

PCL Location	June 97			October 97			February 98		
	0-15 cm	15-30 cm	30-60 cm	0-15 cm	15-30 cm	30-60 cm	0-15 cm	15-30 cm	30-60 cm
1N	1.3	5.5	1.6	2.4	0.9	1.4	2.3	0.7	0.4
1C	1.3	1.8	0.0	1.8	1.1	0.9	1.2	1.1	2.5
1S	0.6	0.0	2.4	2.0	2.3	0.9	1.2	2.3	0.0
3N	1.3	1.5	1.5	1.8	0.7	1.1	1.7	1.7	0.0
3C	0.0	1.7	0.0	2.2	1.4	1.1	N/A	2.1	0.0
3S	1.5	1.5	1.1	1.7	0.5	7.6	1.4	1.1	1.1
5N	1.3	1.7	1.5	2.0	2.1	1.5	1.4	0.4	0.7
5C	0.8	1.8	1.3	0.3	1.6	2.0	2.5	1.4	2.5
5S	1.2	1.5	1.1	1.8	1.4	1.3	3.5	1.4	1.4
7N	1.8	1.1	1.6	1.3	0.9	0.8	3.9	0.4	1.1
7C	1.8	1.3	2.5	1.4	1.9	1.3	1.9	2.8	0.7
7S	1.3	0.8	1.3	2.6	2.1	1.1	4.0	0.4	0.0
2N	2.0	0.8	1.1	3.2	0.7	0.7	0.9	0.7	0.0
2C	2.3	0.9	1.1	3.2	2.3	1.8	1.6	0.4	0.7
2S	2.0	2.0	1.7	4.5	1.4	0.9	3.3	1.1	0.4
4N	2.0	2.0	1.5	1.8	1.6	2.4	2.5	N/A	1.7
4C	1.6	1.2	1.6	1.3	0.9	1.1	0.4	2.5	1.4
4S	1.3	2.0	1.5	1.1	1.9	1.8	1.4	3.2	1.4
6N	1.2	3.1	1.6	1.8	1.2	1.3	0.4	0.0	0.0
6C	6.5	2.0	1.3	0.8	0.0	2.0	1.4	1.1	0.0
6S	1.1	1.8	1.3	31.9	1.9	1.0	2.3	0.0	1.1
8N	1.3	1.1	1.9	1.7	1.4	2.0	2.5	1.4	0.4
8C	1.2	0.8	1.3	1.7	1.1	0.8	0.0	0.7	0.0

¹ The integers represent the cell number, while the letters represent the location within the cells: N = North; C = Center; S = South.

Total N observed in above ground shoot biomass collected from the PCLs.

PCL Location ¹	June 97			October 97			February 98		
	N (%)	DW (g)	N (g)	N (%)	DW (g)	N (g)	N (%)	DW (g)	N (g)
1N	1.7	143.2	2.4	1.4	88.1	1.2	1.5	157.9	2.4
1C	1.8	178.6	3.2	1.2	201.5	2.4	2.1	146.2	3.1
1S	1.3	305.6	4.1	1.3	126.4	1.7	1.6	167.4	2.7
2N	1.5	174.8	2.7	1.5	37.8	0.6	1.9	274.0	5.1
2C	2.0	134.6	2.7	1.1	179.2	2.0	1.5	82.7	1.3
2S	1.8	123.4	2.2	1.4	114.7	1.6	1.6	62.7	1.0
3N	4.1	16.7	0.7	N/A	N/A	N/A	N/A	N/A	N/A
3C	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
3S	3.3	28.2	0.9	N/A	N/A	N/A	N/A	N/A	N/A
4N	2.4	16.2	0.4	N/A	N/A	N/A	N/A	N/A	N/A
4C	2.9	27.4	0.8	N/A	N/A	N/A	N/A	N/A	N/A
4S	1.9	27.3	0.5	N/A	N/A	N/A	N/A	N/A	N/A
5N	2.1	47.6	1.0	2.2	108.5	2.4	2.4	204.4	5.0
5C	2.4	18.0	0.4	N/A	N/A	N/A	N/A	N/A	N/A
5S	2.1	47.9	1.0	2.4	3.4	0.1	1.9	27.5	0.5
6N	3.0	39.8	1.2	2.5	23.2	0.6	1.6	30.3	0.5
6C	4.1	14.6	0.6	N/A	N/A	N/A	N/A	N/A	N/A
6S	2.5	29.4	0.7	2.0	23.9	0.5	2.0	46.4	0.9
7N	2.4	58.3	1.4	2.0	108.9	2.1	N/A	N/A	N/A
7C	2.0	139.3	2.8	1.0	37.7	0.4	2.0	46.0	0.9
7S	2.3	42.5	1.0	1.8	62.0	1.1	N/A	N/A	N/A
8N	2.8	32.1	0.9	1.8	21.4	0.4	N/A	N/A	N/A
8C	2.0	144.0	2.8	1.9	13.7	0.3	1.8	13.0	0.2
8S	2.1	40.7	0.9	1.8	41.8	0.7	N/A	N/A	N/A

¹ The integers represent the cell number, while the letters represent the location within the cells: N = North, C = Center, S = South.

Total N observed in below ground root biomass collected from the PCLs in the 0-15 cm soil segment.

PCL Location ¹	June 97			October 97			February 98		
	N (%)	DW (g)	N (g)	N (%)	DW (g)	N (g)	N (%)	DW (g)	N (g)
1N	1.5	89.9	1.4	2.1	88.5	1.8	2.3	115.0	2.6
1C	1.8	197.2	3.5	1.8	234.5	4.3	2.5	97.0	2.4
1S	1.4	246.5	3.4	2.1	141.2	3.0	2.1	279.0	5.7
2N	2.0	149.8	3.0	2.1	148.8	3.1	1.9	56.0	1.1
2C	1.9	122.6	2.3	2.2	41.3	0.9	2.0	234.0	4.7
2S	2.4	142.3	3.4	2.0	170.9	3.3	2.5	60.0	1.5
3N	1.3	11.5	0.2	N/A	N/A	N/A	1.6	22.0	0.4
3C	1.6	13.8	0.2	0.9	31.5	0.3	N/A	N/A	N/A
3S	1.2	6.7	0.1	1.0	113.6	1.1	N/A	N/A	N/A
4N	0.8	84.9	0.7	N/A	N/A	N/A	N/A	N/A	N/A
5N	0.2	42.5	0.1	1.7	202.0	3.4	2.1	181.0	3.8
5C	1.1	22.8	0.2	0.9	18.6	0.2	N/A	N/A	N/A
5S	1.8	76.0	1.3	1.3	27.8	0.4	N/A	N/A	N/A
6N	1.3	27.9	0.4	N/A	N/A	N/A	1.8	111.0	2.0
6C	1.7	14.2	0.2	1.4	24.9	0.4	1.1	60.0	0.7
6S	1.4	27.3	0.4	2.1	69.0	1.4	2.4	114.0	2.8
7N	1.2	11.9	0.1	1.0	42.3	0.4	N/A	N/A	N/A
7C	1.0	32.7	0.3	0.7	9.8	0.1	1.4	20.0	0.3
7S	0.7	1.5	0.0	1.2	43.0	0.5	1.2	19.0	0.2
8N	0.9	11.3	0.1	0.8	34.3	0.3	N/A	N/A	N/A
8C	0.6	32.1	0.2	1.0	7.4	0.1	N/A	N/A	N/A
8S	0.7	20.6	0.1	1.5	25.2	0.4	N/A	N/A	N/A

¹ The integers represent the cell number, while the letters represent the location within the cells: N = North, C = Center, S = South.

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