PHOSPHORUS REMOVAL AND SOIL STABILITY WITHIN EMERGENT AND SUBMERGED VEGETATION COMMUNITIES IN TREATMENT WETLANDS

By

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Phosphorus (P) removal by treatment wetlands is an integral part of Everglades protection and restoration. The effects of water column shading on P cycling and retention were explored in emergent (*Typha* spp.) and submerged (*Najas guadalupensis*) vegetation communities within a Stormwater Treatment Area (STA-1W) wetland in south Florida. The physicochemical aquatic environments within these two vegetation communities were hypothesized to differentially affect community metabolism, which in turn would affect biological P uptake rates and P stability in accrued soils.

Muck soils beneath emergent aquatic vegetation (EAV) were P-depleted over 8 years of operation in STA-1W, while the muck soils beneath submerged aquatic vegetation (SAV) beds were P-enriched. The stability of P within newly

accrued soils was dependent on macrophyte community type, and was likely increased through water column $CaCO_3$ precipitation. Soils (0-4 cm layer) in SAV communities contained significantly more P in residual, Ca/Mg-bound and fulvic/humic acid-bound pools than soils in EAV. Because of similar pools of exchangeable and Al/Fe-bound P, the two soil types each released P to an oxygenated water column at similar flux rates.

Typha litter and associated microbial biomass retained P mineralized from soils under oxic water column conditions, but retention was lower under anoxic conditions. Dense EAV stands accumulate oxygen demand, reduce light penetration and may have little microbial P uptake and retention capacity due to anoxic conditions. While *Typha* biomass persists as leaf litter and detritus to a greater extent than *Najas* tissues, the extensive *Typha* root system has the potential to hinder long-term storage by mobilizing P from enriched soils.

Community metabolism was influenced by water column shading, which reduced $CaCO_3$ precipitation and Ca-bound P pools; and reduced oxygen supply to microorganisms in EAV communities. Managing for SAV and eliminating dense EAV stands from treatment wetlands may reduce surface water TP concentrations. Phosphorus-enriched areas within the northern Everglades may also be contained or restored by increasing light penetration to the water column, in order to enhance soil P sorption capacity through $CaCO_3$ precipitation and increase photosynthetic oxygen supply to the aquatic microbial communities.

CHAPTER 1
PHOSPHORUS DYNAMICS IN TREATMENT WETLANDS – A REVIEW

Introduction

Phosphorus (P) availability limits primary productivity in many freshwater systems (Boers et al., 1998; Reddy et al., 1999). Cyanobacterial mats are periphyton communities that can maintain high productivity in low P environments due to rapid internal nutrient recycling (Wetzel, 1993). Cyanobacteria (*Schizothrix*, *Scytonema*) within the mats are competitive with emergent macrophytes (i.e., cattails (*Typha* spp.)) in waters with low P concentrations, however they are sensitive to increases in P levels. In contrast, *Typha* grows well in P-enriched soils, and can produce a large biomass annually (Toth, 1988; Davis, 1991; Davis, 1994). A shift from open water areas dominated by calcareous, cyanobacteria periphyton to monoculture stands of *Typha*, in areas downstream of water control structures, has raised concerns among resource managers about the loss of ecological integrity of the oligotrophic Everglades marsh (Davis 1994).

Constructed wetlands have become a practical treatment technology for removing P from surface waters. In south Florida, farmland in the Everglades Agricultural Area (EAA) has been taken out of cultivation and flooded to create wetlands for surface water treatment (Figure 1-1). The created wetlands, or

stormwater treatment areas (STAs), were subsequently colonized by a mixture of emergent, submerged and floating wetland vegetation.

Submerged aquatic vegetation (SAV) including *Najas guadalupensis* Spreng., and emergent cattail (*Typha latifolia* L., and *T. domingensis* Pers.), communities coexist throughout the STAs. Floating macrophytes (*Eichhornia crassipes*, *Pistia stratiotes*) and periphyton appear transient in the upstream and downstream reaches of the wetland flow path, respectively.

Current water management objectives in south Florida for treating agricultural drainage waters (ADW) with treatment wetlands include achieving low effluent total phosphorus (TP) concentrations, maximizing phosphorus removal from the water column, and producing stable soils for long-term P storage. Essential to achieving these objectives is an understanding of P exchange rates between three major storages: soil, biomass and the water column (Figure 1-2). The water column is a compartment of variable P storage that must be minimized for effective P removal. The biomass of macrophytes and microorganisms influences several biogeochemical cycles that control P exchange between compartments. Soils are viewed as the long-term storage compartment, though surface soils and associated P interact with the biomass and water column compartments.

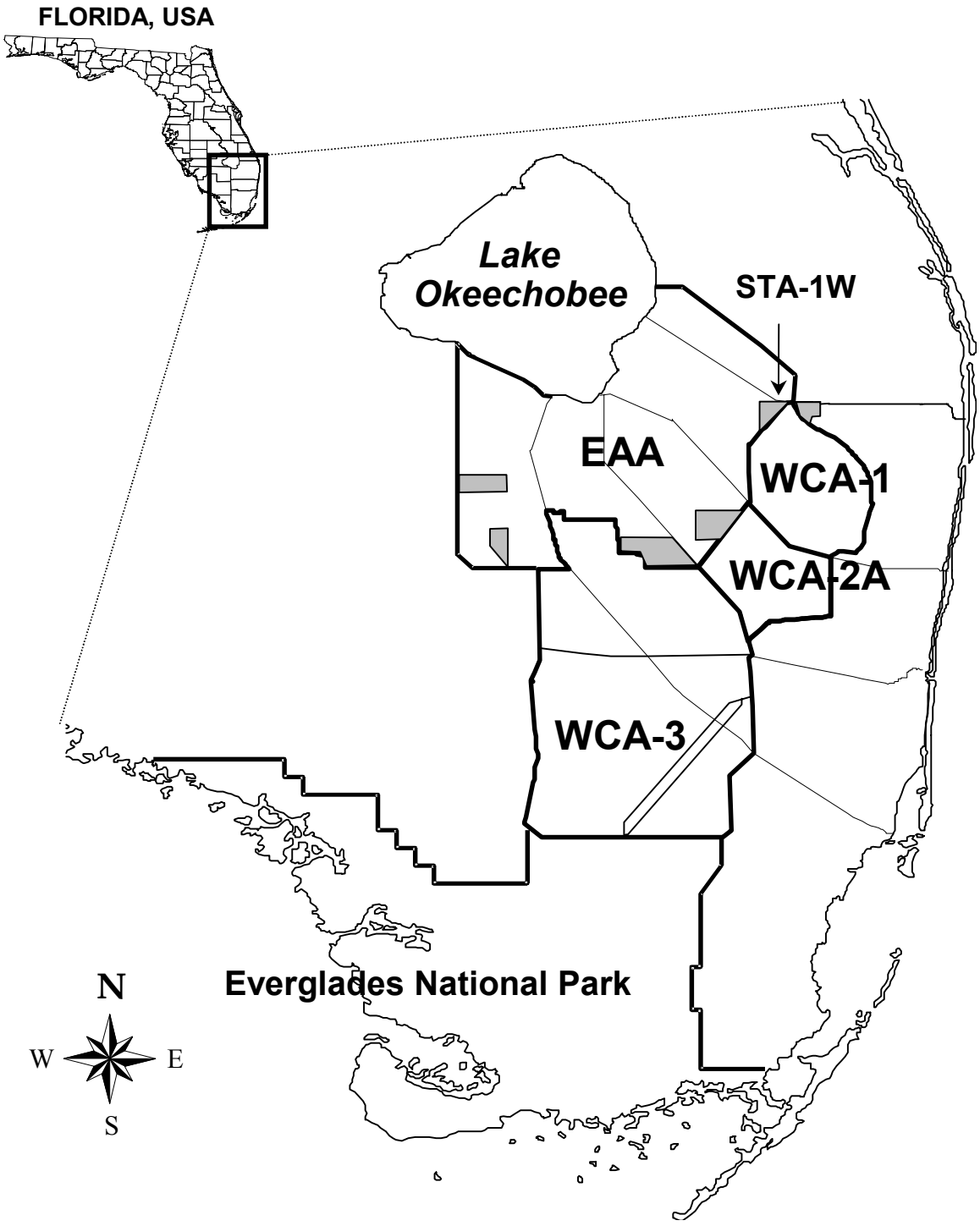

Figure 1-1. The historic Everglades region of south Florida is now three distinct parcels, including the Everglades Agricultural Area (EAA), Water Conservation Areas (WCA) and Everglades National Park. Stormwater treatment areas (STA) are shown (in gray), including STA-1W, where field investigations took place.

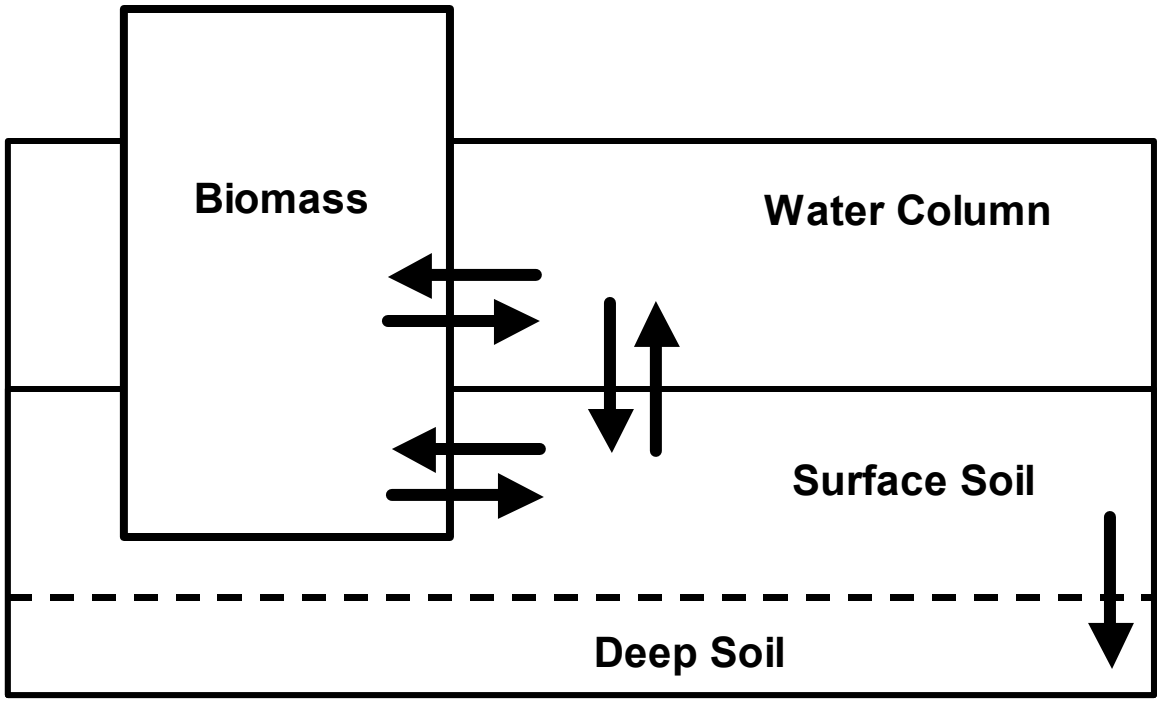

Figure 1-2. General model of phosphorus storages in wetlands. Arrows indicate potential P exchange pathways between storages. Deep soils represent a long-term P storage compartment.

Water column P removal and soil stability (P storage potential) can be understood through examination of biological processes (e.g., plant P uptake, tissue senescence, and microbial decomposition), chemical processes (e.g., adsorption/desorption from surfaces and precipitation/dissolution of P compounds), and physical processes (e.g., particulate settling and resuspension, soil burial). Each of these processes within the treatment system can influence the P retention capacity of the system.

Phosphorus in the Water Column

Phosphorus Species

Phosphorus exists in a variety of chemical forms within natural waters. Compounds can range from dissolved ion species (e.g. $H_2PO_4^-$, HPO_4^{2-}) to large complex molecules. Because of the range in molecular size, it is important to describe two classes of phosphorus compounds, dissolved and particulate species. An operational definition of dissolved P is that which passes through a 0.45 μm membrane filter, whereas the particulate P is retained. Particulates > 0.45 μm in diameter are generally too large for direct uptake by plants, algae and microorganisms. However, particles can be transported by surface waters, and subsequently mineralized downstream.

Phosphorus compounds also exist as organic or inorganic forms, though there can be considerable exchange between the two. Organic P is either derived from cell components or is soluble P that has been complexed by organic matter, and can be resistant to mineralization or dissociation. Inorganic P can be either dissolved phosphate ions or sorbed onto (or incorporated within) a mineral particle.

Soluble reactive phosphorus (SRP) is the dissolved inorganic fraction that is readily bioavailable. Dissolved organic phosphorus (DOP) is less reactive and must be hydrolyzed prior to biological assimilation. The remaining fraction is particulate phosphorus (PP), which is retained by a 0.45 μm membrane.

Phosphorus Sources

Phosphorus in any form can be imported into a wetland through surface inflow waters, precipitation, or groundwater seepage (allochthonous). It can also be transformed within the wetland from one form to another (autochthonous). Internal loading represents P released to the water column from biomass and soil storages. The allochthonous load can be sequestered during the growing season by plant uptake and then released as an internal load during periods of plant senescence. Substantial internal loads can also be attributed to soil P release. Both allochthonous (external) P loads to the Everglades system and autochthonous (internal) loads from nutrient-impacted areas must be reduced in order to achieve management and restoration objectives.

Phosphorus Removal Processes

Biological Processes

Assimilation

Biological P uptake is almost exclusively in the soluble reactive form (SRP). Carignan (1982) correlated 97% of P uptake by macrophytes with changes in soil porewater and water column SRP concentrations. Bacteria and other microorganisms assimilate P and rapidly recycle it to the water column upon death (Currie and Kalff, 1984). Rates of SRP assimilation by macrophytes are slower and highly variable across species. Richardson and Marshall (1986) found that PO_4 additions to emergent macrophyte enclosures were initially assimilated into the microbial biomass and detritus, rather than into the emergent biomass.

Emergent macrophytes and some submerged plants obtain P from the soil porewater, while other submerged macrophytes obtain the nutrient from water directly through stem and leaf tissues. Phytoplankton assimilate P directly from the surrounding water column, and are dependent on mixing to circulate sufficient nutrients for growth. Epiphytes and benthic populations of algae, bacteria and fungi depend on water flow to carry necessary nutrients across the substrate surface, and reduce boundary layers that can otherwise limit nutrient supply to slow rates of molecular diffusion.

Phosphorus uptake rates depend on the microbial, algal or higher plant biomass. Macrophyte biomass P storage increases throughout the growing season as biomass increases, though tissue P concentrations may be variable (Hill, 1979). Cattails can allocate 60% of the total biomass to below-ground tissue in low P environments, but only 40% in P-enriched environments (Miao and Sklar, 1998; Miao and DeBusk, 1999). Biomass P is then subject to recycle into the water column, or incorporation into the soil detritus/microorganism compartment where it is slowly buried into the deep sediments (Richardson and Marshall, 1986).

Nutrient availability in the water column dictates the importance of soil nutrients. Rattray et al. (1991) suggested that in eutrophic, aquatic environments macrophyte growth was not necessarily limited by soil nutrient deficiencies. Using radioisotopes, however, Carignan and Kalff (1980) have shown that SAV rooted in eutrophic waters (167 μg SRP L^{-1}) would still obtain nearly all tissue P

from the soils, and they implicate macrophytes as potential nutrient "pumps" that return P from the soils to the water column. In oligotrophic water, soil nutrition (especially P) can limit growth of rooted, submerged macrophytes (Rattray et al., 1991). Organic matter accumulation on mineral soils was shown to influence SAV species succession in lakes (Barko and Smart, 1983) with maximum growth rates in soils containing 5-20% organic matter. If either SAV species or growth rate affects P removal by SAV beds, organic matter accumulations would influence P removal capacity of the community.

Despite the potential importance of soil P chemistry in rooted macrophyte nutrition, other aquatic plants such as phytoplankton, periphyton, floating plants, unrooted macrophyte species (e.g. *Ceratophyllum demersum*), and SAV fragments depend entirely on nutrient uptake from the water column. Furthermore, water column P uptake beyond the requirements for maximum growth, referred to as "luxury uptake", has been reported for a *Ceratophyllum*/periphyton complex growing in South Florida ADW (Pietro 1998).

Currie and Kalff (1984) suggested similar maximum P uptake kinetics exist for bacteria and phytoplankton. However, they also observed a five-fold greater increase in bacteria intracellular P concentrations over phytoplankton during "P-limited" lab incubations. Algae are therefore capable of greater P uptake per mass in the water column, because its biomass is usually greater than that of bacteria (Currie and Kalff, 1984). However, under the same P-limited

conditions, bacteria are able to incorporate P more readily into cellular tissue, which may affect the rate at which P becomes available again.

Chemical Processes

Adsorption

Phosphorus adsorption onto mineral surfaces is often related to the presence of calcium carbonates and Fe and Al hydroxides in soils (Patrick and Khalid, 1974; Richardson, 1985; Porter and Sanchez, 1992). Patrick and Khalid (1974) demonstrated the influence of iron chemistry on P sorption and release from flooded soils. In the oxidized water column, dissolved iron is low because Fe^{3+} dominates and is precipitated as ferric oxyhydroxides. Under reducing conditions after oxygen is consumed by chemical and biological oxidation, iron is solvent as the Fe^{2+} ion.

In acid soils, the phosphorus adsorption potential can be estimated accurately from the extractable aluminum and iron contents in the soil (Richardson, 1985). In soils of higher pH, carbonates control P sorption capacity and availability. Due to relatively low Fe and Al contents of EAA histosols, P sorption isotherms were unrelated to Al levels and weakly correlated to Fe, but were significantly correlated with total Ca and free carbonates (Porter and Sanchez, 1992).

Richardson and Vaithiyanathan (1995) examined P sorption along a gradient of soil-P enrichment in the northern Everglades. They found the P adsorption coefficient (a measure of P sorption capacity) was lower in Everglades

histosols than for mineral wetland soils where iron regulates P sorption characteristics. In Everglades soils where anoxic, reducing conditions are common, P sorption is controlled by $CaCO_3$ (Koch and Reddy, 1992) rather than iron hydroxides (Reddy et al., 1993; Richardson and Vaithiyanathan, 1995).

Precipitation

In addition to biological uptake and chemical adsorption processes, $CaCO_3$ precipitation is an important P removal mechanism in SAV systems, either through $CaCO_3$ sorption of P or direct coprecipitation (Gumbricht, 1993). $CaCO_3$ compounds are not subject to dissolution and subsequent P release when reducing conditions develop, but are sensitive to change in CO_3^{2-} equilibria, pH and temperature.

$$CaCO_3(s) \rightarrow Ca^{2+} + CO_3^{2-} \qquad K_{sp} = 10^{-8.4} \text{ (at 25°C)} \qquad [1]$$

In Fe and Al-dominated systems, Fe and Al-oxide precipitates play a similar role by incorporating phosphate from the water column into insoluble precipitates (Richardson, 1985). Insoluble calcium phosphate minerals such as apatite can only form under high concentrations of P, well above the levels observed in most surface waters (Golterman, 1998). Porewater P concentrations are frequently higher than those in the water column, however, and precipitation of metastable Ca-P minerals (e.g., tricalcium phosphate) may occur within the soil environment.

Physical Processes

Settling

Newly accrued wetland soil is a particulate matrix comprised of microbial biomass, macrophyte detritus, and inorganic solids settled from the water column. Settling of suspended particles occurs faster in emergent and submerged macrophyte communities than in open water (Madsen et al., 2001), due to reduced flow velocities and mixing. Wetland substrata - including standing shoots and litter in *Typha* stands, SAV biomass, and the soil surface - collect flocculent material as it settles onto surfaces which expands the "active" surface into three dimensions. Recently settled particles are in dynamic exchange with the water column via diffusion gradients, decomposition and leaching, bioturbation and resuspension. They can also be buried deeper into the soil profile. Surface soil P is still potentially bioavailable to rooted macrophytes, benthic algae, and soil microbial populations.

Surface soil is subject to resuspension by turbulent, high-velocity flow, as well as through bioturbation. Soil resuspension increases turbidity and decreases light penetration through the water column (Bloesch, 1995), which affects community metabolism and P uptake. Since P concentrations are generally lower in the water column than in the soil porewater, resuspension of sediment particles increases desorption from solid phases into the bulk solution.

Unvegetated reaches of a wetland flow path may dramatically alter physical soil stability. In addition to faster flow velocities and shorter hydraulic

retention times, unvegetated areas lack roots to stabilize the soil. High flow conditions may scour the bottom, suspending these soils in the water column for transport downstream. This process maintains deep-water conditions relative to nearby vegetated areas. Emergent and submerged macrophyte communities stabilize the water column and reduce soil resuspension (Gumbricht, 1993; Madsen et al. 2001), compared to open water sites.

Burial

Whether initial removal processes are biological, chemical, or physical, long-term storage of P in wetlands requires burial into deep sediments. Burial by organic matter accumulation occurs as the net result of a productive community metabolism – or whenever primary production exceeds respiration. Excessive external P loading to the Everglades marsh has resulted in long-term accumulation of P in the sediments and biomass storages (Reddy et al. 1993, Reddy et al. 1998). Using ^{137}Cs dating techniques, Reddy et al. (1993) estimated P accumulation rates in Everglades soils from 0.11 to 1.14 g m^{-2} yr^{-1}. They also identified higher nutrient retention by *Typha* than *Cladium* (sawgrass) communities.

Long-term burial rates are also influenced by temperature, hydrology, and fire regime (Reddy et al. 1993), as each of these factors affects the rate at which accumulating organic matter is oxidized to CO_2 and water. Decomposition of organic matter increases with temperature due to increased microbial metabolism. The warm temperatures in south Florida allow rapid

decomposition, yet flooded conditions reduce the supply of oxygen to decomposers. Alternate electron acceptors (other than O_2) are used in oxidation reactions under flooded conditions, and may control the overall rates of organic matter decomposition/ accumulation and associated P burial in wetlands (White and Reddy 2001). Periodic fires, often the result of lightning strikes, can rapidly oxidize organic matter and affect long-term rates of organic matter accumulation.

Everglades Research Site Description

Located between the EAA and Water Conservation Area (WCA) – 1, STA-1W was the first of six STAs to begin flow-through operations in 1994. Wetland environments in STA-1W that were dominated by SAV and cattail communities were examined in this study with respect to phosphorus removal and retention into new soils. Currently managed as the Arthur R. Marshall Loxahatchee National Wildlife Refuge, WCA-1 represents the northern extent of the largely unaltered Everglades land.

To the south, WCA-2 has been the focal point of much research on the impacts of P on wetland processes. A clear and well-documented transition from *Cladium jamaicense* prairies to *Typha* spp.-dominated wetlands exists south and west from canal discharge points. Data from this study are compared to the many relevant studies that were conducted within WCA-2A along the northern eutrophication gradient.

Directly south of WCA-2, and between urban Miami and the Everglades National Park, is WCA-3. This region is a mosaic of pristine Everglades ecotypes,

from wet sloughs dominated by *Nymphaea* spp. and *Utricularia* spp. to sawgrass (*Cladium*) ridge and tree island communities. Isolated stands of *Typha* exist across the landscape, and are associated with alligator holes.

Need for Research

Emergent and submerged vegetation communities differ in physical and chemical structure, yet the effects of these differences on P removal and soil stability characteristics within the treatment wetland context are largely unexplored. Until recently, the two vegetation types were each considered a part of the naturally-recruited wetland community, and little effort was made to separate the effects of one independent of the other. For example, the biomass storage potential of cattails has been viewed as an asset to wetland treatment, and many *Typha* wetlands function well as wastewater polishing areas (Kadlec and Knight 1996). However the contribution towards P removal and retention of the submerged macrophytes, epiphyton and microbial community, is unknown.

In eutrophic, aquatic environments continuously loaded with P, cattails are capable of creating large monoculture stands (Davis 1991). Since, they are ubiquitous, aggressive colonizers, the differences between SAV and cattail communities in providing different P removal and effluent TP concentrations are important. My research, therefore, compared emergent and submerged macrophyte communities with respect to P interactions within the water column and the development of new soils.

CHAPTER 2
PHOSPHORUS STABILITY IN ACCRETED WETLAND SOILS

Introduction

Long-term retention of phosphorus (P) in wetlands can occur through the accretion of new soil (Reddy et al., 1993). Treatment wetlands have been constructed for P removal from surface waters, and accrete soils to store P and protect downstream waters from eutrophication (Kadlec and Knight, 1996). Stability of newly accrued soil-P is an important issue in such wetlands, as internal P cycling can elevate water column P above target outflow concentrations. Newly accreted surface soils are in contact with both the water column and macrophyte roots, thus soil-P can be released into the water column. Soil P retention depends on the characteristics of the wetland vegetation as well as the chemical environment within the water column and surface soil layer.

Aquatic photosynthesis elevates water column pH levels during daylight hours due to consumption of dissolved inorganic carbon species (i.e.CO_2, HCO_3^-) that equilibrate with solid carbonates. When water column calcium and alkalinity levels are sufficiently high, pH elevations lead to calcium carbonate supersaturation and precipitation (Otsuki and Wetzel, 1972). The precipitate, in turn, provides sorption sites for dissolved inorganic P, and results in a calcium-

and phosphorus-enriched soil. At 25 °C, the following equilibrium reactions govern $CaCO_3$ solubility in water:

$$CaCO_3(s) + CO_2(g) + H_2O \rightarrow Ca^{2+} + 2HCO_3^- \qquad K = 10^{-6.03} \qquad [2]$$

$$2HCO_3^- \rightarrow 2H^+ + 2CO_3^{2-} \qquad K_{sp} = 10^{-10.33} \qquad [3]$$

$$CaCO_3(s) + CO_2(g) + H_2O \rightarrow Ca^{2+} + 2H^+ + 2CO_3^{2-} \qquad K = 10^{-26.63} \qquad [4]$$

Calcium carbonate chemistry may play an important role in wetland treatment of drainage water from the Everglades Agricultural Area (EAA), a region productive in sugarcane and winter vegetables. The EAA has muck soils underlain with calcareous limestone (Gleason and Stone, 1994). Shallow surface waters coupled with a high water table increases surface water and groundwater interactions, which elevate Ca and carbonate (alkalinity) concentrations in irrigation waters, agricultural drainage water (ADW), and in the soil itself.

Excessive phosphorus (P) loading from EAA ADW discharges has been identified as the primary cause for an observed eutrophication gradient in the northern Everglades. Changes observed near the discharge structures include increased water column, soil, and plant tissue P concentrations, and change in ecosystem function, relative to the interior marsh (Craft and Richardson, 1993; DeBusk et al., 1994; Reddy et al., 1993; Reddy et al., 1998). Phosphorus enrichment has led to increased *Typha* spp. above-ground biomass and shoot density (Grimshaw et al., 1997; Wu et al., 1997; Miao et al., 2000), and reduced PAR levels at the air-water interface, relative to levels at nearby open slough

sites. Reduced PAR may limit aquatic photosynthesis, which could reduce $CaCO_3$ precipitation and soil P retention.

Rates of P diffusion from soils to overlying water can be controlled by the strength of the ion activity gradient, temperature and soil porosity. Unequal distribution of phosphate, due to biological uptake mechanisms or sorption onto solid minerals such as $CaCO_3$ and FeOOH (Golterman, 1998), can establish an ion activity (related to ion concentration) gradient. Moore and others (1991) found a diffusive P flux rate of 1.69 mg m^{-2} day^{-1} for sediments in a hypereutrophic freshwater lake (Lake Apopka, FL). The magnitude of internal nutrient flux can equal external loads to such a system, and maintain water column P above 30 µg L^{-1} , concentrations typical of eutrophic systems (Nurnberg, 1996).

Internal loading can potentially impair treatment wetland performance. As P-enriched soils accumulate, the potential for diffusive flux to mobilize soil-P into the water column may increase. Additionally, emergent macrophyte shade may limit water column photosynthesis and $CaCO_3$ precipitation, relative to submerged macrophyte areas, thereby producing soils of different P retention capacity and internal P flux rates. The influence of macrophyte vegetation shading of the water column on $CaCO_3$ precipitation and accrued soil stability is currently unknown.

The objectives of this study were to:

- Characterize the stability of P within accreted soils of emergent and submerged macrophyte stands through sequential P extraction,

- Determine the potential for $CaCO_3$ formation in emergent and submerged macrophyte communities, both in the water column and the soil porewater,
- Estimate the potential internal P flux to the water column from newly accrued soils formed within emergent and submerged macrophyte stands.

These research objectives will address the stability of P in soils formed in emergent and submerged macrophyte-dominated wetlands, which is essential to the long-term management of STAs as well as the nutrient-impacted northern Everglades.

Materials and Methods

This investigation was pursued through bench–scale incubations, in field-operated mesocosms, and *in situ* within the full-scale wetland environment. Bench-scale studies took place at DB Environmental, Inc., in Rockledge FL; and at the Wetland Biogeochemistry Laboratory at the University of Florida in Gainesville, FL. Outdoor mesocosms were located next to the STA-1W inflow canal on an experimental platform provided by DB Environmental.

STA-1W Site Description

Agricultural runoff from the Everglades Agricultural Area (EAA) Basin S-5 (Figure 2-1) is pumped via canals to STA-1W, a full-scale (2699 ha) treatment wetland operated by the South Florida Water Management District (SFWMD) to reduce P loadings to the Everglades (SFWMD, 2003). Everglades muck soils were drained for agricultural production decades ago. Nearly 50, 000 acres of former ag land have recently been reflooded to create STAs, of which the

Everglades Nutrient Removal project, now part of STA-1W, was the prototype. A detailed spatial analysis of phosphorus and other constituents of the pre-existing farm soils in the STA 1W footprint was conducted prior to flooding (Reddy and Graetz, 1991).

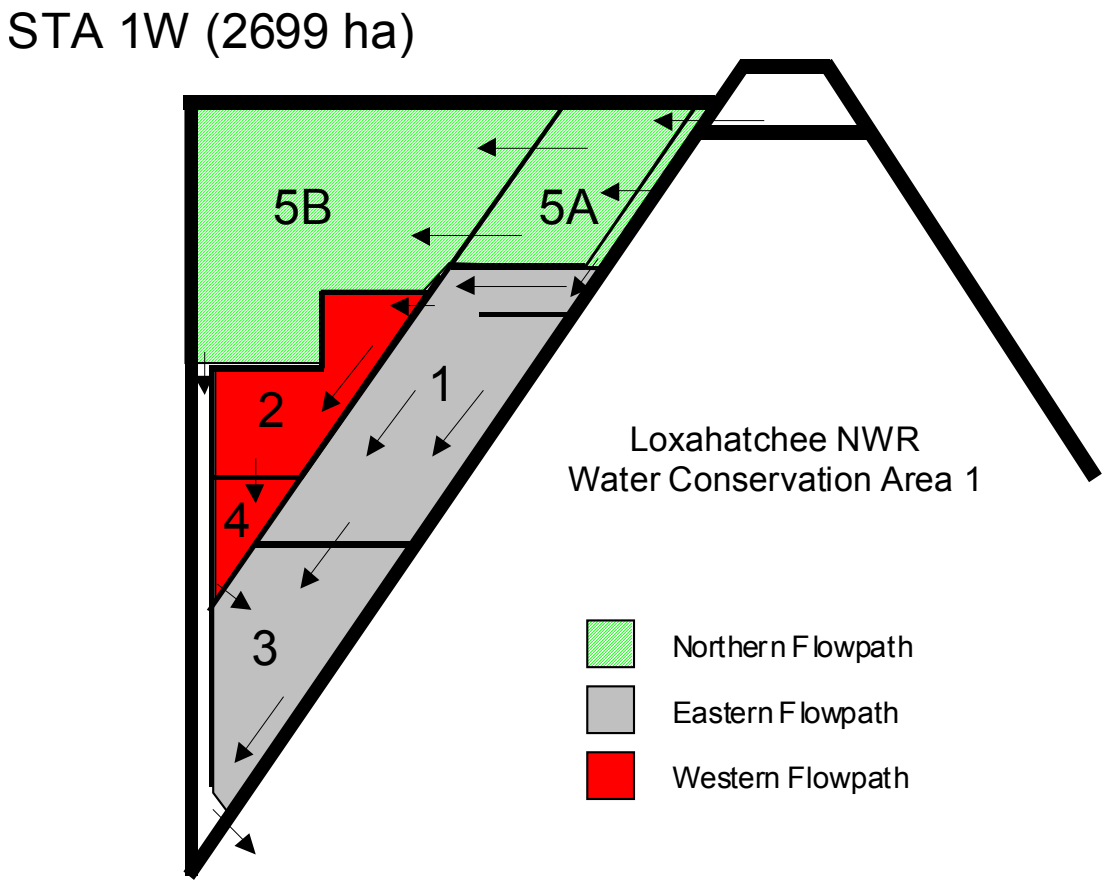

Figure 2-1. Stormwater Treatment Area 1W in Palm Beach county, Florida. Three flowpaths receive surface water drained from adjacent agricultural soils. The wetland functions to reduce water phosphorus concentrations prior to discharge into Water Conservation Area 2A. Cells 1, 2 and 3 are comprised of mixed emergent, submerged and floating vegetation, while Cells 4, 5A, and 5B are primarily submerged and floating vegetation. Arrows indicate general direction of flow.

Cell 1 is the first cell of the eastern flowpath, and is comprised of emergent aquatic vegetation, or EAV, (*Typha* spp.) in the inflow region and along the

eastern edge (Figure 2-2). A mixture of emergent, floating, and submerged aquatic vegetation (SAV) occupy the downstream reaches of the cell. This distribution of community types has remained relatively constant in Cell 1 since the wetland was first flooded in 1994 (Newman and Pietro, 2001). The juxtaposition of areas dominated by submerged and emergent vegetation in the Cell 1 outflow region suggests that the two community types have developed under similar hydraulic and nutrient loadings. Eight years of flow-through operations in this wetland have been sufficient to allow accretion of soils in both community types, and to establish a record of long-term effectiveness of P sequestration from the water column.

Two sampling sites were selected in the outflow region of Cell 1. One station (26.6292°N, 80.4219 °W) represented EAV, *Typha domingensis*, while SAV species *Najas guadalupensis* and *Ceratophyllum demersum* occupied the second station (26.6278°N, 80.4352 °W). The longevity of the contrasting vegetation communities observed at the two locations was verified with aerial photos provided by SFWMD, as well as through personal communication with field personnel.

Stability of recently accreted wetland soil-P was characterized through bulk density analysis and sequential extractions. Vertical profiles of water column chemistry were constructed for mesocosms dominated by emergent and submerged macrophytes and for emergent and submerged macrophyte communities in southeastern Cell 1 of STA-1W. Profiles of soil porewater

constituents were also constructed for the STA soils. For each profile, the $CaCO_3$ saturation index was calculated to determine whether conditions for precipitation were present.

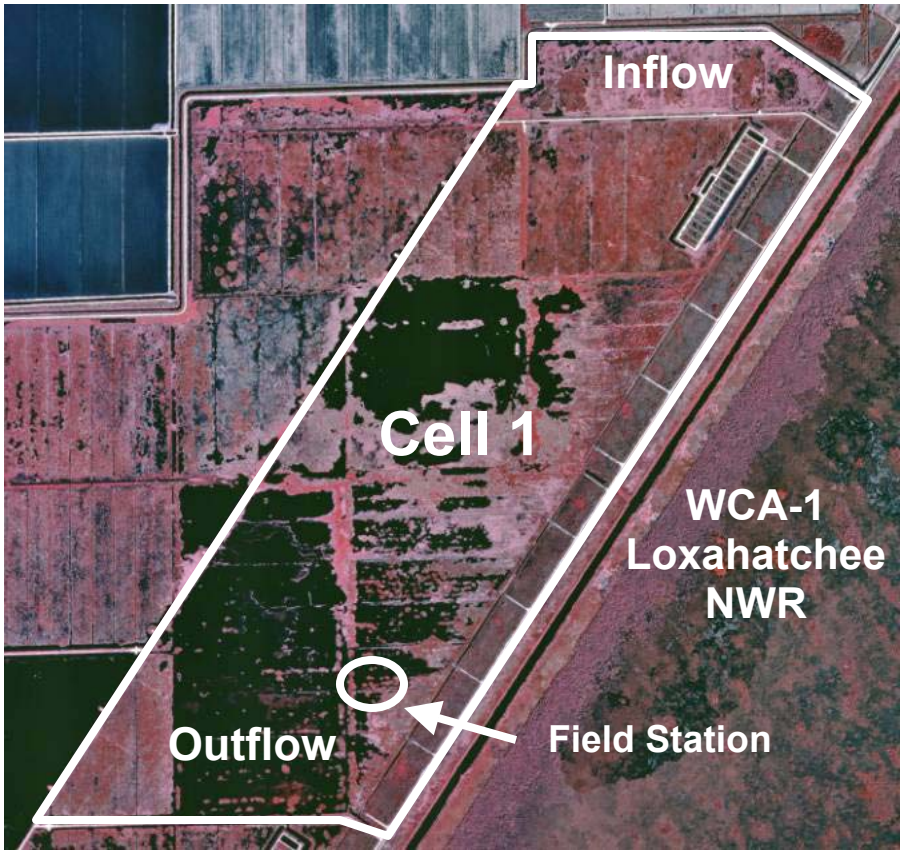

Figure 2-2. Aerial photograph of STA-1W Cell 1, first cell of the eastern flowpath, in November 2000 (courtesy SFWMD). Also shown is the location of the field station in emergent (pink) and submerged (grey) vegetation stands.

Phosphorus diffusion flux rates across the sediment-water interface were calculated based on porewater concentration gradients. Potential P flux from these soils was then determined experimentally using intact cores incubated under lab conditions.

Soil Collection and Analysis

Soil cores were retrieved from both field sites on July 3 and 4, 2002. Each core was retrieved by pushing acrylic core tubes (7 cm dia.) through the accrued soil layer into the underlying native farm muck to a minimum depth of 10 cm. The top end of the core was sealed with a #13½ rubber stopper prior to soil core extraction. The horizon dividing new wetland soils from the native muck soil was determined by differences in color and texture, and the depth of accrued wetland soil was recorded.

Cores were sectioned in the field at 2 cm depth intervals. The 0-2 cm, 2-4 cm, and 4-6 cm intervals were retained for analysis, along with the underlying muck. Like depth increments from five replicate cores were composited and homogenized before analysis to account for field variability. This procedure was repeated three times at both sites, and resulted in triplicate samples for each of the four soil layers. A fresh 90 cc subsample of composite soil samples was dried to constant weight (65°C) for bulk density. Flocculent surface (0-2 cm) samples were allowed to settle in graduated cylinders overnight (dark, 4 °C) and the excess water was discarded prior to analyses.

Sequential Extractions for Inorganic Phosphorus Pools

Inorganic P pools in the soil surface intervals of 0-2 cm and 2-4 cm were characterized by sequential extraction (modified from Hieltjes and Lijklema, 1980) and contrasted to the underlying muck soils. Fresh soil samples were weighed (5g wet) into 50 mL centrifuge tubes. To each centrifuge tube, 40 mL of

1M NH_4Cl was added. The slurry was then shaken for two hours. Samples were then centrifuged for 15 minutes at 2800 rpm before the supernatant was decanted and filtered (0.45 μm) for SRP analysis. The NH_4Cl extraction was repeated, and the supernatant was added to that of the first extraction before analysis. The tube and sample residue were then weighed, and 40 mL of the next extractant was added (Table 2-1).

Sodium hydroxide (NaOH) and HCl extractions were shaken for 17 and 24 hr, respectively. Both SRP and TP analysis were performed on NaOH extractions, while only SRP analysis was performed on the HCl extracts. Soil residue remaining after HCl extraction was subjected to TP analysis. TP samples were filtered through Whatman 41 qualitative filters. Extractants were refrigerated at 4°C until analysis. Soluble reactive phosphorus colorimetric analysis (potassium antimony tartrate, sulfuric acid, ammonium molybdate, ascorbic acid) was performed on a Spectronics Genesys 5 spectrophotometer. Total P analysis included a persulfate digestion and neutralization prior to SRP analysis.

Using the residue weights recorded after each extraction, an estimation of P carry-over (the extractant volume and P mass carried over between extractions) was calculated and used to adjust each P pool.

Flux Study Using Intact Soil Cores

Three additional replicate intact soil cores were retrieved from the SAV and EAV communities for intact core incubations. Each soil core was topped off

with site water, then sealed with rubber stoppers. An opaque shroud minimized solar heating and blocked light during transport to the lab.

Table 2-1. Sequential extraction of phosphorus from wetland soils using a method adapted from Hieltjes and Lijklema (1980).

Sediment P pool	Extractant	Shake Time	Analysis	Stability
Exchangeable	1.0 *M* NH_4Cl at pH 7	Two 2 hr periods	SRP	Readily bioavailable
Fe- and Al-bound	0.1 *N* NaOH	17 hr	SRP	↕
Humic and Fulvic Acid-bound			TP-SRP	
Ca-bound	0.5 *N* HCl	24 hr	SRP	
Residue	Remainder digested for TP			Recalcitrant

Overlying water was replaced with 1.15 L filtered (0.45 μm) STA-1W Cell 4 outflow water. The 30 cm water column was aerated and incubated in a dark water bath at ~22°C for 28 days. Water samples (30 mL) were withdrawn at Δt = 0, 0.5, 1, 1.5, 2, 4, 6, 10, 14, 20, and 28 days, and analyzed for SRP and pH. Representative samples were also analyzed for TSP, dissolved calcium, total alkalinity, and DOC. Reflood water was added (30 mL) after each sampling to maintain water volume.

Samples for SRP and TSP were filtered through a 0.45 μm polyether sulfone filter immediately after sample collection. Persulfate digestion followed by neutralization was performed on TSP samples. SRP and TSP water determinations followed the ascorbic acid-molybdenum blue method (EPA 365.2;

EPA 1979) using a Spectronics Genesys 5 spectrophotometer. Dissolved calcium was determined using flame atomic absoption spectroscopy (EPA 215.1; EPA 1979) on a Perkin-Elmer 3110. Alkalinity was titrated with 0.02N H_2SO_4 (EPA 310.1; EPA 1979). Dissolved organic carbon analysis was on acidified, filtered (0.45 μm) samples, and measured with a Shimadzu TOC-5050A (Duisburg, Germany) TOC analyzer equipped with an ASI-5000A autosampler (5310-A; APHA 1992).

Sample pH was recorded immediately following collection, using a 3 in 1 gel filled combination pH electrode and Corning 313 pH meter. Water bath temperature was continuously recorded by a StowAway Tidbit® logging probe (Onset Computer; Bourne, MA) as well as monitored periodically with a thermometer.

At the conclusion of the P Flux study, the water column of each core was spiked with 100 μg P L^{-1} (as KH_2PO_4). The water volumes above each core differed slightly (± 5 mL) from the original water volume of 1.15 L added one month prior, likely due to different evaporation rates induced by the aerators. These differences were recorded but volumes were not adjusted at that time. Water samples were withdrawn Δt = 0, 4, 8, 24 and 53 hours after the amendment, and analyzed for SRP. Each core received 30 mL of unamended reflood water after sampling to maintain water volume, and was kept under an opaque shroud.

Mesocosm Design and Background

One SAV-dominated and two EAV-dominated mesocosms (4.2m L x 0.79m W x 1.0m D) were maintained for 2.7 years at a hydraulic loading rate of 10 cm day^{-1}, as part of another study (DBEL, 2001). These systems were inoculated with *Najas guadalupensis* and *Typha* spp. plants, respectively, which were collected from within STA-1W Cell 4. Weekly monitoring of inflow and outflow TP concentrations, temperature and pH, and periodic monitoring of other constituents (TSP, SRP, dissolved Ca (dCa), total alkalinity (TA), specific conductance) was performed from December 29, 1998 through August 8, 2001.

Shade Effects within EAV and SAV Mesocosm Communities

In order to investigate the effects of shade on P removal in the EAV mesocosms, I examined the ambient light regime and calcium carbonate saturation index (SI). Duplicate SAV mesocosms with similar operational history were sampled for comparison.

Incident light was measured at 2m above the water surface (above cattail canopy), and at the water surface. Two 4π spherical quantum sensors recorded available PAR during peak daylight hours (1000-1400), using a Li-COR LI-1000 data logger (Lincoln, Nebraska). Eight replicate one-second measurements of photon flux were averaged for each datum value, and recorded when the value had stabilized to roughly within ± 1%. All comparisons were made to the simultaneous "ambient" light levels, to adjust for short-term temporal changes in incident radiation (e.g. change in cloud cover).

Chemical profiles through mesocosm water columns were constructed from *in situ* measurements of dissolved oxygen (DO) concentrations, pH, specific conductance and temperature taken on May 30, 2001. Concurrently, surface (3 cm) and bottom (20 cm) water samples were collected and analyzed for SRP, dCa, TA concentrations. Water samples were withdrawn from the inflow and outflow regions of each tank (approximately 10 cm from end wall), as well as from each inflow stream.

The SI of $CaCO_3$ was calculated from water column and porewater chemistry profiles of submerged and emergent macrophyte communities. Specific conductance values provided an approximation for ionic strength. Hydrogen ion concentration was calculated from pH values. Calcium and alkalinity concentrations were used to calculate activity products of the Ca^{2+} CO_3^{2-} and HCO_3^- ions. Temperature values were used to adjust all solubility constants. The $CaCO_3$ SI was then computed for water at the surface and at 20 cm depth according the following relationship:

$$SI = \frac{\gamma_{Ca^{2+}}\left[Ca^{2+}\right]\gamma_{HCO_3^-}\left[HCO_3^-\right]K}{\left[H^+\right]K_{S0}} \qquad [5]$$

where: γ indicates the activity coefficient of Ca^{2+} and HCO_3^-
[ ] indicates the concentration of Ca^{2+} , HCO_3^- and H^+
K is the acidity constant of HCO_3^-, and
K_{S0} is the solubility constant of $CaCO_3$ at equilibrium.

Diel monitoring of STA-1W *Typha* and *Najas* communities

The mesocosm platforms provided a controlled environment for examining the influence of macrophytes on the water column chemistry. However, the water column environment in a full-scale STA during operations (e.g. high flow events) may be different than that observed at the small-scale. To investigate STA water column chemistry, diurnal monitoring of emergent and SAV communities took place in the southeastern region of STA-1W Cell 1 (Figure 2-2) in July 2002. Two Datasonde Hydrolab multiprobes were deployed to record pH, temperature, D.O. and specific conductance at 15 min intervals during a two-week period of high flows (up to 1200 cfs through STA 1W). Each device was suspended from a tripod to an initial probe depth of 10 cm below the water surface.

Dissolved oxygen, temperature and pH profiles through the water column were recorded for the same SAV and emergent stations on July 27 and September 29, 2002, using Yellow Springs Instrument dissolved oxygen meter and Corning pH meter. In September, the water column light regime was also assessed in SAV and emergent macrophyte stands, as well as in open water reaches of STA 1W Cell 1. Incident light was recorded simultaneously above the water column (ambient level), in surface waters (3 cm), at mid-depth and 10 cm above the substrate, using the methodology described for mesocosm light measurements.

Flux Study Using Porewater Equilibrators

Porewater was collected using porewater equilibrators, or peepers, modeled after Hesslein (1976) (Figure 2-3). Triplicate peepers were deployed in Cell 1 (~ 1 m apart) within emergent and submerged vegetation on June 10, 2002, and retrieved 17 days later. Three peepers at each station were inserted vertically through the accrued soil into the underlying muck soils to a depth of ~25 cm.

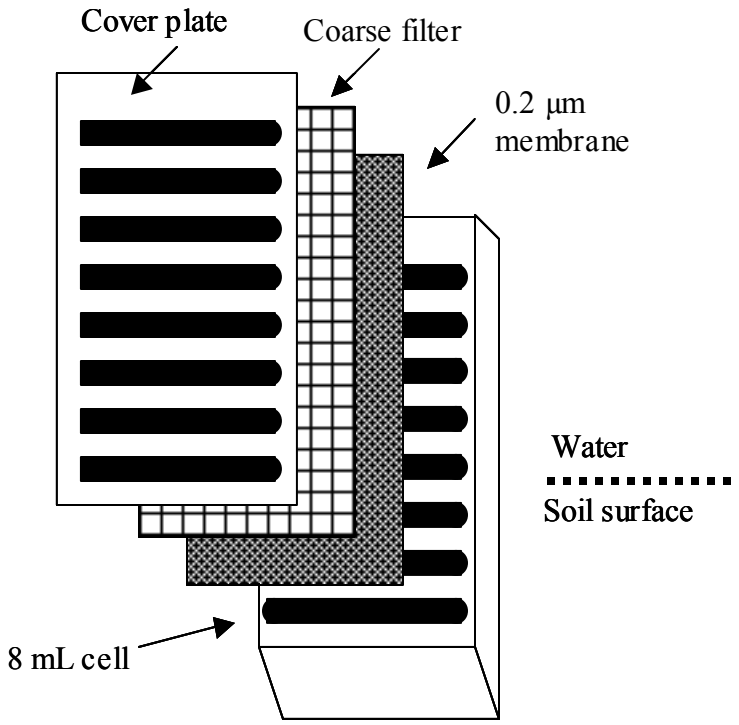

Figure 2-3. Schematic of porewater equilibrator used in estimates of P flux from STA-1W Cell 1 soils into the overlying water column. A 0.2 μm polyether sulfone membrane (Supor® 200) was inserted between a coarse particle filter and the 8 mL sampling cells. Some sample cells are above the soil-water interface, while others equilibrate with porewater below the soil surface.

The soil-water interface bisected the peeper such that some sampling chambers would equilibrate with soil porewater, and others would equilibrate with the water column above the interface. Specific conductance, SRP, TSP,

dissolved calcium and iron, and alkalinity concentrations and pH levels were measured above and below the soil-water interface. Dissolved iron determinations were made by a bathophenanthroline method, modified from APHA 3500-Fe D (APHA, 1992) for small sample volume and using a Spectronics Genesys 5 spectrophotometer.

Diffusive fluxes of P (i.e. $H_2PO_4^-$ and HPO_4^{2-}) at each coring location were calculated based on changes in SRP concentration with depth. The concentration gradient of soil porewater constituents with distance can drive diffusive flux according to Fick's first law, where:

$$J = -\Phi D_s \frac{dC/dz}{-0.47\Phi + 1.91} \qquad [6]$$

where J = diffusive flux, mg m^{-2} s^{-1}

Φ = porosity

D_s = the sediment diffusion coefficient, cm^2 sec^{-1}, and

dC/dz is concentration change, µg L^{-1}, per depth interval cm.

The final term accounts for sediment porewater tortuosity, a parameter that is linearly related to porosity (Sweerts et al., 1991). The concentration gradient *dC/dz*, was determined using the slope of a linear regression through the +5 to –5 cm depth interval.

Temperature was recorded during the equilibration period with max/min thermometers deployed at the soil-water interface adjacent to one peeper at each station. Alkalinity concentrations and pH levels were recorded in the field immediately after peeper retrieval. The thermodynamic potential for calcium

carbonate and calcium phosphate saturation and mineral formation was then calculated for the water column-porewater continuum at each site.

Statistical analyses on experimental data were performed using MSExcel® (v. 2000 ©Microsoft Corp.) ANOVA and t-test macros. Error around mean values is presented as ± one standard deviation for replicate samples.

Results and Discussion

Soil Characterization

STA-1W was previously farmland, with irrigation canals transecting the acreage. Since the land was flooded in 1993, cattail preference for shallow waters has encouraged growth along old canal banks. Dredged spoil from pre-flooding canal maintenance resulted in raised soil surfaces, and shallow water depths. This pattern is evident in aerial photographs taken of the wetland in November 2000, after six years of flow-through operation (Figure 2-2).

Measured from water surface to the top of the litter (or accumulated soil) layer, free-water depths on June 10, 2002, ranged spatially in the outflow region of STA-1W Cell 1 from 0 to 56 cm. Additional water column was occupied by *Typha* leaf litter, with up to 77 cm from air to soil surface. Based on probing-rod sounding measurements, litter accrued to depths ranging from ~ 0 to 30 cm.

Wetland soils had accrued above the native muck soils to variable depths beneath the emergent (6-14 cm) and submerged (4-20 cm) macrophyte communities. The accrued material was flocculent organic matter of lower bulk density than the underlying muck soils (Figure 2-4). Accrued soil bulk density in

the two community types was not significantly different for any given depth ($p > 0.05$). Muck soils were near the surface in some cores, while other cores were characteristic of deeper (> 6 cm) soil accrual, which may explain wide variability in the 4-6 cm layer bulk density measurements at both the SAV station (0.169 ± 0.049 g cm^{-3}) and *Typha* station (0.223 ± 0.093 g cm^{-3}).

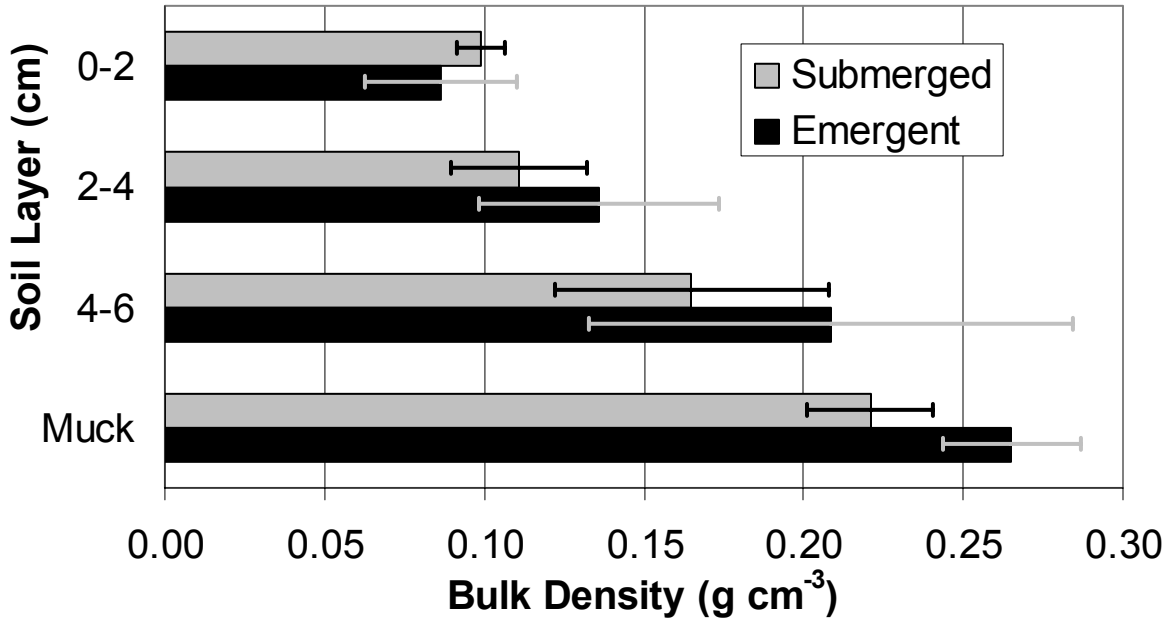

Figure 2-4. Average bulk density values for accrued sediments at depth intervals below the sediment water interface. The underlying farm muck was collected from below the horizon of the accrued sediment. Error bars indicate ± 1 s.d. for triplicate samples, and each sample is a composite of five discrete soil samples.

Soil Phosphorus Pools

A sequential extraction was used to characterize the relative bioavailability of P within the surface soils and the underlying muck soils. Reddy and Graetz (1991) used a similar sequential extraction, except 1M KCl was substituted for NH_4Cl to characterize the readily available P pool. Soil TP

decreased significantly from surface soils (0-2 cm and 2-4 cm layers) in the emergent stand to the underlying muck soils($p < 0.05$), while the SAV 2-4 cm soil layer showed a slight increase above the surface layer in each of three replicate composites (Figure 2-5). In the upper 4 cm, soils from the *Typha* community were P-enriched (584 mg P kg^{-1}) relative to the native muck soils (334 mg P kg^{-1}, $p < 0.05$), but enriched less than 0-4 cm SAV soils (813 mg P kg^{-1}; $p < 0.01$).

There was a significant difference in muck soil P levels beneath the SAV and the emergent communities. SAV-region muck soils were 2-3 times higher in P than the emergent muck soils (500 ± 212 and 168 ± 94 mg P kg^{-1}, respectively). In comparison to the native farm (Knight's Farm) soils (335 ± 31 mg P kg^{-1}) and soils 10 months after flooding (358 ± 35 mg P kg^{-1}), the muck in the emergent region has become depleted in P over the 7 years of operation, while submerged macrophyte muck has been P-enriched (Figure 2-5). The residual P pool was increased 10 months after flooding, and in recent samples from all depths.

Variation in soil TP on Knight's farm prior to flooding was greater than variation for other soil parameters (Reddy and Graetz, 1991), yet no soil-P value was reported lower than those observed below the emergent stand in this study. Local variation in P distribution was accounted for with composite soil samples from multiple cores, taken several meters apart. The SAV and emergent stations were several 100 meters apart.

Compositing vertically, however, may complicate the interpretation of observed differences between this study and that of Reddy and Graetz (1991).

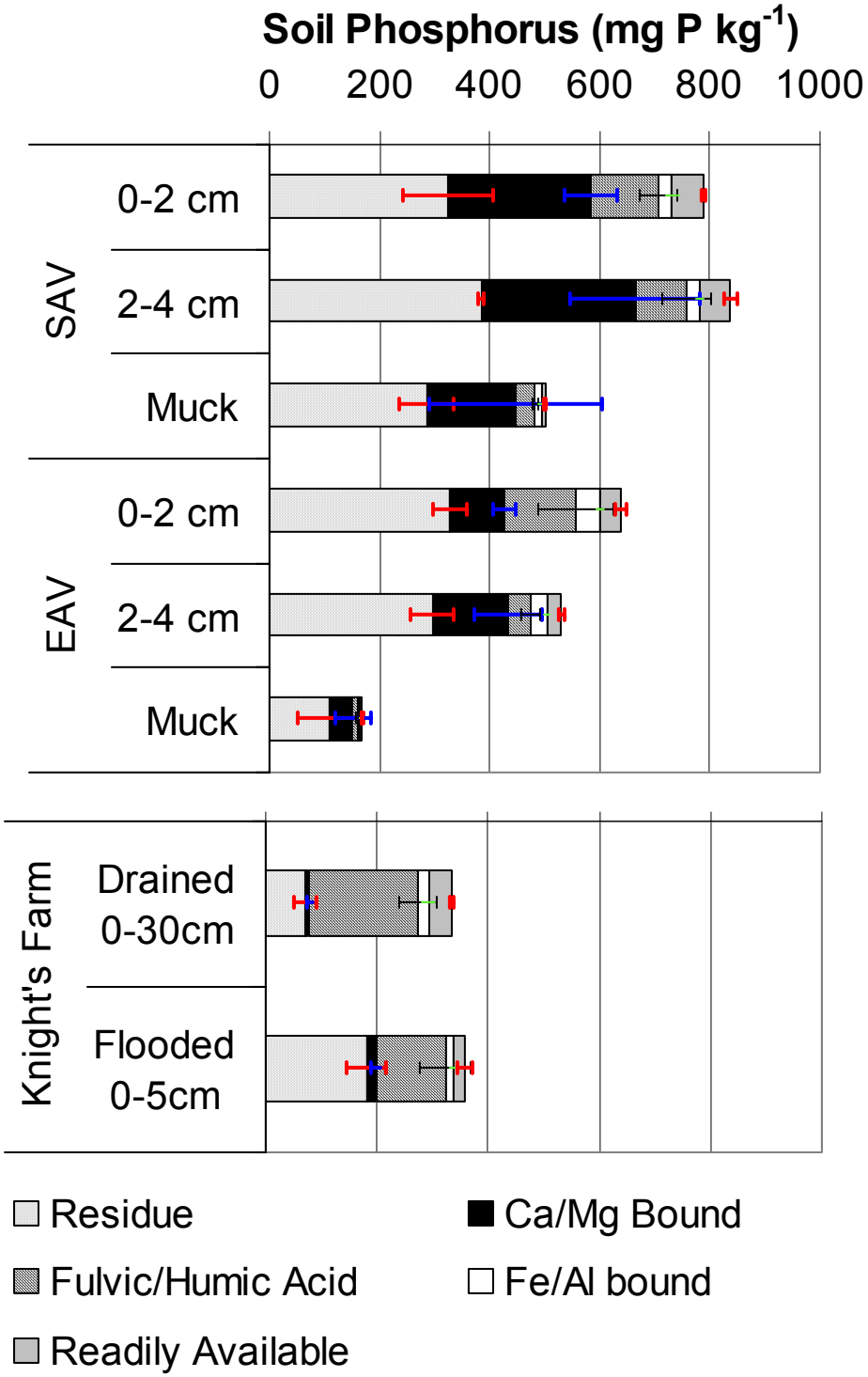

Figure 2-5. Soil phosphorus pools determined through a sequential extraction procedure using 1.0 M NH_4Cl, 0.1 M NaOH, and 0.5 M HCl. The residual P pool remained associated with the soil through all three extractions. Knight's Farm data from Reddy and Graetz, 1991.

Those investigators homogenized soils at 30 cm increments before flooding, and 5 cm increments after flooding. During agricultural production surface soils are mixed through tillage. However, wetland soils lower in the profile may have lower P concentrations than surface soils, and large depth intervals do not adequately describe surface soil chemistry. Thus, small depth increments were used in this study.

Accrued soil-P occurred primarily (34-60%) as residual P, a highly recalcitrant form. Calcium-bound HCl-extractable P was also a major pool in new wetland soils (15-45%), especially in the SAV surface 0-4 cm soils (mean 33%, or 271 mg HCl-extractable P kg^{-1}). This is a substantial increase from 8 mg HCl-extractable P kg^{-1} found in the drained surface (0-30 cm) soils (Reddy and Graetz, 1991).

Significantly lower fractions of total soil P were associated with fulvic and humic acids in accrued soils (5-26%), relative to drained farm muck (58%) ($p < 0.05$). This pool of organic, moderately available P characterized by 0.1 M NaOH extraction was highest in the *Typha* surface 0-2 cm soils (19%, or 128 mg NaOH-extractable organic P kg^{-1}), and decreased with depth at both stations.

The Fe- and Al-bound P pool was relatively small in accrued wetland soils, representing 2-8% of soil TP. Nevertheless, emergent surface 0-2 cm soils contained 45 ± 5 mg NaOH-extractable inorganic P kg^{-1}, nearly twice that of SAV surface soils (26 ± 11 mg kg^{-1}). If such a pool was primarily P associated with Fe-hydroxides, it would be subject to mobilization during prolonged anoxic

conditions. Release of the whole 0-2 cm pool into a 1-m deep water column, assuming equal soil bulk density of 0.1 g cm^{-3}, would elevate TP concentrations by 52 µg L^{-1} in SAV communities, and 90 µg L^{-1} in the emergent region. Muck soils below emergent vegetation were three-fold lower in Fe- and Al-bound P (4 ± 3 mg kg^{-1}) than muck below SAV (12 ± 5 mg kg^{-1}).

The NH_4-extractable P pool (readily available fraction) was higher in the SAV accrued soils (56 ± 8 mg kg^{-1}) than in emergent accrued soils (32 ± 10 mg kg^{-1}). Muck soils were low in NH_4Cl-extractable P, with 5 ± 2 mg kg^{-1} readily bioavailable. These values are within the range of values reported by Reddy and Graetz (1991) for farm soils prior to flooding (0-30 cm, 42 mg kg^{-1}), and muck soils after eight years of submergence (0-5 cm, 2 mg kg^{-1}).

Accumulation of organic matter, P enrichment of surface soils and detritus, and subsequent humification of the organic material, have been viewed as positive wetland attributes for P removal treatment applications. Phosphorus can be removed from the ambient water and become concentrated in recalcitrant organic soil components. When the availability of soil-P was characterized by sequential extraction, however, it was shown that the "bioavailable" and "recalcitrant" P pools alike were reduced in the soil beneath emergent vegetation, relative to either pre-flooded conditions or a contrasting community type, namely submersed macrophytes (*Najas guadalupensis*) (Figure 2-5). It seems likely that recalcitrant P compounds may be susceptible to mobilization through biotic mechanisms such as organic acid mineralization or enzymatic hydrolysis.

In contrast to the *Najas* community soils, the accrued soil beneath *Typha* was likely in close contact with, and under the influence of, extensive below ground biomass.

Water Column Profile and $CaCO_3$ Saturation Index

Each mesocosm received a mean inflow P loading of 3.5 g P m^{-2} yr^{-1} (average inflow concentration = 96 µg TP L^{-1}) over the 2.7-year period of operation. Total P concentration reduction was significantly greater ($\alpha = 0.05$) in the SAV mesocosm, with an average outflow concentration of 26 µg L^{-1}, than in the cattail mesocosms, whose outflows averaged 42 and 62 µg L^{-1}. Differences between cattail communities were not significant with respect to P removal performance.

Calcium and alkalinity concentration reductions were also observed in SAV-mesocosm surface waters, but not in the cattail stands. Between February 15, 2000 and August 8, 2001, Ca levels in inflow waters (72 mg L^{-1}) common to both SAV and cattail mesocosms were reduced to 47 mg L^{-1} at the outflow of the SAV mesocosm. Cattail mesocosm outflows averaged 69 and 70 mg Ca L^{-1}. Likewise, alkalinity was reduced from 206 to 135 mg $CaCO_3$ L^{-1} by SAV while cattail outflows averaged 202 and 204 mg $CaCO_3$ L^{-1}.

On May 30, 2001, SAV mesocosm surface water outflow SRP concentrations were low (5 µg L^{-1}), relative to the inflow region (20-43 µg L^{-1}), and Post-BMP inflow waters (49-58 µg L^{-1}) (Table 2-2). In the *Typha*-dominated mesocosms, SRP concentration reductions were smaller, with surface outflow

waters of 18 and 37 μg L^{-1}. Water at 20 cm depth had higher SRP concentration than surface waters in all mesocosms, perhaps a result of internal P loading from the soil to the bottom waters.

Similar trends were seen in the Ca and alkalinity concentrations (Table 2-2). For example, surface outflow Ca levels (43 and 45 mg L^{-1}) from SAV mesocosms were appreciably lower than those of the ADW inflow water (61 mg L^{-1}). Water at 20 cm depth in both community types was nearly equal in Ca and alkalinity concentration, and similar to the inflow waters (Table 2-2). In the SAV systems, constituent concentrations declined between inflow and outflow, but reductions were more apparent in surface water than at 20 cm depth. Calcium, alkalinity, pH and temperature levels in the *Typha* mesocosms were uniform internally, and showed little change with depth or with distance, compared to the SAV mesocosms (Table 2-2). Surface waters within the SAV mesocosms had elevated pH levels and temperatures, compared to the surface waters shaded by *Typha*.

Calcium carbonate precipitation was thermodynamically favored (SI>1) only in SAV surface waters (Table 2-2). *Typha* surface waters and waters at 20 cm depth in either community were unsaturated with respect to $CaCO_3$, and dissolution of the mineral was favored (SI < 1). While $CaCO_3$ formation may have occurred in the surface waters, the cooler temperatures and lower pH levels of waters at 20 cm depth created an unsaturated environment. Calcium and alkalinity concentrations were higher at depth than in surface waters, which

suggests that bottom waters may be in equilibrium with settled precipitates in the surface soils. Calcium carbonate precipitates may slowly dissolved in the $CaCO_3$ -undersaturated bottom water environment.

The influence of shade on aquatic photosynthesis was apparent in the D.O. concentrations of the two community types (Table 2-2). One replicate cattail mesocosm contained more *Najas* than the other, and had a more open *Typha* canopy. Increased light penetration was observed for this mesocosm, with an 87±8% reduction in incident irradiance at the water surface compared to 97±2% reduction within the more dense *Typha* community. SAV surface water D.O. concentrations were 16.2 mg O_2 L^{-1} or greater, whereas *Typha* surface waters were 0.5 – 3.5 mg O_2 L^{-1}. Waters at depth were lower in D.O. than surface waters in both SAV and *Typha*, due to lower aquatic productivity and greater distance from the air-water interface.

Diel Water Quality Monitoring of Emergent and Submerged Communities

Since it was created in 1994, surface water in Cell 1 has been maintained at an average stage of 3.64 ± 0.16 m NGVD. During the 2002 deployments of the porewater equilibrators (June 10- 27) and hydrolabs (July 3 – 17), the cell stage was only slightly higher than average (3.89 m) (Figure 2-6). Prior months had declining water levels and low (<10 cfs) hydraulic loads, typical flows during the spring dry season.

Table 2-2. Water column characteristics at 3 cm and 20 cm depths in duplicate mesocosms (2.2m L x 0.79m W x 0.4m D) dominated by emergent and submerged aquatic vegetation (EAV and SAV, respectively) on May 30, 2001. Water from STA-1W inflow canal was delivered to each mesocosm at 10 cm day^{-1} between December 1998 and August 2001.

			Rep	Time	DO mg L^{-1}	$CaCO_3$ SI	Alkalinity mg $CaCO_3$ L^{-1}	Diss. Ca mg L^{-1}	SRP µg L^{-1}	pH	Temp °C
Inflow		EAV	1	13:22			148	59.6	58	7.36	31.0
			2	13:22			150	60.5	56	7.59	33.3
		SAV	1	13:40			142	61.1	52	7.37	33.5
			2	13:40			148	62.7	49	7.56	34.3
3 cm	Inflow Region	EAV	1	10:00	3.5	0.7	134	60.1	14	7.38	25.7
			2	11:20	1.5	0.4	146	58.2	36	7.18	27.3
		SAV	1	11:40	16.2	**15**	144	59.7	29	8.60	31.5
			2	12:00	20+	**32**	108	41.6	11	9.26	31.9
	Outflow Region	EAV	1	10:24	2.3	0.8	146	58.5	18	7.42	25.4
			2	10:45	0.3	0.3	147	59	37	6.99	25.1
		SAV	1	12:20	20+	**103**	122	42.7	5	9.90	33.2
			2	12:35	20+	**105**	111	42.3	5	9.91	33.9
20 cm	Inflow Region	EAV	1	10:15	0.5	0.3	146	58.4	20	7.09	24.5
			2	11:30	0.2	0.2	164	59.5	49	6.89	24.7
		SAV	1	11:50	0.8	0.6	172	67.7	56	7.22	24.8
			2	12:10	0.1	0.6	154	59.1	30	7.33	25.0
	Outflow Region	EAV	1	10:28	0.5	0.3	150	58.5	25	7.00	24.6
			2	10:55	0.2	0.2	140	59.4	40	6.94	24.2
		SAV	1	12:28	0.1	0.7	176	60.5	7	7.30	25.2
			2	12:55	0.5	0.7	162	60.7	5	7.34	25.4

Fluctuating water levels exposed the Hydrolab probes to air twice during the two-week deployment, and only a subset of the data (July 9 – July 17, 2002) was used in this discussion. Differences in bottom elevation between stations meant that water depth was on average 0.3 m deeper at the SAV station than at the nearby emergent station (Figure 2-6).

Water quality monitoring in the Cell 1 outflow region revealed similar temperature, dissolved oxygen saturation and specific conductance levels between emergent and SAV communities (Figure 2-7, Figure 2-8, and Figure 2-9). The emergent community, however, had pH levels roughly 1 pH unit lower than the submerged community (Figure 2-10). Average (± one s.d.) pH level from July 9 – 16, 2002 was 8.05 ± 0.17 in the SAV community, as compared to 7.10 ± 0.34 in the emergent community. Such a difference in pH may result from inorganic carbon uptake by SAV for aquatic photosynthesis.

Conversion of HCO_3^- and CO_2 into organic cell components likely depleted these constituents despite the evidence of well-mixed conditions provided by other parameters. Calcium carbonate equilibria can buffer water column pH around 8.3. In the SAV community, sustained underwater photosynthesis would tend to increase local pH levels, drive $CaCO_3$ precipitation, and create a $CaCO_3$ –buffered environment.

The lack of a diel pattern in pH levels suggests that during the high flow event in June, little photosynthesis was occurring at either station. The high flows may have suspended bottom sediment, and water depths were substantially

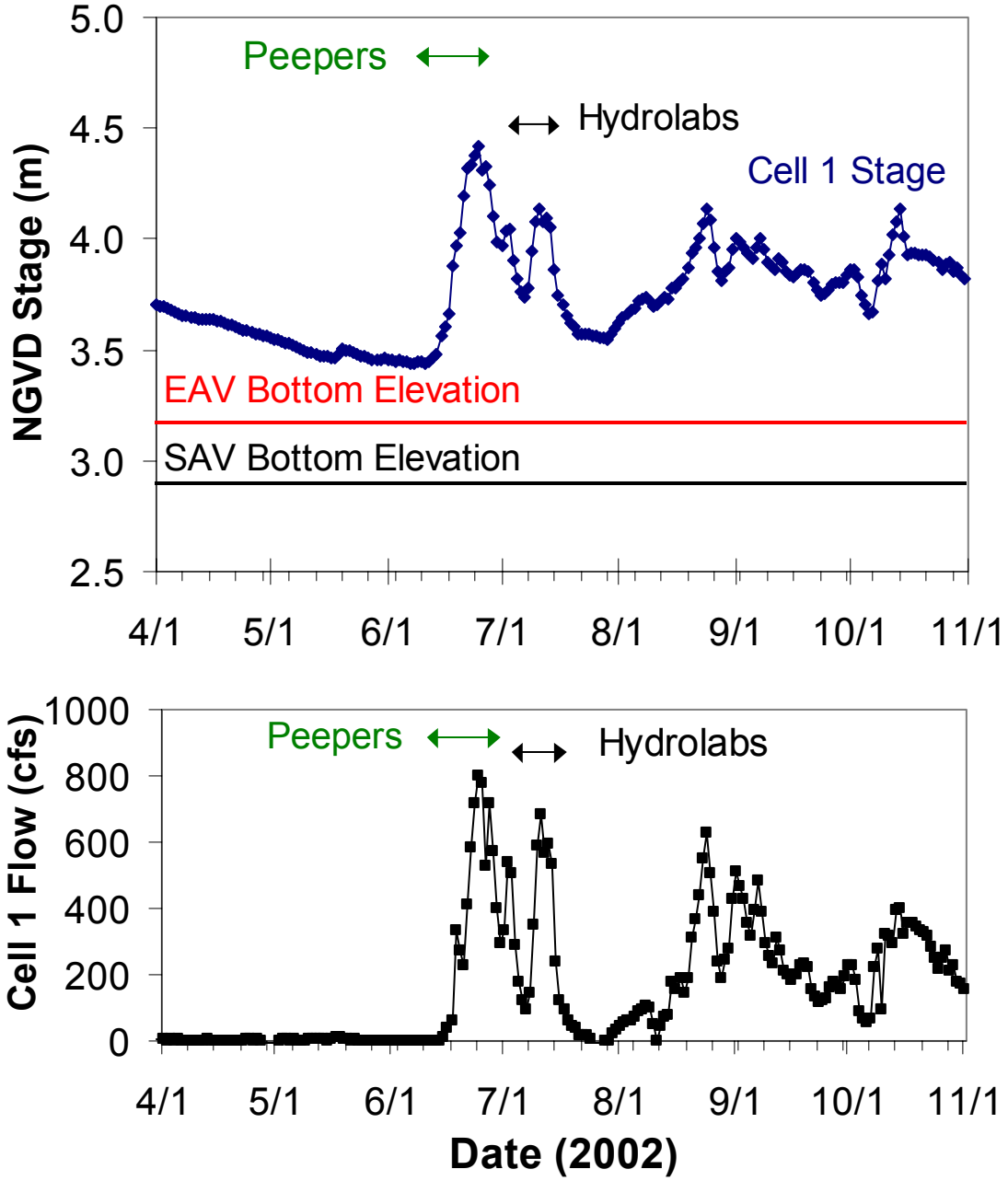

Figure 2-6. Inflow rates and stage level of water in the outflow region of Cell 1 compared to bottom elevations at the emergent and submerged aquatic vegetation (SAV) stations, prior to and during peeper and Hydrolab deployments.

increased. The dissolved organic carbon in wetland surface waters can attenuate light in a few meters depth (Krause-Jensen and Sand-Jensen, 1998). While D.O.

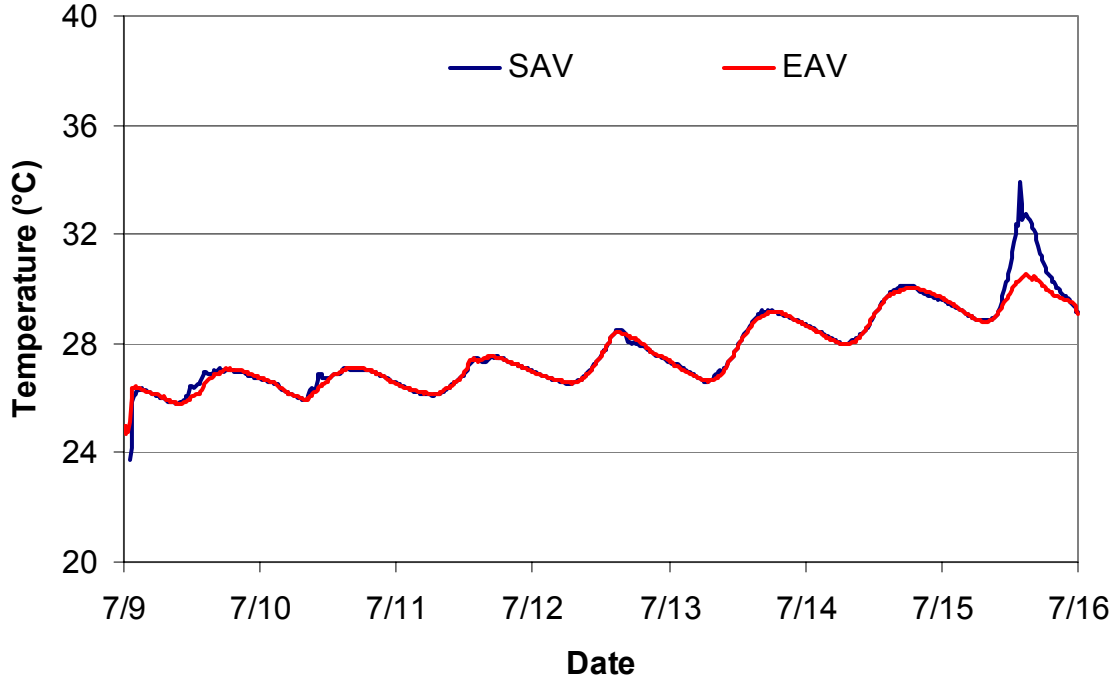

Figure 2-7. Water column temperature within Cell 1 emergent and SAV communities during the Hydrolab monitoring period (July 3 – 17, 2002).

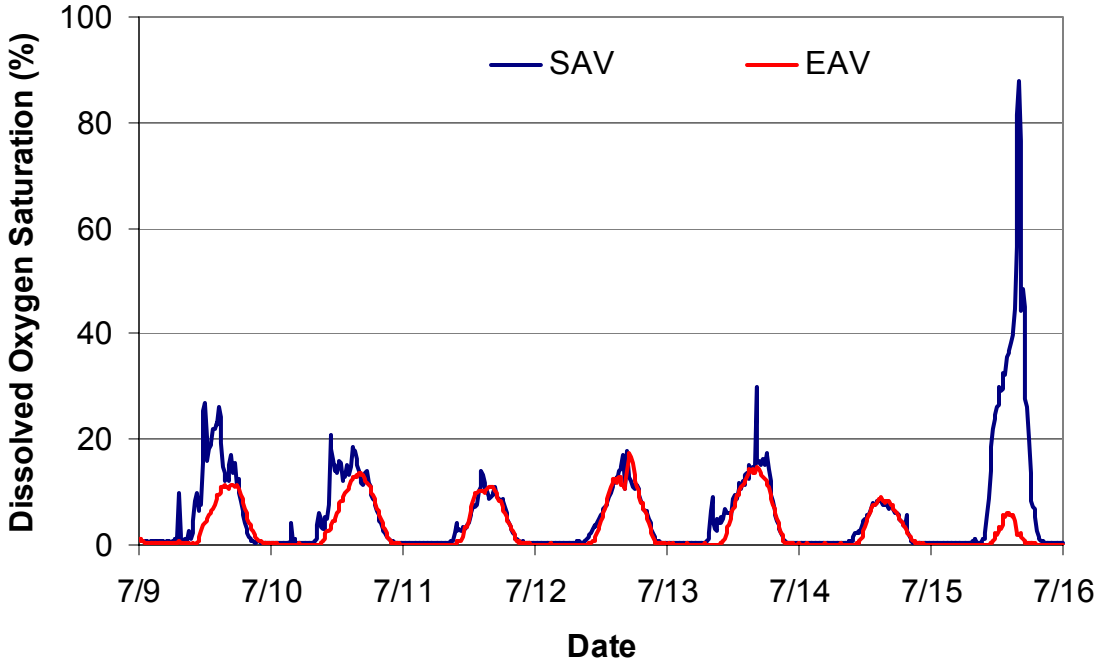

Figure 2-8. Water column dissolved oxygen saturation levels within Cell 1 emergent and SAV communities during the hydrolab monitoring period (July 3 – 16, 2002).

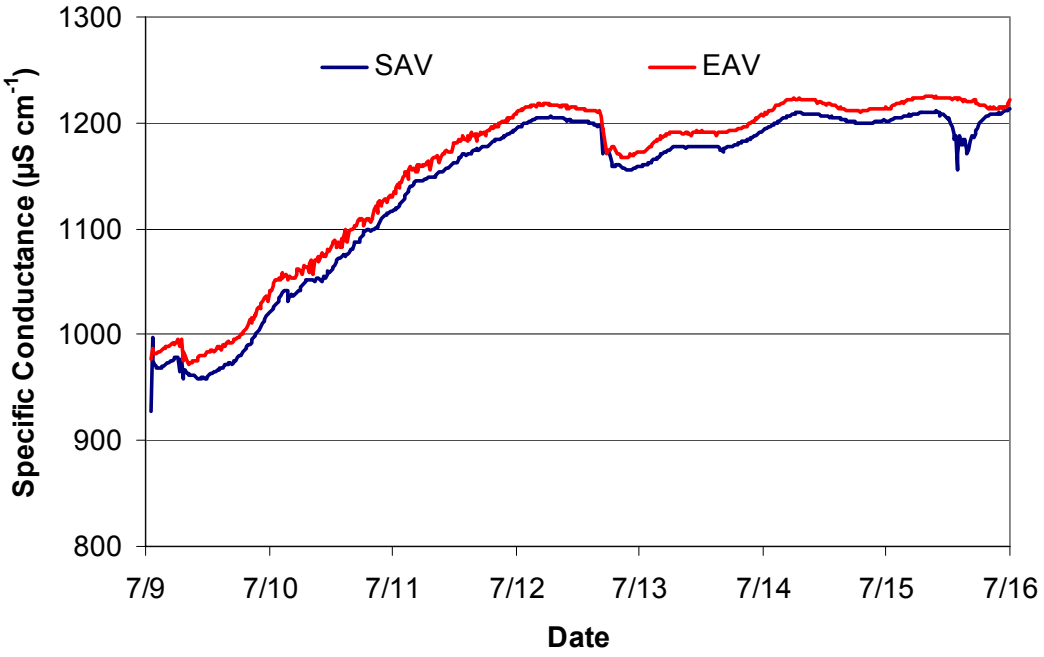

Figure 2-9. Water column specific conductance levels within Cell 1 emergent and SAV communities during the hydrolab monitoring period (July 3 – 16, 2002).

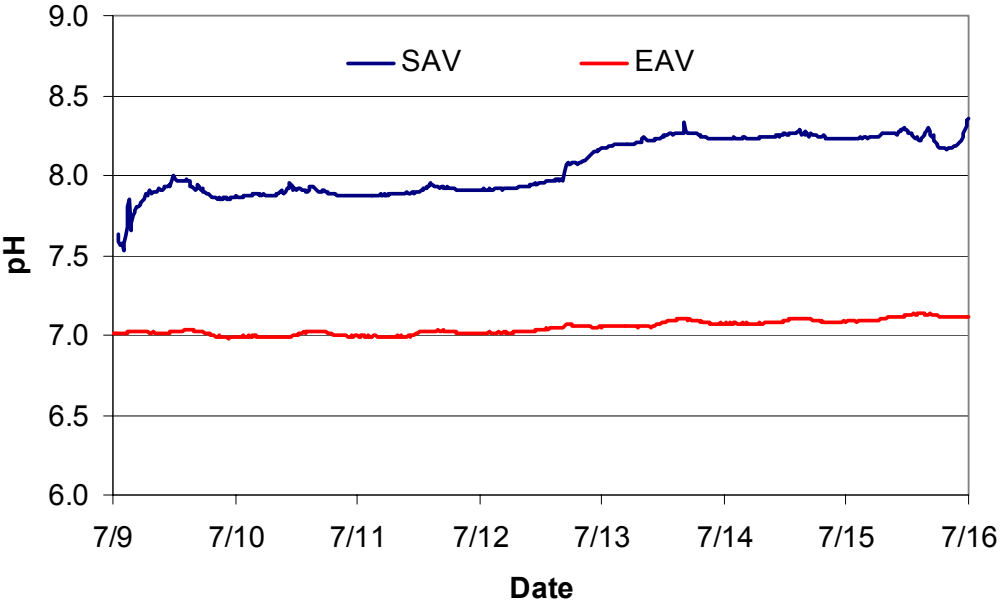

Figure 2-10. Water column pH levels within Cell 1 emergent and SAV communities during the hydrolab monitoring period (July 3 – 16, 2002).

concentrations did increase from 0 to 20% saturation during daylight hours at each station, this is lower than the levels observed in SAV communities under more typical, quiescent conditions (DBEL, 2001) or as observed in the mesocosms (Table 2-2). Oxygen concentrations in June were lower than those observed on September 29, 2002 at the same Cell 1 stations (Table 2-3).

In September, D.O. concentration profiles at the SAV station revealed supersaturated conditions in the SAV surface waters (10.6 mg L^{-1}, 31.8°C) and lower levels (0.65 mg L^{-1}) near the bottom of the water column (0.65 m from water surface and 0.1 m above the sediment surface). Emergent-stand surface waters were less saturated than the SAV station with respect to D.O. (2.15 mg L^{-1}, 31.5°C), but waters at depth were similar (0.60 mg D.O. L^{-1}). Surface water pH levels at that time were 8.29 and 7.66 for SAV and emergent stations, respectively.

Light penetration into the water column is essential for aquatic photosynthesis. On September 29, 2002, below-surface light levels were reduced in emergent stands relative to open water and SAV beds (Figure 2-11). In the SAV and open water areas, light available just below the surface was substantially reduced (~65%) from ambient light 2 m above the surface. Light reflecting off the water surface may have increased the "ambient" measurements and decreased the submerged surface measurements. Further light reduction within the open water column may have resulted from attenuation and scattering by dissolved organic matter or suspended particulate matter.

Table 2-3. Water column dissolved oxygen (D.O.) concentrations, pH, and temperature profiles in the STA-1W Cell 1 surface waters at the time of peeper retrieval on June 27, and on September 29, 2002.

		EAV		SAV	
Parameter	**Depth**	June	Sept.	June	Sept.
Dissolved oxygen, mg L^{-1}	Surface	0.30	2.15	1.1	10.6
	Mid	0.25	0.55	1.1	3.1
	Bottom	0.15	0.60	0.60	0.65
pH	Surface	7.50	7.66	7.38	8.29
	Mid	7.33	7.34	7.26	7.34
	Bottom	7.47	7.15	7.22	7.26
Temperature, °C	Surface	31.0	31.5	30.3	31.8
	Mid	29.7	28.9	29.3	29.0
	Bottom	29.7	29.1	28.3	28.8
Water Depth, m		1.10	0.74	1.45	0.70

The *Typha* canopy drastically reduced the amount of light available just below the water surface. SAV tissues reduced available light by shading the waters below the leaf canopy. Interesting to note, however, is the near total light extinction (>99% attenuated) at bottom depths (62 - 65cm) in both SAV and emergent communities, regardless of the shallow water column (75 cm). The phototrophic benthos at the open water stations, in contrast, had 7.7 ± 0.3 % (as mean ± 1 s.d.) of the incident photon flux available as light energy.

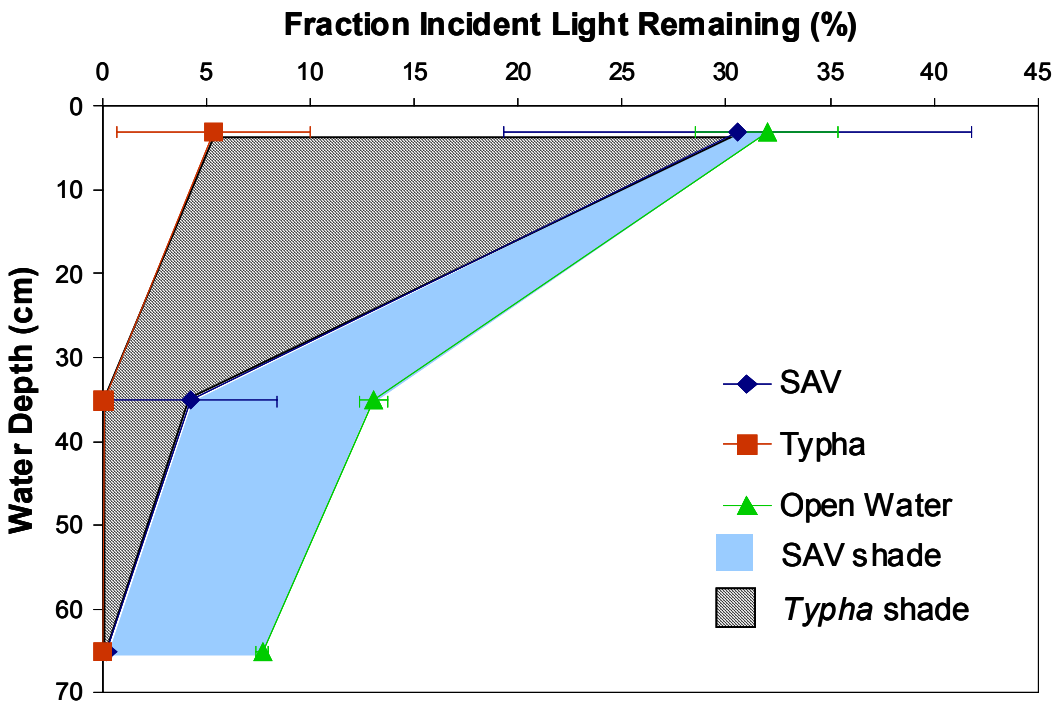

Figure 2-11. Incident light (measured as photon flux) remaining at water depths of 0+, 35 and 65 cm in emergent and submerged macrophyte communities, as well as within open water reaches, of STA 1W, Cell 1, in September 2002. The shaded regions represent the contribution of SAV and *Typha* canopy shading to light reductions, beyond the attenuation attributed to the water column alone.

Even in shallow (<1m) treatment wetlands, the sediment-water interface and a portion of the water column may be below the euphotic zone. Light availability controls aquatic photosynthesis and alters surface water chemistry. The influence of light and aquatic photosynthesis on water column P dynamics, then, is greater near the air-water interface when the area is vegetated, while the remainder of the water column is influenced by respiration processes.

Porewater Chemistry

Significant rainfall within the S-5 basin during the 17-day equilibration required pumping of ADW into STA-1W. Water levels increased approximately 0.9 m between deployment June 10, and retrieval June 27, 2002 (Figure 2-12). At the time of retrieval, recent inflow waters had likely influenced surface water chemistry (Table 2-3).

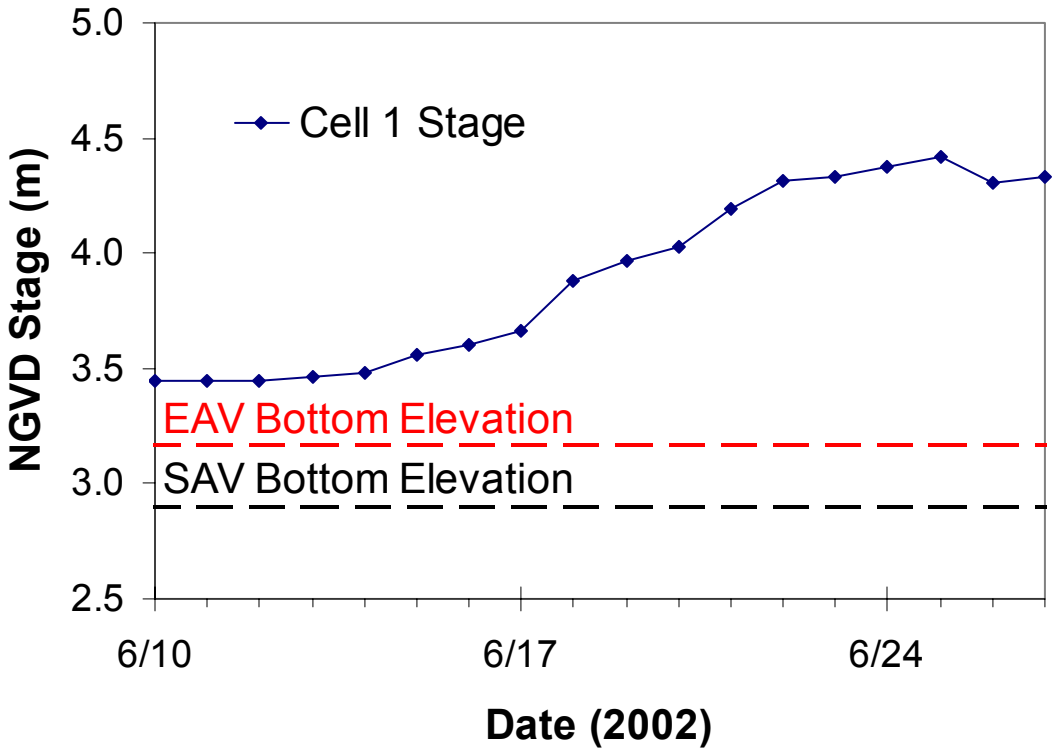

Figure 2-12. Stage level recorded during the seventeen day equilibration period for porewater samplers deployed in the outflow region of STA-1W Cell 1, during June 2002. The bottom depths for the SAV and Emergent vegetation stations are shown for reference.

Profiles of porewater pH levels associated with emergent and submerged vegetation were circumneutral (6.77 –7.57) in the both sediment types (Figure 2-13) and below the pH levels in the overlying water column. While pH values

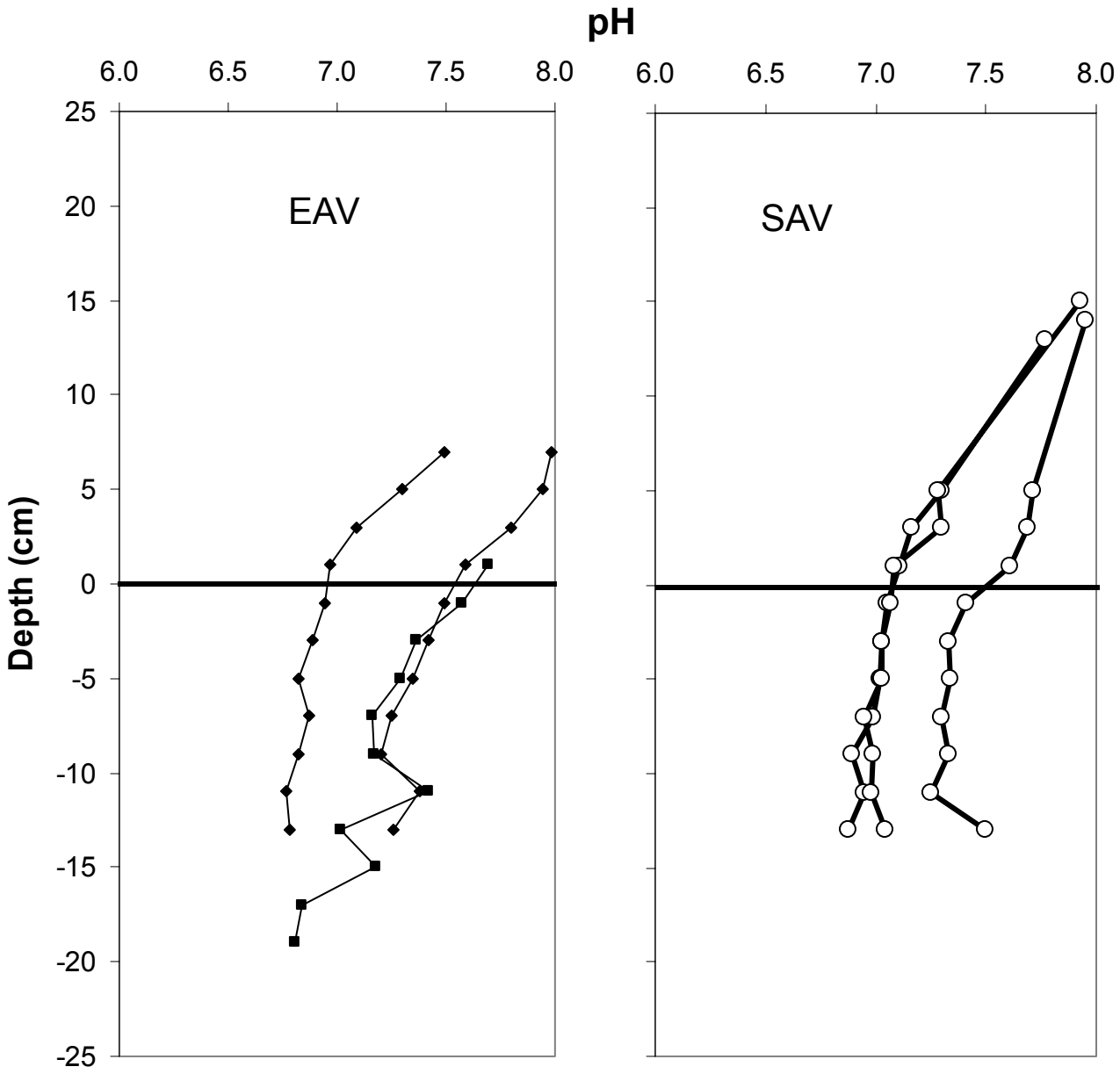

Figure 2-13. Vertical profiles of porewater pH values from soil collected from emergent and submerged vegetation. Porewater equilibrators were deployed for 17 days in June 2002, in the outflow region of STA-1W Cell 1.

were measured immediately after peeper retrieval, the disparity between replicates may be an artifact of oxygen reintroduction into, and/or CO_2 outgassing from, porewater samples. However, the disparity is more likely due

to the natural spatial variability of the soils. All three replicate peepers were retrieved at once at each site, and the first station's samples were processed before the second station peepers were withdrawn. Those replicates processed first had lower pH values than replicates sampled last. All pH determinations were made within 4 hours of peeper retrieval.

Profiles of SRP concentrations were different between emergent and submerged community sediments (Figure 2-14). Emergent aquatic vegetation (EAV) sediments exhibited slightly higher mean porewater SRP concentrations (0-6 cm depth, 996 ± 131 $\mu g\ L^{-1}$) and lower overlying water column concentrations (0+6 cm depth, 402 ± 293 $\mu g\ L^{-1}$) than the submerged plant communities (0-6 cm depth, 728 ± 279 $\mu g\ L^{-1}$; 0+6 cm depth, 616 ± 256 $\mu g\ L^{-1}$). The resulting diffusive P flux was greater from the emergent sediments (0.39 ± 0.11 mg P $m^{-2}\ d^{-1}$) than from SAV sediments (0.07 ± 0.05 mg P $m^{-2}\ d^{-1}$).

Porewater DOP concentrations in EAV soils ranged from 0 to 350 $\mu g\ L^{-1}$, with one measurement at 658 $\mu g\ L^{-1}$, 18-20 cm below the soil surface (Figure 2-15). The SAV soil DOP profile ranged from 0-830 $\mu g\ L^{-1}$, with maximum concentrations 2-6 cm below the soil surface in all three replicates. DOP may diffuse upward into the water column, or downward into deeper soils, where further transformations may increase P recalcitrance, or conversely make it bioavailable. Surface soils also contained greater fulvic- and humic acid-associated P in both soil types than the muck soils lower in the soil profile (Figure 2-5). Microbial biomass and activity have been reported higher in WCA-

2A surface soils than in deeper soils (White and Reddy, 2001), and may influence both the porewater DOP pool and the soil fulvic- and humic acid-P pools through release of metabolic by-products or mineralization of organic matter.

Water columns at both EAV and SAV stations were likely anoxic during the 17-day peeper equilibration period. High flows (Figure 2-6) likely

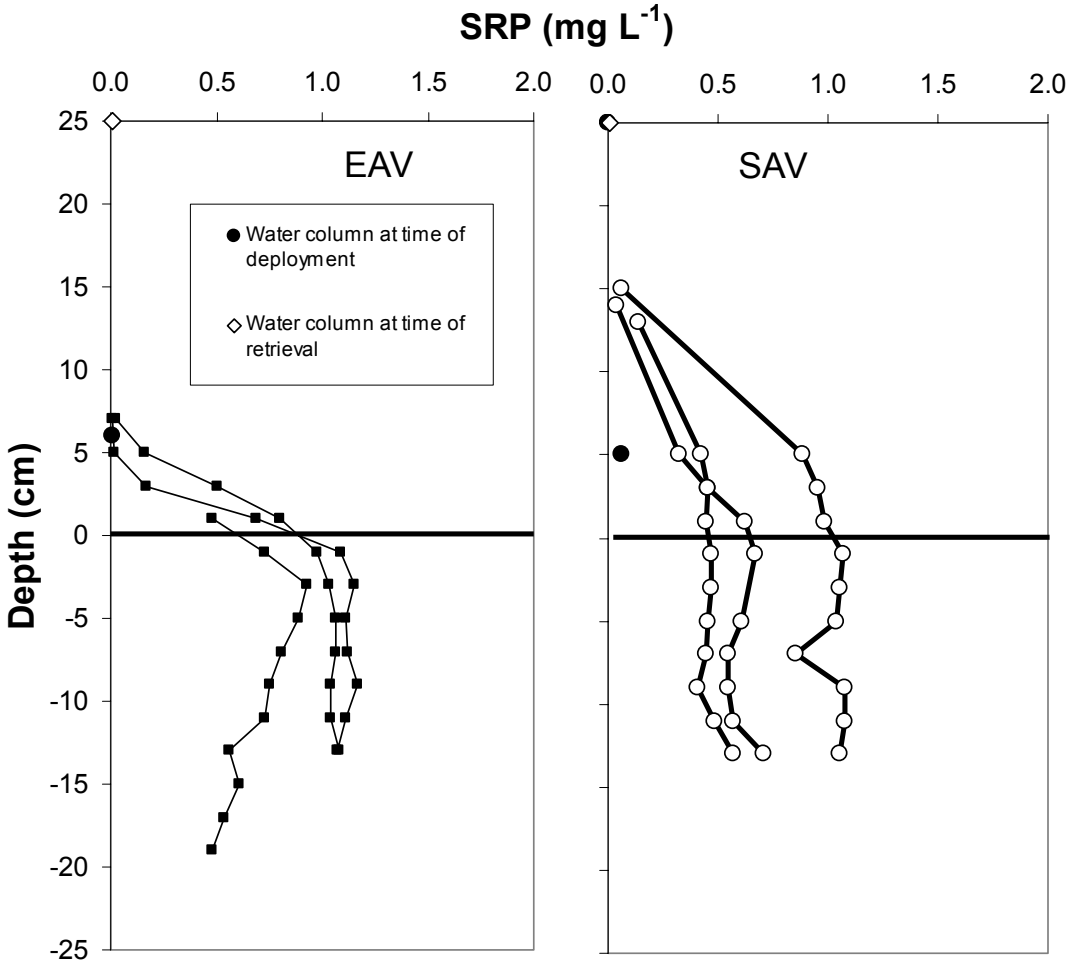

Figure 2-14. Vertical profiles of soluble reactive phosphorus concentrations in soil porewater collected from emergent and submerged vegetation. Porewater equilibrators were deployed for 17 days in June 2002, in the outflow region of STA-1W Cell 1.

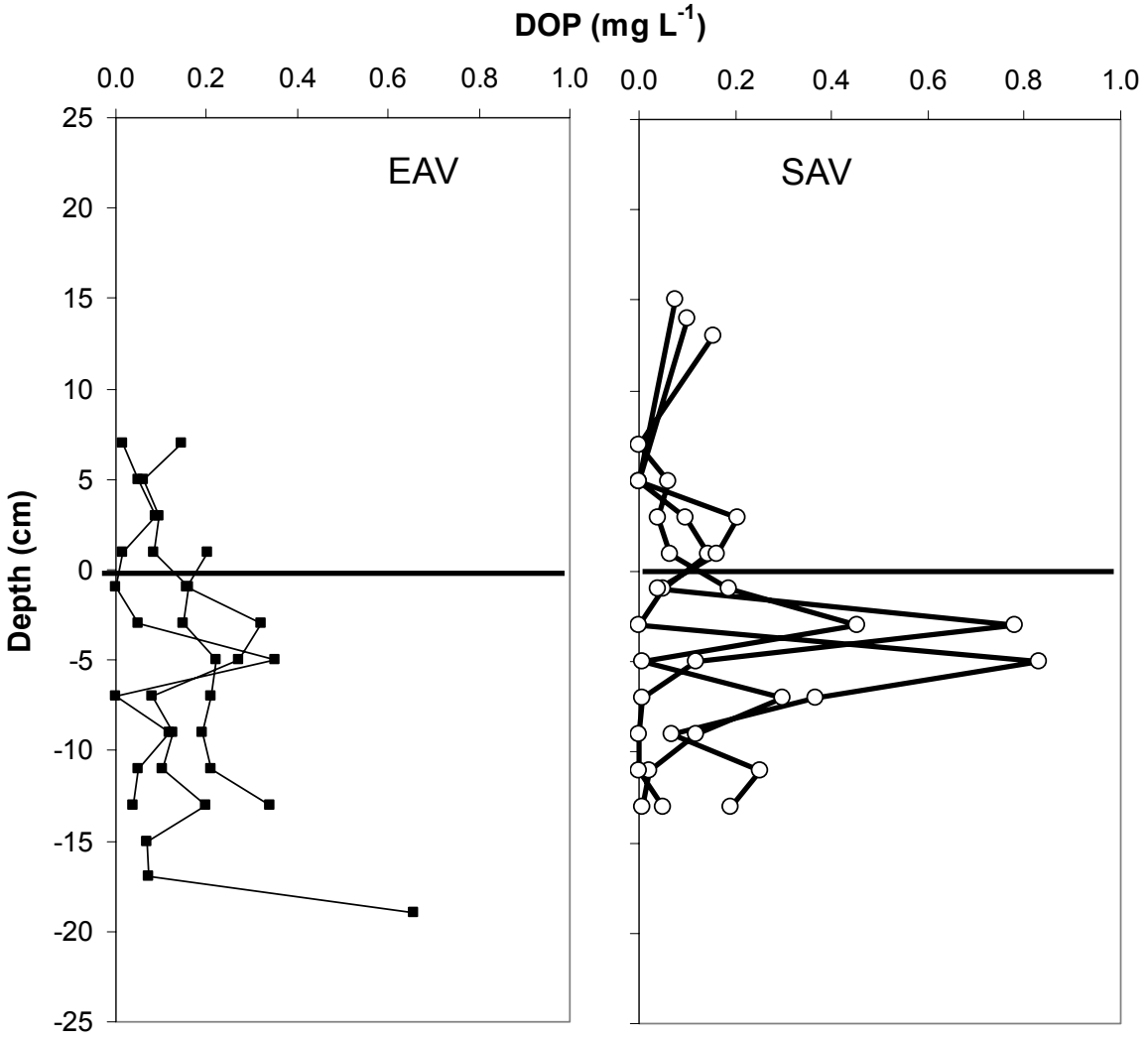

Figure 2-15. Vertical profiles of dissolved organic P concentrations in soil porewater collected from emergent and submerged vegetation. Porewater equilibrators were deployed for 17 days in June 2002, in the outflow region of STA-1W Cell 1.

resuspended surface sediments, increasing light attenuation and decreasing photosynthesis. Dissolved oxygen concentrations on June 27, 2002 (retrieval) were low even in surface waters (0.30 and 1.1 mg L^{-1}) within the SAV and EAV communities, respectively (Table 2-3).

Under low-oxygen conditions, Fe-oxides can be reduced to soluble Fe^{2+}. This was observed, as dissolved Fe concentrations increased with depth through the emergent macrophyte porewater (Figure 2-16). The profile in submerged community was more uniform with depth. All concentrations were low compared to those typical (50-100 mg L^{-1}) of reduced mineral soil environments (Patrick and Khalid, 1974). Newman and Pietro (2001) reported similarly low total Fe concentrations (0.15-0.25 mg L^{-1}) in STA-1W Cell 4 in 1993-1994 surface water samples taken just after field flooding. Even under oxygenated conditions, such low Fe concentrations may not provide the P sorption capacity characteristic of acidic mineral wetland soils (Richardson, 1985).

Calcium, specific conductance and especially alkalinity concentration profiles all increased with depth in the EAV profiles, but were constant or decreasing with depth in the submerged profiles (Figure 2-17 to Figure 2-19). Calcium carbonate precipitation in surface waters within the SAV community may have led to elevated values, while dissolution of the underlying limerock may have influenced the porewater concentrations in both soil types. The EAV community porewater exhibited a clear gradient of alkalinity concentrations, suggesting upward diffusive flux of these components through the soil profile, and potentially into the water column.

Calcium carbonate precipitation is concomitant with aquatic photosynthesis in hard waters. Everglades ADW was often saturated with $CaCO_3$ before entering STA 1W, as observed in mesocosm inflow waters.

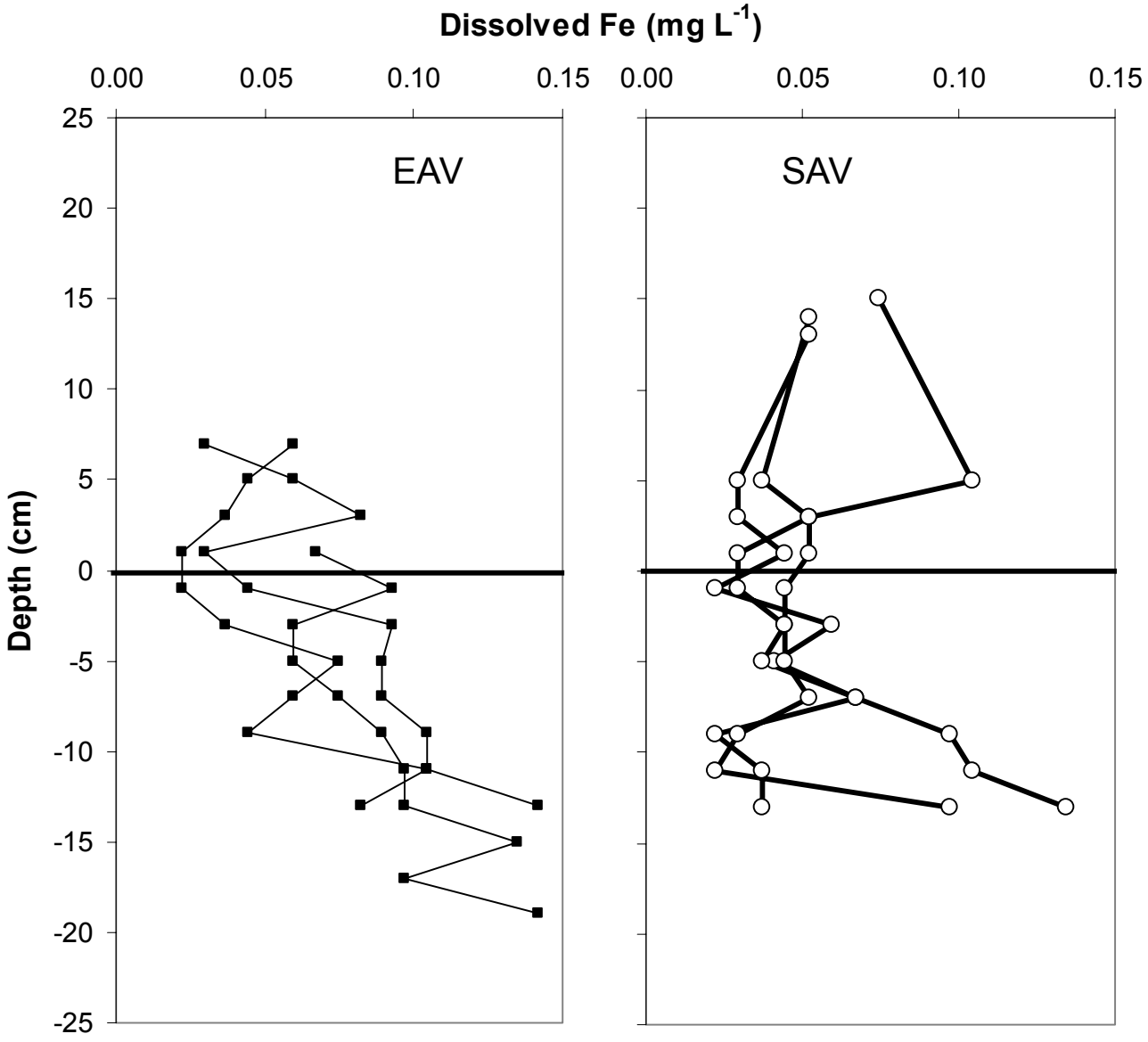

Figure 2-16. Vertical profiles of dissolved iron concentrations in sediment porewater collected from emergent and submerged vegetation. Porewater equilibrators were deployed for 17 days in June 2002, in the outflow region of STA-1W Cell 1.

Daytime aquatic photosynthesis elevated pH levels in the mesocosms, and the water column became supersaturated. The formation of $CaCO_3$ precipitates is therefore an important mechanism for P sorption capacity of newly accrued soils.

Saturation index values indicated $CaCO_3$ precipitation was favored in the porewater of both community types and at all depths (Figure 2-20). Over the long term, water column $CaCO_3$ precipitation may be less important to soil sorption capacity than previously thought. Differences in the water column

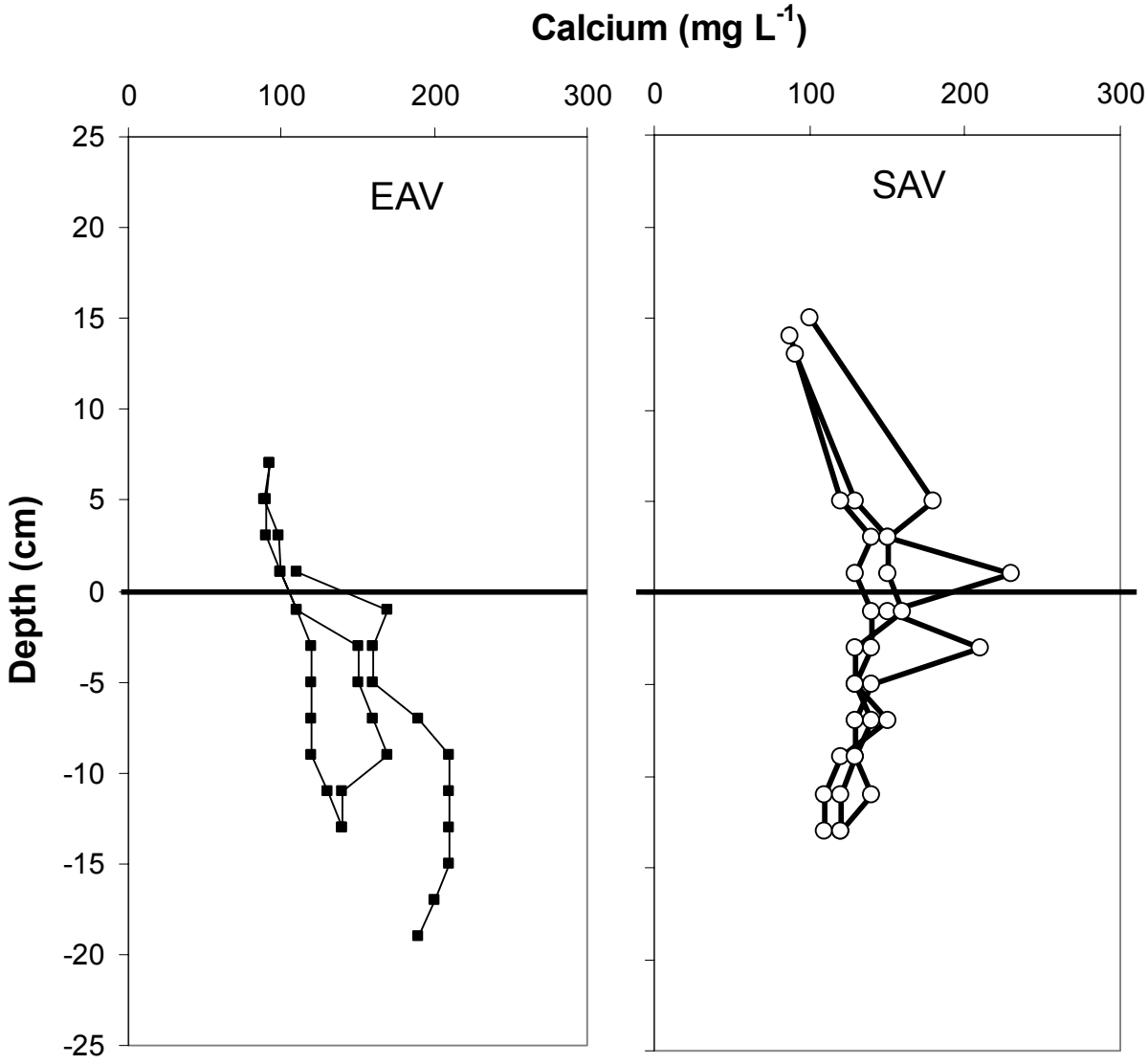

Figure 2-17. Vertical profiles of dissolved calcium concentrations in sediment porewater collected from soils below emergent and submerged vegetation. Porewater equilibrators were deployed for 17 days in June 2002, in the outflow region of STA-1W Cell 1.

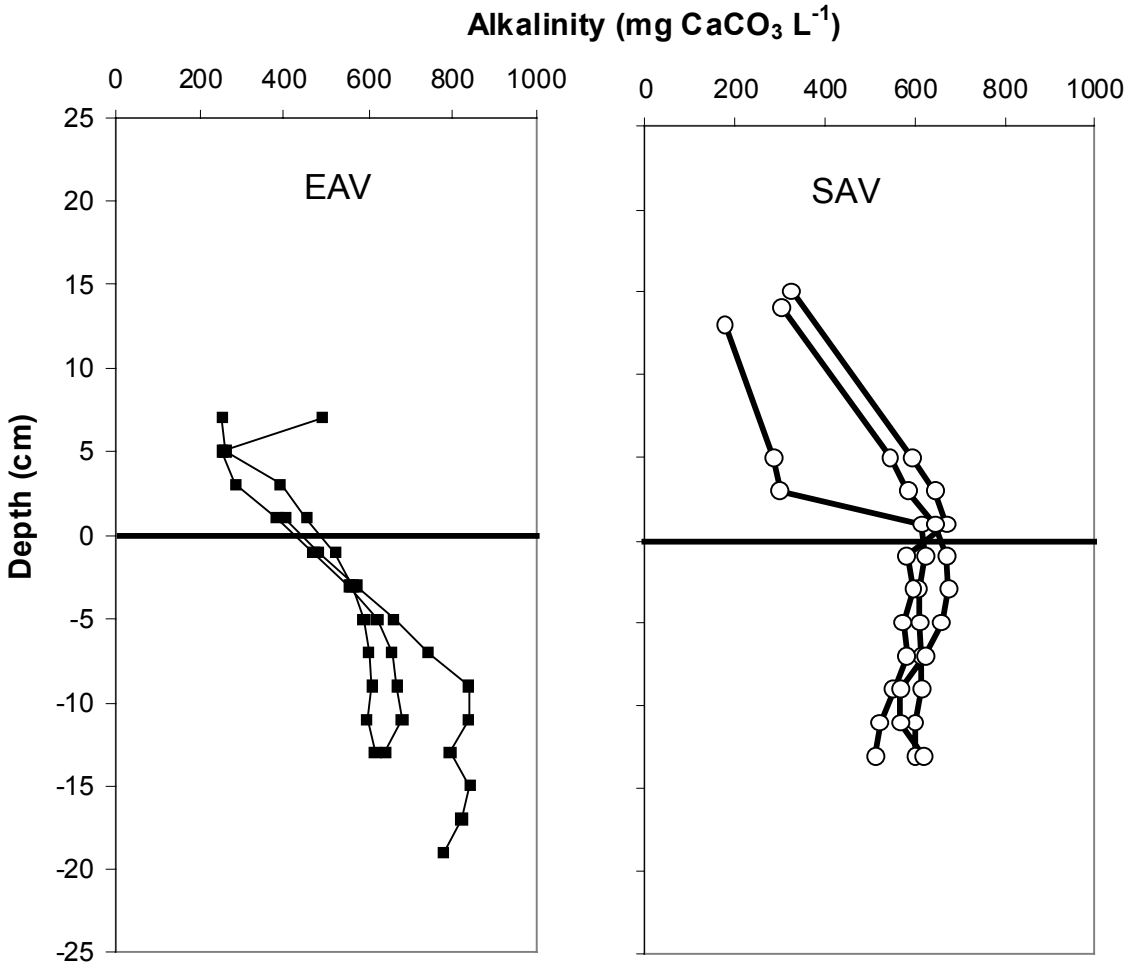

Figure 2-18. Vertical profiles of alkalinity concentrations (as $CaCO_3$) in sediment porewater collected from soils below emergent and submerged vegetation. Porewater equilibrators were deployed for 17 days in June 2002, in the outflow region of STA-1W Cell 1.

chemistry observed for submerged and emergent macrophyte stands were not as apparent in the porewater. Influence of the native calcareous substrata on emergent community porewater may account for P storage in the Ca-bound, HCl-extractable P pool in those soils, despite the lack of evidence for calcium

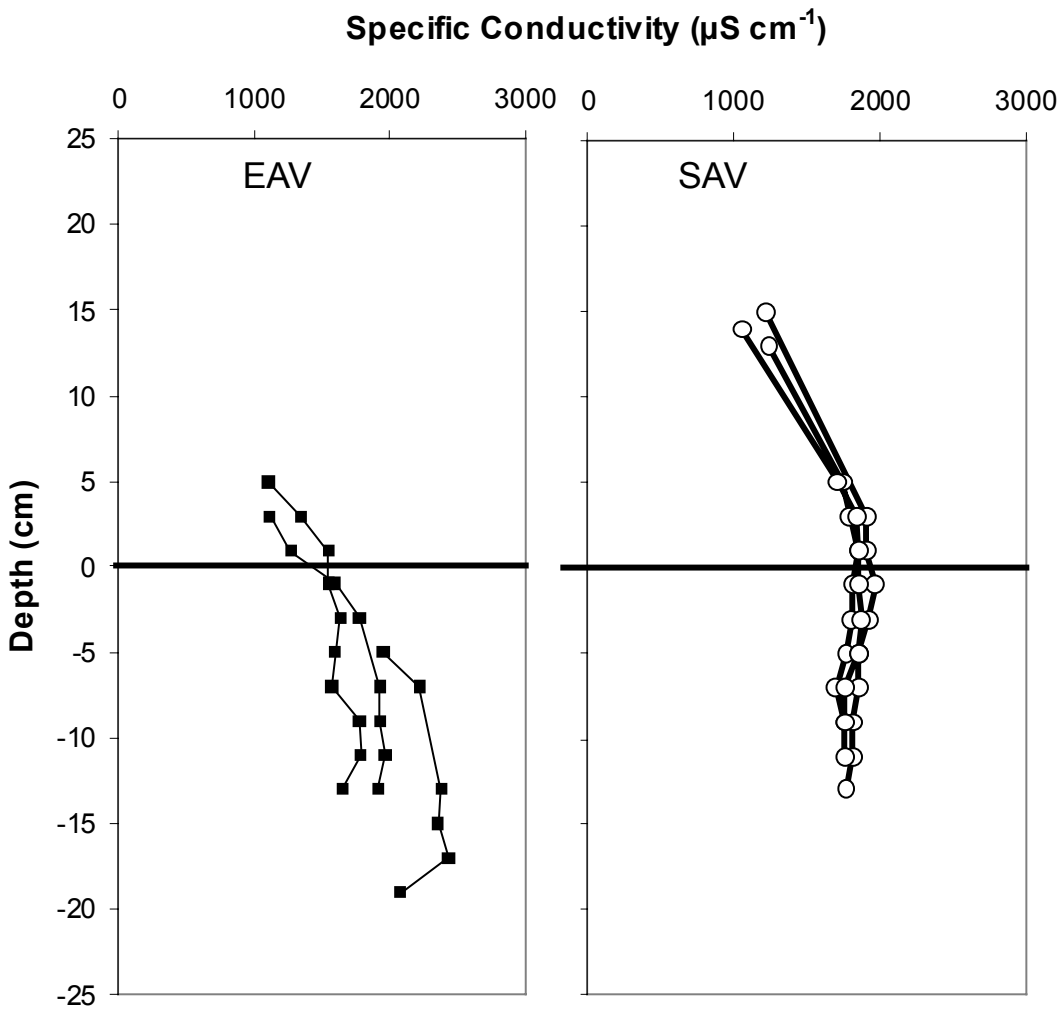

Figure 2-19. Vertical profiles of specific conductance values in sediment porewater collected from soils below emergent and submerged vegetation. Porewater equilibrators were deployed for 17 days in June 2002, in the outflow region of STA-1W Cell 1.

carbonate precipitation in the EAV water column. Water column $CaCO_3$ precipitation in the SAV beds probably explains the higher HCl-bound P pool found in those soils (271 ± 80 mg P kg^{-1}) than those of the *Typha*-region soils (119 ± 45 mg P kg^{-1}).

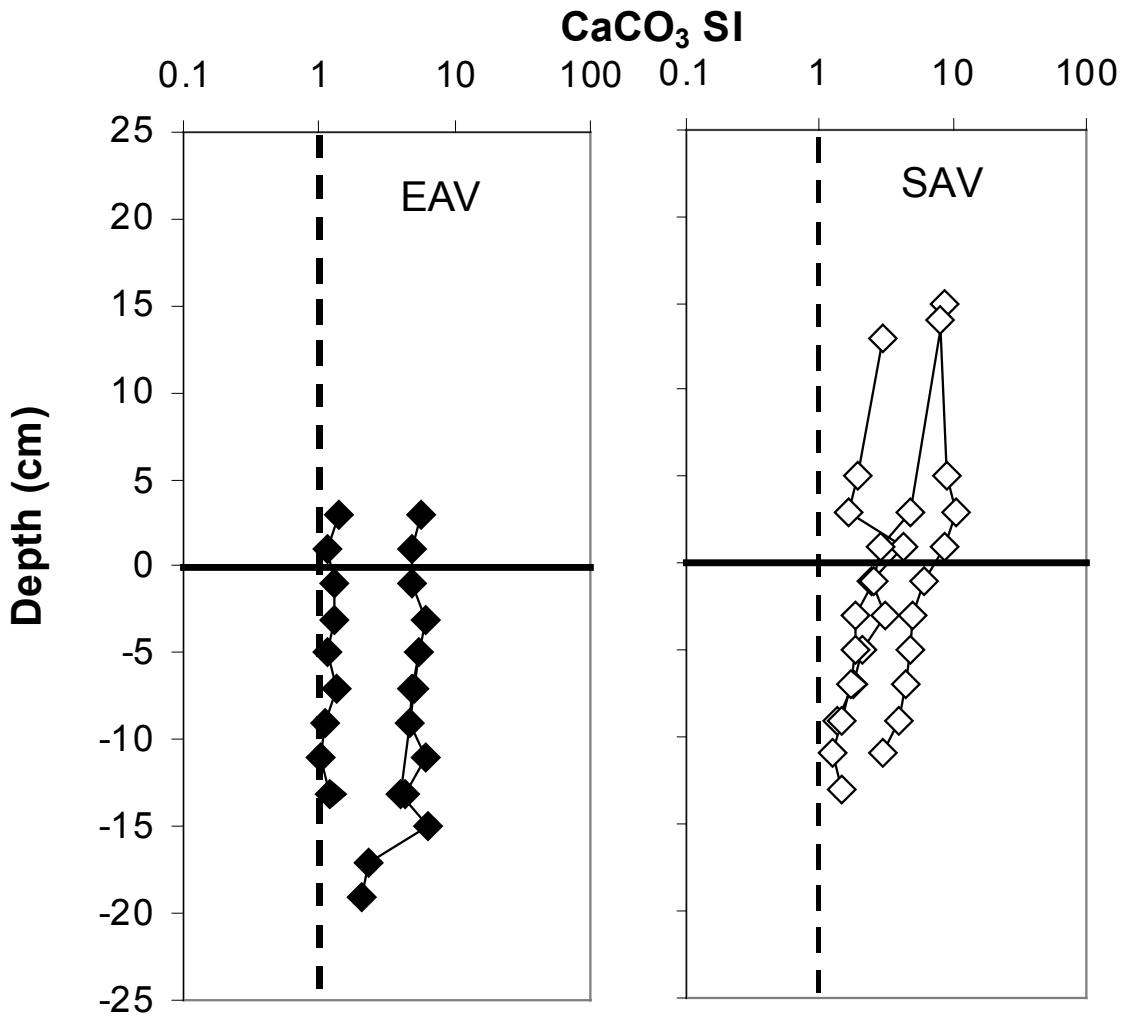

Figure 2-20. Vertical profiles of calcium carbonate saturation index (SI) values in porewater collected from soils beneath emergent and submerged vegetation. All index values were greater than one, indicating supersaturated conditions with respect to $CaCO_3$. Note log scale on x-axis.

Intact Core P Flux Study

Soils collected from beneath the *Typha* and SAV communities released P into overlying water under dark, oxic conditions (Figure 2-21). Initial floodwater was particle free, and contained 13 µg DOP and 3 µg SRP L^{-1}. After 28 days of

incubation, SRP concentrations were highest in two of the three replicate waters overlying SAV soils (98 ± 60 µg L^{-1} for all three SAV cores). The cattail soils also released P into the water column at 49 ± 49 µg L^{-1}.

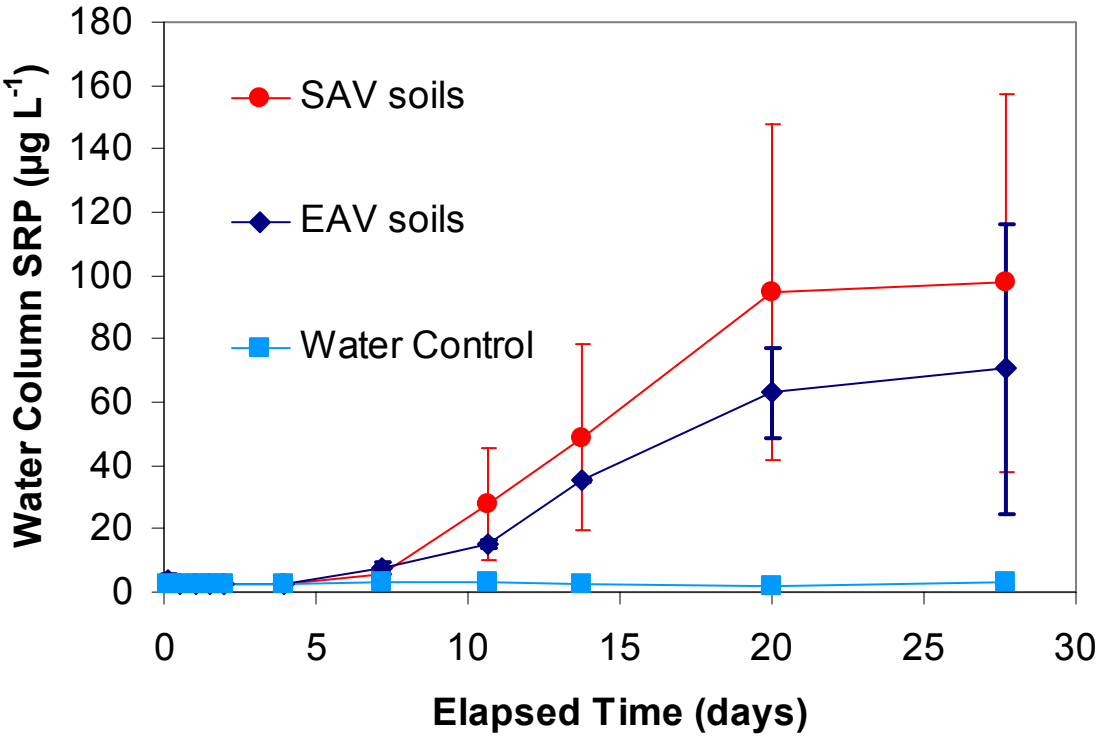

Figure 2-21. Soluble reactive phosphorus (SRP) concentrations in the water columns of SAV- and *Typha*- region soils and control columns (no soil) during 28-day dark laboratory incubation. Error bars for each treatment indicate ± one standard deviation between triplicate cores. Note: *Typha* soil treatment values are from duplicate cores, while control and SAV soil values were from triplicate cores.

Of the three un-amended cattail cores, one replicate had visible fragments of leaf litter within the surface soils. The results observed from this core were excluded from flux calculations, due to the possible effects of the litter fragments on the results. Phosphorus movement from the soil to the water during the 7-20 day period of linear SRP concentration increase was greatest from SAV soils.

Corresponding flux rates from SAV soil, adjusted for change in control column concentrations, was 1.84 ± 1.04 mg P m^{-2} day^{-1}. Emergent soils showed slightly less P release (Table 2-4), though the difference was not significant ($p > 0.05$). Near-zero (-0.02 to –0.03 mg P m^{-2} day^{-1}) flux was observed in the control cores.

Water column pH and temperature values stayed consistent throughout the 28-day incubation. Initial flood water pH (8.06) increased slightly to 8.36 ± 0.06 before the 8-hour sampling, and remained similar in all treatments (8.41 ± 0.09) during the remaining 28 days. Temperature was moderated by the incubation water bath, and averaged 24 ± 3 ° C during the incubation period.

Table 2-4. Flux estimates from intact SAV and Emergent soil cores kept under dark conditions for 35 days. The 30 cm water column was continuously aerated. A 100 µg L^{-1} phosphorus spike was added after 30 days to measure short-term uptake rates. All values are means (± st. dev.) of triplicate cores minus mean flux in control cores, in mg P m^{-2} day^{-1}.

	Period of flux estimate	**SAV soil**	**EAV soil**
Release	Day 7 to Day 20	1.84 ± 1.04	1.20 ± 0.27
Uptake	0 – 8 hours after spike	4.97 ± 12.6	0.17 ± 2.7

Similarly, dissolved calcium (Ca) concentrations were stable after a short initial equilibration period (Figure 2-22). Alkalinity concentrations increased marginally in the *Typha* soil treatment over the 28 days (Figure 2-22). SAV soil and water control treatments were constant with respect to dissolved calcium and alkalinity concentrations.

Dissolved organic P concentrations were slightly elevated in SAV-soil incubations, relative to control treatments or treatments with *Typha* soils (Figure 2-23). During the incubation period, mean DOP concentrations for both soil

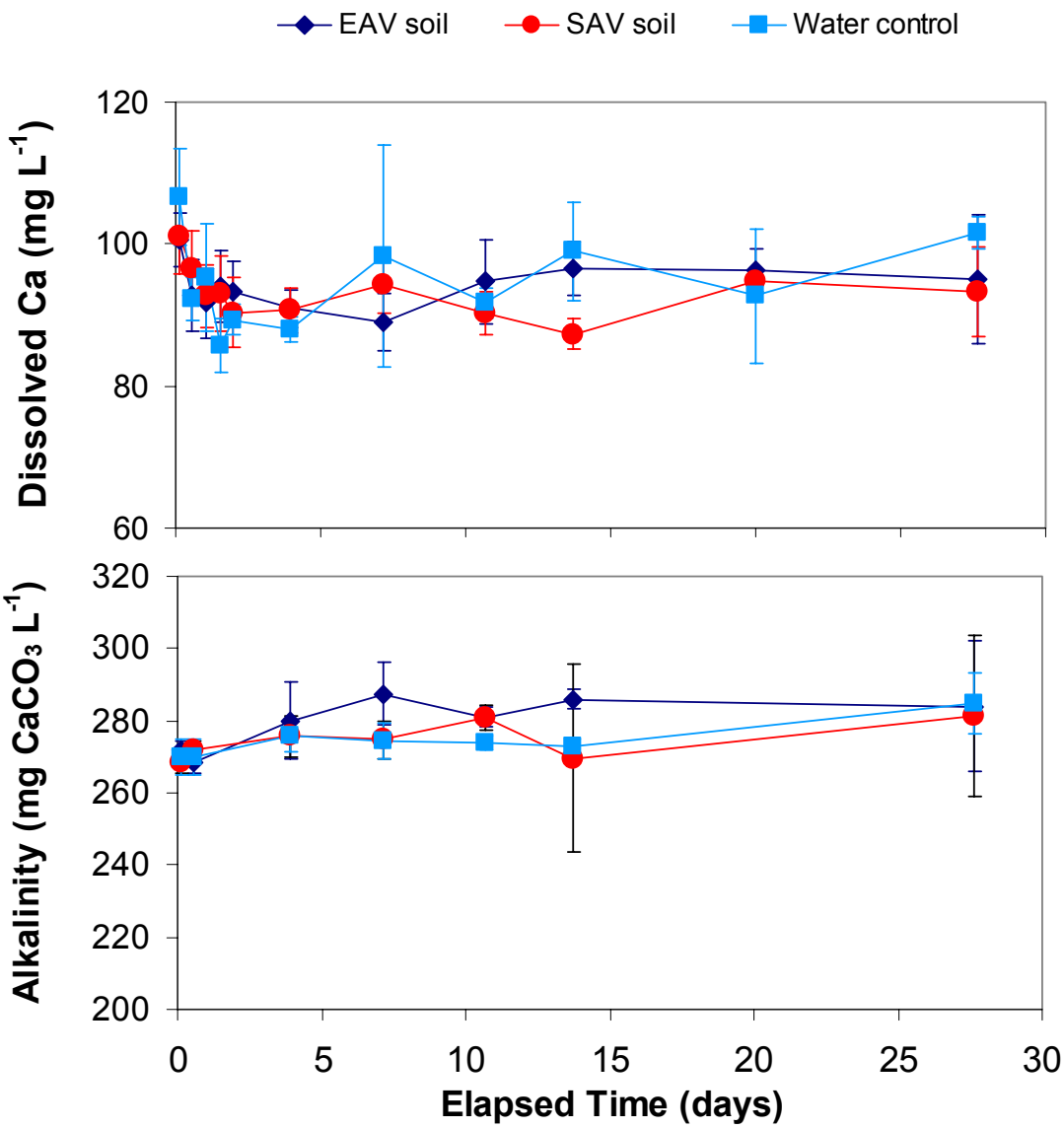

Figure 2-22. Mean dissolved calcium and alkalinity concentrations in the water columns of three treatments (see text for details) during 28-day dark laboratory incubation. Error bars indicate ± 1 standard deviation from the mean of three replicates.

treatments decreased slightly over the first 24-hours, then increased for the next 27 days. After 28 days, water column DOP concentrations above SAV soils (28 ± 3 µg L^{-1}) were higher than in waters above cattail soil (22 ± 3). These concentrations are very low relative to those observed in the porewater (e.g. as high as 830 µg L^{-1} in the surface 2-6 cm SAV soils), which were also higher in the SAV soils (Figure 2-15). Concentrations of dissolved organic carbon (DOC) were similar during the period of linear P flux (monitored only between 4 and 14 days of incubation) (Figure 2-24).

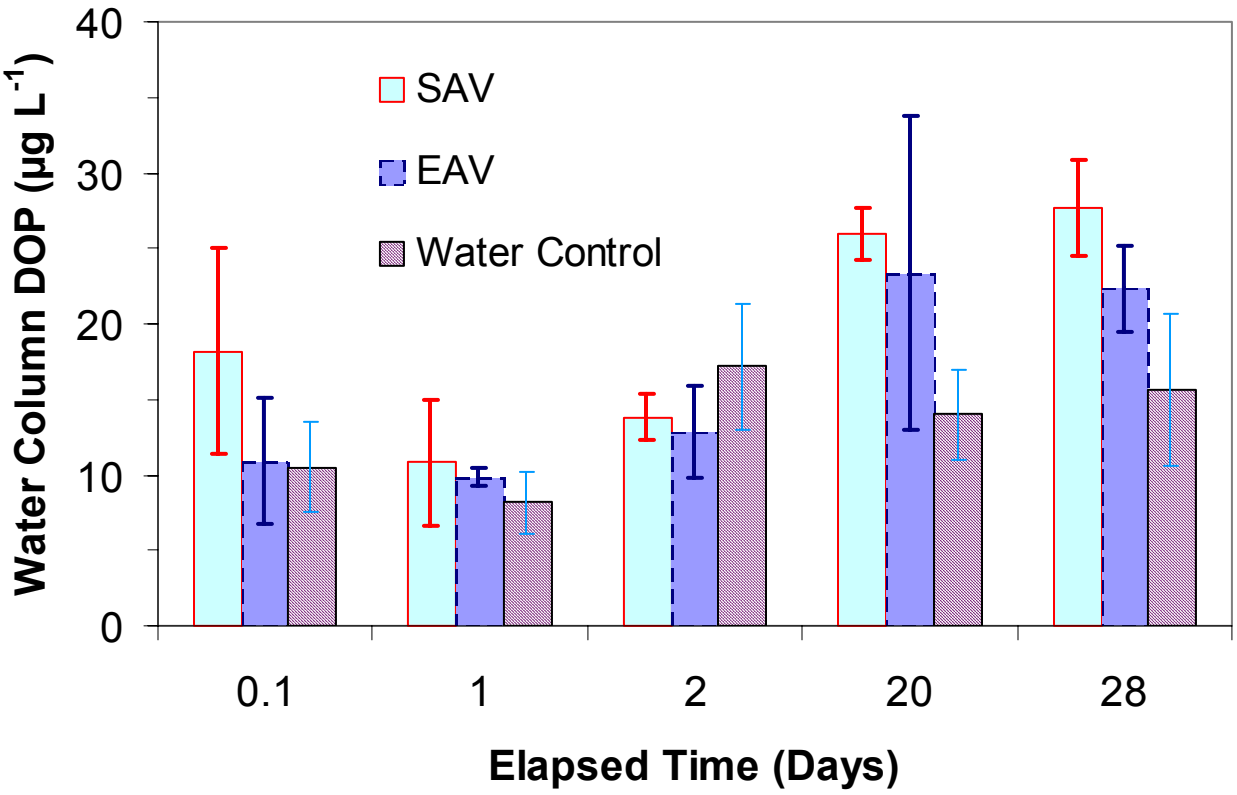

Figure 2-23. Mean dissolved organic phosphorus concentrations in the water columns above SAV and *Typha*-region soils and in control (water only) columns during 28-day dark laboratory incubation. Error bars indicate ± 1 standard deviation from the mean of three replicates.

Greater P flux from SAV soils compared to the *Typha*-region soils, though insignificant ($p > 0.05$), was supported by greater total and exchangeable P in the SAV soils (Figure 2-5). The potential diffusive P flux of 0.39 mg P m^{-2} day^{-1} in the *Typha* region, as calculated from porewater gradients, was a small contributor to overall flux (1.20 mg P m^{-2} day^{-1}).

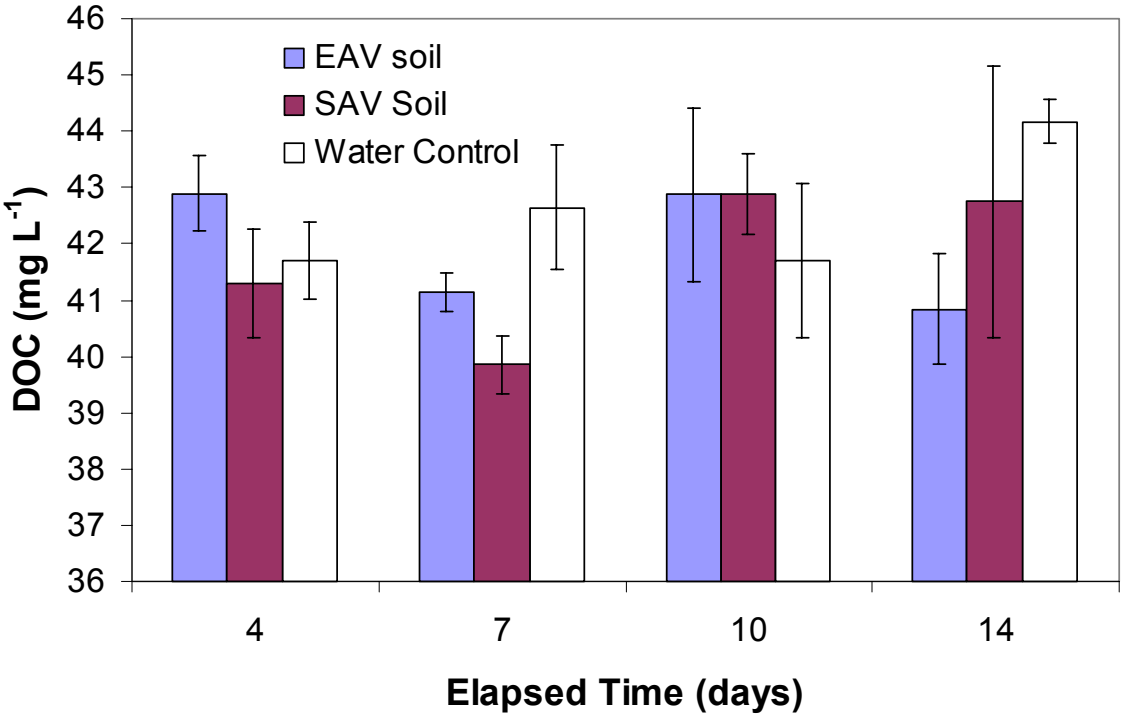

Figure 2-24. Dissolved organic carbon concentrations in water columns over *Typha* soils, SAV soils and control columns (no soil) after 4, 7, 10 and 14 days of dark laboratory incubation. Error bars indicate ± 1 standard deviation from the mean of three replicates.

Rapid biological P uptake occurred during the 8 hours following P amendment (100 µg P) (Figure 2-25). Control columns showed a small decrease in SRP concentration, then leveled off near 60 µg L^{-1}. Phosphorus in the control

waters may have been absorbed by bacteria in the water column or attached to core walls, aerator, etc., and converted to non-reactive phosphorus.

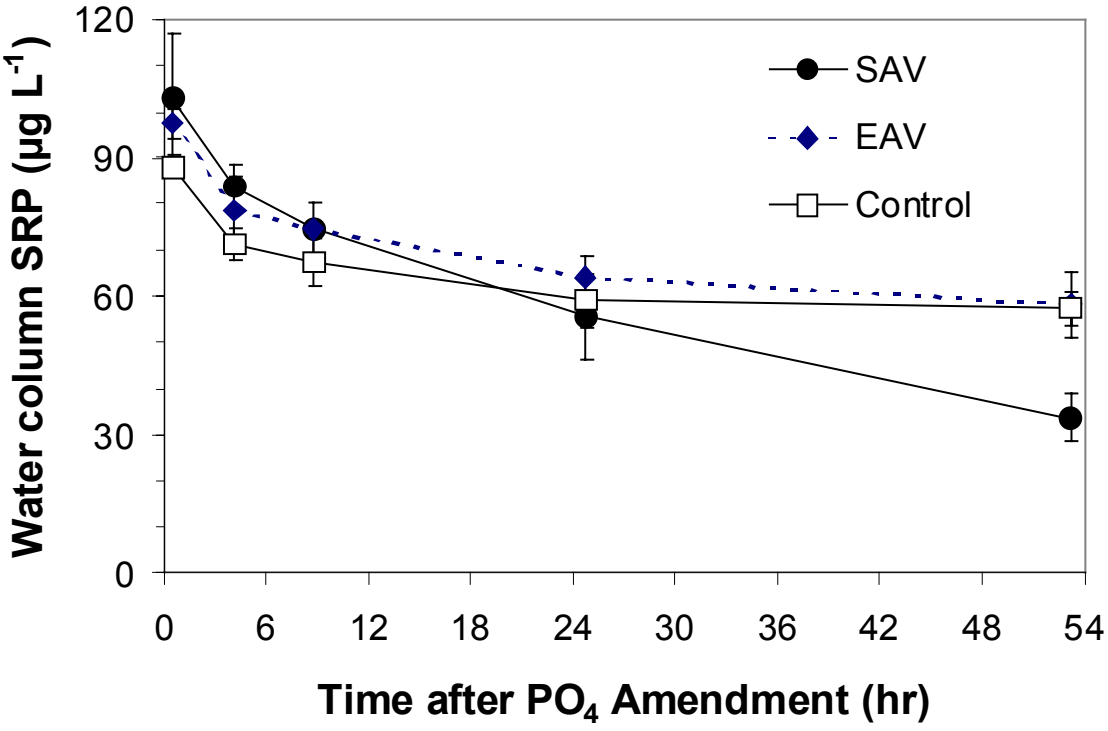

Figure 2-25. Water column SRP concentrations above a "background" concentration as determined before a 100 µg spike was added to each column. Background concentration was assumed stable through the 53-hour period. Triplicate water columns were assembled with or without soils from cattail or SAV communities and aerobically incubated for 30 days prior to the P-spike amendment.

The change in the control water concentration was assumed to be an experimental artifact and was subtracted from the observed concentration for each treatment. The effects of soil type on P flux could then be evaluated directly. In the eight hours following P additions, SAV soils showed greater potential for P uptake than the EAV soils (Table 2-4). The EAV soil cores provided almost no uptake capacity above that observed for the control cores. This may be indicative

of higher P sorption capacity and the influence of $CaCO_3$ in the SAV surfaces soils, compared to EAV soils.

Implications for STA and WCA Management

In both STA-1W and WCA-2A, recently accreted soils were enriched in P relative to deeper soils (Craft and Richardson, 1993; Reddy et al., 1993; Reddy et al., 1998; Koch-Rose et al. 1994; Newman and Pietro, 2001). In this study, SAV soils were enriched relative to soils from EAV areas within STA-1W Cell 1. Surface-soil P enrichment creates a potential for diffusive flux both upward into the water column, as well as downward into underlying soil material. Upward flux can increase outflow water P concentrations and reduce wetland removal efficiency. Downward flux, on the other hand, may be a beneficial process for long-term storage.

SAV soils (0-2 cm layer) contained more P in stable pools (705 ± 68 mg P kg^{-1}) than EAV soils (555 ± 111 mg P kg^{-1}) after eight years of flow-through STA operations. Labile pools of exchangeable and Al- and Fe-bound P were similar in both surface-soil types (84 ± 8 mg P kg^{-1} for SAV; 83 ± 15 mg P kg^{-1} for *Typha*). Because of similar labile-P pools, the two soil types each released P to an oxic water column at similar flux rates, with slightly greater flux rates coming from the SAV soils. SAV soils also reduced P concentrations following water column P amendments, whereas *Typha* soils did not.

Within the submerged community, uptake mechanisms for water column P reduction are mostly associated with soil sorption and leaf surfaces. Leaves are

concentrated towards the top of the canopy to intercept light, and senesce from older, lower portions of the plant. A *Najas* canopy submerged under increasing water depths would isolate water beneath the submerged canopy and close to the soil surface from the overlying water column. This lack of water exchange above the soil-water interface could allow bottom water to reach P concentrations similar to those of the porewater through diffusion.

Removal mechanisms were not effectively maintaining water column concentrations lower than the porewater in SAV beds during the high flow event of June 2002. Net removal of SRP did not occur in the water column within 5 cm of the soil-water interface, which in effect increased the diffusion distance and reduced the overall flux rate. Had the peeper incubation period occurred under conditions of a shallow water column, biological P uptake near the soil-water interface may have resulted in a higher P flux.

Potential diffusion rates appeared to account for only a portion (4-33%) of the flux measured in intact cores. Other studies estimating P flux in wetlands have found diffusive flux rates lower than intact core flux or *in situ* benthic chamber measurements (Fisher and Reddy, 2001). Burrowing macroinvertebrate activity may cause advective water exchange from soils into overlying water. Additionally, uptake of water column P near the soil surface by macrophytes and their epiphytes, phytoplankton and benthic microorganisms is necessary to maintain strong concentration gradients over short diffusion distances. Inorganic

retention of P through adsorption and Fe precipitate formation also influence the distribution of dissolved P along the water column-porewater gradient.

In Everglades soils, soil respiration rate (as CO_2 production) and decomposition of litter/detritus was correlated to soil P content along a nutrient enrichment gradient in the northern Everglades (Davis, 1991; DeBusk and Reddy, 1998; Qualls and Richardson, 2000). Thus increased P mobilization may be expected in P-enriched areas, relative to areas of lower P levels. The rate of organic matter mineralization for soils along a eutrophication gradient in WCA-2A was controlled primarily, however, by the availability of electron acceptors (White and Reddy, 2001), of which oxygen is the most efficient. The nutrient-impacted region of WCA-2A is frequently anoxic due to high sediment oxygen demand and shade-induced limitations of aquatic photosynthesis. Therefore, organic matter mineralization rates may be higher in P-enriched areas dominated by submerged macrophytes that are capable of supplying O_2 to the water column, than in those areas dominated by dense *Typha* stands.

The nutrient-impacted areas of WCA-2A and STA-1W are near monospecific stands of *Typha*. While SAV communities seem to be outcompeted in these inflow regions, they do exist in STA-1W further downstream. Data collected from other porewater studies in STA-1W (DBE, unpublished data) along with the results presented here, suggest diffusive flux rates decrease with distance through the wetland for the SAV communities (Figure 2-26). Such a relationship is useful for predicting internal P loading at various locations along

the treatment gradient. However, the diffusive flux is likely only a fraction of the total mobilization of P from soils to overlying water.

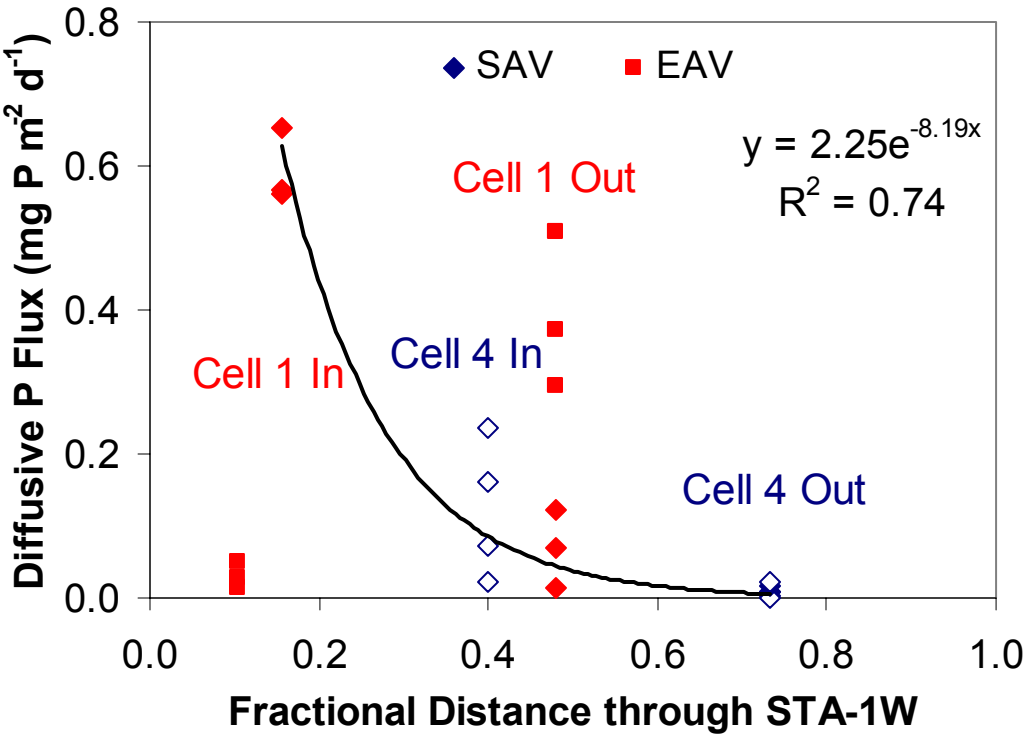

Figure 2-26. Phosphorus diffusive flux estimates based on 22 porewater equilibrators deployed in STA-1W Cell 1 (June-July 2002; closed markers) and Cell 4 (November-December 2001; open markers), as a function of distance through the entire wetland. Diamond markers indicate SAV; square markers indicate EAV. The regression line was calculated using only SAV diffusive flux rates.

Emergent vegetation diffusive P flux did not show similar dependence on fractional distance. This difference may have been due to the influence of root uptake on soil porewater SRP concentrations, which was greater for soils in the outflow *Typha*-dominated region than for soils from dense *Typha* stands near the inflow region. Porewater SRP concentrations in the upper 10 cm of emergent

sediments were much lower in the inflow region (0.18 mg P L^{-1}) than near the Cell 1 outflow (0.99 mg P L^{-1}).

The Cell 1 inflow emergent station may also have been isolated from the primary flow paths through the dense emergent in the inflow region, whereas the outflow station was adjacent to open water. This difference in proximity to flow paths may have subjected the inflow-region *Typha* station to lower P loads than the outflow-region *Typha* station. These data suggest that the internal P load from porewater diffusion depends on vegetation, distance from the inflow, and the proximity to the main flow paths through the wetland.

Conclusions

Managers of treatment wetlands must consider options for reducing internal P loading, which can occur through diffusion and other processes (e.g. resuspension, dissolution), by creating stable soil-P pools. At a minimum, internal P loading should be minimized in outflow regions of the STAs to achieve low TP concentrations in surface water outflows. Options may include maximizing natural processes of new soil humification and Ca-P mineral formation, or enhancing stability through chemical additions. Areas dominated by SAV may provide a benefit during early stages of wetland creation and soil development through water column $CaCO_3$ precipitation, which appeared to increase the Ca-bound P pool in SAV soils, relative to EAV soils. Soils beneath EAV were P-depleted relative to soil P concentrations shortly after flooding, which suggests that pools operationally defined as "more recalcitrant" are still

subject to mobilization. Over time and especially in inflow regions, high rates of soil accrual may necessitate mechanical soil removal to reduce internal P loads, which occurred regardless of vegetation type.

CHAPTER 3
BIOMASS PHOSPHORUS STORAGES AND DYNAMICS

Introduction

Tracts of the northern Everglades were transformed into three water conservation areas (WCAs) in the 1940-50's by surrounding the vast marshes with earthen berms (Figure 3-1). These areas provided water storage and flood control for urban and agricultural development in South Florida. To the northwest of the WCAs lies the Everglades Agricultural Area (EAA), a 200,000 ha region of drained Everglades soils in sugarcane and winter vegetable production.

WCA-2A has received agricultural drainage water (ADW) discharges since 1955 (Bartow et al., 1996). Excessive phosphorus (P) loading from these discharges has been identified as the primary cause for an observed eutrophication gradient. Changes observed near the discharge structures include increased water column, soil, and plant tissue P concentrations, and change in ecosystem function, relative to the interior marsh (Craft and Richardson, 1993; DeBusk et al., 1994; Reddy et al., 1993; Reddy et al., 1998).

In order to reduce P loads to the Everglades, stormwater treatment areas (STAs) were constructed to intercept P in EAA drainage waters before it enters the WCA marsh. The STAs have performed well since construction, reducing TP concentrations to below 50 $\mu g\ L^{-1}$ as required by the Everglades Forever Act

(EFA) of 1994. The EFA mandates further P load reductions, requiring TP concentrations of 10 μg L^{-1} for discharges into the WCAs.

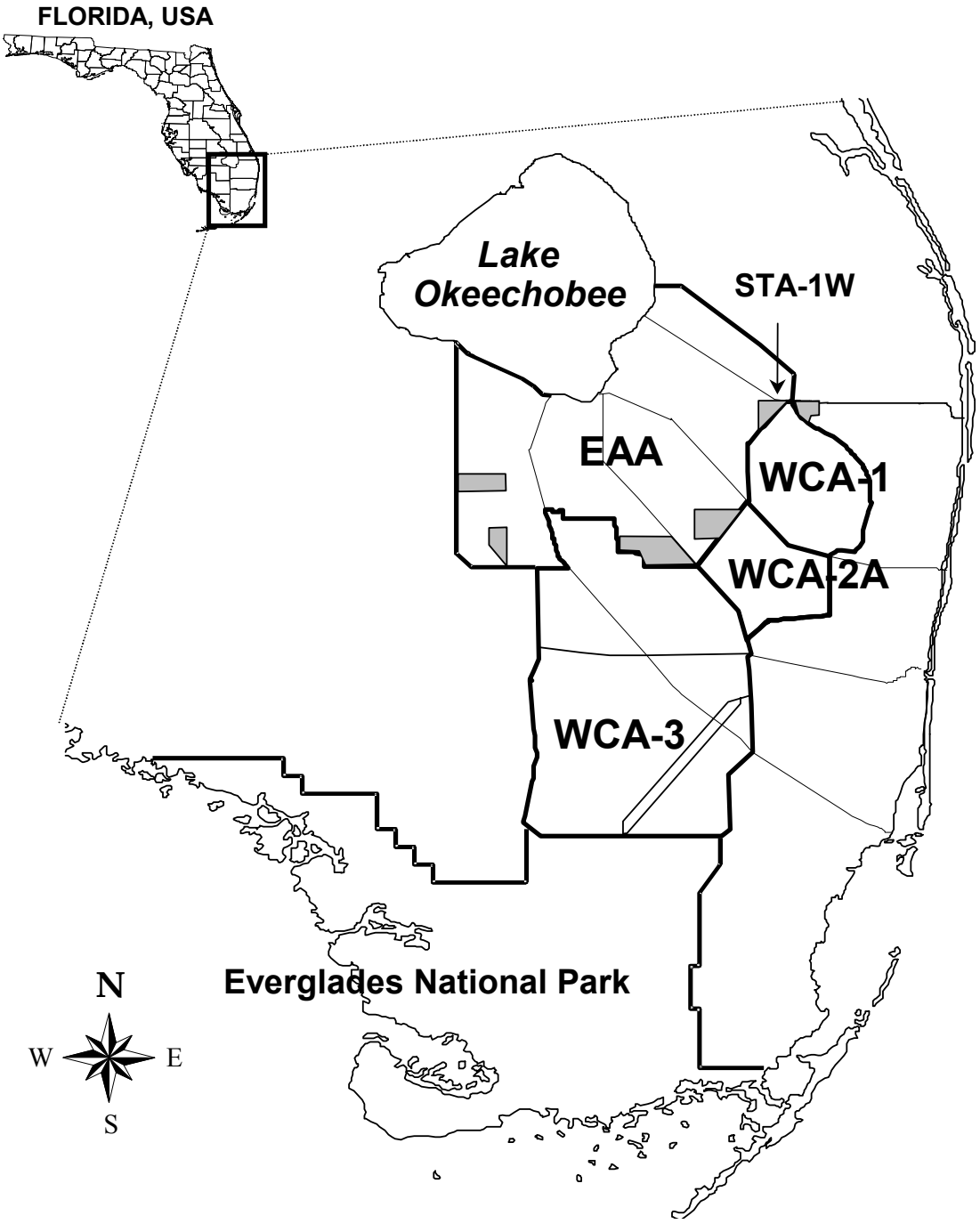

Figure 3-1. The historic Everglades region of south Florida is now three distinct parcels, including the Everglades Agricultural Area (EAA), Water Conservation Areas (WCA) and Everglades National Park. Stormwater treatment areas (STA) are shown (in gray), including STA-1W.

Macrophytes influence P removal performance in treatment wetlands through direct P uptake as well as by influencing the physicochemical characteristics of its environment. *Typha domingensis* can grow in dense stands when nutrient and hydrologic conditions allow. Phosphorus enrichment in the northern Everglades has increased *Typha* above-ground biomass and shoot density, and slough communities of WCA-2A were replaced by *Typha* near inflow structures (Wu et al., 1997; Miao et al., 2000). Dense *Typha* stands and open water areas differ in rates of water exchange and community metabolism (Belanger et al., 1989; Brix, 1997; McCormick et al., 1997). In WCA-2A, *Typha* canopy shading of the water column has reduced aquatic photosynthesis and wind-induced mixing, leading to reduced oxygen supply relative to nearby open water areas (Belanger et al.,1989; Grimshaw et al., 1997). Dense stands of macrophytes can have large accumulations of organic materials, either settled from flowing water or produced on-site. These accumulations increase heterotrophic oxygen demand which, coupled with canopy shading, can result in anoxic conditions (Belanger et al., 1989; McCormick et al., 1997).

Also important to P dynamics in wetlands are microbial processes in the soil and water column, which respond quickly to system inputs or environmental change. Qualls and Richardson (2000) reported *Typha* litter in WCA-2A flumes sequestered SRP up to 10 times the original litter content over a one-year period, with little net change to the macrophyte P content. Microbial biomass on the leaf litter increased in P content, yet the factors controlling

microbial P uptake and release from wetland biomass remain unidentified. Specifically, the effect of community metabolism on biomass P uptake within *Typha* leaf litter is unknown.

As an open water community transitions towards a dense macrophyte stand, such as occurred in WCA-2A, P cycling and removal processes within the water column are likely affected. A similar increase in *Typha* stand density likely occurs within the STAs due to high rates of nutrient loading, yet the effects of water column shading by *Typha* on P cycling process remains unknown. Through field observations, mesocosm experiments, and controlled laboratory incubations, P storage, uptake and release were examined from *Typha* tissues as well as from microbial populations associated with *Typha* leaf litter. Specific objectives included:

- Estimating the relative partitioning of P between live and dead components of *Typha* stands (Mesocosm Phosphorus Storage)
- Investigating the role of leaf litter accumulations on P release from the soil (Flux Study Using Intact Soil Cores)
- Quantifying the rate of P uptake from the water column by microbial populations associated with *Typha* litter under oxygenated and anoxic conditions (Litter Incubations)

Materials and Methods

Bench-scale studies took place at the Wetland Biogeochemistry Laboratory at the University of Florida in Gainesville, FL. Outdoor mesocosms were located next to the STA-1W inflow canal on an experimental platform provided by DB Environmental. Field investigations took place in STA-1W Cell 1.

STA-1W Site Description

Agricultural runoff from the Everglades Agricultural Area (EAA) Basin S-5 (Figure 3-1) is pumped in canals to STA-1W, a full-scale (2699 ha) treatment wetland operated by SFWMD to reduce P loadings to the Everglades (SFWMD, 2003). Everglades muck soils were drained for agricultural production decades ago, and were reflooded in 1994. The wetland contains three flowpaths of two cells each (Figure 3-2). Cell 1 is the first cell of the eastern flowpath, and contains emergent vegetation (*Typha* spp.) in the inflow region and along the eastern edge (Figure 3-3). A mixture of emergent, floating, and submerged vegetation occupy the downstream reaches of the cell. This distribution of community types has remained relatively constant in Cell 1 since the wetland was first flooded (Newman and Pietro, 2001). During eight years of flow-through operations, the wetland has accrued new wetland soils.

Mesocosm Phosphorus Storage

Two outdoor mesocosms (2.2m L x 0.79m W x 1.0m D) located near STA-1W received STA-inflow water for 2.7 years at a hydraulic loading rate of 10 cm day^{-1}, as part of another study (DBEL, 2001). Muck soils in these systems were inoculated with *Typha* collected from within STA-1W.

The vegetation and soils in the two cattail mesocosms were sampled destructively in late August and early September 2001, at the end of the water quality monitoring period (December 29, 1998 through August 14, 2001). All vegetation was collected from each mesocosm and segregated into inflow and

outflow regions. Cattail shoots were cut at the soil surface, and separated into live (green) and dead shoots. Triplicate soil cores (38.5 cm^2 each) were retrieved from the inflow and outflow regions. The underlying muck soil was discarded.

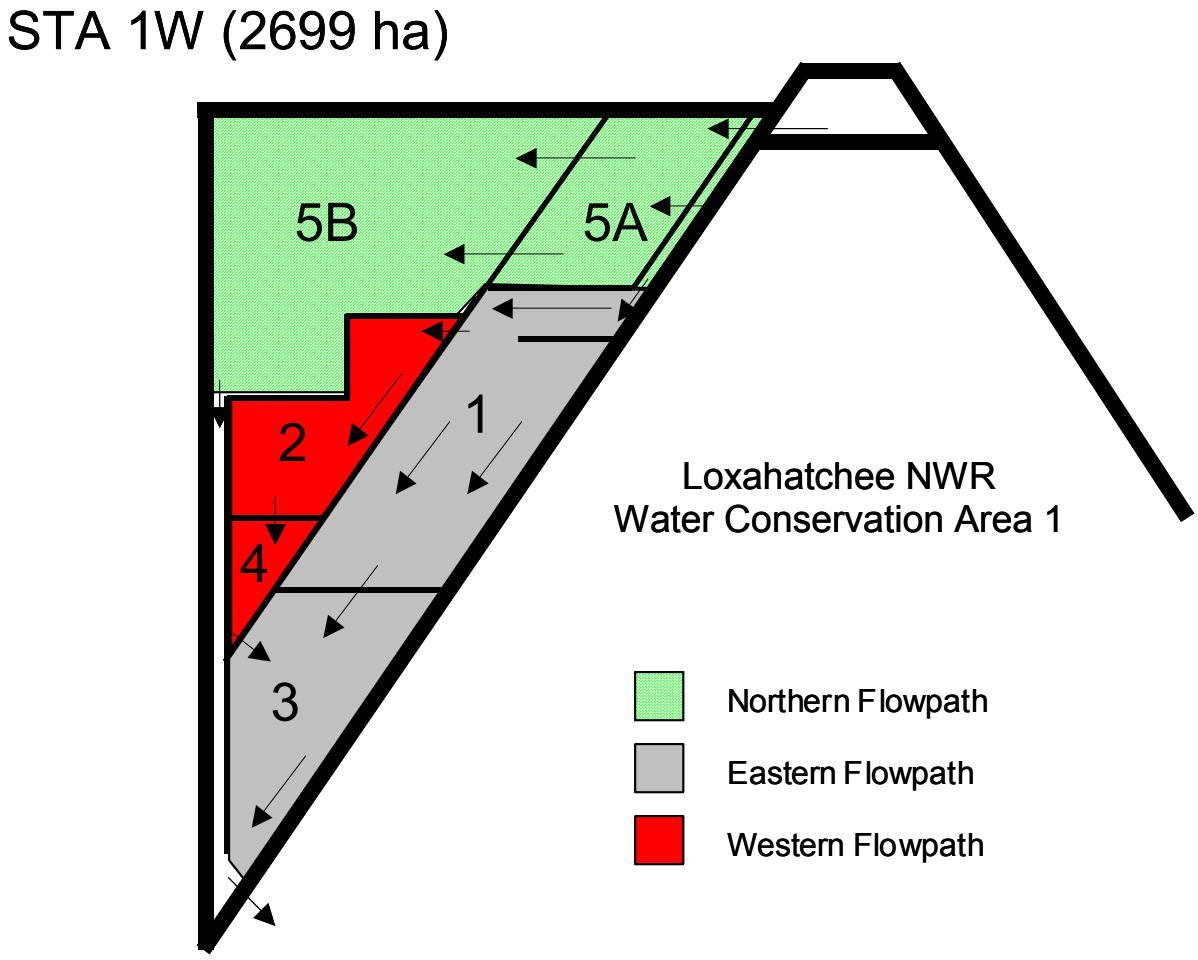

Figure 3-2. Stormwater Treatment Area 1 W in Palm Beach county, Florida. Three flowpaths receive surface water drained from adjacent agricultural soils and reduce the phosphorus load to Water Conservation Area 2A. Cells 1, 2 and 3 are comprised of mixed emergent, submerged and floating vegetation, while Cells 4, 5A, and 5B are primarily submerged and floating vegetation. Arrows indicate general direction of flow.

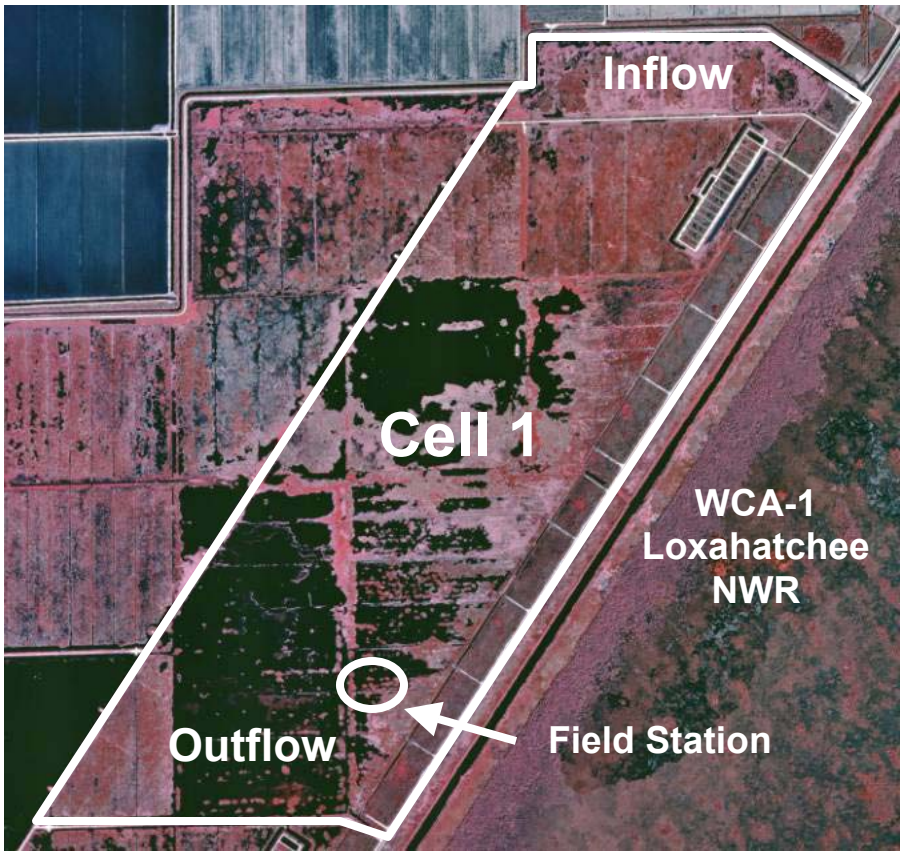

Figure 3-3. Aerial photograph of STA-1W Cell 1, first cell of the eastern flowpath, in November 2000 (courtesy SFWMD). Also shown is the location of the emergent vegetation field station.

Belowground cattail biomass and submerged macrophyte tissues were collected after soil sampling was complete. Wet weights for vegetation samples were recorded in the field. Bulk density was determined for accrued soils. Vegetation and soil samples were oven dried (65°C), weighed, and homogenized for analysis. TP content was determined by digesting 50 mg sample in concentrated nitric acid, followed by perchloric acid digestion at 210 °C (COE 3-227, Plumb 1981).

Phosphorus mass removals were calculated for each mesocosm based on inflow and outflow TP concentrations of weekly or bi-weekly grab samples and a constant hydraulic loading rate of 10 cm day^{-1} (DBEL, 2001). The P mass recovered in accrued soil, together with the change in biomass P storage, was compared to mass removal based on water column concentration reductions.

Plant Tissue Desiccation Study

Biomass components within the water column of a treatment wetland are largely comprised of live SAV in submerged macrophyte beds, and both live and dead shoots in emergent stands. Samples of fresh *Najas* tissue and dead *Typha* shoots (leaf litter) were collected from the outflow region of Cell 1 on May 29, 2002. Triplicate vertical acrylic columns (7 cm i.d.) received 13 g tissue (wet wt.), along with 0.75 L of reflood water collected from the STA-1W Cell 4 outflow weir. A pair of control columns contained no plant tissue. All treatments and controls were placed in a water bath, continuously-aerated, and covered with an opaque shroud. Water samples were withdrawn at times 0, 1 and 9 days, and filtered (0.45 μm). SRP and acidified TSP samples were analyzed colorimetrically (potassium antimony tartrate, sulfuric acid, ammonium molybdate, ascorbic acid) on a Spectronics Genesys 5 spectrophotometer (EPA 365.2; EPA 1979). Analysis for TSP was preceded by persulfate acid digestion and neutralization.

Flux Study Using Intact Soil Cores With and Without *Typha* Litter

Six replicate intact soil cores were retrieved from a *Typha* stand (26.6292°N, 80.4219 °W) in the outflow region of STA-1W Cell 1 on July 4, 2002.

Each core was retrieved by inserting an acrylic core tube (38.5 cm^2) through the newly accrued soil layer into the underlying native farm muck to a minimum depth of 10 cm. The top end of the core was sealed with a #13½ rubber stopper prior to extraction. Each of the six intact cores was completely filled with site water and sealed with a rubber stopper. An opaque shroud minimized solar heating and blocked light during transport to the lab.

Typha leaf litter was collected from STA-1W Cell 1 at the time of soil core collection, and was kept in site water on ice for transport to the lab. Litter consisted of intact shoots that had little to no visible damage (from grazers, breakage, etc.), yet had become neutrally- or negatively buoyant. This phase of shoot decomposition was chosen because of three attributes:

- Fresh live shoots would likely leach tissue P rapidly after being collected from the source plant
- Ample time in the water column for colonization by aquatic microorganisms
- Negative buoyancy facilitates the experimental incubation design

Each shoot was divided into uniform lengths of ~5 cm. Pieces from all shoots were mixed at random, then added to six cores, three with soil and three with reflood water only (litter controls). Overlying water was replaced with 1.15 L filtered (0.45 µm) STA-1W Cell 4 outflow water (15 µg TSP L^{-1}). The 30 cm water column was aerated and incubated in a dark water bath at ~22°C for 28 days.

Water samples (30 mL) were withdrawn at Δt = 0.1, 0.5, 1, 1.5, 2, 4, 6, 10, 14, and 20 days, and analyzed for SRP and pH. Representative samples were also analyzed for TSP, dissolved calcium, total alkalinity, and dissolved organic carbon (DOC).

Dissolved calcium was determined using flame atomic absoption spectroscopy (EPA 215.1; EPA 1979) on a Perkin-Elmer 3110. Alkalinity was titrated with 0.02N H_2SO_4 (EPA 310.1; EPA 1979). Dissolved organic carbon was on acidified, filtered (0.45 μm) samples, and measured with a Shimadzu TOC-5050A (Duisburg, Germany) TOC analyzer equipped with an ASI-5000A autosampler (5310-A; APHA, 1992).

Sample pH was recorded immediately following collection, using a 3 in 1 gel filled combination pH electrode and Corning 313 pH meter. Water bath temperature was continuously recorded by a StowAway Tidbit® logging probe (Onset Computer; Bourne, MA) as well as monitored periodically with a thermometer.

After 30 days, an amendment of 100 μg P L^{-1} (as KH_2PO_4) was added to each core. The water volumes above each core differed slightly (± 5 mL) from the original water volume of 1.15 L added one month prior, likely due to different evaporation rates induced by the aerators. These differences were recorded but volumes were not adjusted at that time. Water samples (30 mL) were withdrawn Δt = 0, 4, 8, 24 and 53 hours after the amendment, and analyzed for SRP. Each core received 30 mL of P-unamended reflood water after sampling.

Typha Litter Incubations

Through a series of oxic and anoxic incubations, P uptake and release rates were determined for freshly submerged *Typha* litter and associated microbial biomass. Senescent submerged plant tissue from a stand of *Typha* ~30 m in diameter (25° 50.275' N; 80° 42.991' W) were collected in southern WCA-3A, and kept in site water on ice for transport to the lab. Directly south of WCA-2 and the EAA, west of urban Miami and north of Everglades National Park (Figure 3-1), WCA-3A is a mosaic of pristine Everglades ecotypes, from wet sloughs dominated by *Nymphaea* spp. and *Utricularia* spp. to sawgrass (*Cladium*) ridge and tree island communities. The slough surrounding the *Typha* stand was dominated by *Nymphae oderata*, has historically low water TP and SRP levels of 9 ± 2 μg L^{-1} and <2 μg L^{-1}, respectively (Swift and Nicholas, 1987), and was the source of water for the incubations.

Part I. P uptake by *Typha* litter

Incubation water was filtered (0.45 μm) and amended with KH_2PO_4 to concentrations of 0, 10, 30, 100 and 300 and 1000 μg P L^{-1} above background SRP concentrations. This concentration range was representative of the P gradient to which litter is exposed: low-P surface water to high-P porewater.

Short acrylic columns received 25 g litter (wet wt.) and 0.50 L P-amended water (Figure 3-4). Cores were sealed with rubber stoppers at each end. The top stopper was penetrated by two glass tubes which permitted sparging with air for oxic trials, or N_2 + 0.03% CO_2 for anoxic trials. Gas flow mixed the water column

during sparge (0.5 - 3hrs), after which gas lines were clamped shut. Water samples were withdrawn after 0.3, 1, 2, 3, and 5 days of incubation, and analyzed for SRP, pH and DOC.

Dissolved oxygen concentrations were monitored periodically during sampling, and measured for each replicate at the end of the incubation to ensure experimental conditions were achieved.

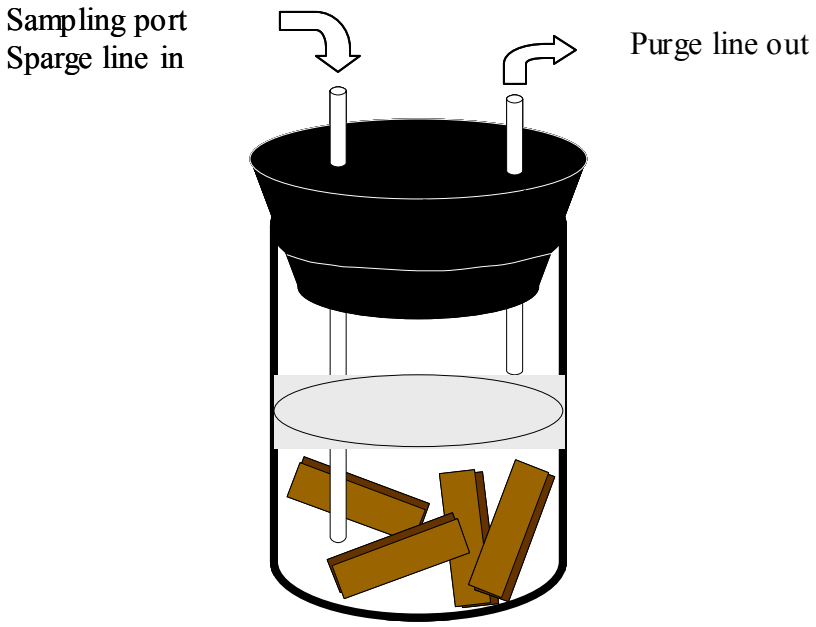

Figure 3-4. Schematic of incubation design, containing *Typha* litter and P-amended surface waters from WCA 3A. Not to scale: Water volume was 0.5 L, head space was ~ 0.05L.

Part II. P release from enriched litter

After seven days of incubation, the incubation waters for each litter sample were replaced by un-amended site water. Water column SRP concentrations were measured immediately ($\Delta t=0$), and at 1, 2, 3, and 5 days after the water exchange. Water SRP concentrations were used to calculate oxic and

anoxic P uptake and release rates for *Typha* litter across the range of initial water P concentrations.

Part III. Microbial Biomass Phosphorus

At the conclusion of the release phase, *Typha* litter subsamples of ~10 g wet weight were subjected to either 24-hour $CHCl_3$ fumigation and 16 hour 0.5 M $NaHCO_3$ (pH 8.5) extraction on a reciprocating shaker, followed by centrifugation (10 min, 6000 rpm), or direct extraction without fumigation. Additional P extracted after fumigation was considered the contribution of the microbial biomass phosphorus (MBP) pool that was present and susceptible to chloroform (Hedley and Stewart, 1982). *Typha* tissue samples were analyzed in their original condition (preserved field-moist in the dark at 3.5°C) for comparison.

Extracts were analyzed for SRP and TP using the ascorbic acid-molybdenum blue method (EPA 365.2; EPA 1979) on a Technicon Autoanalyzer. TP samples were prepared by persulfate digestion at 150°C, increasing to 380°C, prior to analysis.

Statistical Methods

Statistical analyses on experimental data were performed using MSExcel® (v. 2000 ©Microsoft Corp.) ANOVA and t-test macros. Error around mean values is presented as ± one standard deviation of replicate measurements.

Results and Discussion

Mesocosm Phosphorus Storages

The following narrative describes observed changes to vegetation in the mesocosms, though no quantitative measurements of biomass occurred prior to August 2001. In February of 1999, the surface of the water in rep 1 was covered in *Lemna*, which likely reduced light penetration and gas exchange to the water column. *Lemna* persisted throughout 1999 in rep 1, with none noted in rep 2. Dense mats of floating *Lemna minor* were shown by Ngo (1987) to inhibit phytoplankton growth by shading the water column, causing algae to die and settle out. Water column chemistry can also be affected below dense mats of floating macrophytes. Low pH and oxygen levels can develop, and increased nutrient levels may result from internal loading (Gopal et al., 1984).

Greater *Najas* biomass was observed in rep 2 than rep 1 in December 1999. Through the first year of operation, rep 2 appeared to outperform rep 1 with respect to water column P reduction (Figure 3-5). From December 29, 1998 through December 21, 1999, mean inflow TP concentration (106 $\mu g\ L^{-1}$) was reduced to mean outflow concentrations of 70 and 33 $\mu g\ L^{-1}$ for rep 1 and rep 2, respectively.

Lemna may have created anoxic conditions and internal P loading from the initial muck substrate, while *Najas* in rep 2 may have provided oxygenated water column conditions which can minimize soil-P release (Gonsiorczyk et al., 2001). By September 2000, rep 1 appeared to have more *Najas*, while rep 2 had

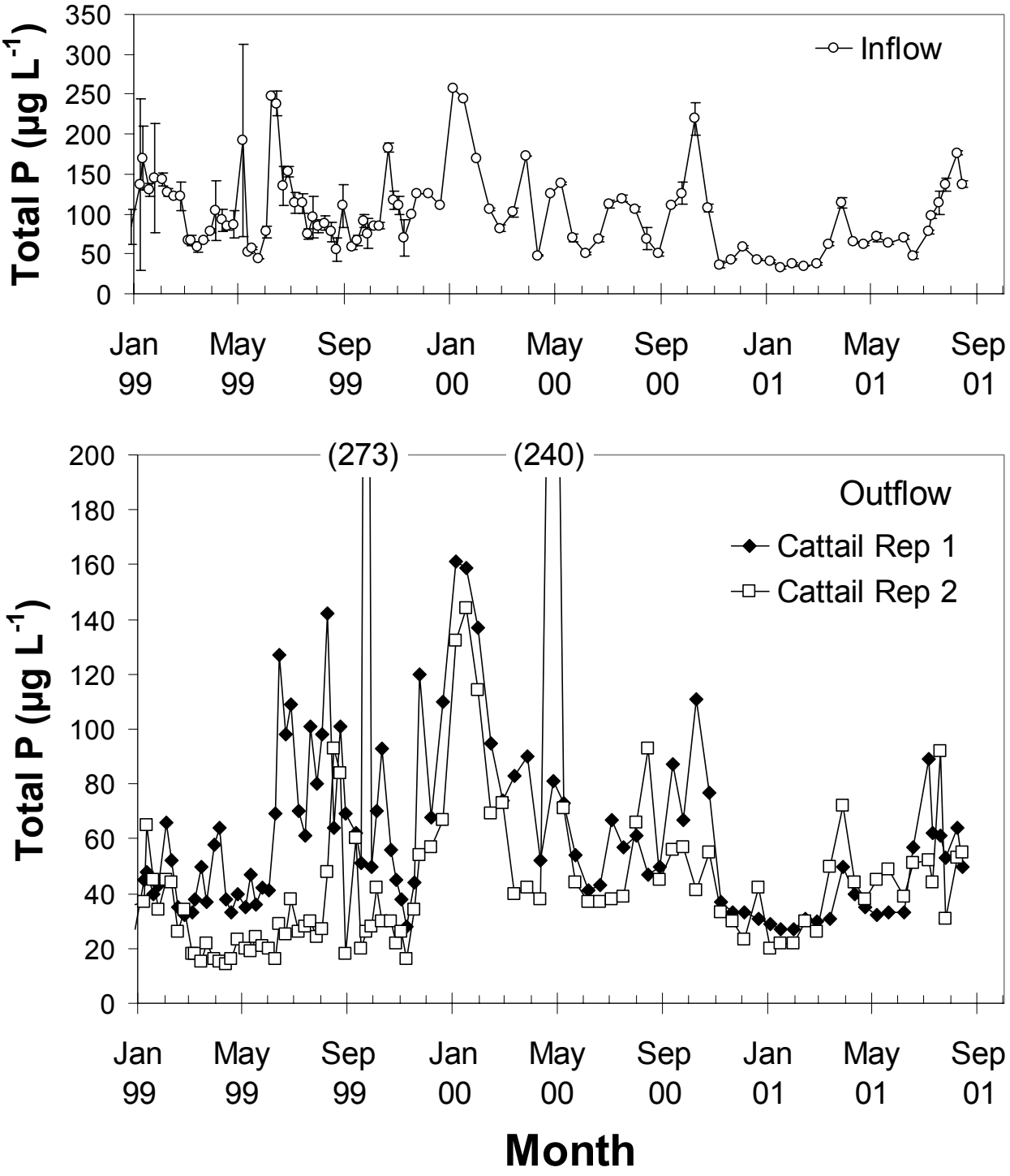

Figure 3-5. Inflow and outflow TP concentrations for mesocosms dominated by *Typha* and operated from December 1998 through August 2001.

developed an underwater periphyton mat. At the time of the whole-mesocosm biomass sampling in August 2001, no periphyton was found in either mesocosm, and rep 1 contained more *Najas* (289 g dry wt.) than rep 2 (12.4 g dry wt.). Mean surface water TP concentration reductions were greater for the latter mesocosm, which had a more dense *Typha* canopy (rep 2), though variability in vegetation and P removal performance occurred throughout the period of record (Figure 3-5). It is not clear whether fluctuations in biomass and species composition of SAV, floating plants, and periphyton within and between the two cattail mesocosms contributed to variability in P removal performance.

Based on mean inflow and outflow water TP concentrations, the two cattail mesocosms removed 1.2 (rep 1) and 2.0 (rep 2) g P m^{-2} yr^{-1} of an average annual loading rate of 3.5 g P m^{-2} yr^{-1}. Phosphorus removal resulted in an increased biomass P pool and accrued soils. Of the total P mass recovered in biomass and new soils, 66 (rep 1) and 96% (rep 2) was attributed to TP removal from the water column. Unfortunately, the change in P in the initial muck substrate was not quantified for the period of record. The difference between water column P mass removal and P recovered in biomass and new-soil P reflected experimental error inherent in sampling, but P mobilization from the initial muck substrate (Table 3-1) was also likely. The substrate in one mesocosm, therefore, may have had only a minor net P contribution (0.2 g P m^{-2}), while the other mesocosm substrate provided nearly a third (1.5 g P m^{-2}) of the P mass accrued in biomass and new soil storages.

Typha P concentrations for live tissues, dead leaves and root/rhizomes were comparable to those reported for the Everglades (e.g. Toth, 1988) (Table 3-2). Belowground cattail tissues (roots+rhizomes) comprised 39% and 65% of the total recovered biomass P in rep 1 and rep 2, respectively (Figure 3-6). Previous Everglades studies estimate 30-40% of total *Typha* biomass P was associated with belowground tissues in P-enriched areas, while 60% was belowground in low-P environments (Toth, 1988; Miao and Sklar, 1998).

The disparity in the below-ground cattail P storage between the replicates in this study was echoed by the above-ground standing crop (Figure 3-6). Greater biomass P storage observed in replicate 2 agreed with greater reductions between inflow and outflow water TP concentrations over the 2.7-year study period.

Table 3-1. Phosphorus removed from the water column over the 2.7-year study period and recovered in the vegetation (*Typha* + *Najas*) and soils upon termination of the study on August 20, 2001.

	Rep 1	**Rep 2**	**Average**
Biomass Storage, g P m^{-2}	1.2	1.8	1.5 (29%)
New Soil Storage, g P m^{-2}	3.6	3.9	3.7 (71%)
Total Recovered P, g P m^{-2}	4.9	5.6	5.2 (100%)
Inflow-Outflow Water Mass P Removed, g P m^{-2}	3.2	5.4	4.3
Potential net P contribution from muck substrate into biomass and new soils, g P m^{-2}	1.6	0.2	0.9

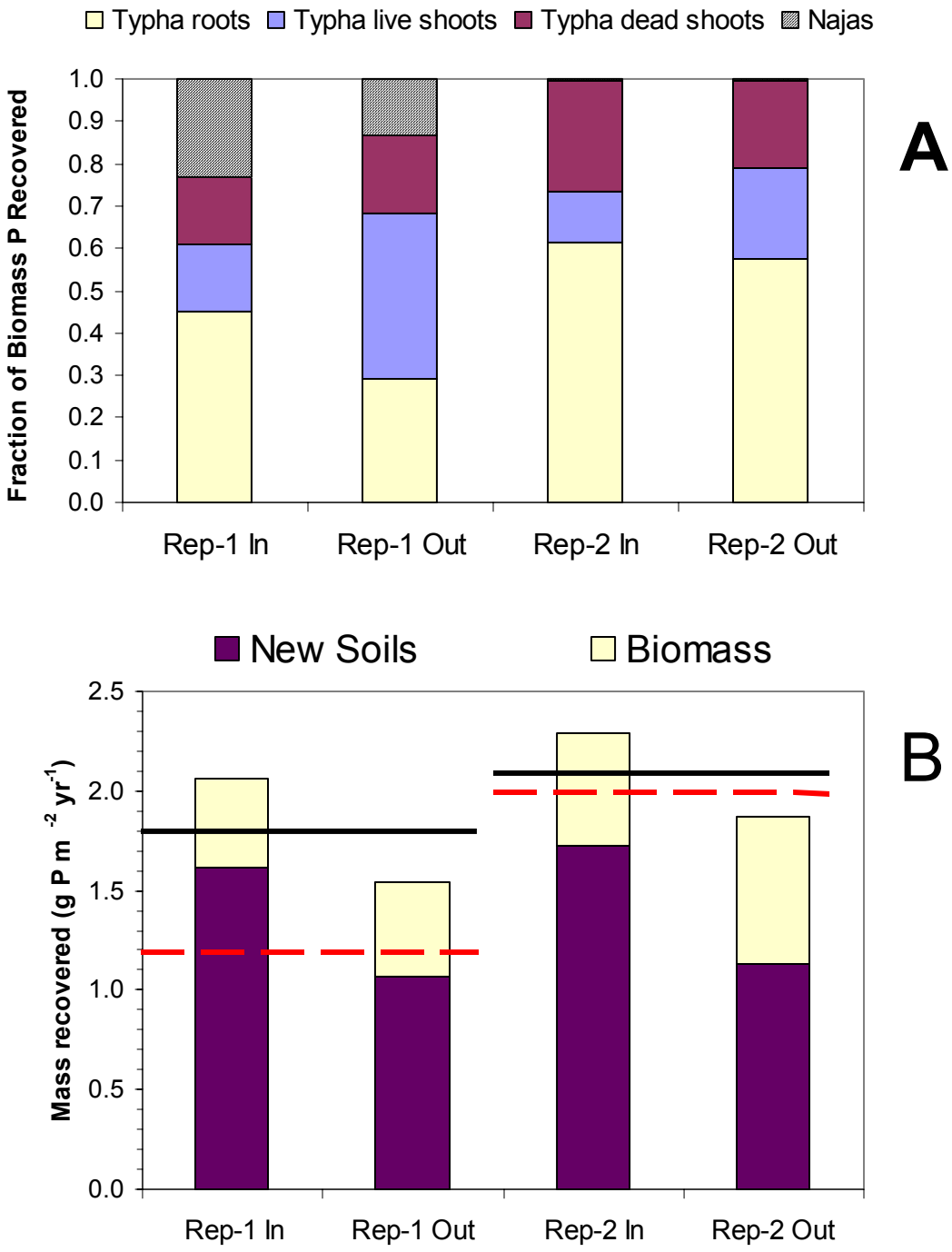

Figure 3-6. A: Partitioning of recovered *Najas* and *Typha* tissue P within the inflow (In) and outflow (Out) halves of two mesocosms operated from December 1998 through August 2001. B: Total P mass recovered (solid line) in biomass and sediments relative to annual mass removal rates (dashed line) calculated from water column TP concentration reductions.

Table 3-2. Dry matter and P concentrations in accrued soil and tissue storages (live and dead *Typha* shoots, below-ground *Typha* (roots and rhizomes), and *Najas* tissues) retrieved from mesocosms on August 20, 2001. *Typha* tissue P concentrations are comparable to values reported by Toth (1988).

	Live *Typha* shoots	**Dead *Typha* shoots**	**Below-ground *Typha***	**All *Najas* Tissues**	**Newly accrued soil**
			(g dry m^{-2})		
Rep 1	416	1163	745	41	3932
Rep 2	466	2141	1616	1.8	5426
			(mg P kg^{-1})		
Rep 1	790	180	620	5300	924
Rep 2	710	190	630	3300	747
Toth 1988	580-1000	160-260	570-740		

High biomass in rep 2 caused the belowground portions of the *Typha* stand to become space limited, and root structures extended upwards into the water column. These structures may have provided a pathway for direct P uptake from the water column, more so in rep 2 than in the less dense rep 1, since greater P reduction was observed over the 2.7-year period of record in the mesocosm with greater cattail density and less SAV biomass. Root and rhizome biomass in rep 2 (1.6 kg m^{-2}) was more than double that of rep 1 (0.75 kg m^{-2}), but root TP concentrations were similar (Table 3-2).

Additionally, rep 2 had twice the dead *Typha* biomass (2.1 kg m^{-2}) that was present in rep 1 (1.2 kg m^{-2}). Dead biomass in rep 2 may have provided substrate for a larger microbial community than rep 1, and increased P removal from the water column in that mesocosm. Live above-ground *Typha* biomass in this study was similar in the two mesocosms (0.42 and 0.47 kg m^{-2}, for rep 1 and rep 2

respectively), and with similar TP contents (Table 3-2), the two had similar P mass stored in that compartment.

Substantial fine-grained organic soil (not fibrous cattail detritus) was produced in the 2.7-year study period. Total P content of the accrued soils was more uniform within rep 1 (inflow, 922 mg kg^{-1}; and outflow, 926 mg kg^{-1}) than rep -2 (inflow, 640 mg kg^{-1}; outflow, 854 mg kg^{-1}).

When P associated with the dead *Typha* leaves and accrued soil is compared to P in live plant biomass (*Najas* + live *Typha* leaves, roots), nearly four-fold greater P mass was sequestered into "dead storage" (4.05 g P m^{-2}) than into "live storage" (1.19 g P m^{-2}). If it is assumed that P found in the soils which accrued during the period of record resulted from the live biomass (SAV tissues, epiphytes, and live *Typha* leaves),

$$\text{Net turnover rate for Live Biomass P, yr}^{-1} = \frac{(\text{Soil, g P})}{(\text{Live Biomass, g P})(\text{POR, years})}$$

then a turn-over rate of 4 yr^{-1} for the live macrophyte P storage is approximated. This calculation is subject to several errors (e.g. (it ignores direct sedimentation and P adsorption by the soil matrix), but approximates the rate of P movement from biomass to soil storages.

In a full-scale wetland, mass balance of P into biomass would be more complex. Non-uniform soil accretion can occur when a high flow event scours the soil or introduces suspended solids. Macrophyte tissue senescence occurs inconsistently throughout the year. Cattails in temperate wetlands may senesce

annually (Kadlec and Knight, 1996), but the mild subtropical climate of South Florida does not necessarily cause *Typha* die-back in the winter months.

Tissue Desiccation Study

Emergent plant tissue has high proportions of structural (lignin and cellulose) tissues in order to support the aboveground portions of the plant (Debusk and Reddy 1998). Submerged vegetation, in contrast, relies on buoyancy within the water column for vertical support. Lignin and cellulose components of macrophyte tissues are slowly decomposed in wetlands (Godshalk and Wetzel, 1978), which results in slower decomposition of *Typha* tissues than SAV tissues. *Typha* tissues persisted in litter bags for several years in WCA 2A (Davis, 1991), with approximately 50% weight remaining after two years. In contrast, Dierberg (1993) found only 9-14% of the original mass of the submerged macrophytes *Hydrilla verticillata* Royle and *Vallisneria americana* Michx. remained after three weeks in the Lake Okeechobee littoral zone following a decline in lake level. Tissues in that study, however, desiccated when exposed to the air, and decomposition in the water column may be less rapid or incomplete, allowing the SAV fragment to regrow.

Upon senescence, macrophyte tissue (litter) undergoes a series of P exchanges before incorporated into the soil. Submerged surfaces become sites for epiphyte attachment. Microbial "attack" on the leached litterfall acts to enrich litter P content, as organisms sequester soluble P from the surrounding water to maintain suitable ratios of C, N, and P. As the available carbon and P sources are

depleted, recalcitrant organic-P is partially mineralized by extracellular enzyme production in the low-P environment. Over time (several months to years) an apparent increase in litter P content may also be due to losses of other elements (e.g., carbon to CO_2 through respiration, N through denitrification) relative to P within decomposing litter (Davis, 1991). In the case of *Typha*, where senescent tissue was used for incubation, labile phosphorus would have been translocated into the plant base during senescence or was rapidly leached into the surrounding water prior to collection (Christiansen et al., 1985, Davis, 1991).

Initial floodwaters of the desiccation study were low in bioavailable SRP (3 µg L^{-1}), and contained 24 µg TP L^{-1} (Table 3-3). Soluble reactive phosphorus in the floodwater in the *Najas* treatment increased markedly above initial levels over the nine-day incubation, but not in either the *Typha* incubations or the control waters (Figure 3-7). Because of the variability in *Najas* SRP release, however, differences in SRP between treatments were not significant ($p > 0.05$) after either 1 or 9 days. DOP concentrations in floodwater from the *Najas*

Table 3-3. Mean (± 1 s.d.) water quality parameter values measured in the Cell 4 outflow water used in the preliminary desiccation experiment.

Temperature	**pH**	**TP**	**SRP**	**DOP**	**PP**
° C		µg L^{-1}	µg L^{-1}	µg L^{-1}	µg L^{-1}
28.0	8.73	24	3	11	10

treatment increased significantly ($p < 0.05$) above control water concentrations after both 1 and 9 days, from 11 to 29 µg L^{-1} over the nine days, while floodwater

DOP from the *Typha* treatment (17 µg L^{-1}) was not significantly higher than control concentrations (14 µg L^{-1}) or significantly different from *Najas* treatment ($p > 0.05$).

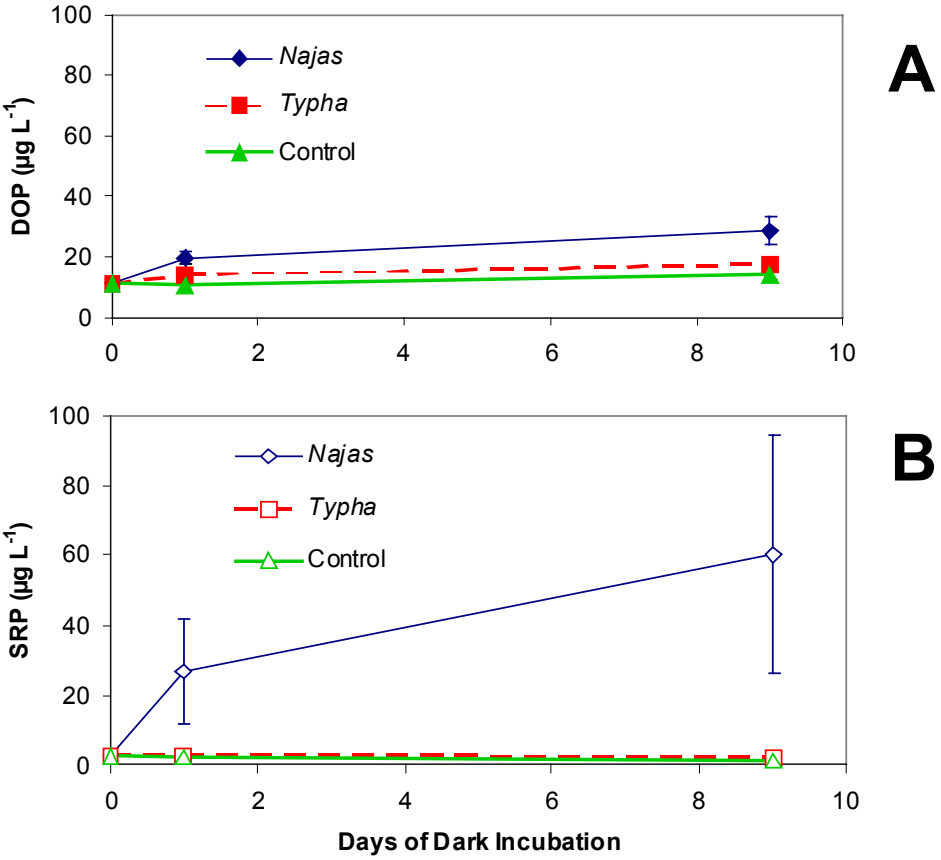

Figure 3-7. A: Dissolved organic phosphorus (DOP) and B: soluble reactive phosphorus (SRP) concentrations in floodwaters containing fresh *Najas* tissues and *Typha* litter, compared to control waters containing no plant tissues, during a nine day dark incubation. Error bars indicate ± one standard deviation from triplicate (experimental) or duplicate (control) cores.

Dierberg (1993) reported that the majority of tissue-P was rapidly (days) released from desiccating SAV exposed by lake level draw-down. The released P was largely bioavailable. In this study, the soluble P released from SAV over nine

days of dark conditions was 77% SRP. Vascular aquatic plants translocate some nutrients, including P, from senescing older tissues to new leaves to avoid such losses (Christiansen et al., 1985).

Najas tissue became chlorotic and fragmented during the nine-day incubation, while *Typha* litter remained intact. Light appeared to be important in maintaining P content in SAV, and the plant may be susceptible to soluble P release during short-duration turbid events which inhibit light penetration into the water column.

Phosphorus stored in submerged dead *Typha* tissues was not sensitive to dark conditions, as the tissues did not release SRP or DOP to the water column. Since the *Najas* community produces little discernible "leaf litter," and fresh *Najas* tissue rapidly senesced in dark lab conditions, only the effects of *Typha* litter on P flux were explored with intact soil cores.

Flux Study using Intact Soil Cores

Soils collected from beneath the *Typha* community in STA-1W Cell 1 outflow region released P into overlying water under dark, aerobic conditions (Figure 3-8). Initial floodwater was particle free (filtered 0.45 μm), and contained 13 μg DOP L^{-1} and 3 μg SRP L^{-1}. Of the three cattail soil cores without litter amendments, only one replicate had visible fragments of leaf litter within the surface soils. The results observed from this core were more similar to the litter amended treatments than the replicate treatments without litter, and therefore were excluded from this discussion.

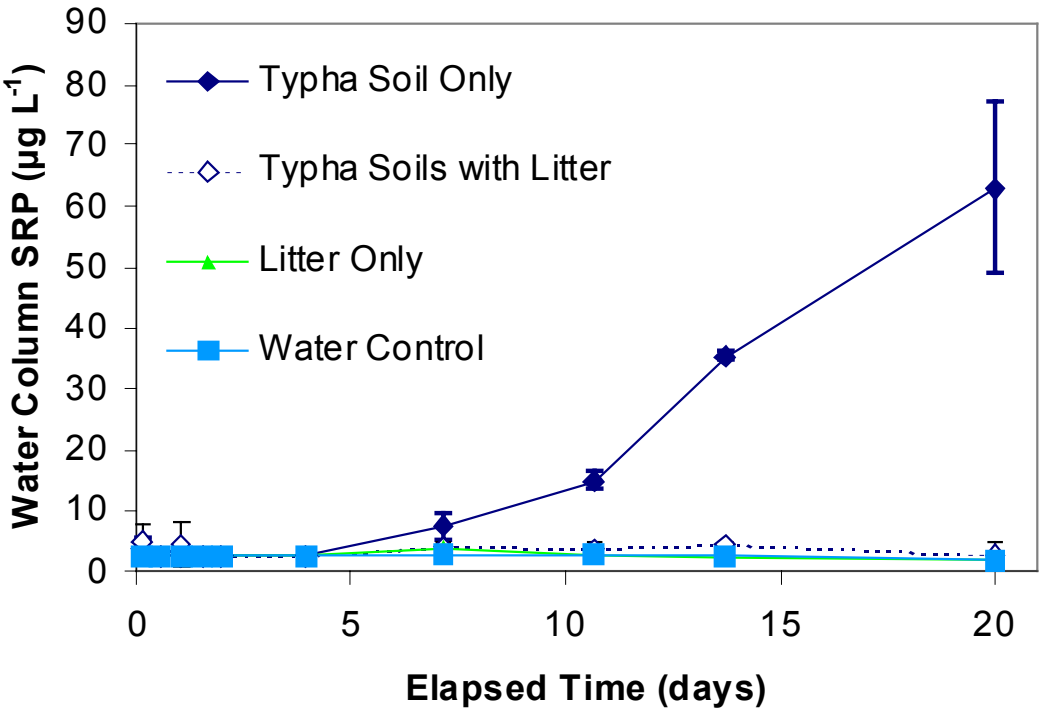

Figure 3-8. Soluble reactive phosphorus concentrations in the water columns of *Typha*-region intact soil cores with and without *Typha* litter amendments during 20-day dark laboratory incubation. Litter and water control cores contained no soil. Error bars for each treatment indicate ± one standard deviation between triplicate cores. *Typha* soils with litter, litter only, and water control treatment SRP concentrations were each near the detection limit of 2 µg L^{-1} for 28 days. *Typha* soil treatment values are from duplicate cores.

Between 7 and 20 days of incubation, SRP concentrations were significantly higher ($p < 0.05$) in the remaining two replicate waters overlying *Typha* soils without litter amendments (mean at 20 days= 63 µg L^{-1}; n=2), than in litter only and water control treatments (mean at 20 days = 2 µg L^{-1}; n=6). After 10 days, SRP concentrations in the water columns above soil-only treatments were significantly higher (15 µg L^{-1}) than those with litter plus soil (4 µg L^{-1}) ($p< 0.05$). *Typha* litter amendments (25g wet wt.) had a marked effect on SRP

concentration in the floodwater during the 20-day incubation. Concentrations never exceeded 4 μg SRP L^{-1} in any replicate without soils (litter only and water control), indicating that there was no net P flux out of the litter amendments. When leaf litter was added to cattail soils that alone had shown a positive flux into the water column, the flux was absent. Instead, SRP concentrations in the overlying water were maintained throughout the 20-day incubation (4 ± 1 μg L^{-1}), or as low as the control treatments.

Initial flood water pH (8.06) increased slightly to 8.36 ± 0.06 before the 8-hour sampling, and was similar among all treatments (8.41 ± 0.09) during the remaining 20 days. Water bath temperature averaged 24 ± 3 ° C during the incubation period.

Water column DOP concentrations for the two *Typha* soil treatments after 20 days (20 and 35 μg L^{-1}) were higher than the *Typha* soil + litter, litter only and incubation water control, which after 20 days were 15 ± 1, 15 ± 2, and 14 ± 3 μg L^{-1}, respectively, though differences were not significant ($p > 0.05$) (Figure 3-9). All treatment and control water DOP concentrations were not significantly different from one another or different over time during the 20-day incubation ($p > 0.05$).These results agree with the results of the 9-day tissue desiccation experiment: *Typha* litter does not release DOP under dark aerobic conditions.

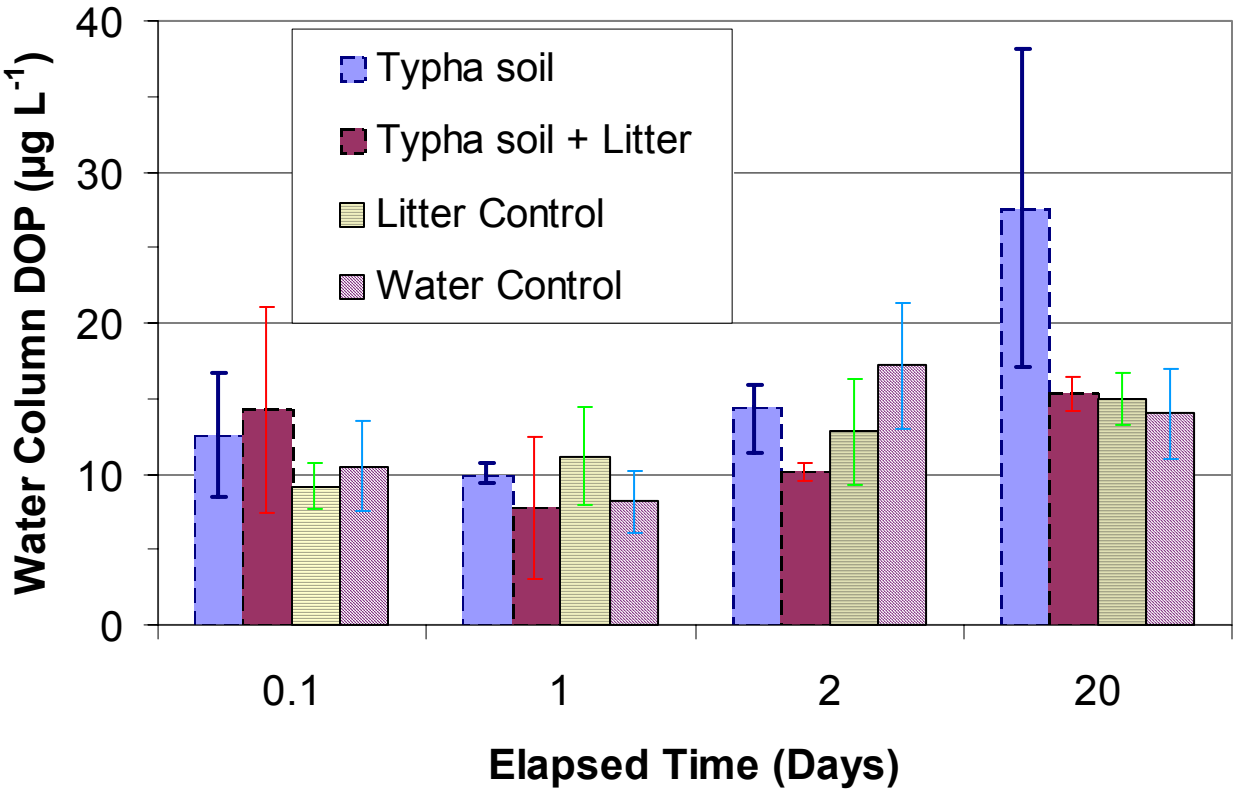

Figure 3-9. Mean dissolved organic phosphorus concentrations in the water columns of four treatments (see text for details) during 28-day dark laboratory incubation. Error bars indicate ± 1 standard deviation from the mean of three replicates.

Mean calcium values for the soil + litter and litter only treatments were significantly higher (123 mg L^{-1}) than those of the treatments without litter (104 mg L^{-1}) after 0.1 days of incubation ($p < 0.05$). After 0.5 days, however, no significant differences were observed in dissolved calcium concentrations between any treatment. Calcium concentrations were stable after that initial equilibration period (Figure 3-10). From 7 to 20 days of incubation, water column Ca concentrations in *Typha* soil + litter treatments (85 ± 5 mg L^{-1}) were

lower than litter only and water control treatments, but not significantly different.

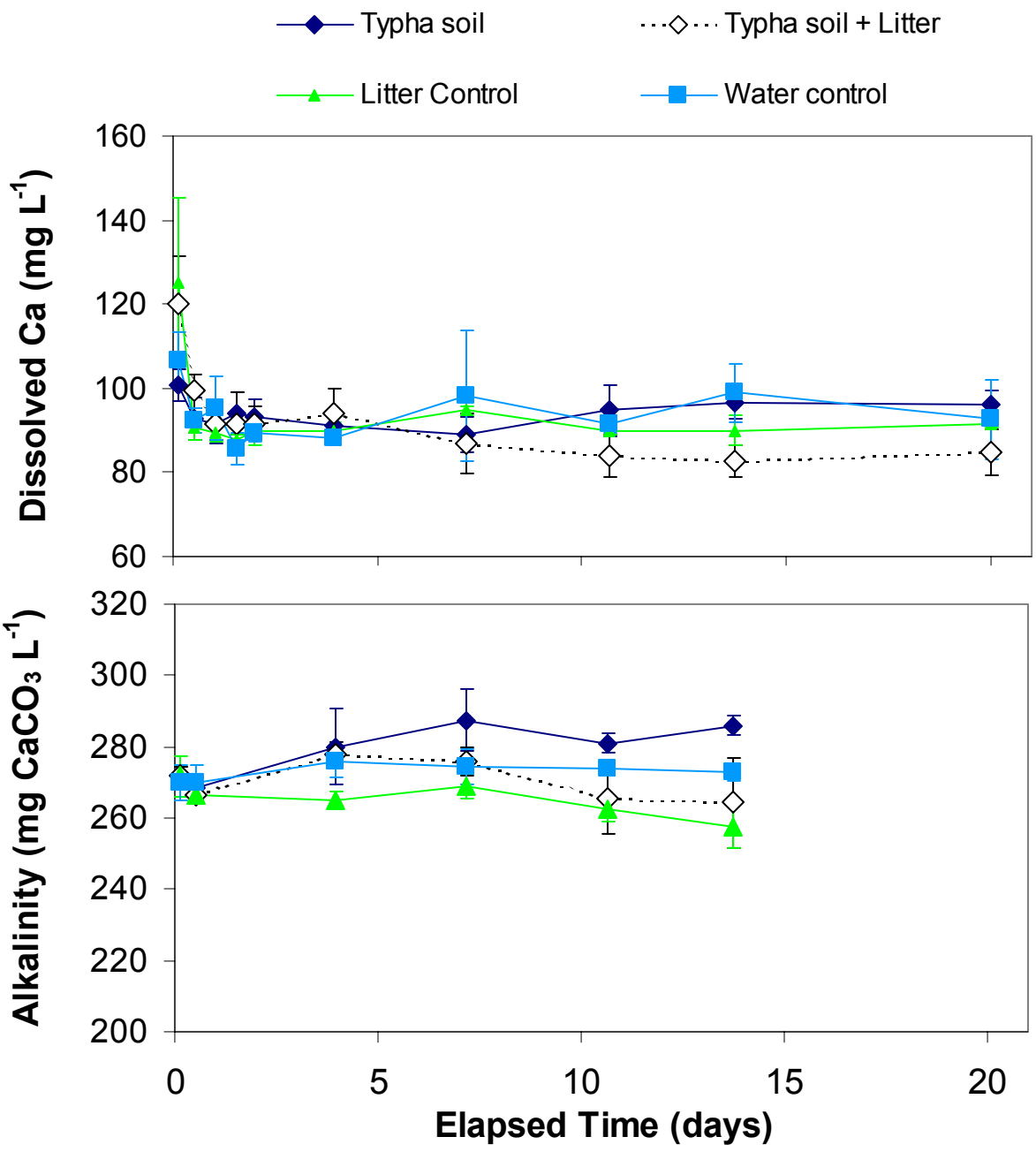

Figure 3-10. Mean dissolved calcium and alkalinity concentrations in the water columns of four treatments (see text for details) during a 28-day dark laboratory incubation. Error bars indicate ± 1 standard deviation of the mean from three replicates.

Alkalinity concentrations decreased significantly ($p < 0.05$) in litter control treatments from initial floodwater concentrations of 271 ± 6 mg L^{-1} to 258 ± 6 mg $CaCO_3$ L^{-1} after 14 days (Figure 3-10). After 14 days, the litter only treatment was lower in alkalinity (258 ± 6 mg $CaCO_3$ L^{-1}) than the *Typha* soil-only treatment (286 ± 3 mg $CaCO_3$ L^{-1}). All other treatments were constant with respect to alkalinity concentrations.

Dissolved organic carbon (DOC) concentrations were significantly higher after 4, 7, 10, and 14 days in treatments containing litter amendments (mean ± s.d. for all time intervals, 46 ± 1.9 mg L^{-1}), relative to soil-only and water-only control treatments (42 ± 1.3 mg L^{-1}) ($p > 0.05$) (Figure 3-11). Release of DOC from *Typha* litter may explain the lack of increased water column P, as microbial P uptake in the soil-only treatment could have been carbon-limited.

The method of DOC quantification used here did not describe the quality of carbon for microbial utilization, however there may be additional differences in the carbon quality between treatments that influenced P removal from the water column (Koshmanesh et al., 1999).

Rapid biological P uptake during the half-hour following the P amendment of 100 μg L^{-1} may explain an observed increase in SRP concentrations (94 μg L^{-1} for all columns, 88 μg L^{-1} for the control columns) of less than 100 μg L^{-1} (Figure 3-12). The water-only control columns showed a small decrease in concentration, then leveled off near 60 μg L^{-1}. Phosphorus in the control waters may have been absorbed by bacteria in the water column or

attached to core walls, aerator, etc., and converted to non-reactive phosphorus. The change in the control water P concentration was assumed to be an experimental effect and was subtracted from the observed concentration for each treatment.

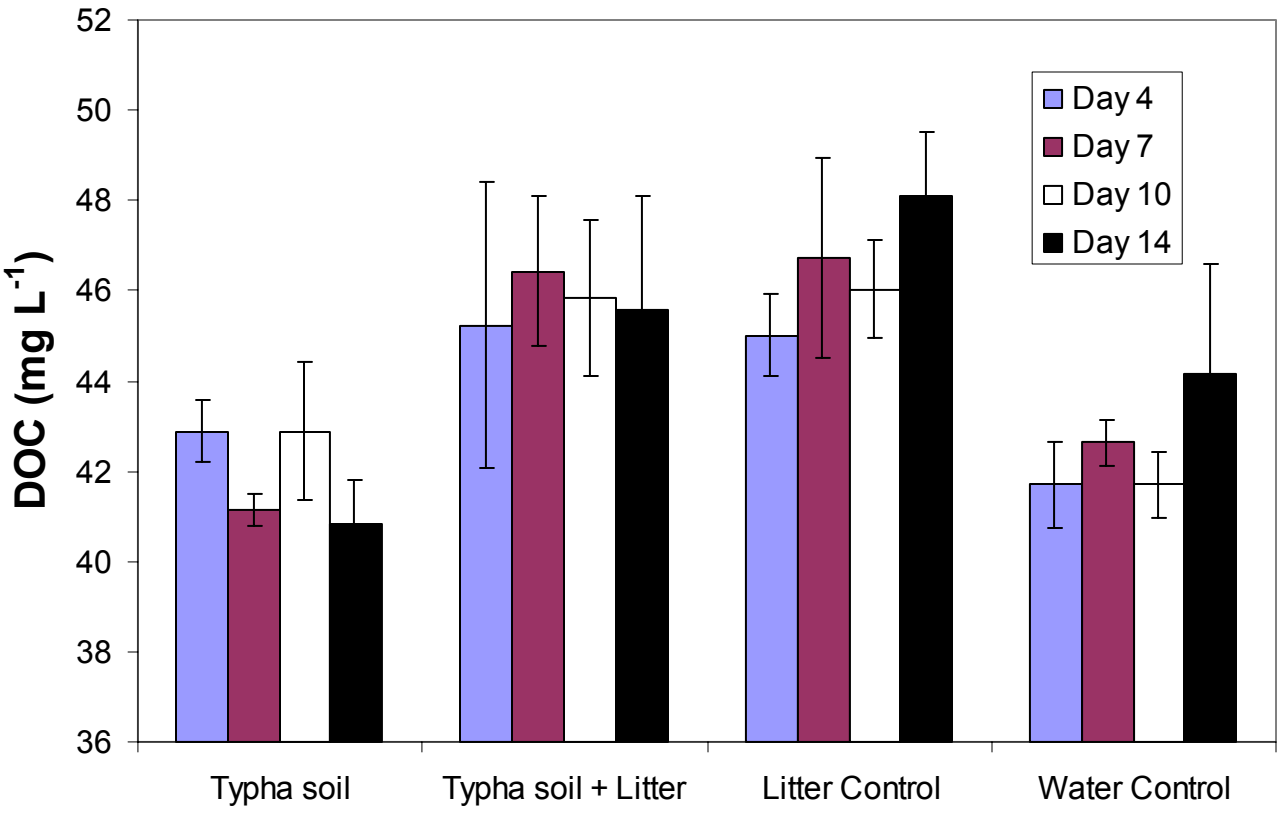

Figure 3-11. Dissolved organic carbon concentrations in water columns of four treatments (see text for details) after 4, 7, 10 and 14 days of a dark laboratory incubation. Error bars indicate ± 1 standard deviation from the mean of three replicates.

The *Typha* soil cores provided no significant P uptake capacity when compared with the water-only control cores ($p > 0.05$), while litter-amended treatments decreased the SRP concentrations to 3-11 µg L^{-1} in 53 hours. Eight hours after the amendment, concentrations in the litter-only treatment were

significantly lower than the other treatments, including the *Typha* soil + litter treatments (Figure 3-12). In the eight hours following the P-amendment, litter-only columns exhibited the greatest uptake rate of any of the controls or treatments (27.3 ± 2.3 mg m^{-2} day^{-1}), followed by the litter-amended cattail soils (22.5 ± 2.4 mg m^{-2} day^{-1}) (Table 3-4). The significant difference ($p < 0.05$) between P uptake rates of either litter treatment and the soil-only treatment is likely due to continued P flux out of the soil into the water column, as observed during the initial 20-day release period.

Soil N release into the water column increased the TN content of the litter over the 32-day experiment, from 0.71 ± 0.04 to 1.22 ± 0.11%, while the TN content of litter without soil decreased slightly to 0.66 ± 0.11%. Total C content of the litter was similar before (45.5 ± 1.1 %) and after incubation (44.5 to 46.6 %) in both litter treatments. There was likely some conversion of *Typha* tissue biomass into microbial biomass carbon. In decomposition studies on *Typha* biomass, half of the live tissue carbon may remain after a two year period of decay (Davis, 1991). Thus the litter carbon pool may be stable relative to the nutrient pools.

Phosphorus Uptake by Litter Microbial Populations

After sampling events at ΔT = 1, 2 and 3 days, three anoxic flasks and three oxic flasks were monitored for D.O. concentration. Short daily sparge periods (0.5 to 3 hr) during the 5-day P uptake monitoring period with N_2 + 0.03 % CO_2 (anoxic) or air (oxic) followed by stagnant incubation conditions resulted in low D.O. concentrations in anoxic treatments (0.59 ± 0.13 mg O_2 L^{-1}) relative to

oxic treatments (4.63 ± 0.86 mg O_2 L^{-1}). After 5 days, D.O. measured in all flasks was still significantly higher ($p < 0.05$) for oxic treatments (2.92 ± 1.00 mg O_2 L^{-1}) than for anoxic treatments (0.21 ± 0.13 mg O_2 L^{-1}), even though no flask was continuously purged.

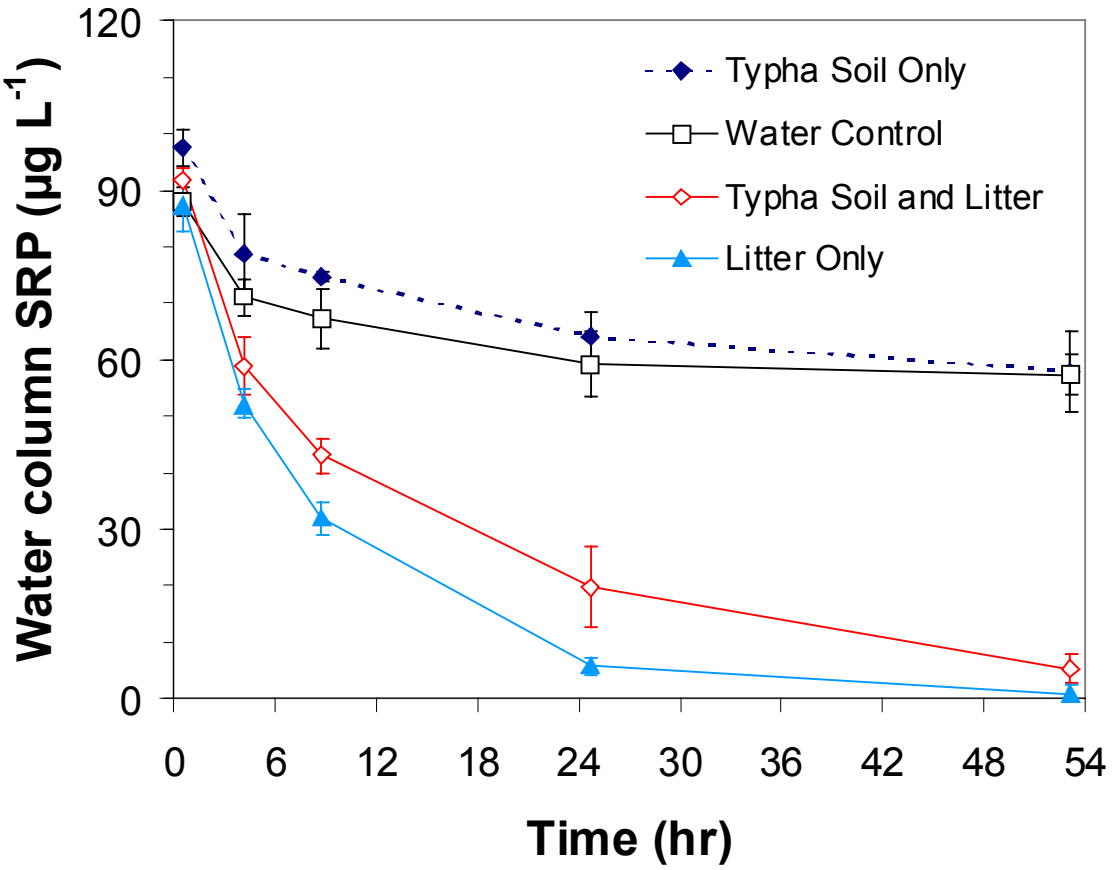

Figure 3-12. Water column SRP concentrations above a "background" concentration as determined before a 100 µg spike was added to each column. Background concentration was assumed stable through the 24-hour period. Triplicate water columns were assembled with or without soils from a cattail stand in STA-1W Cell 1, and aerobically incubated for 30 days prior to the P-spike amendment.

Table 3-4. Flux estimates from intact *Typha*-region soil cores and submerged *Typha* litter kept under dark conditions for 35 days. The 30 cm water column was continuously aerated. A 100 µg L^{-1} phosphorus spike was added after 30 days to measure short-term uptake rates. All values are means (± st. dev.) of triplicate cores minus mean flux in control cores, in mg P m^{-2} day^{-1}.

	Period of flux estimate	***Typha* soil**	***Typha* soil *Typha* litter** (mg P m^{-2} day^{-1})	***Typha* litter**
Release	7 – 20 days incubation	1.20 ± 0.27	0.00 ± 0.05	-0.01 ± 0.01
Uptake	0 – 8 hours after spike	0.17 ± 2.7	22.5 ± 2.4	27.3 ± 2.3

Rapid P uptake occurred under oxic conditions, reducing water SRP concentrations from 1000 µg L^{-1} initial concentration to 7 ± 3 µg SRP L^{-1} (Figure 3-13). The maximum uptake rate (56 µg P g dry matter $^{-1}$ $d^{-1)}$ in the 1000 µg L^{-1} oxic treatment was observed prior to the first 8 hour sampling. Based on field estimates of litter accumulations in STA-1W Cell 1 of 0.93 kg dry matter m^{-2}, the equivalent areal uptake rate would be 52 mg m^{-2} d^{-1}, nearly twice the rate observed in the intact core incubation P uptake period (Table 3-4). This potential rate may not be sustainable over the long term, however short-term uptake at that rate appears possible if another element is not limiting, and if adequate P was loaded to the system. For treatments that began the 5-day P uptake incubation at lower concentrations (100, 30, 10 and 0 µg P L^{-1} amendments), SRP concentrations were reduced to below the analytical detection limit of 2 µg L^{-1}

(Figure 3-13) in as little as 8 hours. Because of lower P mass added, uptake rates were slower in those treatments.

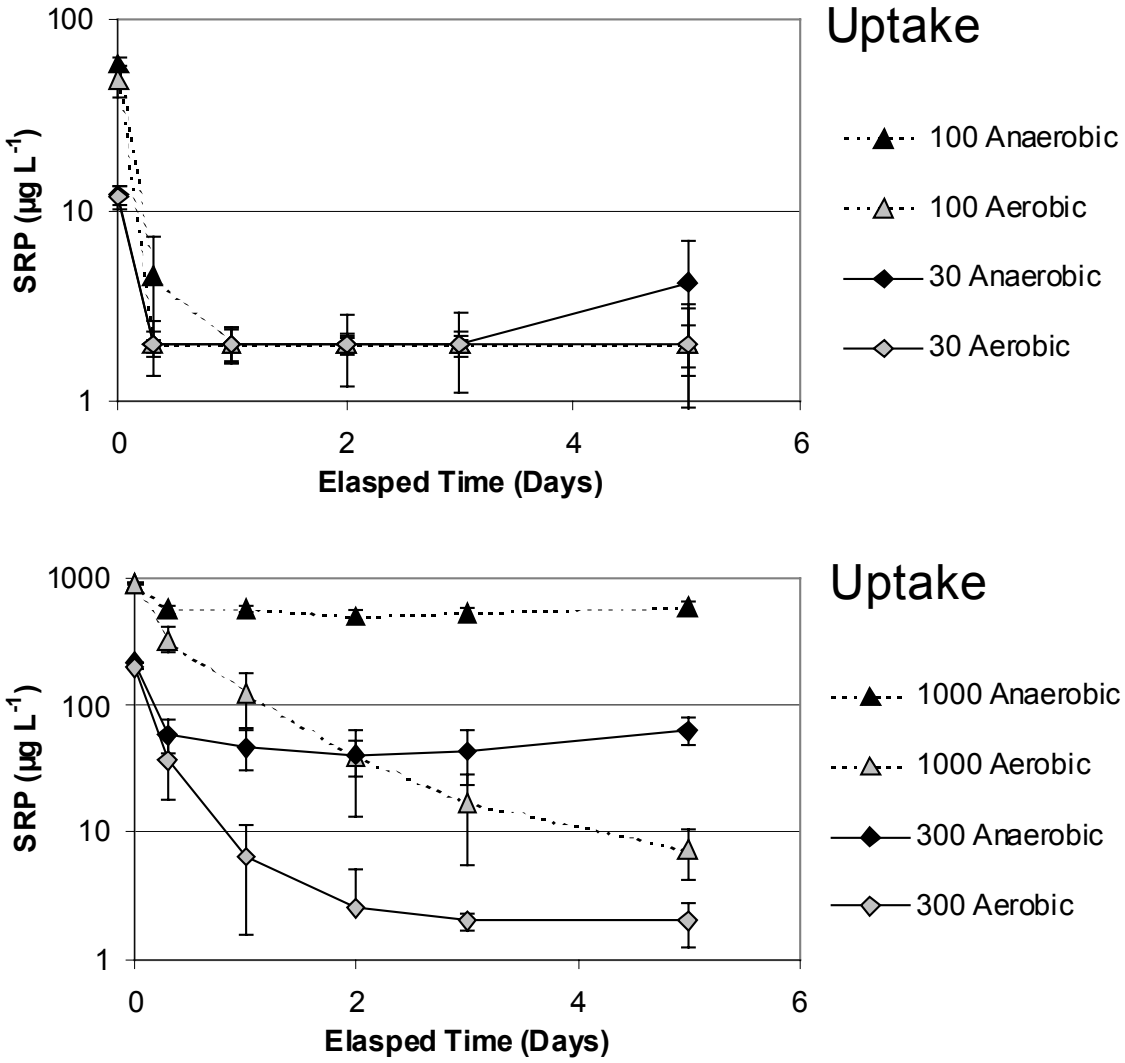

Figure 3-13. Mean soluble reactive phosphorus (SRP) concentrations in triplicate 0.5 L flasks, containing *Typha* litter incubated in the dark under oxic or anoxic conditions for 5 days. Initial concentrations of 1000, 300, 100 and 30 µg L^{-1} above background SRP level (< 2 µg L^{-1}) were achieved with KH_2PO_4 amendments at Time = 0 days. Note SRP concentrations are shown on a logarithmic scale.

When waters with initial P concentrations of 1000 or 300 µg L^{-1} above background were sparged with N_2 + CO_2 gas, P uptake was slower and less

complete than in the corresponding oxic treatments. Final SRP concentrations for the 1000 and 300 anoxic treatments were 580 and 63 μg P L^{-1}, representing reductions of 42 and 79%, respectively. Anoxic P uptake by litter in initial P concentrations lower than 300 μg L^{-1} was no different than oxic P uptake, and SRP concentrations were maintained below 2 μg L^{-1} after the 8 hour sampling.

Phosphorus-amended waters were replaced with fresh unamended water (<2 μg SRP L^{-1}) after seven days of incubation. Concentrations of SRP measured immediately after the exchange were below detection for all treatments, with the exception of the 1000 μg L^{-1} anoxic treatments (3-4 μg L^{-1}) (Figure 3-14). The minor amount of P detected was possibly residual from the wetted flask and litter surfaces, which were drained of P-amended water, but not rinsed before new low-P water was added. In the same treatment (1000 μg L^{-1} anoxic), SRP concentrations reached 23 ± 6 μg L^{-1} 24 hours after water exchange. Concentrations remained similar over the next five days (19 ± 6 μg L^{-1}) in each of the replicate samples.

Phosphorus concentrations also increased in the 300 μg L^{-1} anoxic treatment, rising steadily over the 5-day release monitoring period. Anoxic treatments initially incubated with 100 or less μg P L^{-1} showed no detectable P release over five days of incubation in un-amended waters. While the oxic litter treatments exhibited greater P uptake than the comparable anoxic treatments, no P release was observed from any oxic treatment. Even the 1000 μg L^{-1} oxic *Typha*

litter maintained water column SRP concentrations at 2 µg L^{-1} for five days in low-P reflood waters (Figure 3-14).

At the end of the entire 12-day incubation period (uptake followed by release), *Typha* litter showed an increase in microbial biomass P (MBP) in the 1000 and 300 µg L^{-1} oxic treatments (104.4 ± 29.2 and 65.8 ± 11.1 mg P kg dry $litter^{-1}$, respectively), relative to control (40.0 ± 5.9 mg P kg^{-1}) tissues preserved in the dark at 3.5 °C during the study (Figure 3-15).

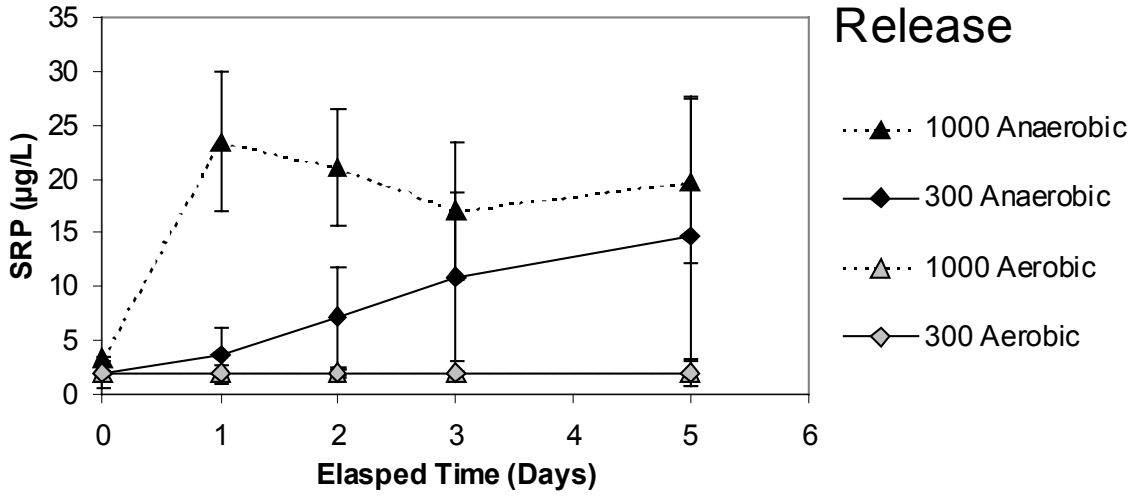

Figure 3-14. Mean soluble reactive phosphorus (SRP) concentrations in triplicate 0.5 L flasks, containing *Typha* litter incubated in the dark under oxic or anoxic conditions. After incubation under initial concentrations of 1000 or 300 µg L^{-1} above background SRP level (< 2 µg L^{-1}), water was replaced with fresh low-P water (< 2 µg SRP L^{-1}) at Time = 0 days.

Typha litter exposed to lower initial water P concentrations under aerobic conditions showed similar MBP levels to control litter. Microbial biomass P levels for the anoxic *Typha* incubations were at or below levels measured in the control samples. While small decreases in MBP were observed in the 30 and 1000

anoxic litter treatments (27.4 ± 7.8 and 27.6 ± 4.5 mg P kg^{-1} dry litter) relative to control tissues, differences in MBP for all anoxic tissue incubations did not appear to depend on initial water P concentration (Figure 3-15).

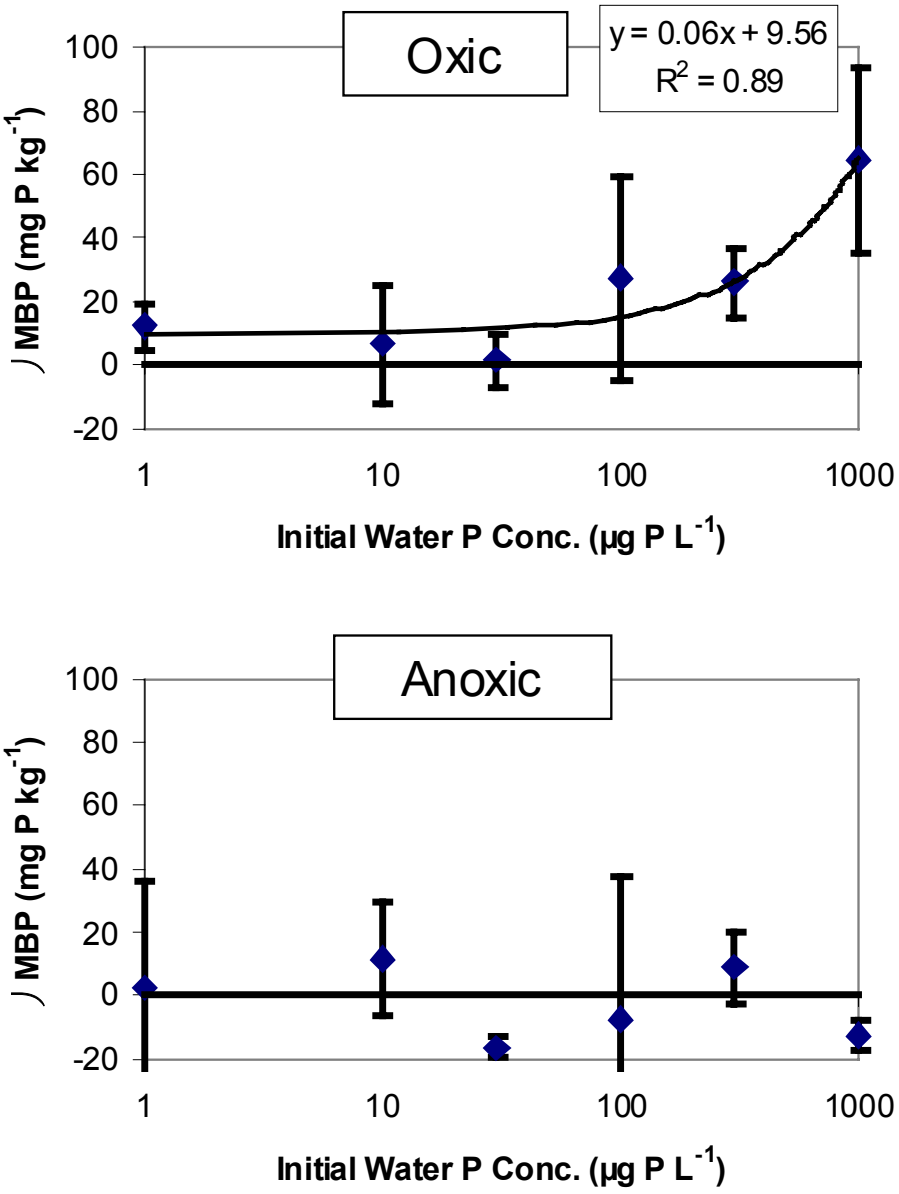

Figure 3-15. Change in microbial biomass phosphorus (MBP) of *Typha* litter incubated for seven days at initial water concentrations of 0, 10, 30, 100, 300 and 1000 µg L^{-1} SRP, under oxic (top) and anoxic (above) conditions.

Koshmanesh and others (1999) showed the importance of carbon substrate quality in the uptake and retention of P by soil microbes in an Australian constructed wetland receiving ADW. Bioavailable carbon additions (glucose) to the soil increased P uptake by 35-45%, and was attributed to increased microbial uptake and growth. In that study, sequestered P was retained under anoxic conditions. Acetate amendments increased P uptake to rates comparable to those from glucose addition, but the acetate-induced "luxury P uptake" as polyphosphate was subject to release again during anoxic conditions. Utilization of acetate as a carbon source was likely less efficient than glucose, and therefore P incorporation into cell tissue was lower for the same incubation period (Koshmanesh et al., 1999).

Implications for STA management

Reduced efficiency of microbial P uptake and retention under anoxic conditions has important implications for both STA management and remediation of the nutrient-impacted WCA-2A marsh. Several studies implicate microbial P acquisition as the primary mechanism of P immobilization in wetlands (Brix, 1997; Grimshaw et al., 1997), especially those with soils of low sorption capacity (Qualls and Richardson, 2000). *Typha domingensis* in outdoor enclosures retained P within the litter layer through microbial uptake processes (Richardson and Marshall, 1986; Richardson, 1999), and P content increased up to ten-fold above live tissue concentration during long-term (2 yr) nutrient enrichment studies.

Long-term nutrient loading has resulted in the continued expansion of a P-enriched, low-oxygen zone extending southward into WCA-2A from ADW discharges (Belanger et al., 1989; Reddy et al., 1993; Reddy et al., 1998). Similar conditions may develop within *Typha*-dominated regions of STA-1W Cell 1 inflow. Surface water TP reductions in WCA-2A are gradual with distance from the inflow, until a point where a mixture of sawgrass and open-water sloughs compete with encroaching *Typha* (Reddy et al., 1998). High soil-P levels in the WCA-2A impacted area adjacent to inflow structures may represent deposition that occurred before *Typha* colonized to the density observed in recent surveys. Phosphorus remains mobile in the impacted region because the water column lacks sufficient oxygen supply to support rapid microbial sequestration. In contrast, oxygen supply from slough areas adjacent to the low-oxygen region allows rapid microbial P uptake along the transition zone between slough and cattail-dominated areas. Total P concentration reduction rates with distance increase dramatically through the transition region of WCA-2A (Reddy et al., 1998).

An analysis of the soil sorption capacity along the WCA-2A eutrophication gradient has shown that soil sorption capacity was not saturated even in the enriched areas, yet elevated SRP concentrations persist through that region (Richardson and Vaithiyanathan, 1995). This suggests that the water column may not be in contact with the sorption sites, possibly due to the litter accumulations or macrophyte P uptake depleting exchangeable soil-P storages.

More important are the uptake mechanisms of microbial biomass within the litter layer and in contact with the water column. Due to O_2 limitation, however, this mechanism is inefficient in dense stands. One possibility for controlling oxygen levels may be periodic drawdown. The drawdown would consolidate and oxidize newly accreted soil material. A subsequent reflood may allow SAV and algal communities to colonize and oxygenate the water column, and microbial communities to rapidly sequester released nutrients. Oxygen supply to the water column is not expected to significantly increase mobilization of soil-P through decomposition as long as sufficient dissolved carbon is available as an energy source for metabolism.

Conclusions

The majority of P removed from the water column by *Typha* mesocosm communities was incorporated into newly accreted soils (78%) during 2.7 years, though the new soil did not appear to be of cattail origin. *Typha* incorporated 39 and 65% of total mesocosm biomass P to belowground tissue. Healthy *Najas* tissues became chlorotic after nine days in the dark, and may be a source of SRP or DOP to the water column during events of high turbidity. In contrast, *Typha* litter material was relatively stable in dark conditions. While *Typha* biomass appears to persist as leaf litter and detritus to a greater extent than SAV tissues, the extensive root system has the potential to hinder long-term storage by mobilizing P from enriched soils.

Typha leaf litter at the soil surface may limit diffusive flux of soil-P into the water column if *Typha* stand density is low and the water column is oxygenated by aquatic autotrophs. Similarly in treatment wetlands, the litter layer can become P-enriched from external loading to the water column through microbial and algal uptake mechanisms. However, at higher stand densities, P removal from surface water is limited to soil sorption and microbial uptake mechanisms because primary productivity and oxygen supply is light-limited. When oxygen is depleted in the highly enriched regions near ADW discharge structures, P uptake by litter-associated microbial biomass is likely oxygen limited. Treatment wetland managers may need to monitor oxygen levels in inflow regions with dense *Typha* stands, and provide dissolved oxygen through SAV aquatic photosynthesis or other means to ensure low outflow water TP concentrations. Further research will likely identify the long-term sustainability of P uptake processes by leaf litter, and conditions under which the sequestered P is re-mobilized into surface waters.

CHAPTER 4
SYNTHESIS

Constructed treatment wetlands are useful tools for removing phosphorus (P) from agricultural drainage waters (ADW). Besides being a cost-effective treatment alternative with secondary benefits (e.g., wildlife values, aesthetics), wetland communities typically reduce P concentrations in surface waters between inlet and outflow. However, characteristics of submerged aquatic vegetation (SAV) and emergent wetland communities, including water column shading, oxygen supply, and community metabolism, can influence treatment wetland P removal performance.

Emergent and submerged vegetation communities each remove P from the water column, though the dominant processes are different. There is significant debate in the literature over the uptake of P through SAV roots vs. shoots (Denny, 1972; Bole and Allen, 1978; Hill, 1979; Carignan and Kalff 1980; Barko and Smart, 1980,1981; Carignan, 1982; Carpenter, 1983; Moeller et al., 1988; Rattray et al., 1991; Gumbricht, 1993; Stephen et al., 1997; Kufel and Kufel, 2002). Many of these studies, including those using radiotracers, maintained SAV tissue cultures in P-free overlying water and concluded sediments can act as a source for nearly all P taken up by rooted macrophytes. However, fragments of rooting macrophytes, as well as rootless species and macroalgae (e.g., charophytes) can

grow well in soil-free cultures of agricultural drainage waters (Pietro, 1998, Kufel and Kufel, 2002, and personal observation). It is generally agreed that the source of P to *Typha* spp. is through root uptake (Davis, 1991; Newman et al., 1996; Lorenzen et al., 2001), and below-ground *Typha* biomass can potentially deplete soil P.

In a review on the effects of SAV on nutrient dynamics during sediment deposition and resuspension, Barko and James (1998) concluded that SAV beds efficiently trap suspended matter from the water column by lowering water velocities and reducing resuspension. The SAV community itself also can quickly produce appreciable sediment. Mesocosms vegetated with SAV accumulated deep organic sediment layers over three years of operations, at rates of up to 3.1 cm yr^{-1} under high hydraulic loads (DBEL 2001). It is possible that deposition of P-laden organic matter resulted in P-enrichment of soils beneath submerged vegetation observed in this study.

The *Typha* community likely relies on slow flows and rapid microbial uptake mechanisms to retain P "on-site." Emergent biomass-P is subject to rapid leaching from senescent tissues and can be transported downstream. To counteract such losses, emergent litter accumulation appears to provide some capacity for assimilating and retaining P. *Typha* leaf litter can provide substrata for microbial growth and P uptake, where incorporation of inorganic P into organic P pools during soil formation delays recycling into a bioavailable inorganic form. Without continuous additions of new leaf litter, bacterial films

and algae become space-limited on substrata, and overextended colonies slough particulates into the water column under high flows.

Phosphorus uptake by the litter layer may also function as a driver of diffusive flux from the soil. Litter/microbial P uptake mechanisms were able to maintain low water column SRP concentrations, which led to strong diffusion gradients and higher potential diffusive P flux in the emergent macrophyte stand, relative to SAV. This flux may not impact water column P concentrations if the nutrient is incorporated into the litter layer at the soil surface, but would mobilize P from long-term storage in the accrued soils.

Water column P removal is thus aided by the biological P cycling that occurs primarily in association with submerged surfaces. Humification of organic detritus and leaf litter results in lower P bioavailability than in the original tissue and is an important process in soil development. It is within the soil matrix that physicochemically stable P forms must be sequestered over the long term for wetland water treatment to be successful.

Irradiance and Water Column Shading

Solar UV irradiance of the water column is moderated by turbidity (inorganic particles, planktonic algal cells, etc.), color from dissolved organic matter (DOM), and canopy shading (Wetzel et al. 1995). In the absence of these controlling factors, UV–B irradiance (280-320 nm) can reach depths of 5 m or more. During periods of high flows, low-density organic matter can become suspended in the water column, increasing turbidity and decreasing light

penetration. After short periods under aphotic conditions, SAV tissues can atrophy and release soluble reactive P (SRP) and dissolved organic P (DOP). Meanwhile, dead EAV tissues can persist in the dark without releasing P into surrounding waters.

Dissolved organic compounds are protected from photolysis by the emergent canopy. Rose and Crumpton (1996) found standing live and dead *Typha* shoots, leaf litter, and ephemeral floating plant populations combined to reduce light levels, measured at 5 cm below the water surface, to 2% of ambient levels. In contrast, less dense emergent macrophyte stands allow UV penetration into the water column for photolysis of organic compounds into bioavailable SRP. Submerged macrophytes can similarly reduce light penetration, photosynthesis and dissolved oxygen generation in waters below the leaf canopy. Depending on current water depth and recent depth changes, the proportion of the water column thus isolated from the euphotic zone can be variable.

Wetzel et al. (1995) examined with laboratory experiments the UV-B photolysis of DOM compounds derived from *Typha latifolia* and *Juncus effusus.* Total DOM concentration was found to change little under natural irradiance levels. However, a conversion of recalcitrant humic substances to labile compounds did occur, which increased the bioavailability of those organic compounds to further microbial degradation.

In the absence of dense vegetation, shallow wetlands (<2m deep) are sometimes entirely euphotic, with benthic algae capturing incident light and reflecting the excess. Shading of incident radiation by an emergent *Typha* canopy was observed to reduce the net primary productivity of periphyton within the stand by 80%, relative to adjacent unshaded communities (Grimshaw et al., 1997). Irradiance can control the distribution of algal species because of differences in optimum light levels between species. In oligotrophic waters, planktonic bacteria and algae are subject to short-term negative effects of high ultra-violet (UV) irradiance (Karentz et al., 1994), but are otherwise stimulated by increased light availability. Bunn et al. (1999) estimate that diatoms are in optimal light regime where shading by riparian canopy exceeds 80% (< 9 mol m^{-2} day^{-1}), while the filamentous alga *Cladophora* requires less than 50% shading (26 mol m^{-2} day^{-1}) (Graham et al., 1995).

Submerged macrophytes are typically able to grow in full sunlight. Some researchers have reported photoinhibition in marine macrophytes within intertidal areas (King and Schramm, 1976), when falling water levels expose macrophytes to increasing light. The SAV canopy can also preclude light penetration to the soil surface if dense beds develop under static flow conditions and static or decreasing water depths. Under dynamic water levels and flow conditions, however, the submerged macrophyte bed is continuously reshaped and increases light penetration.

In the northern Everglades mixed-marsh community of sawgrass prairies and wet sloughs, periphyton dominates productivity in the sloughs, while the sawgrass community dominates marginally higher land elevations. Emergent vegetation is typically found in more shallow waters than SAV sloughs, and as a result, soil and detritus may be exposed more frequently. In a shallow water column the oxygen demands of the soil, detritus and biota are concentrated in a smaller water volume than under deep-water conditions, and the water column can become anoxic.

Oxygen Supply

In the continuously flooded environment of a treatment wetland, oxygen supply comes primarily from aquatic photosynthesis and secondarily from pressure-induced oxygen transport through macrophyte aerenchyma tissues, or physical (e.g. wind-induced) mixing. Oxygen demand is high in the flooded wetland environment. Biological oxygen demand by heterotrophic organisms and respiring plant cells, and chemical oxygen demand of reduced soil compounds, all deplete dissolved oxygen (DO) levels.

Water circulation is important at the site of oxygen consumption. Boundary layers develop along soil and detritus surfaces which limit oxygen supply to consumers and decomposers even in aerated water. Jorgensen and Revsbech (1985) described boundary layers adjacent to coastal sediments and detritus as 0.2 to > 1 mm in thickness, depending on flow velocity and surface roughness. Diffusion times for O_2 passing through the layer were 1.2 to 9 min.

Aquatic photosynthesis, meanwhile, increases DO concentrations and potential for metabolic activity (Karjalainen et al., 2001). Atmospheric oxygen diffuses into still waters at slow rates (0.72 g O_2 m^{-2} day^{-1}) relative to potential oxygen production rates via aquatic photosynthesis (Odum, 1956). Wind-induced mixing of surface waters can increase diffusion rates, though emergent macrophyte stands reduce wind velocities near the water surface.

Aquatic photosynthesis by algae, cyanobacteria, and macrophytes can be extremely high. Submerged macrophytes in Florida spring runs can generate as much as 64 g O_2 m^{-2} day^{-1}. This rate is among the highest reported for any system (Duarte and Canfield, 1990). The high rates of productivity observed in Everglades cyanobacterial mats (wet season, 3 to 23 g O_2 m^{-2} day^{-1}) (McCormick and Laing, 2003) are possible only through intense recycle pathways of nutrients, carbon and gases (Wetzel, 1993).

Rooted emergent macrophytes (Jespersen et al., 1998) and submerged aquatic plants (Karjalainen et al., 2001) both can transfer oxygen from shoots to roots through aerenchyma tissues, at rates which sometimes exceed that required for root respiration. Excess oxygen is lost to the surrounding soil environment. The microenvironment around root hairs, or the rhizosphere, can experience increased redox potential and dissolved oxygen levels, relative to the bulk soil (Wium-Anderson and Anderson, 1972; Flessa 1994; Wigand et al. 1997). The effects of an oxidized rhizosphere on soil processes include coupled nitrification/denitrification reactions, increased microbial metabolism (including

the activation of aerobic extracellular enzymes), and concentration of Fe along the oxic-anoxic interface.

The importance of oxic surface sediments in maintaining oligotrophic conditions was described for Lake Stechlin (Germany) by Gonsiorczyk et al. (2001). In that study, oxidized Fe hydroxides within the surface sediments provided P sorption sites and microbial activity acted to minimize P release from sediment porewater. Similarly in this study, leaf litter and detritus surfaces above wetland soils minimized P loading to the water column, but was sensitive to oxygen supply. Due to the low Fe concentrations in Everglades soils, Fe hydroxide formation may not be as important to maintaining low water column P concentrations as for lakes with mineral sediments.

Community Metabolism

Wetland community metabolism has a fundamental influence on P cycling and stability in treatment wetlands. Light penetration into the water column provides energy for aquatic photosynthesis and organic matter provides energy to heterotrophic organisms (Odum 1956). In shaded areas, heterotrophic respiration exceeds autotrophic photosynthesis, and the community metabolism is said to be negative. Negative metabolism results in an absence of diurnal increases in dissolved oxygen and pH levels, as well as reduced rates of $CaCO_3$ precipitation.

Limited available P in the freshwater environment drives intense competition for this essential nutrient between all life forms. Through biological

uptake, chemical sorption and precipitation, and physical settling processes, P is concentrated on solid surfaces, and diluted in the ambient water column. In beds of the submerged macrophyte, *Najas guadalupensis*, surfaces consist of live photosynthetic leaf and shoot tissues, inorganic precipitates and settled particles. The emergent macrophyte, *Typha* spp., has stands of live shoots, along with dead non-photosynthetic shoots, decomposing leaf litter, and settled particles in contact with the water column.

Epiphytic and microbial colonies utilize submerged surfaces, especially those which provide necessary nutrients and carbon. Nutrients absent in the substrate must be acquired from the ambient water solution. Due to low P availability relative to carbon, oxygen, nitrogen, and other essential elements, P uptake can be rapid. In some systems, uptake of P from the water column by epiphytes and microbes could equal or exceed direct uptake by the macrophytes which are used to describe the community. However, microbes and metaphyton are well-known to internally cycle nutrients many times faster than macrophytes because of their faster turnover rates.

Due to rapid desiccation and inadequate sampling methodology, microbial and epiphytic biomass is often considered small if not insignificant to the P mass balance of the larger community. Nevertheless, P sequestration to surfaces by epiphytes and microbes may be an important link between the ambient water column and the limitations of direct uptake mechanisms of macrophytes. In this way, the aquatic epiphytes, bacteria and fungi are

analogous to mycorrhizae which extend the P depletion zone in soils around terrestrial plant roots. In fact, aquatic mycorrhizae associated with the freshwater submerged macrophyte, *Vallisneria americana*, were observed to enhance P uptake by 85% compared to plants without active mycorrhizae (Wigand and Stevenson, 1997).

Microbial activity in treatment wetlands is controlled by numerous factors including the abundance of available oxygen and alternative electron acceptors. In this study, microbial P uptake was dependent on DO concentrations. At low oxygen concentrations, soluble P concentration reductions by *Typha* litter were slower and less complete than in similar treatments incubated under higher DO levels. As an emergent stand increases in density, shading of the water column may reduce photosynthetic oxygen re-supply to detritus- and leaf litter-associated heterotrophs. At the same time, accumulations of litter in dense stands increase the biological oxygen demand. Less dense stands of emergent macrophytes that allow light penetration to aquatic autotrophs are likely to host microbial colonies capable of reducing P concentrations to very low levels (SRP < 2 $\mu g\ L^{-1}$).

Phosphorus sequestered into microbial biomass under low oxygen conditions is subject to release into the water column. This suggests that under stagnant conditions, water beneath the canopy of emergent vegetation where respiration exceeds photosynthesis, dissolved oxygen levels are reduced and water column P concentrations can become elevated.

The differences in morphology between the emergent and submerged macrophytes therefore may be generalized in terms of metabolism. Dense emergent stands contain many surfaces that consume oxygen, while SAV communities provide more surfaces that are oxygenated through primary productivity.

Implications for STA Management

The uptake potential of the *Typha* litter may account for the water column P reduction capacity observed in *Typha*-dominated wetlands. Cell 1 of a Stormwater Treatment Area (STA-1W) wetland removed an average 0.860 g P m^{-2} yr^{-1} during 2002 (SFWMD, 2003), which equates to 2.4 mg m^{-2} day^{-1}. In comparison, SAV-dominated Cell 4 is a smaller cell that has consistently provided higher P reductions than Cell 1. In water year 2002, Cell 4 P reductions averaged 2.0 g P m^{-2} yr^{-1}, or 5.4 mg P m^{-2} day^{-1}. The P removal rate for either cell is much lower than the 27.3 mg P m^{-2} day^{-1} taken up by fresh *Typha* litter during short-term (8 hours) intact soil core incubations, and the 56 mg P m^{-2} day^{-1} estimated from soil-less *Typha* litter incubations. Compared to *Typha* soils without fresh *Typha* litter, the addition of litter increased the rate of uptake by two-fold. With litter accumulating available P from the water column as well as any mobilized soil-P from below, a P-enriched litter layer will result.

Uptake rates measured in the cores containing *Typha* litter cannot be extrapolated to the entire cell. The inflow P load to an STA is not entirely bioavailable, nor is Cell 1 a uniform *Typha* stand with bare soils or accumulated

fresh litter. Instead, Cell 1 is a patchwork of floating, submerged and emergent vegetation. Sparse and dense stands of each are found throughout the cell. Water column conditions at the center of a *Typha* stand can be anoxic, and litter-associated microbial P uptake is slower under low oxygen conditions.

The region of a wetland that receives point discharge of ADW can be densely colonized by cattails. It is not clear, however, if cattail vegetation colonize in response to elevated water column P conditions, or elevated soil P conditions, or a combination of both. After a dense stand of the emergent macrophyte has formed however, little P removal from external loads is expected, due to anoxic conditions typical within such a stand (Figure 4-1). The slough communities of the Everglades are shallow (< 1m) systems which contain considerable DOM released from vegetation. High light levels in the slough communities probably helps drive primary production by autotrophs (e.g., algae, SAV) that assimilate the photolysate P.

Calcium enrichment of soils through water column $CaCO_3$ precipitation may have increased the soil Ca-bound P pool in SAV soils, but not to the extent that P mobilization from the soil was negated. Reddy et al. (1998) found that along a eutrophication gradient in Water Conservation Area (WCA)-2A, calcium enrichment in surface soils was thirty times greater than P enrichment.

When ADW containing high concentrations of Ca^{2+} passes through a photosynthetically-active wetland, $CaCO_3$ precipitation occurs. Suspended particles in ADW that contain calcium may also settle within the wetland

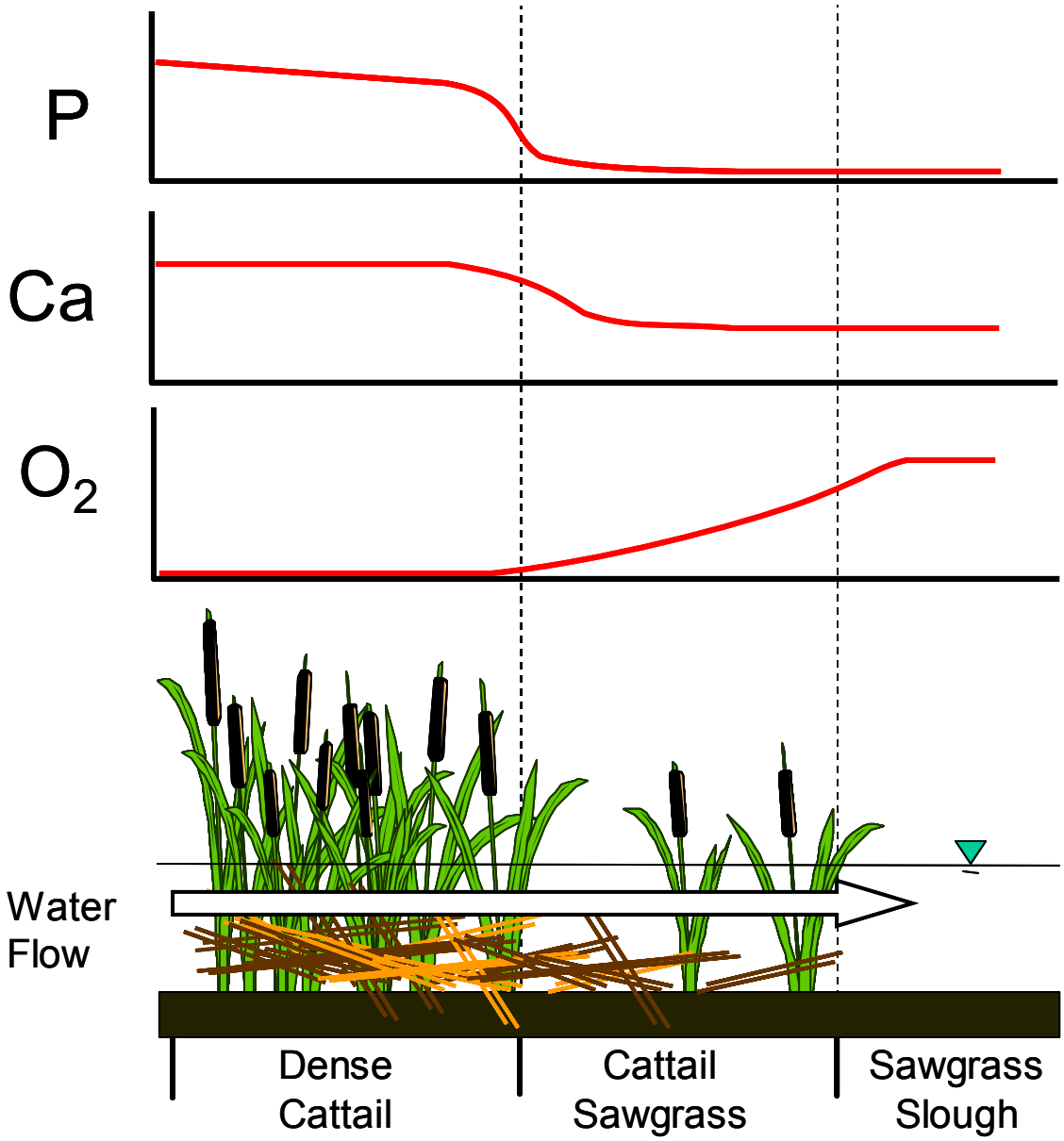

Figure 4-1. Conceptual diagrams of phosphorus, calcium and oxygen levels in the water column in relation to vegetation type. Dense cattail stands are found near point discharges of nutrient and Ca-enriched agricultural drainage waters, while sawgrass slough communities are un-enriched (with respect to Ca and P) areas of the wetland.

environment to enrich soil calcium contents. The sorption capacity of $CaCO_3$ is finite, however, and lower than that of Fe and aluminum hydroxides

(Richardson, 1985). Continuous P loadings to treatment wetland soils may exhaust existing or newly-formed calcareous sorption sites.

Soils below cattail stands may also become Ca-enriched through interactions with groundwater. Since $CaCO_3$ precipitation is not likely to occur in dense cattail stands such as those near the inflow of WCA-2A, the aforementioned Ca-enrichment observed at those locations either occurred prior to cattail invasion, during periods of lower stand density, or through interactions with the groundwater. Calcium deposition would likely have occurred immediately upon commencement of ADW discharges to the WCA, because the slough environment present at that time would have been capable of precipitating $CaCO_3$.

Over time, the calcium precipitate may have increased soil sorption capacity, and led to P enrichment of WCA-2A inflow-region soils. Cattails that colonized the enriched soils may have altered the P retention process at work in that region. Cattail roots mobilize P from the soils (White et al., in press), canopy shade hinders the regeneration of soil sorption capacity by reducing $CaCO_3$ precipitation, and oxygen demand slows microbial activities. An STA inflow region colonized by cattails through a similar process may provide some initial P removal, but as stand density increases, removal efficiency is likely to decline. Open water areas may therefore be essential to successful P removal from surface waters and long-term P retention in the newly accreted soils.

Conclusions

Availablility of light, carbon, oxygen and other essential nutrients creates an intense pressure for organisms to acquire and conserve P to effectively maximize growth when favorable conditions arise. As a controlling nutrient in many freshwater systems, P is closely associated with macrophyte and microbial biomass in wetlands. Macrophytes incorporate soil-P into biomass, then when tissues senesce, P is released into the water column in both organic and inorganic water-soluble forms.

Macrophyte vegetation also plays an important role in structuring the physicochemical underwater environment. Adsorptive surfaces of amorphous $CaCO_3$ precipitates can effectively retard the movement of P through the landscape, and may enhance Ca-bound P pools in newly accrued soils. SAV beds oxygenate the water column which allows increased microbial metabolism and P uptake. Oxygen demand maintains anaerobic conditions in deep soils and dense macrophyte stands. The greatest consequence of vegetation type on P removal performance of a wetland may be the effect the macrophyte has on the microbial biomass which it supports. The synthesis of these P cycling processes can aid wetland managers in achieving water quality treatment objectives and ultimately aid in the restoration of P-impacted marshes.

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BIOGRAPHICAL SKETCH

Kevin Grace was born in Philadelphia, PA in 1976, and grew up in Silver Spring, MD. At the age of 16, Kevin left Maryland to attend the Florida Institute of Technology, in Melbourne, FL, where he received a BS degree in environmental science. After graduation, Kevin remained in Melbourne, a fair city by any measure, and began working for DB Environmental. In 2001, he moved to Gainesville, FL to pursue his interests both at the university and the greater north-central Florida region, where hiking, camping and canoeing opportunities abound. With luck, Kevin will continue to interact with good people through interesting work.