Phosphorus Removal and Accumulation by Swiss chard (*Beta vulgaris*) Grown in Floating Treatment Wetlands

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Significance to Industry: Hyper-eutrophication occurs when surface waters are enriched with excessive nutrients and results in excessive primary production or algal and cyanobacteria blooms. The overabundance and decomposition of these organisms leads to decreased water clarity, hypoxic conditions, and fish kills. Phosphorous (P) availability typically limits the rate of eutrophication in freshwater systems. Some growers apply more P than is necessary to grow healthy crops. Irrigation and rain move excess P into surface water flows, where it may promote the rate of eutrophication. Phosphorus is essential to all life forms, and technologies that help prevent eutrophication and/or help recycle P are needed. We examined the potential of floating treatment wetlands established with *Beta vulgaris* (Swiss chard) to remediate P from simulated nursery runoff. Swiss chard displays low to moderate potential for P remediation. Yet if P accumulation can be paired with a saleable end product, use of FTW can reduce environmental impacts while enhancing grower profitability.

Nature of Work: Floating treatment wetlands are one option for remediating excess nutrients from water. In floating treatment wetlands, plants are placed directly into floating mats with roots extending directly into the water column. Floating treatment wetlands (FTWs) provide similar benefits as constructed and natural wetlands, namely filtration of particulates and removal of excess nutrients. However, FTWs are somewhat less costly than constructed wetlands and are more adaptable, as FTWs are not permanent installations, require no additional land, and plant choice is not restricted to aquatic macrophytes alone. Plants can be chosen for their nutrient removal characteristics, aesthetic quality, or even edibility.

Research by Glenn et al. (1) indicated that Golden Canna (*Canna flaccida*) grown in a FTW is capable of aiding P removal from simulated nursery runoff. The purpose of this study was to assess the potential for a common vegetable crop, Swiss chard (*Beta vulgaris*), to remove P from simulated nursery runoff by evaluating the influence of cultivar and nutrient load on plant growth, phosphorus uptake, and phosphorus removal.

Swiss chard plants used in this experiment were either started from seed or procured as mature plugs (*Beta vulgaris* 'Bright Lights') from Hi Cotton Greenhouses, (St. Matthews, SC). On 6 March 2013, all plants were of sufficient size to incorporate into the floating treatment wetlands and were individually placed in 5 cm diameter aerator cups, which were then seated within a 1 cm thick, solid core foam mat (60 cm x 60 cm) precut with 10 holes for each plant per experimental unit. Swiss chard plants were grown in the floating treatment wetlands for 9 weeks before they reached mature (edible size) and were harvested on 7 May 2013. Sampling was initiated 13 March 2013, and recurred on a weekly basis (for 8 weeks) until harvest.

The controlled experimental factors of fertilizer load (moderate and high) and species mix [*B. vulgaris* (SCS), *B. vulgaris* 'Bright Lights' (SCB), and a 50% SCS and 50% SCB mixed planting (SCM)] were subdivided among experimental units (EUs), which consisted of 16, 380-liter (100 gal.) Rubbermaid® tanks. Moderate (12.6 g) and high (24.6 g) fertilizer treatments were assigned across EUs (8 per level) using a 15-5-15 commercial grade soluble fertilizer (The Scotts Company LLC, Marysville, OH). Within the moderate treatment, 3 EUs were assigned to SCB, 4 to SCS, and 1 to SCM; within the high treatment 3 EUs were assign to SCB and SCS, with 2 EUs assigned to SCM.

The sixteen EUs were individually spiked with fertilizers, drained, refilled, and respiked on a weekly basis, to simulate a static system with a 7-day hydraulic retention time. Approximately 7-days after each spiking event, water samples were collected, and dissolved oxygen (DO, mg/L), pH, oxidation-reduction potential (ORP, mVolts), and temperature (ºC) measurements were recorded. Water samples were analyzed for dissolved anion and mineral content. Anions concentrations (NO_3-N, NO_2-N, NH_3-N, PO_4-P, and SO4-S) were determined using a Dionex AS50 ion chromatograph (Dionex Corp., Sunnyvale, CA), and mineral concentrations (total P, K, Ca, Mg, Zn, Cu, Mn, Fe, S, Na, B, Al) were analyzed via inductively coupled plasma emission spectrophotometer (ICP-ES, 61E Thermo Jarrell Ash, Franklin, MA).

Measurements of pH, ORP, and temperature were measured with a YSI Professional Plus meter, and DO was measured using a YSI Pro ODO meter (YSI Incorporated, Yellow Springs, OH). Shoot height (cm) and root length (cm) were measured on a weekly basis. On the first day of sampling, three plants from each EU were chosen for repeated measurement of plant growth over the duration of the experiment. On 7 May 2013, after final plant measures were made, three plants were harvested from each EU. The fresh weight (g) and dry weight (g) for the roots and shoots of each plant were recorded. Plants were submitted to the Clemson Agriculture Service laboratory for tissue analyses. Data were analyzed using the GLM procedure and when appropriate, means separated using the Students *t* –test (α = 0.05) within JMP v10.0 (SAS Institute Inc. Cary, NC).

Results and Discussion: Swiss chard established in FTWs removed P from simulated nursery runoff (Figure 1). Effluent P concentrations were lower than influent levels for both high and moderate fertilizer loads (Figure 1). The concentration of P was greatest in shoot tissue for both high and moderate fertilizer loads ($P < 0.0001$; Table 1, Figure 2). Shoot tissue comprised the majority of total plant tissue mass, and so greater accumulation of P in shoot tissues was expected. Harvest of plant biomass from floating treatment wetlands may be an important aspect of their management, because removing nutrients from the internal cycle of the water body helps to reduce primary productivity. However, tissue allocations differ among plant species. White and Cousins 2013 (unpublished data) observed P and nitrogen fixation was greatest in the below-mat biomass of their two plants of study, *Canna flaccida* and *Juncus effusus*. Consequently, for effective nutrient remediation, harvesting the whole plant was recommended. These results, which differ from our current study, indicate that plant choice may dictate management of FTWs.

All three species mixes grew well over the 8 weeks of sampling (Figure 3). Static shoot growth during the first few weeks of the experiment, for all species mixes, may be an indication of a shift in resource allocation to root growth initially, as roots grew consistently over the first 3 weeks (Figure 4). During this time the plants were acclimating to their new environment, as consistent shoot growth was recorded over the remaining weeks. 'Bright Lights' (SCB) was significantly shorter than the straight species (SCS) or a mix of SCB and SCS over the experiment ($P < 0.0001$; Table 2). However, these differences in shoot height could be an artifact of the method used for measurement. Shoot growth was measured from the top of the aerator cup to the top of the tallest standing leaf. Shoots and leaves were not extended vertically for measurements of height. As leaf and stalk growth continued, some plants lost height as their leaves and stalks sagged from their own weight. When additional experiments are conducted with these species, or other plants with similar growth habits, plant leaves should be extended fully for measurement, as this will likely result in a more accurate measurement of shoot growth and correlate more accurately with mass allocation to either shoot or root biomass.

Root lengths were longer for species spiked with the moderate fertilizer load (Table 3). One explanation for this partitioning of metabolic energy towards root growth is that plants residing in nutrient rich environments have no need to expend energetic resources to extend their roots in search of nutrients, which are readily available to them. For example, canna exposed to low nutrient concentrations drive nutrient accumulation into root tissue, as metabolic resources are directed to these organs for growth (2). In a study which analyzed allocation of stem, leaf, and root biomass of twenty seven herbaceous clonal plants grown at either a high or low nutrient environment, Müller et al. (3) reported that the fraction of biomass in roots was typically greater for plants grown in the low nutrient environment. Future experiments may focus on exposing Swiss chard plants to nutrient poor environments and determining if partitioning patterns change to greater loading of P within root tissues rather than shoot tissues. Understanding these changes in partitioning in nutrient rich and nutrient poor environments could alter floating treatment wetland management for species that concentrate nutrients in shoot tissue. If such a perennial species were used in an environment where influent nutrient levels fluctuate seasonally from high to low concentrations, whole plant harvesting may be crucial for effective nutrient remediation, due to nutrient accumulation in root tissue during times of low influent nutrient levels.

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Literature Cited

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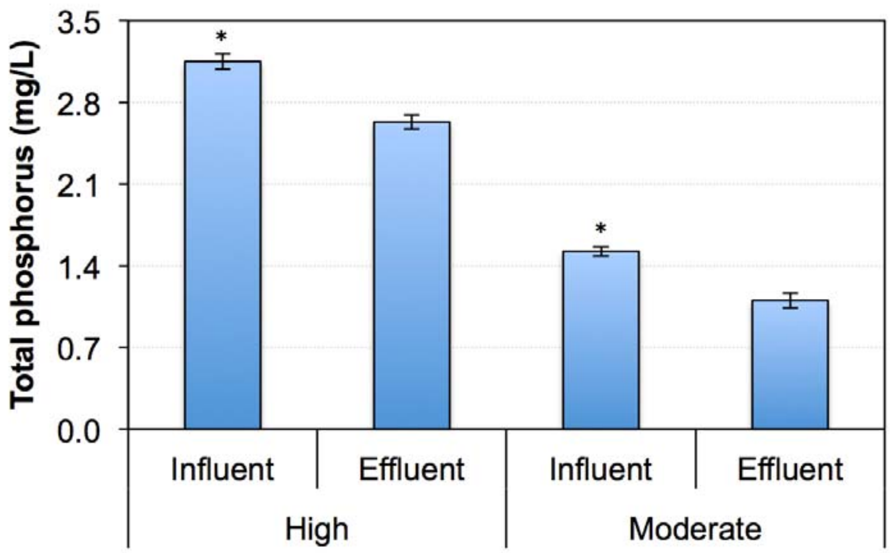

Figure 1: Change in total P concentration of influent and effluent as influenced by floating treatment wetlands established with Swiss chard (*Beta vulgaris*). High and Moderate fertilizer loads were refreshed weekly and the experiment was conducted over an 8 week period. Bars represent mean values for influent and effluent P concentration over time ± the standard error of the mean. The * designates a significant difference between influent and effluent values (p = < 0.0001) for both Moderate and High fertilizer treatments.

Table 1: Changes in P accumulation (g/m^2/8 weeks) and allocation within Swiss chard (*Beta vulgaris*), grown in floating treatment wetlands over an 8-week period, as influenced by fertilizer (2, high, moderate), species mix (3, *B. vulgaris* 'Bright Lights'; *B. vulgaris*; and an even mix of *B. vulgaris* and *B. vulgaris* 'Bright Lights'), and organ (2, shoot, root).

3-Way ANOVA	DF	*P > F*
Fertilizer	1	< 0.0001
Species mix	2	0.0624
Organ	1	< 0.0001
Fertilizer x species mix	2	0.2205
Fertilizer x organ	1	< 0.0001
Species mix x organ	2	0.1255
Fertilizer x species mix x organ	2	0.5439

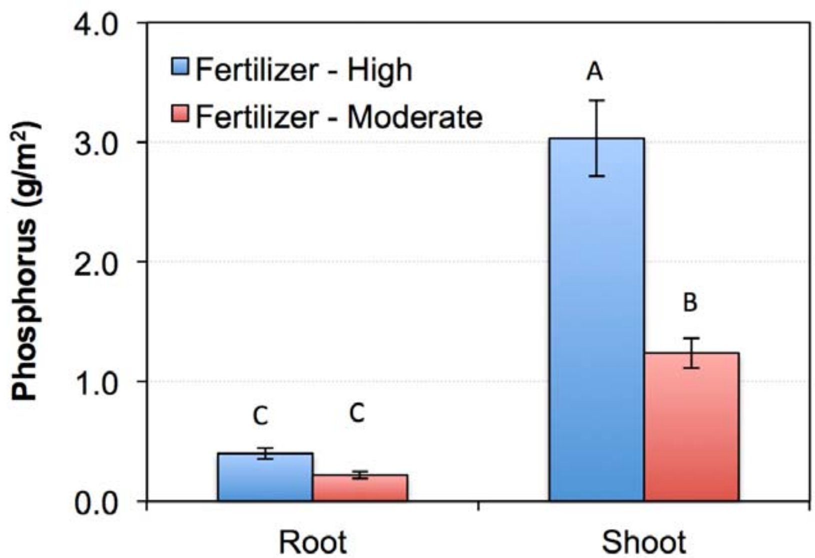

Figure 2: Total phosphorous content (mean ± standard error) of shoots and roots of Swiss chard (*Beta vulgaris*) grown in floating treatment wetlands and exposed to either high or moderate nutrient loads over an 8-week experimental period. Means with different letters are significantly different, students *t* test ($P \leq 0.05$).

Table 2: Shoot height of Swiss chard (*Beta vulgaris*) established in floating treatment wetlands for 8-weeks as impacted by fertilizer (high, moderate) and species mix (SCB = *B. vulgaris* 'Bright Lights'; SCS = *B. vulgaris*; and SCM = even mix of SCB and SCS). Means with the same letters are not significantly different, students *t* test (α ≤ 0.05).

2-Way ANOVA	DF	*P* > *F*
Species	2	< 0.0001
Fertilizer	1	0.2238
Species x fertilizer	2	0.3688

Species	LS Mean	
SCB	16.6	A
SCM	12.6	B
SCS	12.3	B

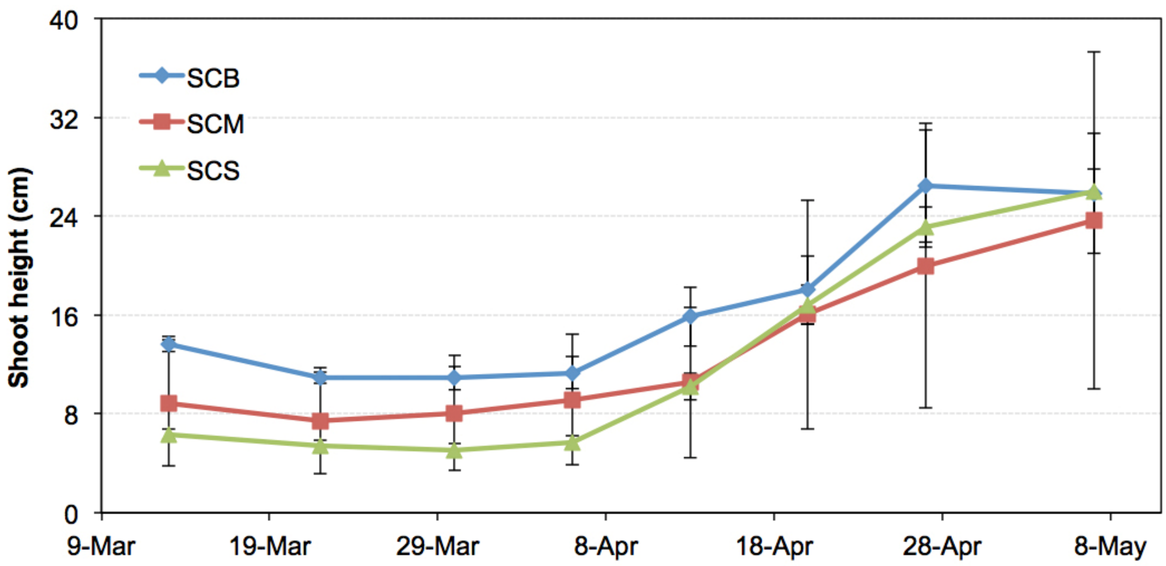

Figure 3: Shoot growth (mean ± standard error) of three species mixes (SCB = *B. vulgaris* 'Bright Lights'; SCS = *B. vulgaris*; and SCM = even mix of SCB and SCS) of *Beta vulgaris* (Swiss chard) established in floating treatment wetland over an 8-week experimental-period.

Table 3: Root length of Swiss chard (*Beta vulgaris*) established in floating treatment wetlands for 8-weeks as impacted by fertilizer (high, moderate) and species mix (SCB = *B. vulgaris* 'Bright Lights'; SCS = *B. vulgaris*; and SCM = even mix of SCB and SCS). Means with the same letters are not significantly different, students *t* test (α ≤ 0.05).

2-Way ANOVA	DF	*P > F*
Species	2	< 0.0001
Fertilizer	1	0.0003
Species x Fertilizer	2	0.2049

Species	LS Mean	
SCS	15.7	A
SCB	12.2	B
SCM	10.6	B

Fertilizer	LS Mean	
Moderate	14.3	A
High	11.3	B

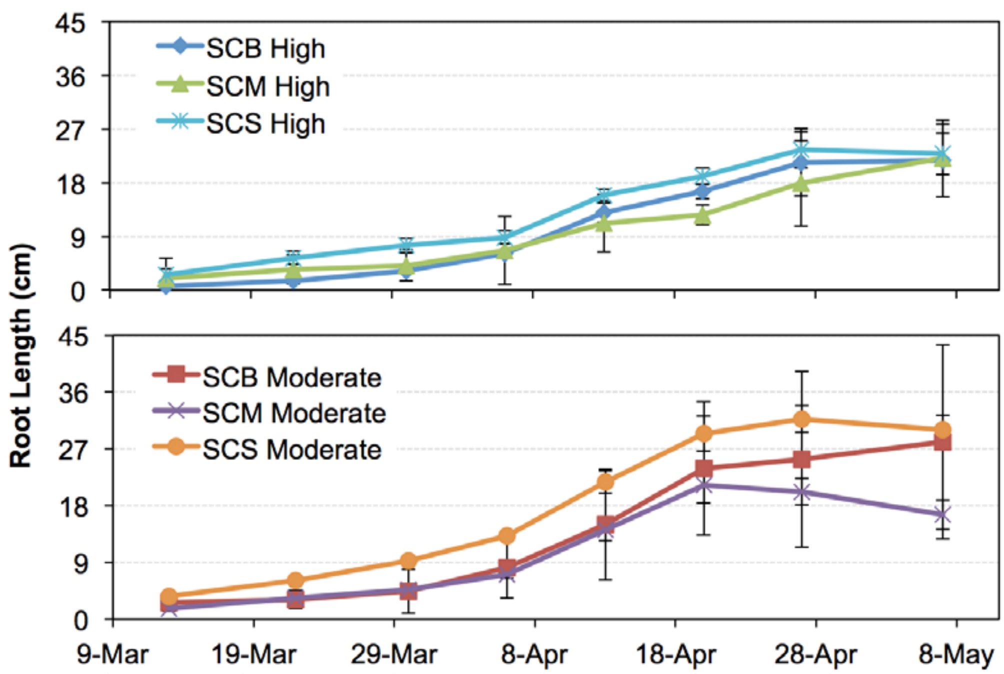

Figure 4: Root length (mean ± standard error) of *Beta vulgaris* (Swiss chard) grown in floating treatment wetlands over an 8-week period, exposed to two fertilizer levels (Moderate, High) and three species (SCB = *B. vulgaris* 'Bright Lights'; SCS = *B. vulgaris*; and SCM = even mix of SCB and SCS).