Water Air Soil Pollut (2009) 197:223–232
DOI 10.1007/s11270-008-9805-x

Nitrogen and Phosphorus Remediation by Three Floating Aquatic Macrophytes in Greenhouse-Based Laboratory-Scale Subsurface Constructed Wetlands

Robert F. Polomski · Milton D. Taylor · Douglas G. Bielenberg · William C. Bridges · Stephen J. Klaine · Ted Whitwell

Received: 13 February 2008 / Accepted: 15 July 2008 / Published online: 8 August 2008

Abstract In the greenhouse and container nursery production industry there is potential for runoff of nitrogen (N) and phosphorus (P), which may contaminate surface and groundwater. Since the 1950s constructed wetlands (CWs), as a simple, low-technology method, have been shown to effectively treat agricultural, industrial, and municipal wastewater. We investigated the N and P attenuating potential of three floating hydrophytes planted in a laboratory-scale subsurface flow (SSF) CW system. Over an 8-week period plants were supplied with N and P (0.39 to 36.81 $mg{\cdot}L^{-1}$ N and 0.07 to 6.77 $mg{\cdot}L^{-1}$ P) that spanned the rates detected in nursery runoff between the discharge and inflow locations of a commercial nursery currently employing CWs. Whole plant dry weight was positively correlated with N and P supplied. Highest N recovery rates were exhibited by water hyacinth (*Eichhornia crassipes* [Mart.] Solms.) and water lettuce (*Pistia stratiotes* L.). P recovery rates were similar for water hyacinth, water lettuce, and dwarf redstemmed parrotfeather (*Myriophyllum aquaticum* [Vell.] Verdc.). These floating hydrophytes can be cultivated in a SSF CW to remediate runoff losses of N and P. The possibility exists for integrating them into a polycultural remediation system that includes emergent aquatic macrophytes for processing and polishing nursery/greenhouse wastewater.

R. F. Polomski (✉) · D. G. Bielenberg · W. C. Bridges · S. J. Klaine · T. Whitwell
Clemson University,
Clemson, SC, USA
e-mail: bplmsk@clemson.edu

M. D. Taylor
InsectiGen, Inc.,
Athens, GA, USA

Keywords Nursery runoff · Nutrient contaminants · Parrotfeather · Phytoremediation · Water hyacinth · Water lettuce · Water quality

1 Introduction

Irrigation of nursery and greenhouse container crops may lead to leaching and loss of fertilizers and other agricultural chemicals. This can pose a threat to groundwater and result in surface water contamination. Runoff containing nitrate–nitrogen (NO_3^-–N) and soluble reactive phosphate ($H_2PO_4^-$, HPO_4^{2-}, and PO_4^{3-}) may lead to excessive algal and aquatic plant growth in surface waters, resulting in accelerated eutrophication, primarily in freshwater streams, rivers, lakes, and reservoirs (Carpenter et al. 1998).

The US Environmental Protection Agency (US EPA) has established the maximum contaminant level for NO_3^-–N in drinking water to be 10 $mg{\cdot}L^{-1}$ (US EPA 1986). No federal limits on P contaminant levels in freshwater exist; however, the US EPA recommends that total P not exceed 0.10 $mg{\cdot}L^{-1}$ in streams or other flowing waters and 0.05 $mg{\cdot}L^{-1}$ in any streams that enter lakes or reservoirs (US EPA 1986).

Greenhouse crop production may result in NO_3–N runoff levels of 100 mg·L^{-1} NO_3–N (Wood et al. 1999). Nitrate–nitrogen concentrations in nursery crop runoff can range from 0.1 to 135 mg·L^{-1} (Alexander 1993; Yeager et al. 1993; Taylor et al. 2006) and P concentrations from 0.01 to 20 mg·L^{-1} P (Alexander 1993; James 1995; Taylor et al. 2006). To reduce the discharge levels of these nonpoint source pollutants and to comply with increasingly stringent environmental regulations at state and federal levels, CWs have been promoted as inexpensive, low-technology alternatives to conventional water treatment systems. Similar to natural wetlands, CWs treat wastewater with physicochemical and biological processes that involve vegetation, soils, and associated microbial populations in a controlled environment. These engineered wetlands are defined by their vegetation: free-floating, floating-leaved, emergent, and submerged plants (Vymazal 2007). In temperate regions emergent macrophytes are commonly used in surface-flow (SF) and SSF CWs to treat agricultural wastewater (Arnold et al. 1999; Berghage et al. 1999; Taylor et al. 2006). Due to the large land area required by typical SF CWs and the concomitant loss of production area, SSF CWs have been recommended as a viable alternative for greenhouse and nursery water treatment (Arnold et al. 1999; Berghage et al. 1999).

In tropical and subtropical regions free-floating hydrophytes are the dominant vegetation in CWs because of their ability to overwinter (Nahlik and Mitsch 2006). Details of floating aquatic plant CWs are described by DeBusk and Reddy (1987) and Vymazal et al. (1998). Many studies have documented their ability to remediate various anthropogenic pollutants that include nutrients (Gopal 1987; Vymazal 2007), herbicides (Wilson et al. 2001; Knuteson et al. 2002), heavy metals (Odjegba and Fasidi 2004; Liao and Chang 2004; Padmavathiamma and Li 2007) and antibiotics (Gujarathi et al. 2005). The high rates of biomass production by floating hydrophytes necessitates periodic harvesting to prevent the export of nutrients, particularly P, via vegetative decomposition and to maintain open water areas to permit increased oxygen exchange (Masifwa et al. 2004; Kadlec 2005).

Recently, Polomski et al. (2007) proposed a sustainable nutrient remediation strategy that involves the production of economically important emergent macrophytes in a SSF CW that remediates wastewater runoff. The objective of this study was to take an unconventional approach and determine the ability of water hyacinth, water lettuce, and parrotfeather, to thrive and recover nursery runoff levels of N and P in a similarly constructed laboratory-scale subsurface CW system.

2 Methods

Experimental procedures were similar to those described by Polomski et al. (2007). However, a brief description follows with an emphasis on the experimental setup and nutrient solution treatments.

2.1 Plant Characterization and Culture

This greenhouse study was conducted from 2003–2004 at Clemson University's Biosystems Research Complex (Clemson, SC, USA; latitude 34°40′8″; longitude 82°50′40″). Water hyacinth, water lettuce, and parrotfeather were selected for their remediating ability and their commercial importance as biological filters in water gardens (Speichert and Speichert 2004). Water hyacinth is a free-floating plant comprised of a rosette of petiolate leaves, an attractive purple inflorescence, and extensive submerged roots (Gopal 1987). Despite its free-floating habit, water hyacinth can also root in substrate, which has been postulated as an ancestral trait (Gopal 1987). Water hyacinth rapidly propagates vegetatively and sexually, although vegetative propagation via fragmentation is the primary form of reproduction. The free-floating, stoloniferous water lettuce produces a rosette of light to lime-green velvety leaves; it can reach a mature height of 30.5 cm (Speichert and Speichert 2004). It reproduces by offsets that grow from the base of the mature plant. Dwarf redstemmed parrotfeather is a compact selection with bright red prostrate or ascending stems bearing whorls of gray-green feathery leaves (Speichert and Speichert 2004). This creeping emergent roots freely in floating mats or anchored in substrate where it reproduces primarily by stem fragmentation (Sytsma 1989).

Water lettuce and dwarf red-stemmed parrotfeather (Charleston Aquatic Nursery, Johns Island, SC) were floated in tapwater-filled 3.8 L aquatic pots, fertigated with 20–20–20 (Peter's Professional®) water-soluble fertilizer as needed, and maintained in the greenhouse.

Water hyacinth stock plants were collected from drainage canals near Cape Coral, Florida, USA, transferred to 60 L containers and fertigated as needed with 20–20–20 water soluble fertilizer.

Two to 4 weeks prior to the start of an experiment, 40 to 50 plants were removed from their containers, their roots washed in running tapwater to remove microalgae, and weighed. They were transplanted into the simulated laboratory subsurface CW comprised of two polyethylene pots: a 16.5-cm diameter "azalea" pot filled with pea gravel and placed inside a 16.7-cm diameter aquatic pot (3.8 L pot with no drainage holes) so their rims were even. Single ramets (vegetatively produced plants) of water hyacinth, individual plantlets of water lettuce, and five 14 cm long rooted stem fragments of parrotfeather were planted in each pot. After fitting the azalea pot into the aquatic pot, ~1.350 L of a 10% modified Hoagland's solution (21.57 $mg \cdot L^{-1}$ N and 3.63 $mg \cdot L^{-1}$ P; Hoagland and Arnon 1950) was added to each pot until water appeared at the gravel surface. During the acclimation period, plants were watered every 2 or 3 days to maintain the water level just below the gravel surface. The average daily temperatures, relative humidity, and daily light integral are listed in Table 1. A 16:8 h light/dark photoperiod was maintained during the winter months with 1,000 W metal halide lights.

2.2 Nitrogen and Phosphorus Treatment Solutions

Five treatment levels of 0.1%, 1%, 5%, 10% and 20% modified Hoagland's solution ("Solution 1" using NO_3–N) were prepared and contained the following mean concentrations of N and P ($mg \cdot L^{-1}$): (1) 0.39 N; 0.07 P; (2) 1.75 N; 0.18 P; (3) 10.44; 1.86 P; (4) 21.57 N; 3.63 P; and (5) 36.81 N; 6.77 P. These concentrations encompassed the typical range of nutrients found in nursery CW discharge and nursery runoff, and used in nursery irrigation. The initial pH of the nutrient solution was adjusted to 6.2 with 6 N H_2SO_4.

At the start of the experiment, 30 acclimatized plants and six gravel-only pots were removed from their aquatic containers, flushed with deionized water, and then returned to the aquatic pots that had been emptied and rinsed with deionized water. The appropriate treatment solution was batch-loaded into the pots with plants until it was visible at the gravel surface. Gravel-only pots received 10.44 and 1.86 mg L^{-1} N and P, respectively. Thereafter, nutrient solution was supplied every 2 days to maintain the water level at the gravel surface. Containers were arranged in a randomized complete block design with six replicates. Experiments were replicated twice for each species during the time periods listed in Table 1.

2.3 Plant and Water Analysis

Over the course of each experiment the volume of nutrient solution supplied to each wetland unit was recorded over the 8-week period. When the experiment was terminated, each plant was severed at the gravel surface and the above- and below-ground portions were weighed. The below-ground portions, which included

Table 1 Experiment dates and selected environmental variables (mean ± SE) for the two replicates of each species conducted in the Biosystems Research Complex greenhouses, Clemson University, Clemson, SC

Species	Experiment 1			Experiment 2		
	Temperature (°C)	Relative humidity (%)	Daily light integral (mol m^{-2} d^{-1})	Temperature (°C)	Relative humidity (%)	Daily light integral (mol m^{-2} d^{-1})
Eichhornia crassipes	27.3±0.07[a]	72.3±0.3[a]	25±1[a]	24.5±0.2[b]	61.5±1.1[b]	19±1[b]
Myriophyllum aquaticum	27.3±0.07[c]	74.5±0.3[c]	22±1[c]	22.2±0.2[d]	48.0±1.3[d]	12±1[d]
Pistia stratiotes	24.6±0.2[e]	61.0±1.1[e]	15±1[e]	24.3±0.2[f]	61.0±1.2[f]	16±1[f]

[a] 25-Jun-2003 to 20-Aug-2003

[b] 9-Sept-2003 to 3-Nov-2003

[c] 26-Jun-2003 to 21-Aug-2003

[d] 20-Jan-2004 to 15-Mar-2004

[e] 17-July-2003 to 11-Sep-2003

[f] 24-Oct-2003 to 18-Dec-2003

roots that had grown through the drainage holes of the gravel-filled azalea pots, were placed over a screen and washed with tapwater, rinsed with distilled water, and then weighed. Dried roots and shoots (80°C to constant dry weight) were ground separately in a Wiley Mill® (Thomas Scientific, Swedesboro, NJ) to pass through a 40-mesh (0.425-mm screen). N and P tissue concentrations were determined as described by Polomski et al. (2007). N and P content was calculated by multiplying plant part dry weight by nutrient concentration. Whole plant N and P content was derived from combining above- and below-ground mineral content.

The water that remained in the aquatic pots was sampled and stored at 4°C prior to anion analysis with a Dionex AS50 IC with AS50 autosampler (Dionex Corp., Sunnyvale, CA). Percentage of recovered nutrient was determined with the following equation: (mg N or P supplied − mg nutrient remaining in solution ÷ mg N or P supplied)×100.

2.4 Statistical Analysis

Data from both replicated experiments were pooled because analysis of variance indicated no significant treatment interactions with rep and block. Changes in biomass and nutrient recovery relative to N or P supplied for each species was determined by regression analyses. For each species the analyses indicated significant slope for biomass and nutrient uptake efficiency (i.e., amount of nutrient supplied that is assimilated by the plant). Linear contrasts and F tests compared slopes among the species. Differences between shoot and root concentration means and content means of each species were determined by Student's *t* tests. All analyses were performed with SAS (version 9.1 for Windows; SAS Institute, Cary, NC), and all tests were conducted with α=0.05.

3 Results and Discussion

3.1 Biomass Accumulation

Over the 8-week period the growth rates of the three species increased linearly and were highly correlated with increasing levels of nitrogen and phosphorus (Fig. 1a,b). Due to its higher evapotranspiration rate (Gopal 1987), water hyacinth was supplied with greater amounts of N and P than the other two species, which yielded the highest rate of dry weight accumulation. Water lettuce received the least amount of N and P over the 8-week period but had a similar growth rate to water hyacinth (Fig. 1a,b). Parrotfeather had the lowest growth rate compared to water hyacinth and water lettuce. Gravel-only pots receiving 10.44 and 1.86 mg L^{-1} N and P, respectively, were supplied with 62% to 86% of N and 52% to 86% of P than planted pots receiving the same level of N and P (data not presented).

At the lowest treatment level, all species exhibited visual nutrient deficiency symptoms that included marginal to complete foliar necrosis, chlorotic, senescent leaves, and spindly growth. Some water hyacinths produced inflorescences, which was not unexpected since water hyacinth has been reported to survive and grow under a wide range of water nutrient concentrations as low as 0.05 mg L^{-1} nitrogen supplied either as nitrate (Shiralipour et al. 1981) or ammonia (Tucker 1981) and 0.1 mg L^{-1} P, which Haller et al. (1970) determined as the lower critical level for growth of water hyacinth in a hydroponic environment.

3.2 N and P Recovery

Nitrogen and phosphorus recovery rates of the three species were evaluated by comparing the amount of N or P supplied and assimilated in whole plant tissues to an optimal recovery rate where the amount of N or P supplied equaled the amount of N or P recovered in the tissues. Nitrogen and P content of whole plant tissues for all three species increased linearly with increasing concentrations of N and P and was highly correlated with the amount supplied to each species (Fig. 2a,b). The N recovery rate of water hyacinth and water lettuce was similar to the optimal recovery rate of N (mg N supplied = mg N in tissues) and higher than the N assimilation rate of dwarf red-stemmed parrotfeather (Fig. 2a). Since nutrient recovery is a product of biomass and tissue nutrient concentration, the low biomass productivity of parrotfeather contributed to its lower N assimilative rate. Compared to similar studies with herbaceous emergent aquatic plants, water hyacinth and water lettuce had N uptake efficiencies similar to Louisiana iris hybrid 'Full Eclipse,' *Pontederia cordata* 'Singapore Pink,' *Thalia geniculata* f. *rheumoides* Shuey, *Rhyncospora colorata* (L.) H. Pfeiffer, and *Oenenathe javanica* (Blume) DC. 'Flamingo' (Polomski et al. 2007, 2008).

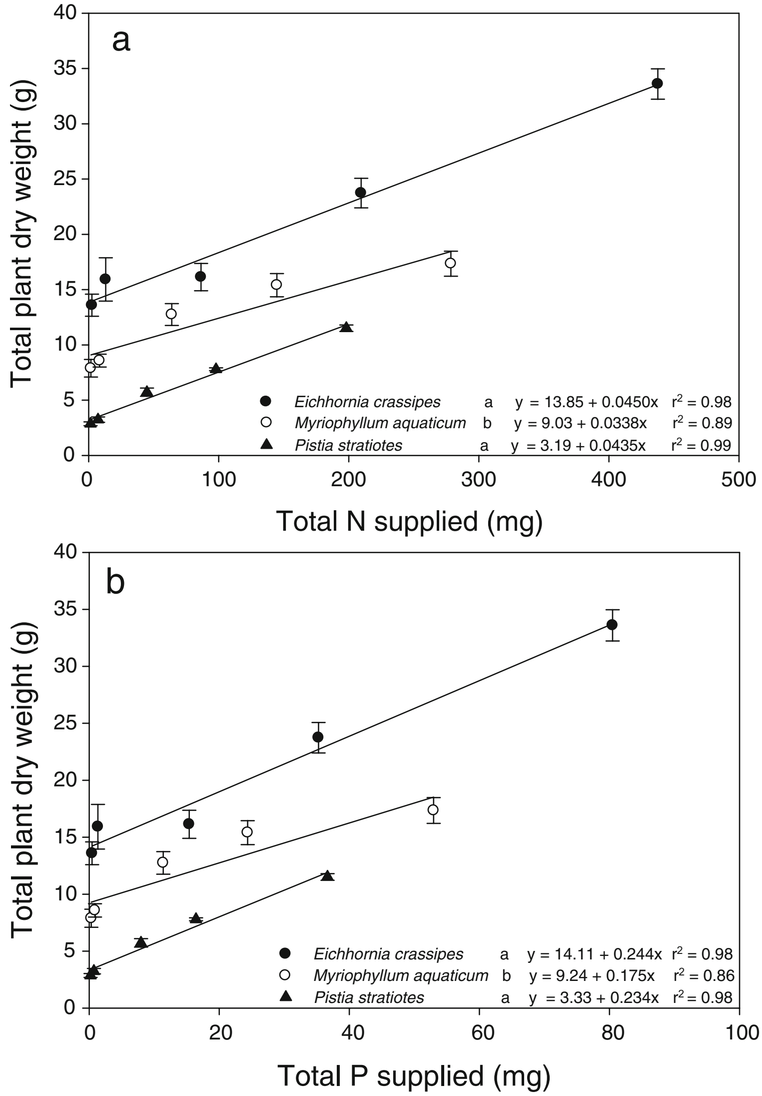

Fig. 1 The effect of N (**a**) and P (**b**) on whole plant dry weight of three, greenhouse-grown floating hydrophytes growing in a laboratory scale subsurface flow constructed wetland over an 8-week period. Five concentrations of modified Hoagland's solution [N and P ($mg{\cdot}L^{-1}$): (1) 0.39 N; 0.07 P; (2) 1.75 N; 0.18 P; (3) 10.44; 1.86 P; (4) 21.57 N; 3.63 P; and (5) 36.81 N; 6.77 P] were initially batch-loaded and then supplied every 2 days to maintain the water level at the gravel surface. *Vertical bars* represent standard error of N or P content. *Data points* are means of 12 plants. Slopes of regression lines were compared using linear contrasts and *F* tests; *species with different letters* have significantly different slopes ($P \leq 0.05$)

None of the three species had P assimilation rates that were similar to the optimal P recovery rate (Fig. 2b). The P recovery rates were similar for water hyacinth, parrotfeather, and water lettuce. Their P recovery rates were similar to *Canna* x *generalis* Bailey (pro sp.) 'Bengal Tiger,' *Peltandra virginica* (L.) Schott, *Pontederia cordata* L. 'Singapore Pink,' and *Thalia geniculata* f. *rheumoides* (Polomski et al. 2007, 2008). Phosphorus uptake by these floating macrophytes may have been affected by the N/P ratio of treatment solutions (Reddy et al. 1989, 1990; Jayaweera and Kasturiarachchi 2004) or the pea gravel medium, which may have altered root architecture and P acquisition.

An analysis of the water that remained in the pots after 8 weeks revealed no significant differences between species and treatment levels in the concentration of leftover N and P. Less than 4% and 7% of the original amount of N and P supplied to the plants, respectively, was detected in the remaining solution (data not shown). Of the original amount of N and P supplied to gravel-only pots, 38% to 48% N and 22% to 58% P remained (data not shown). Depletion of P in the gravel-only pots could have resulted from assimilation by the thin film of algae present near the gravel surface and from microorganisms in biofilm. N depletion may have occurred via denitrification processes. It is unlikely that P precipitation occurred

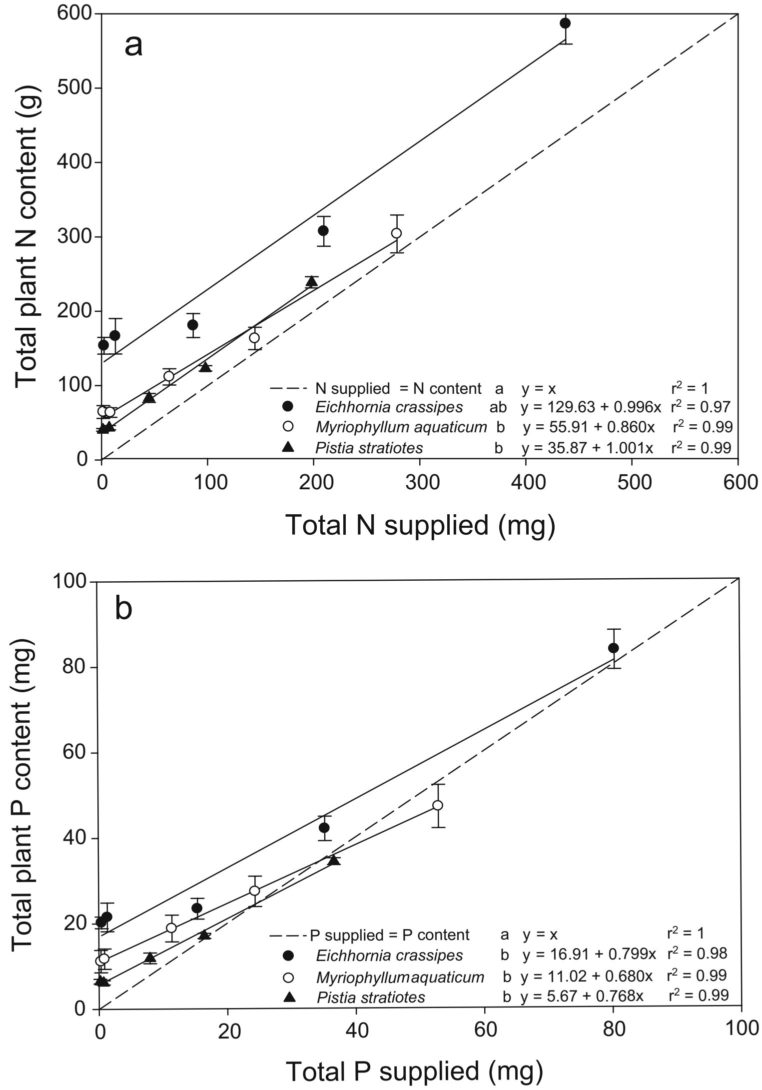

Fig. 2 Nitrogen (**a**) and phosphorus (**b**) recovered in whole plant tissues of three greenhouse-grown floating hydrophytes growing in a laboratory scale subsurface flow constructed wetland over an 8-week period. Five concentrations of modified Hoagland's solution [N and P ($mg \cdot L^{-1}$): (1) 0.39 N; 0.07 P; (2) 1.75 N; 0.18 P; (3) 10.44; 1.86 P; (4) 21.57 N; 3.63 P; and (5) 36.81 N; 6.77 P] were initially batch-loaded and then supplied every 2 days to maintain the water level at the gravel surface. *Vertical bars* represent standard error of N or P content. *Data points* are means of 12 plants. *Dashed line* represents hypothetical 100% recovery rate. Slopes of regression lines were compared using linear contrasts and *F* tests; species with different letters have significantly different slopes ($P \leq 0.05$)

in the gravel-only pots because the pH was not alkaline enough (mean pH of 7.1) to promote precipitation of insoluble tricalcium-phosphate [$Ca_3(PO_4)_2$] complexes (Richardson 1985). Using Visual Minteq 2.52, a chemical equilibrium computer program that calculates the speciation, solubility, and equilibrium of solid and dissolved phases of minerals in aqueous systems, further confirmed that P precipitation was an unlikely transformation pathway for P removal from the nutrient solution (Gustafsson 2008).

3.3 Nitrogen and Phosphorus Tissue Concentration

Mineral concentrations are typically reported in wetland plant nutrient recovery research, although the contents or the weights of nutrients reflect differences in nutrient accumulation by plants. As expected, the differences in allocation of the nutrients to shoot and roots within species varied by the method in which the results were expressed, i.e., concentration vs. content. N concentration of water hyacinth shoots was greater than roots at the two highest N treatment levels (Table 2). A similar trend was observed with P as water hyacinth shoots exhibited a higher sink strength with increasing P treatment levels. Allocation of N and P to above- rather than below-ground water hyacinth parts with increasing N and P levels has been observed by other researchers in free-floating hydroponic experiments (Shiralipour et al. 1981; Reddy and Tucker 1983; Xie

Table 2 Mean (n=12) nitrogen (N) and phosphorus (P) concentration and content of shoots and roots of three floating hydrophytes grown for 8 weeks in a laboratory-scale subsurface constructed wetland and receiving five treatment levels of N or P incorporated in a modified Hoagland's nutrient solution containing all other nutrients at levels to support normal plant growth

Treatment level (mg L^{-1})		Concentration (mg g^{-1})				Content (mg)			
		N		P		N		P	
N	P	Shoot	Root	Shoot	Root	Shoot	Root	Shoot	Root
Eichhornia crassipes									
0.39	0.07	11.51	10.70	1.54**	1.29	134.400**	19.144	17.954**	2.287
1.75	0.18	10.16	10.19	1.30	1.18	143.480**	22.865	18.804**	2.708
10.44	1.86	11.29	10.38	1.45**	1.26	158.060**	22.749	20.647**	2.763
21.57	3.63	13.042*	11.55	1.78	1.67	284.000**	23.470	38.617**	3.429
36.81	6.77	17.78**	13.08	2.53**	1.71	559.790**	26.784	80.134**	3.502
Myriophyllum aquaticum									
0.39	0.07	8.45	7.39	1.19	1.31	48.538**	15.935	7.304	3.865
1.75	0.18	7.59	7.48	1.23	1.33	43.174**	20.568	6.968	4.775
10.44	1.86	9.28**	7.52	1.45	1.26	84.083**	27.607	13.323**	5.505
21.57	3.63	11.30*	9.28	1.81*	1.41	130.990**	32.313	21.189**	6.211
36.81	6.77	18.15	16.76	2.73*	2.18	257.480**	46.522	38.721**	8.397
Pistia stratiotes									
0.39	0.07	13.93	16.46	2.20	2.66	33.011**	7.472	5.401**	1.097
1.75	0.18	13.19	15.97**	1.88	2.04	34.102**	9.263	5.053**	1.137
10.44	1.86	14.14	16.78**	1.96	2.33*	68.688**	14.718	9.843**	2.039
21.57	3.63	15.75	16.36	2.18	2.22	107.720**	15.392	14.927**	2.111
36.81	6.77	21.08	19.08	3.07**	2.35	217.790**	21.090	31.736**	2.624

Treatments were initially batch-loaded and then supplied every 2 days to maintain the water level at the gravel surface.

*$P \leq 0.05$, **$P \leq 0.01$, mean separation by t test comparing N and P in shoots and roots within species at each treatment level with significant differences

et al. 2004). Agami and Reddy (1990) also found N concentration of water hyacinth shoots was ~2-fold higher than roots, but P accumulation was evenly distributed between roots and shoots. Our water hyacinth total N and P concentrations were comparable to values reported in dairy lagoon wastewater (DeBusk et al. 1995; Tripathi and Upadhyay 2003) and free-floating hydroponic studies (Boyd 1976; Tucker and DeBusk 1981).

Parrotfeather shoot N was higher than root N at 10.44 and 1.86 mg L^{-1} N and P, respectively, and 21.57 and 3.63 mgL^{-1} N and P, respectively, suggesting N partitioning to shoots with increasing N treatment levels. N concentration of dwarf red-stemmed parrotfeather at the highest treatment level was comparable to the N concentration of field-collected parrotfeather sampled from natural stands growing in agricultural drainage canals or from creeks and pools receiving agricultural runoff in central California (Rejmankova 1992). Phosphorus concentration in parrotfeather was higher in shoots rather than roots at the two highest treatment levels, which indicated an increasing allocation of P to shoots than to roots with increasing P levels.

No N concentration trend was evident with water lettuce, but P concentration indicated a greater allocation of P to shoots with increasing P treatment levels. Shoot P was greater in shoots than roots at 10.44 and 1.86 mg·L^{-1} N and P, respectively, and at the highest treatment level. Our N and P concentrations in water lettuce were similar to other studies (Tucker and DeBusk 1981; Agami and Reddy 1990) and in CWs (Greenway and Woolley 1999).

N concentration was greater in water lettuce than water hyacinth (data not presented), which was similar to other studies (Tucker 1981; Reddy and DeBusk 1985). We attribute the difference to N dilution caused by water hyacinth's growth rate—

among the highest of any plant known (Gopal 1987); its greater biomass production diluted N assimilated by water hyacinth. Contrary to these findings, Aoi and Hayashi (1996) reported greater (~1.5 times) N and P concentrations in water hyacinth than water lettuce in an outdoor study in Japan involving a continuous flow and batch culture system. Upadhyay et al. (2007) reported “initial” P concentrations of water hyacinth and water lettuce that were 1.3- and 2-fold greater in leaves and roots, respectively, and 1.8- and 3.5-fold higher in water lettuce leaves and roots, respectively, compared to our highest treatment level, but “initial” N concentrations were comparable to ours. The discrepancies in N and P concentration could have resulted from variations in experimental design that includes plant density, temperature, duration of the experiment, solar radiation, and the concentration and ratio of nutrients. The effect of mechanical impedance by the pea gravel substrate on root architecture and nutrient absorption warrants further investigation.

3.4 Nitrogen and Phosphorus Tissue Content

Nitrogen content (plant dry weight × tissue N concentration) of the three species was higher in shoots than roots at every treatment level. Water hyacinth allocated ≥86% N to shoots compared to roots (Table 2). Greatest amount of assimilated N was in water lettuce and parrotfeather shoots (≥78% and ≥59%, respectively) than roots. This dominant sink strength of shoots at every N treatment level was observed in the marginal aquatic garden plants Louisiana iris hybrid ‘Full Eclipse,’ *Pontederia cordata* L. ‘Singapore Pink,’ *Oenenathe javanica* (Blume) DC. ‘Flamingo,’ *Phyla lanceolata* (Michx.) Greene, *Rhyncospora colorata* (L.) H. Pfeiffer, and *Thalia geniculata* f. *rheumoides* Shuey (Polomski et al. 2007, 2008).

Phosphorus content was also greatest in above-ground organs at every treatment level for the three species. Water hyacinth, water lettuce, and parrot-feather shoots contained ≥87%, 79%, and 59% P, respectively, compared to roots. Also, we observed this partitioning of P to shoots instead of roots with increasing levels of P in *Canna* x *generalis* Bailey (pro sp.) ‘Bengal Tiger’ and *Colocasia esculenta* (L.) Schott var. *antiquorum* (Schott) Hubbard and Rehd. ‘Illustris (Polomski et al. 2007).

The potential application of water hyacinth, parrotfeather, and water lettuce for nutrient attenuation of nursery/greenhouse wastewater must be tempered by their well-documented reputations as noxious weeds in certain regions and ecosystems. Water hyacinth, in particular, possesses a dichotomous nature: one of the world’s worst weeds that devastates environmental systems, but demonstrates substantive phytoremediating ability (Holm et al. 1997; Mehra et al. 1999).

4 Conclusions

Over an 8-week period water hyacinth, water lettuce, and parrotfeather thrived in a gravel-based, laboratory-scale subsurface CW receiving nursery runoff levels of N and P. Nitrogen uptake efficiency was highest in water hyacinth, and N content was greatest in above-ground tissues at every treatment level. Phosphorus recovery rates were similar for the three species and P was preferentially stored in shoots.

Similar to the recommendations of Hadad and Maine (2007), our study supports the possibility of integrating floating aquatic macrophytes with emergent macrophytes in a self-contained polycultural SSF CW system that can be used to remediate runoff from nursery and greenhouse operations. Floating macrophytes may have an important role in greenhouse production in temperate areas where they can be cultivated indoors in SSF CWs to assimilate NO_3^-, and soluble PO_4^{3-}, and heavy metal trace elements, which are often applied year-round (Biernbaum 1992). In addition, their ability to process high volumes of nutrient-rich water reduces the amount of effluent that has to be discarded.

Acknowledgments This work received financial support through the Floriculture and Nursery Research Initiative for Environmental and Resource Management Practices and Strategies, USDA Agriculture Research Service Ft. Pierce, Florida and is gratefully acknowledged. Technical contribution no. 5386 of the Clemson University Experiment Station. The authors wish to thank Sarah White, Deidre Jones, and Robby Taylor for their invaluable assistance, and Carolina Nurseries Inc. and Fafard Inc. for their donations of plants and soilless media.

References

Agami, M., & Reddy, K. R. (1990). Competition for space between *Eichhornia crassipes* (Mart.) Solms and *Pistia* *stratiotes* L. cultured in nutrient-enriched water. *Aquatic Botany*, *38*, 195–208. doi:10.1016/0304-3770(90)90005-6.

Alexander, S. (1993). Pollution control and prevention at containerized nursery operations. *Water Science and Technology*, *28*, 509–517.

Aoi, T., & Hayashi, T. (1996). Nutrient removal by water lettuce (*Pistia stratiotes*). *Water Science and Technology*, *34*, 7–8. doi:10.1016/S0273-1223(96)00772-X.

Arnold, M. A., Lesikar, B. J., Kenimer, A. L., & Wilkerson, D. C. (1999). Spring recovery of constructed wetland plants affects nutrient removal from nursery runoff. *Journal of Environmental Horticulture*, *17*, 5–10.

Berghage, R. D., MacNeal, E. P., Wheeler, E. F., & Zachritz, W. H. (1999). "Green" water treatment for the green industries: opportunities for biofiltration of greenhouse and nursery irrigation water and runoff with constructed wetlands. *HortScience*, *34*, 50–54.

Biernbaum, J. A. (1992). Root-zone management of greenhouse container-grown crops to control water and fertilizer use. *HortTechnology*, *2*, 127–132.

Boyd, C. E. (1976). Accumulation of dry matter, nitrogen and phosphorus by cultivated water hyacinths. *Economic Botany*, *30*, 51–56.

Carpenter, S. R., Caraco, N. F., Correll, D. L., Howarth, R. W., Sharpley, A. N., & Smith, V. H. (1998). Nonpoint source pollution of surface waters with phosphorus and nitrogen. *Ecological Applications*, *8*, 559–568. doi:10.1890/1051-0761(1998)008[0559:NPOSWW]2.0.CO;2.

DeBusk, T. A., Dierberg, F. E., & Reddy, K. R. (1995). Use of aquatic and terrestrial plants for removing phosphorus from dairy wastewaters. *Ecological Engineering*, *5*, 371–390. doi:10.1016/0925-8574(95)00033-X.

DeBusk, T. A., & Reddy, K. R. (1987). Wastewater treatment using floating aquatic macrophytes; contaminant removal processes and management strategies. In K. R. Reddy, & W. H. Smith (Eds.), *Aquatic plants for water treatment and resource recovery* (pp. 643–656). Orlando: Magnolia.

Gopal, B. (1987). *Water hyacinth*. New York: Elsevier.

Greenway, M., & Woolley, A. (1999). Constructed wetlands in Queensland: Performance efficiency and nutrient bioaccumulation. *Ecological Engineering*, *12*, 39–55. doi:10.1016/S0925-8574(98)00053-6.

Gujarathi, N. P. B., Haney, J., & Linden, J. C. (2005). Phytoremediation potential of *Myriophyllum aquaticum* and *Pistia stratiotes* to modify antibiotic growth promoters, tetracycline, and oxytetracycline in aqueous wastewater systems. *International Journal of Phytoremediation*, *7*, 99–112. doi:10.1080/16226510590950405.

Gustafsson, J. P. (2008). Visual Minteq, ver. 2.52. Dept. of Land and Water Resour. Eng., Stockholm. Retrieved January 25, 2008, from http//www.lwr.kth.se/English/OurSoftware/vminteq/.

Hadad, H. R., & Maine, M. A. (2007). Phosphorus amount in floating and rooted macrophytes growing in wetlands from the Middle Parana River floodplain (Argentina). *Ecological Engineering*, *31*, 251–258. doi:10.1016/j.ecoleng.2007.08.001.

Haller, W. T., Knipling, E. B., & West, S. H. (1970). Phosphorus absorption by and distribution in water hyacinths. *Proceedings—Soil and Crop Science Society of Florida*, *30*, 64–68.

Hoagland, D. R., & Arnon, D. I. (1950). The water-culture method for growing plants without soil. Calif. Agr. Exp. Sta. Circ. 347.

Holm, L. G., Plucknett, D. L., Pancho, J. V., & Herberger, J. P.(1977). *The World's Worst Weeds: Distribution and Biology*. 18th ed. (Honolulu, HI: University Press Publications).

James, E. A. (1995). Water quality of stored and runoff water in plant nurseries and implications for recycling. *Combined Proceedings International Plant Propagators'. Society*, *45*, 117–120.

Jayaweera, M. W., & Kasturiarachchi, J. C. (2004). Removal of nitrogen and phosphorus from industrial wastewaters by phytoremediation using water hyacinth (*Eichhornia crassipes* [Mart.] Solms). *Water Science and Technology*, *50*, 217–225.

Kadlec, R. H. (2005). Phosphorus removal in emergent free surface wetlands. *Journal of Environmental Science and Health. Part A, Toxic/Hazardous Substances & Environmental Engineering*, *40*, 1293–1306. doi:10.1081/ESE-200055832.

Knuteson, S. L., Whitwell, T., & Klaine, S. J. (2002). Influence of plant age and size on simazine toxicity and uptake. *Journal of Environmental Quality*, *31*, 2096–2103.

Liao, S., & Chang, W. (2004). Heavy metal phytoremediation by water hyacinth at constructed wetlands in Taiwan. *Journal of Aquatic Plant Management*, *42*, 60–68.

Masifwa, W. F., Okello, W., Ochieng, H., & Ganda, E. (2004). Phosphorus release from decomposing water hyacinth and effects of decomposition on water quality. *Uganda Journal of Agricultural Sciences*, *9*, 389–395.

Mehra, A., Farago, M. E., Banerjee, D. K., & Cordes, K. B. (1999). The water hyacinth—an environmental friend or pest? A review. *Resource and Environmental Biotechnology*, *2*, 255–281.

Nahlik, A. M., & Mitsch, W. J. (2006). Tropical treatment wetlands dominated by free-floating macrophytes for water quality improvement in Costa Rica. *Ecological Engineering*, *28*, 246–257. doi:10.1016/j.ecoleng.2006.07.006.

Odjegba, V. J., & Fasidi, I. O. (2004). Accumulation of trace elements by *Pistia stratiotes*: Implications for phytoremediation. *Ecotoxicology (London, England)*, *13*, 637–646. doi:10.1007/s10646-003-4424-1.

Padmavathiamma, P. K., & Li, L. Y. (2007). Phytoremedation technology: hyper-accumulation metals in plants. *Water, Air, and Soil Pollution*, *184*, 105–126. doi:10.1007/s11270-007-9401-5.

Polomski, R. F., Taylor, M. D., Bielenberg, D. G., Bridges, W. C., Klaine, S. J., & Whitwell, T. (2007). Nutrient recovery by seven aquatic garden plants in a laboratory-scale subsurface constructed wetland. *HortScience*, *42*, 1674–1680.

Polomski, R. F., Taylor, M. D., Bielenberg, D. G., Bridges, W. C., Klaine, S. J., & Whitwell, T. (2008). Differential nitrogen and phosphorus recovery by five aquatic garden species in laboratory-scale subsurface constructed wetlands. *HortScience*, *43*, 868–874.

Reddy, K. R., & Tucker, J. C. (1983). Productivity and nutrient uptake of water hyacinth, *Eichhornia crassipes*. I. Effect of nitrogen source. *Economic Botany*, *37*, 237–247.

Reddy, K. R., & DeBusk, W. F. (1985). Nutrient removal potential of selected aquatic macrophytes. *Journal of Environmental Quality*, *14*, 459–462.

Reddy, K. R., Agami, M., & Tucker, J. C. (1989). Influence of nitrogen supply rates on growth and nutrient storage by water hyacinth (*Eichhornia crassipes*) plants. *Aquatic Botany*, *36*, 33–43. doi:10.1016/0304-3770(89)90089-2.

Reddy, K. R., Agami, M., & Tucker, J. C. (1990). Influence of phosphorus on growth and nutrient storage by water hyacinth (*Eichhornia crassipes* (Mart.) Solms) plants. *Aquatic Botany*, *37*, 355–365. doi:10.1016/0304-3770(90)90021-C.

Rejmankova, E. (1992). Ecology of creeping macrophytes with special reference to *Ludwigia peploides* (H.B.K.) Raven. *Aquatic Botany*, *43*, 283–299. doi:10.1016/0304-3770(92)90073-R.

Richardson, C. J. (1985). Mechanisms controlling phosphorus retention capacity of freshwater wetlands. *Science*, *228*, 1424–1427. doi:10.1126/science.228.4706.1424.

Shiralipour, A., Garrard, L. A., & Haller, W. T. (1981). Nitrogen source, biomass production, and phosphorus uptake in waterhyacinth. *Journal of Aquatic Plant Management*, *19*, 40–43.

Speichert, G., & Speichert, S. (2004). *Encyclopedia of water garden plants*. Portland: Timber.

Sytsma, M. (1989). A study of growth, resource allocation and nutrient requirements of *Myriophyllum aquaticum*. Technical Progress Report for USGA Grant No. 14-08-0001-G1626. University of California: Davis, CA, 32 pp.

Taylor, M. D., White, S. A., Chandler, S. L., Klaine, S. J., & Whitwell, T. (2006). Nutrient management of nursery runoff water using constructed wetland systems. *Hort-Technology*, *16*, 610–614.

Tripathi, B. D., & Upadhyay, A. R. (2003). Dairy effluent polishing by aquatic macrophytes. *Water, Air, and Soil Pollution*, *143*, 377–385. doi:10.1023/A:1022813125339.

Tucker, C. S. (1981). The effect of ionic form and level of nitrogen on the growth and composition of *Eichhornia crassipes* (Mart.) Solms. *Hydrobiologia*, *83*, 517–522. doi:10.1007/BF02187049.

Tucker, C. S., & DeBusk, T. A. (1981). Productivity and nutritive value of *Pistia stratiotes* and *Eichhornia crassipes*. *Journal of Aquatic Plant Management*, *19*, 61–63.

Upadhyay, A. R., Mishra, V. K., Pandey, S. K., & Tripathi, B. D. (2007). Biofiltration of secondary treated municipal wastewater in a tropical city. *Ecological Engineering*, *30*, 9–15. doi:10.1016/j.ecoleng.2006.09.004.

U. S. Environmental Protection Agency (EPA). (1986). Quality criteria for water. EPA Rpt. 440/5-86-001. U.S. EPA Office of Water Regulations and Standards. U. S. Gov. Print. Office (PB87-226759), Washington, D. C.

Vymazal, J., Brix, H., Cooper, P. F., Haberl, R., Perfler, R., & Laber, J. (1998). Removal mechanisms and types of constructed wetlands. In J. Vymazal, H. Brix, P. F. Cooper, M. B. Green, & R. Haberl (Eds.), *Constructed wetlands for wastewater treatment in Europe* (pp. 17–66). Leiden, The Netherlands: Backhuys.

Vymazal, J. (2007). Removal of nutrients in various types of constructed wetlands. *Science of the Total Environment*, *380*, 48–65.

Wilson, P. C., Whitwell, T., & Klaine, S. J. (2001). Simazine toxicity and uptake by parrotfeather. *Journal of Aquatic Plant Management*, *39*, 112–117.

Wood, S. L., Wheeler, E. F., Berghage, R. D., & Graves, R. E. (1999). Temperature effects on wastewater nitrate removal in laboratory-scale constructed wetlands. *American Society of Agricultural Engineers*, *42*, 185–190.

Xie, Y., Wen, M., Yu, D., & Li, Y. (2004). Growth and resource allocation of water hyacinth as affected by gradually increasing nutrient concentrations. *Aquatic Botany*, *79*, 257–266.

Yeager, T. H., Wright, R., Fare, D., Gilliam, C., Johnson, J., Bilderback, T., & Zondag, R. (1993). Six state survey of container nursery nitrate nitrogen runoff. *Journal of Environmental Horticulture*, *11*, 206–208.