Project Completion Report
Project title
Evaluating Restoration and Mitigation Aquatic Plant Species and Markets to Advance the Commercialization of the Industry
Subaward #
Grant # 2004-38500-14589

PROJECT CODE: **SUBCONTRACT/ACCOUNT NO.:**
PROJECT TITLE: Evaluating Restoration and Mitigation Aquatic Plant Species and Markets to Advance the Commercialization of the Industry
DATES OF WORK: 06/01-2007 to 6/30/2010

PARTICIPANTS:
Andrew Lazur, Court Stevenson, and Erin Markin; University of Maryland Center for Environmental Science
Dan Terlizzi, Doug Lipton, and Don Webster; University of Maryland Extension
Reggie Harrell, University of Maryland Dept. Of Environmental Science and Technology
Mike Pietrak; Maine Aquaculture Association
Todd West and Karen Buzby; West Virginia University
Dennis McIntosh; Delaware State University

REASON FOR TERMINATION:
Project Completed

PROJECT OBJECTIVES:
<u>Assessment of Market Potential and Economic Benefit</u>
1. Determine the current market (quantity and value) for the use of aquatic plants for restoration and mitigation purposes in the northeast United States;
2. Determine the potential growth in the market over the next five years for aquatic plants for restoration and mitigation purposes in the northeast United States;
3. Determine the potential impact of the establishment and implementation of nutrient trading programs on the market for aquatic plants in the northeast United States;
4. Evaluate costs and plant performance of field studies to estimate economic value of various aquatic plant applications

<u>Development and Selection of Potential Plant Species</u>
5. Conduct laboratory nutrient (nitrogen and phosphorus) uptake potential of 12 aquatic plant species (both warm and cold species) identified as having both attractive growth characteristics and strong market potential in the restoration/mitigation and ornamental markets;

<u>Demonstration of Commercial Production Methods</u>
6. Develop and demonstrate technology/procedures and conduct individual plant growth and nutrient uptake performance selection trials for several plant species under field conditions;

7. Verify the growth performance, nutrient remediation, and aesthetic value of several aquatic plant species in various field applications including stormwater management, hatchery and raceway effluent treatment and pond production; and

Grower Outreach and Education

8. Plan and conduct an Extension program that will disseminate research-based information from this project to the plant industry on the economics of the project, provide information on plant species in a performance database and demonstrate culture technology to industry participants and other stakeholders, including terrestrial and aquatic nurseries, restoration/mitigation contractors, natural resource management agencies, contractors/developers, and research and Extension faculty involved in aquatic environment education.

ANTICIPATED BENEFITS:

- First of its kind database of nitrogen and phosphorus uptake of 12 native aquatic plant species for warm and cool climates within NE
- Foundation of plant selection methodology and production information for the top performing of the twelve species advancing the level of technology for plant culture
- Multi-stakeholder education of methodology and specific species selection for restoration/mitigation applications resulting in expansion of sales and degree of applications
- Field verification of aquatic plant integration in effluent mitigation
- Integrated Extension program with objectives of involving producers and prospective new entrants utilizing research-based information derived from this project and with a program developed across the region
- Factsheet publications and web based material on culture species, methods and systems, and market information allowing industry to make informed decisions on entry into this field.

PRINCIPAL ACCOMPLISHMENTS:

Objective: Assessment of Market Potential and Economic Benefit

1. Determine the current market (quantity and value) for the use of aquatic plants for restoration and mitigation purposes in the northeast United States:

A. Identified that approximately 125 plants species are sold as ornamentals; product forms include tubers, bare root plants, 4 and 6 inch and one gallon pots. Wholesale prices ranged from $2-19.00. 270 plant species are sold in the mitigation markets. Major product forms for these species include 2 inch plugs in varied sized trays, pint and 4-6 inch pots. Prices for mitigation species ranged from $0.27 to 3.50 depending on size and species.

B. Identified the top 12 aquatic plants in demand for the water garden market.

2. Determine the potential growth in the market over the next five years for aquatic plants for restoration and mitigation purposes in the northeast United States;

Identified six estuaries in the Northeast which are currently planning total maximum daily load regulations. Further identified two states which have expressed an active interest in biological mitigation techniques.

3. Determine the potential impact of the establishment and implementation of nutrient trading programs on the market for aquatic plants in the northeast United States;

Assuming an average uptake of 13.05 pounds of nitrogen and 1.03 pounds of phosphorus per acre of aquatic plant mitigation, each acre of vegetation planted could qualify for between $53.22 and $135.69 of nutrient credits per year.

4. Evaluate costs and plant performance of field studies to estimate economic value of various aquatic plant applications.

Given the cost of alternative abatement technologies, the mitigation of nutrient loading through the use of aquatic plants is a viable option.

Objective: Development and Selection of Potential Plant Species

5. Conduct laboratory nutrient (nitrogen and phosphorus) uptake potential of 12 aquatic plant species (both warm and cold species) identified as having both attractive growth characteristics and strong market potential in the restoration/mitigation and ornamental markets.

A. Based on phytotron studies of nutrient uptake and growth of the twelve plant species, five were selected for greater N and P uptake:
Decadon verticullatus, swamp willow; *Kosteletzkya*, swamp mallow; *Panicum hemitomon*, maidencane; *Panicum virgatum*, switchgrass; and *Scirpus cypernus*, woolgrass.

B. Nutrient uptake efficiency studies showed:

- B. Of all twelve species examined for ammonia uptake, *S. cyperinus* was found to have the highest affinity for ammonia, while *K. virginica* has the lowest affinity.
- *K. virginica* was found to be the fastest taker-upper of ammonia of all species examined and *D. arundinaceum* is the slowest.
- Our results show that *C. vulpinoidea* does not use NO_3^- and that *S. cyperinus* does not use NH_4^+.
- In nitrate experiments, *A. calamus* was found to have the highest affinity for NO_3^- while *S. sempervirens* has the lowest.
- *S. americanus* takes up NO_3^- at a higher velocity when compared to the other species and *A. calamus* is the slowest species to take up NO_3^-.
- No nitrate data is available for *S. alternifolia* because experiments are in progress.
- *P. virgatum, S. cyperinus* and *A. calamus* are the only species to show evidence of saturation in NO_3^- uptake experiments. All other species show evidence of NO_3^- use (with the exception of *C. vulpinoidea*), but no saturation; therefore, the Michaelis-Menten kinetics do not apply.

Objective: Demonstration of Commercial Production Methods

6. Develop and demonstrate technology/procedures and conduct individual plant growth and nutrient uptake performance selection trials for several plant species under field conditions.

A. In the outdoor studies, it was shown that *Kosteletzkya*, swamp mallow, and *Panicum virgatum*, switchgrass had significantly greater nitrogen uptake, and *Panicum virgatum* had significantly greater phosphorus uptake than the other species.

7. Verify the growth performance, nutrient remediation, and aesthetic value of several aquatic plant species in various field applications including stormwater management, hatchery and raceway effluent treatment and pond production; and

A. In the salmon hatchery effluent treatment system, the amount of nitrogen and phosphorous removed was highest for *Decodon*> *Scirpus*> *Carex*> *Panicum*. Phosphorus removal was very low, 1-3.7g/m^2 of plant area, and therefore the area of aquatic plant treatment needed is immense and it is deemed to be an impractical means of nutrient treatment.
B. Aquatic plant nutrient uptake in stormwater ponds showed nutrient uptake by aquatic plants in the floating wetlands of 146.3 g/m^2 and 11.7g/m^2 for nitrogen and phosphorus respectively.
C. The trout raceway effluent treatment system demonstrated that cardinal flower, *Lobelia cardinalis*, parrot feather, *Myriophyllum aquaticum*, and blue-eyed grass, *Sisyrinchium montanum* produced the greatest biomass and over the growing season parrot feather removed 39% of the NH_3, 22% of NO_3 and 20% of PO_4 while forget-me-not removed only 3% of the NH_3 and there was no reduction in NO_3 or PO_4 concentrations. Study showed with proper plant choice, aquaponics could be integrated into a flow-through raceway system despite low nutrient concentrations and low water temperatures.

Objective: Grower Outreach and Education

8. Plan and conduct an Extension program that will disseminate research-based information from this project to the plant industry on the economics of the project, provide information on plant species in a performance database and demonstrate culture technology to industry participants and other stakeholders, including terrestrial and aquatic nurseries, restoration/mitigation contractors, natural resource management agencies, contractors/developers, and research and Extension faculty involved in aquatic environment education.

The publication, manuscripts and presentation section provides details on the following: Fifteen presentations have been made relating information on integrating aquatic plants in various settings for nutrient remediation. Over 740 people were educated. Tours of the three integrated systems (DSU, ME and WV) and the stormwater ponds in MD, provided technical information and support to over 650 people. Nine publications have been produced with an estimated circulation of over 10,000 people. Web sites at the three universities also include information about this project and its results.

IMPACTS:

- New product developed by industry partner: bio-matrix floating wetland

- Project has attributed to an increase in sales of aquatic plants and floating wetland products for stormwater and other ponds amounting to over $256,000.
- Anticipated to impact the design of effluent treatment system for a new small scale hatchery
- Served as professional development for commercial and public fish hatchery staff
- Aided in evaluation of improved sustainable effluent treatment options leading to eco-certification for a commercial salmon hatchery
- Two state agencies in Delaware, Department of Natural Resources and Environmental Control's Division of Soil and Water Conservation has adopted aquatic plant growout technology from this project to produce plants needed in restoration
- Two producers in Delaware have adopted or have plans to implement similar aquatic plant/fish culture systems
- Several schools have implemented aquaponics in the classrooms
- Project results led to data to support expanded aquatic plant uptake in stormwater ponds including denitrification studies and expanded outreach. PI's were successful in securing a $65,000 grant from the Chesapeake Bay Trust

RECOMMENDED FOLLOW-UP ACTIVITIES:

The results of this project show several applications where integrating aquatic plants is effective in remediating nutrients and offers a source of additional income from sales of plants to both the ornamental and restoration markets. Recommended follow up work includes:
1. Assessing other plant species for nutrient uptake
2. Determining plant density to fish or nutrient source ratios
3. Evaluating genetic influence of nutrient uptake and selection trials
4. Assessing economic returns of various integrated scenarios
5. Expand educational activities in stormwater pond management emphasizing benefits of aquatic plants in nutrient removal, habitat diversity, long term pond maintenance costs and effect on property values
6. Evaluate nutrient uptake via denitrification within floating island matrix/plan roots
7. Assess periodic cuttings of vegetative growth as nutrient export

SUPPORT:

YEAR	NRAC Funding	OTHER					
		University	Industry	Other Federal	Other	Total	Total Support
2008	157,343			-	-		157,343
2009	140,256			-	-		140,256
2010	152,304			-	-		152,304
Total	449,903			-	-		449,903

PUBLICATIONS, MANUSCRIPTS, OR PAPERS PRESENTED:

Publications:

Buzby, Karen. 2008. Aquaponic Research at WVU farm in Wardensville. Fish Tails Newsletter 6(4). West Virginia Extension Service. Circulation ~ 500.

Marcia R. Guedes, Carrie Shoonover, and Daniel E. Terlizzi. 2008. Nitrogen Uptake Kinetics in Aquatic Vascular Plants in vitro and in Recirculating Trout Aquaculture Effluent. Proceedings of the 6th international recirculating aquaculture conference. Roanake, Va.

McIntosh, D., E. Markin, D. Wujtewicz and A. Lazur. 2008. Increasing Economic and Environmental Sustainability of Aquaculture Production Through Aquatic Plant Culture. Aquaculture America 2008 Book of Abstracts, Lake Buena Vista, FL, USA.

Lazur, A. M., A. Hengst, E. Markin, D. Webster, K. Billing, D. Schuck, and H. Buritsch. 2009. Urban and stormwater pond management. Maryland Sea Grant Extension. UM-SG-SGEP-2009-01. 10 pp.

Lazur, A. M., E. Markin and M. Pietrak. 2010. Aquatic plant culture opportunities for restoration and mitigation. 2010 Aquaculture America Abstracts.

Lazur, A. M., G. E. Flimlin, and D. Morse. 2010. Small scale marketing opportunities for aquaculture products in the northeast. Northeast Regional Aquaculture Center Factsheet. In Press.

Pietrak, M., A. Lazur, and W. Otto. 2010. Use of aquatic plants in a freshwater hatchery integrated multi-trophic aquaculture system. 2010 Aquaculture America Abstracts.

Schuck, D. and K. Billing. 2007. Rafted Plants Q&A. PondKeeper Magazine. November issue.

Schuck, Dick. 2009. The best plants to use in a veggie filter. Pond Trade Magazine. November/December 2009.

Presentations:

Billing, K. 2007. Aquatic Weed Control. 2007 LCA Winter Workshop MD, DC, VA.

Billing, K. 2010. Ornamental Pond Pest Management. 2010 15th Annual Procrastinator's Pest Management Conference, Univ. of MD Extension / Montgomery College. Management

Billing, K. Summary of Stormwater Project Results. 2010 Chesapeake Green Conference.

Garcia, V. 2008. Nutrient Uptake Efficiency in Five Wetland Plant Species of the Northeast United States. Maryland Sea Grant Research for Undergraduate final report. Chesapeake Biological Laboratory. Solomons, MD.

Lazur, A.M. 2007. Enhancing water quality, aesthetics and nutrient management of small ponds through use of aquatic plants. Aquaculture America 2007. San Antonio, TX.

Lazur, A.M., 2009. Considerations of selecting a culture species. Aquaculture America 2009. Seattle, WA.

Lazur, A.M. 2010. Enhancing pond management with aquatic plants. Annual Procrastinator's Pest Management Conference, Univ. of MD Extension. Salisbury, MD.

McIntosh, D. 2008. Integrating Aquatic Plant and Finfish Aquaculture. (poster). March 26, 2008. SARE 2008 National Conference, St. Louis, MO.

McIntosh, D., E. Markin, D. Wujtewicz and A. Lazur. 2008. Increasing Economic and Environmental Sustainability of Aquaculture Production Through Aquatic Plant Culture. February 12, 2008. Aquaculture America 2008, Lake Buena Vista, FL.

McIntosh, D. 2008. Increasing Economic and Environmental Sustainability of Aquaculture Systems Through Aquatic Plant Culture. January 9, 2008. DE Ag Week, Harrington, DE.

Pietrac, M., A. Lazur and M. Otto. 2010. Use of aquatic plants in a freshwater hatchery integrated multi-trophic aquaculture system. 2010 Aquaculture America, San Deigo, CA.

Pietrac, M., 2010. 3rd Annual Maine Aquaculture Association hosted Hatchery Round Table: Phosphorous removal from hatchery effluent water. 28 attendees mostly hatchery managers or staff and environmental engineers.

Schuck, D. 2010. Aquatic Plant Rafts for Mitigation. 2010 International Water Lily and Water Gardening Society (IWGS) Annual Symposium in San Angelo Texas.

West, T., 2009 - Tour of the aquaponics facility at the Reymann Memorial Farm in Wardensville, WV. 12 attendees, many of whom are active fish producers interested in exploring aquaponics.

<u>Student Activity/Degrees:</u>

Marcia Guedes. MS Environmental Biology. Hood College 2007.
Project Title: Nitrogen Uptake Kinetics in 3 species of Aquatic Vascular Plants in vitro and in Recirculating Trout Aquaculture Effluent.

Susanne Shoepe. Dec 2008. MS Environmental Biology. Hood College

Project Title: Nitrogen Uptake Kinetics in 6 species of Aquatic Vascular Plants in vitro and in Recirculating Trout Aquaculture Effluent.

Victoria Garcia. REU Internship. May 2008. Nutrient Uptake Efficiency in Five Wetland Plant Species of the Northeast United States. University of Maryland Center for Environmental Science. (REU Abstract and presentation).

Tiffany Brooks. Maryland high school intern completed a science project using aquatic plants for nutrient remediation. 2009.

Attachment 9
Project Completion Report – June 2010
Evaluating Restoration and Mitigation Aquatic Plant Species and Markets to Advance the Commercialization of the Industry
Subaward #
Grant # 2004-38500-14589

Part II

TECHNICAL ANALYSIS AND SUMMARY:

Objective: Assessment of Market Potential and Economic Benefit

1. Determine the current market (quantity and value) for the use of aquatic plants for restoration and mitigation purposes in the northeast United States;

A survey of the northeastern United States resulted in the following information:

Number of species sold	**Product size**	**Wholesale prices**
Ornamentals (125)	Tubers, bare root,4 and 6 inch pot, gallon	$2.00 – 19.00
Restoration/Mitigation (270)	2 inch plugs, pint, 4-6 inch pots	$0.27 – 3.50

Top twelve plant species sold

Rank	**Botanical name**	**Common Name**
1	Nymphaea - Hardy Water Lilies	red, pink, white, yellow, peach
Emergent Species		
	Acorus calamus 'Variegatus'	Variegated Sweet Flag
3	Hibiscus moschuetos	Swamp Hibiscus
4	Iris laevigata; versicolor	Variegated and Louisiana Iris
5	Myriophyllum aquaticum	Parrots Feather
6	Peltandra virginica	Arrow Arrum
7	Pontederia cordata	Pickerel Weed

	Pontederia dilatata	Royal Pickerel
8	Ranunculus flammula	Miniature Spearwort
9	Saururus cernuus	Lizard's Tail
10	Typha latifolia; laxmanni	Variegated Cattail
Submerged Plants		
11	Ceratophyllum demersum	Hornwort / Coontail
11	Egeria densa	Anacharis
Floating Plants		
12	Eichhornia crassipes	Water Hyacinth
12	Pistia stratoides	Water Lettuce

Markets for aquatic plants include; Landscape and environmental restoration/mitigation contractors, government agencies (natural resources and transportation) garden centers, home improvement stores, non-governmental agencies and to a lesser extent, internet sales.

2. Determine the potential growth in the market over the next five years for aquatic plants for restoration and mitigation purposes in the northeast United States;

The demand for nutrient mitigation technologies is steadily increasing. A survey of the Northeast identifies at least six Total Maximum Daily Load (TMDL) allowance systems currently in development. These include regulations for Chesapeake Bay, Long Island Sound, New York - New Jersey Harbor, Buzzards Bay, Cape Cod, and the lower South Shore in Massachusetts[1]. The Long Island and Chesapeake Bay TMDL regulations alone call for a flow decrease of over 84 million lbs of nitrogen and 4.1 million lbs of phosphorus into these estuaries. Although some of this decrease has already been achieved, there still remains a growing demand for mediation technologies. Due to the fact that many of the TMDL regulations are in the early stages of planning and implementation, the incorporation of aquatic plant mitigation and restoration projects

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[1] Buzzards Bay National Estuary Program. Draft Action Plan 1: Managing Nitrogen-Sensitive Embayments. Retrieved from http://www.buzzardsbay.org/newccmp-nitrogen.htm; Massachusetts Bays Program. (2003). Comprehensive Conservation and Management Plan. Retrieved from http://www.mass.gov/envir/massbays/ccmp.htm; New York State Department of Environmental Conservation & Connecticut Department of Environmental Protection. (2000). A total maximum daily load analysis to achieve water quality standards for dissolved oxygen in Long Island Sound.; New York-New Jersey Harbor Estuary Program. Goals of the Nutrient Module. Retrieved from http://www.harborestuary.org/nutrients.htm; U.S. Environmental Protection Agency. (2010). Chesapeake Bay TMDL Retrieved from http://www.epa.gov/chesapeakebaytmdl/.

within these plans is a feasible objective. An interest in the use of biological mitigation techniques has been explicitly expressed in Maryland with the use of cover crops and wetland restoration[2] and in New York through the use of seaweed and shellfish aquaculture[3]. The use of aquatic plants in a similar capacity would therefore be a familiar prospect to many policymakers looking to undertake nutrient mitigation.

3. Determine the potential impact of the establishment and implementation of nutrient trading programs on the market for aquatic plants in the northeast United States;

The generation of nutrient credits through the use of cover crop mitigation has been researched extensively in Maryland, with a projected low price of $3.68 per pound of nitrogen mitigation[4]. Between 2007 and 2009 the posted price per credit of nitrogen on the Maryland Nutrient Trading site NutrientNet was $10.00/lb of nitrogen[5], and we have used this as a high for the price of credit for nitrogen mitigation. In 2007 phosphorus credits traded on the Chesapeake Bay Nutrient Credit Exchange for $5.04/ lb[6]. Because phosphorus credits are a small component of the overall nutrient credit associated with aquatic plants, this is the only price of phosphorus considered. Given the average uptake of 13.05 pounds of nitrogen and 1.03 pounds of phosphorus per acre of aquatic plant mitigation reported below, each acre of vegetation planted could qualify for between $53.22 and $135.69 of nutrient credits per year. It is assumed that nutrient credit trading in other states within the Northeast would produce prices within the range derived for Maryland.

4. Evaluate costs and plant performance of field studies to estimate economic value of various aquatic plant applications.

Given the range of costs defined within this study, and assuming that ponds must be replanted on an annual basis, the mitigation of nutrient loading through the use of aquatic plants could be a cost-effective alternative. For example, assuming that 1% of a wetland acre is planted with a density of .6 plants per square foot, at the current low price of $0.27/plant this would lead to a cost of $72.84/acre planted versus a maximum expected benefit of around $135.69/acre planted. This provides a net benefit of $62.85. At the average cost of $1.89/plant, planting that same 1% area would cost $508.56, for a net loss of $372.87. At the low end of the credit prices projected, the nutrient credits generated by aquatic plants would not be enough to fully offset the cost of planting at current plant prices. An alternative way in which to look at the efficiency of aquatic plant nutrient

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[2] Wieland, R., Parker, D., Gans, W., & Martin, A. (2009). Costs and cost efficiencies for some nutrient reduction practices in Maryland. NOAA Chesapeake Bay Office and Maryland Department of Natural Resources Report.

[3] Long Island Sound Study. (2009). Nutrient Remediation Workshop: Dec. 3-4, 2009. Retrieved from http://longislandsoundstudy.net/2010/02/nutrient-remediation-workshop/.

[4] Hanson, J.C., & McConnell, K.E. (2008). Simulated trading for Maryland's nitrogen loadings in the Chesapeake Bay. Agricultural and Resource Economics Review. 37(2), 211-226.

[5] Maryland Department of Agriculture. Maryland Nutrient Trading. Retrieved from http://www.mda.state.md.us/nutrad/.

[6] Vanotti, M.B., Szogi, A.A., Hunt, P.G., Millner, P.D., & Humenik, F.J. (2007). Development of environmentally superior treatment system to replace anaerobic swine lagoons in the USA. *Biosource Technology*, 98(17), 3184-3194.

mitigation is by looking at cost per unit of nutrients removed. Concentrating on nitrogen and excluding the potential for nutrient credit trading, the average cost of mitigation is \$38.97/lb of nitrogen removed, with a minimum cost of \$5.58/lb. The current average cost of mitigation utilizing cover crops has been calculated to be \$5.83/lb of nitrogen removed[7], comparable to the minimum cost of using aquatic plants. Thus, the judicious choice of aquatic plant species in nutrient mitigation could be comparable in cost to currently employed mitigation technologies. In addition, with a large enough market for aquatic plants used for mitigation, production costs can be expected to decrease and be at the low end of the range examined due to improved production technologies and increased economies of scale.

Objective: Development and Selection of Potential Plant Species

5. Conduct laboratory nutrient (nitrogen and phosphorus) uptake potential of 12 aquatic plant species (both warm and cold species) identified as having both attractive growth characteristics and strong market potential in the restoration/mitigation and ornamental markets;

Nutrient Uptake in Aquatic Plants

Materials and methods:

Multiple flask method:

A 100µM NH_4Cl solution was prepared and Hoagland's (no nitrogen) was added. The solution was diluted to concentrations of 10, 20, 30, 40, 50, 60, 70, 80, 90, 100µM and poured into beakers. A 2mL sample was taken from each beaker (T_0) and diluted to 10mL. One individual plant was placed into each beaker and incubated for a total period of 1hr at controlled temperature (25ºC) and light/dark cycle (L/D). During incubation, 2mL samples were taken from each beaker at 15-minute intervals and diluted to 10mL (T_{15}, T_{30}, T_{45}, T_{60}). Samples were analyzed colorimetrically and plants had their weight taken.

For nitrate experiments, the same procedure described above was used with the exception of the solution, which was KNO_3^- , and the incubation period, which varied from species to species, ranging from 60 to 1399min.

Perturbation method (*Carex vulpinoidea* nitrate experiment only):

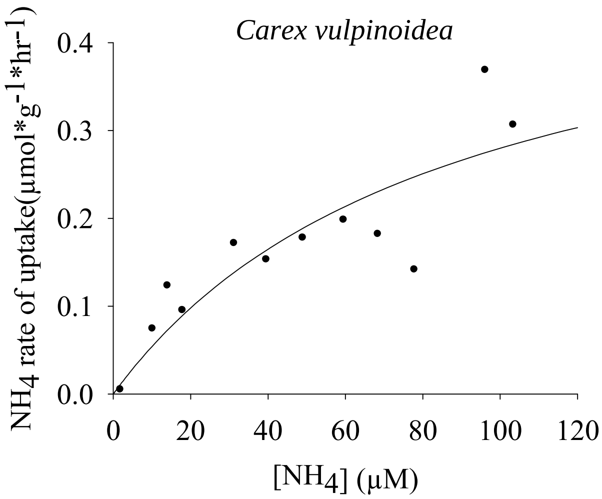
A 100µM KNO_3 solution was prepared and poured into a beaker. Hoagland's (no nitrogen present) was added to the solution and 6 individual plants were placed into the beaker. 2mL samples were taken at 5-minute and 15-minute intervals and diluted to 10mL. Samples were analyzed colorimetrically and the plants had their weight taken. The total incubation time ranged from 30 to 120 minutes.

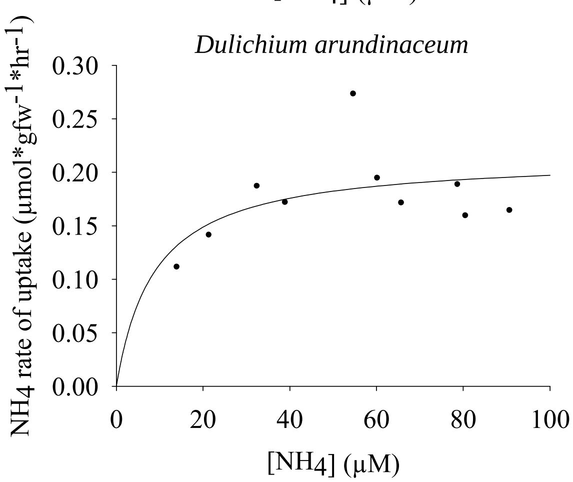
[7] Wieland, R., Parker, D., Gans, W., & Martin, A. (2009). Costs and cost efficiencies for some nutrient reduction practices in Maryland. NOAA Chesapeake Bay Office and Maryland Department of Natural Resources Report.

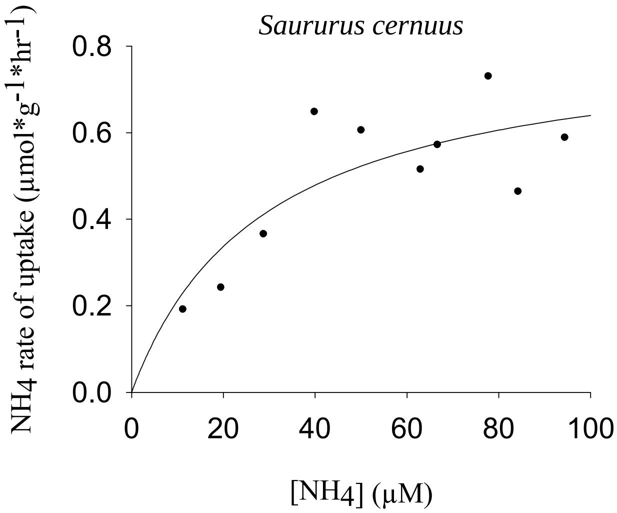
Data analysis:
All data was analyzed using Microsoft Excel and the Enzyme Kinetics module of the Sigma Plot software.

Results
Ammonia experiments:

Year: 2007

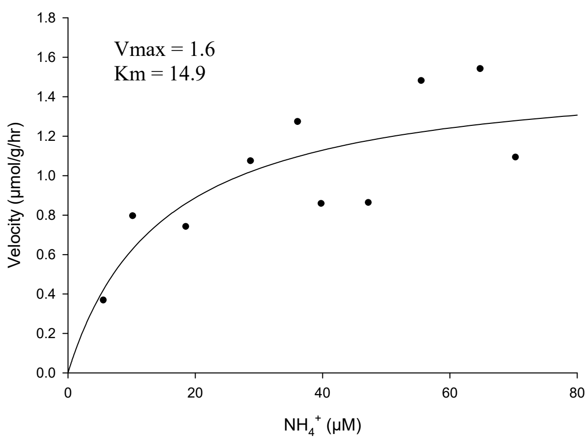






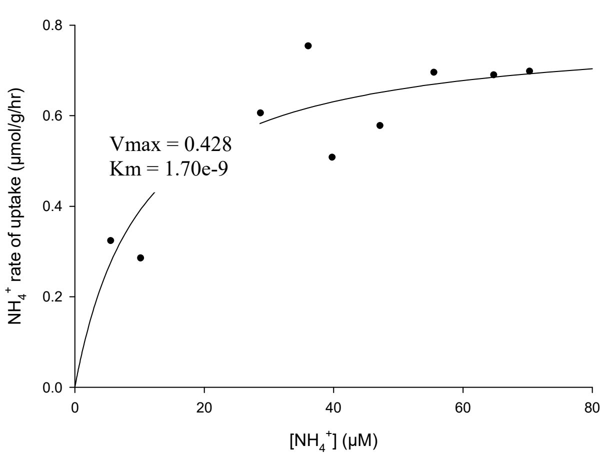
Year: 2008

A. calamus: NH_4^+ Uptake kinetics
in a 30min. interval.



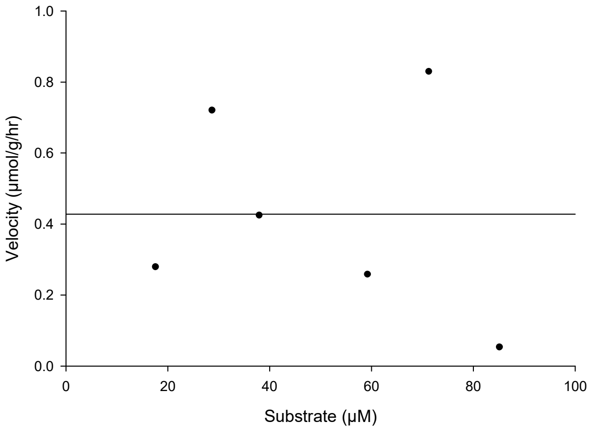
Vmax = 0.7953
Km = 10.4

A. calamus: NH_4^+ Uptake kinetics
in a 60min. interval.

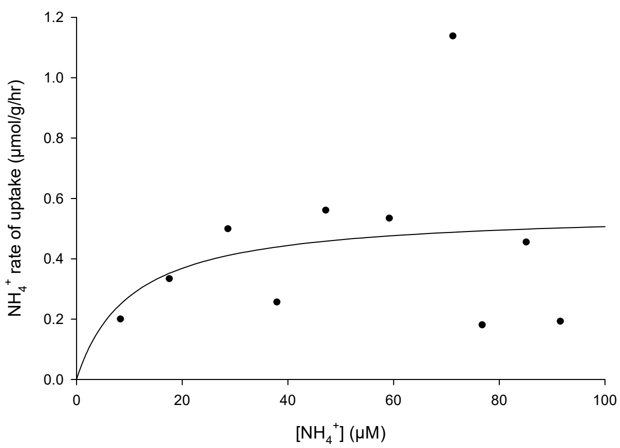


Vmax = 0.5586
Km = 10.3

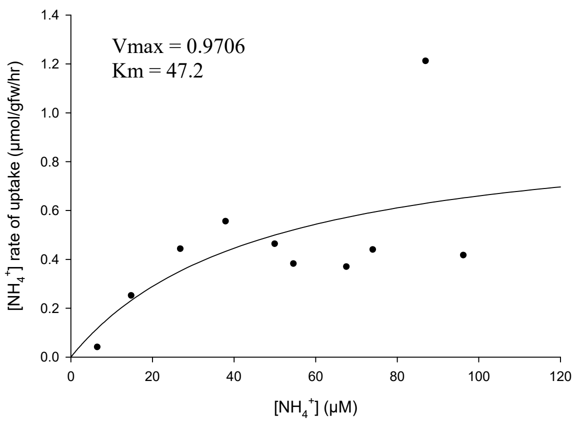
D. verticillatus: NH_4^+ uptake kinetics
in a 30min. interval'



D. verticillatus: NH_4^+ uptake kinetics
in a 60min. interval.

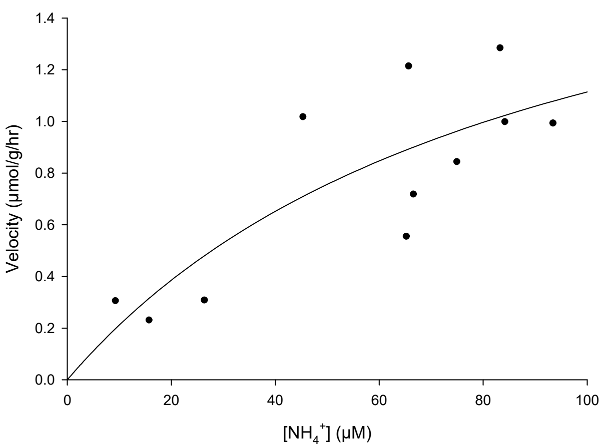


E. angustifolium NH_4^+ uptake kinetics
in a 60min. interval.



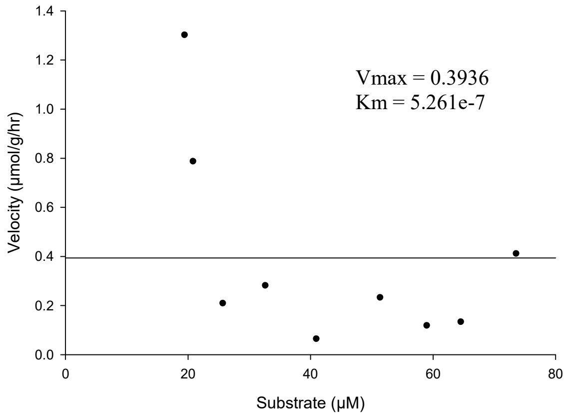
Vmax = 2.1
Km = 89.8

K. virginica NH_4^+ uptake kinetics
in a 15min. interval.

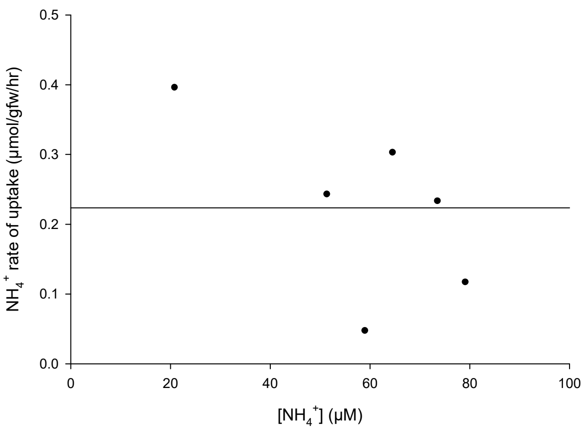


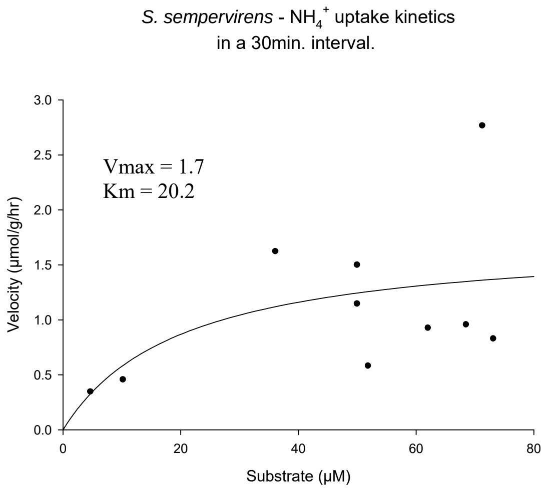
S. cyperinus NH_4^+ uptake kinetics
in a 30min. interval.

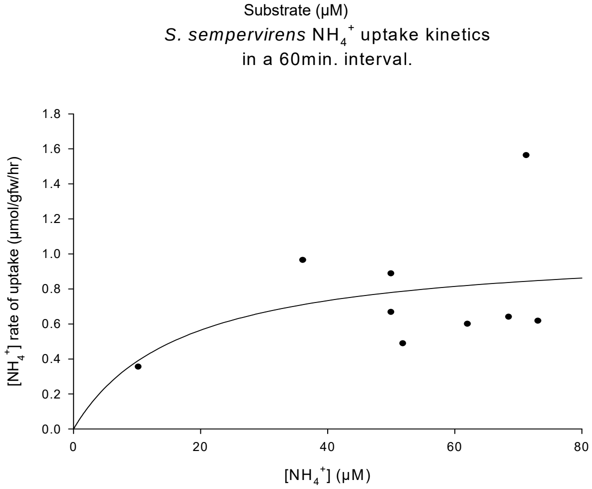
Vmax = 0.2235
Km = 2.647e-7



S. cyperinus NH_4^+ uptake kinetics
in a 60min. interval.



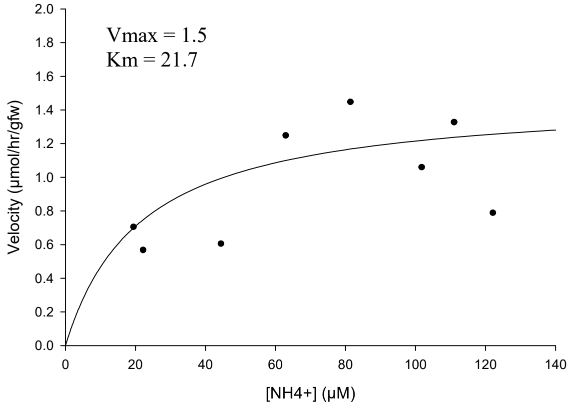




Vmax = 1.0
Km = 17.0

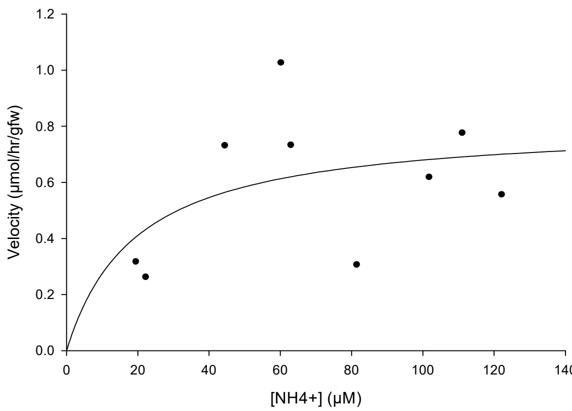
<u>Year 2009:</u>

P. virgatum - NH_4^+ uptake kinetics
in a 30min. interval

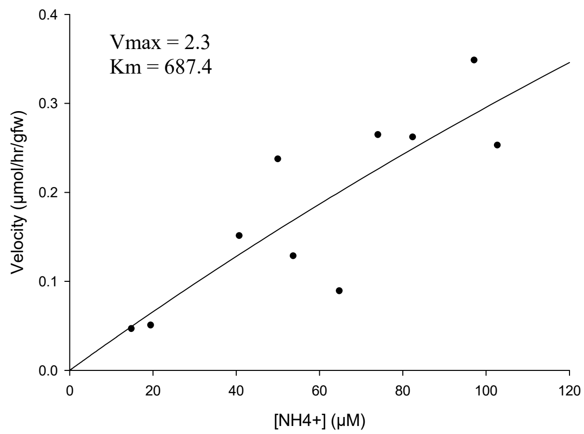


Vmax = 0.8123
Km = 19.5

P. virgatum - NH_4^+ uptake kinetics
in a 60min. interval

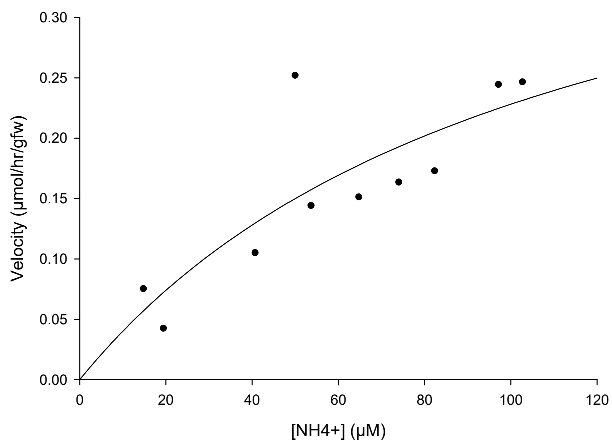


S. alternifolia - NH_4^+ uptake kinetics
in a 30min. interval.

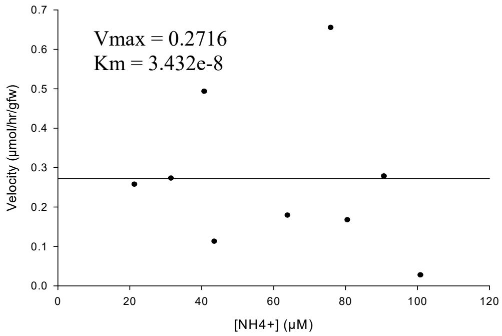


Vmax = 0.4782
Km = 108.7

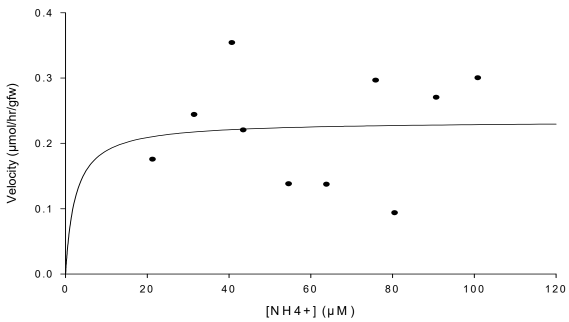
S. alternifolia - NH_4^+ uptake kinetics
in a 60min. interval.



S. americanus - NH_4^+ uptake kinetics
in a 30min. interval.



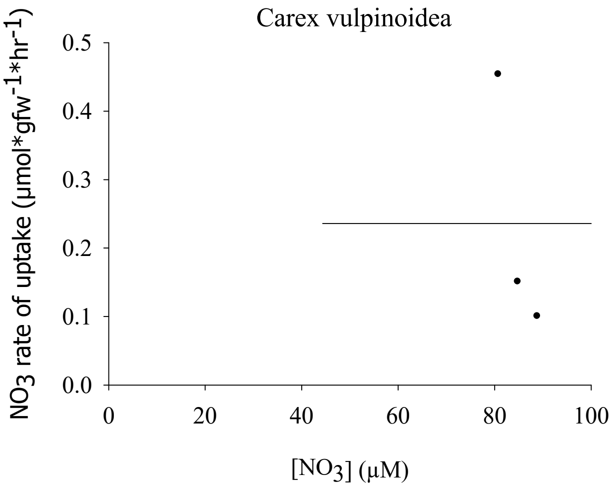
S. americanus - NH_4^+ uptake kinetics
in a 60min. interval.

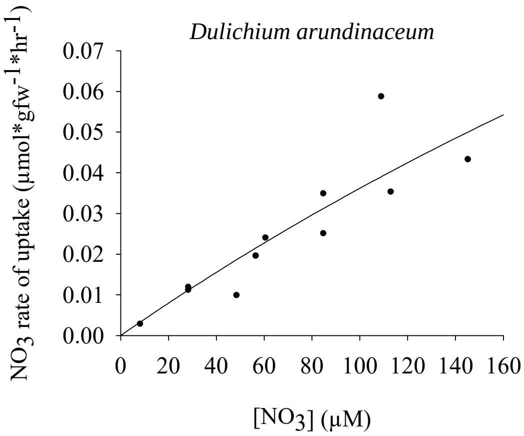


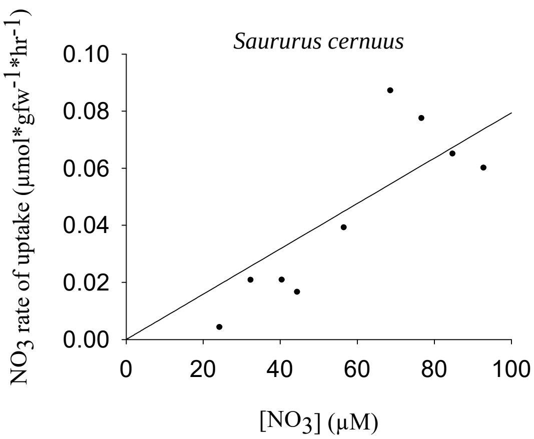
Vmax = 0.234
Km = 2.4

<u>Nitrate experiments:</u>

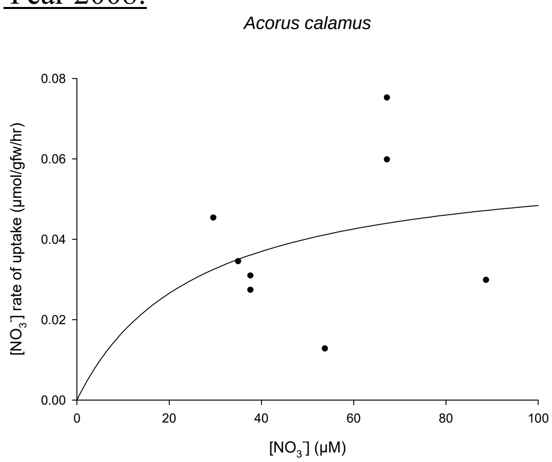
<u>Year 2007:</u>



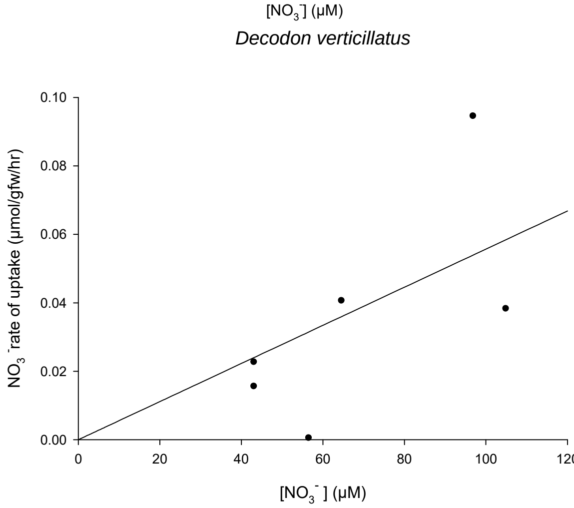




Year 2008:

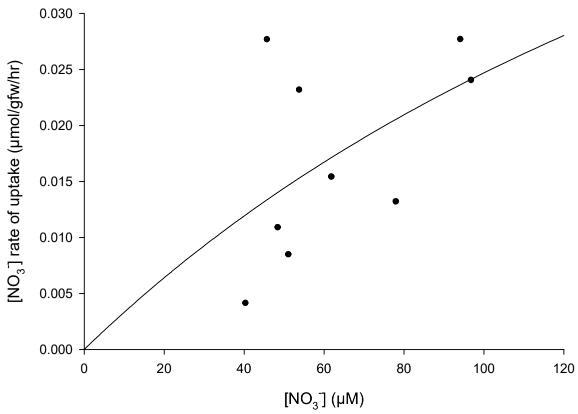


Acorus calamus
0.08
0.06
0.04
0.02
0.00
[NO3-] rate of uptake (µmol/gfw/hr)
0
20
40
60
80
100
[NO3-] (µM)

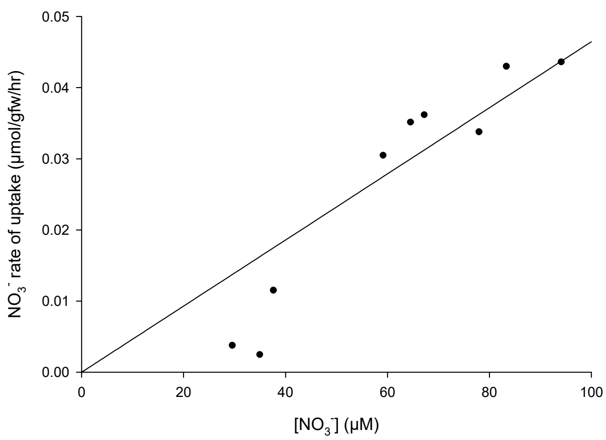


Decodon verticillatus
0.10
0.08
0.06
0.04
0.02
0.00
NO3- rate of uptake (µmol/gfw/hr)
0
20
40
60
80
100
120
[NO3-] (µM)

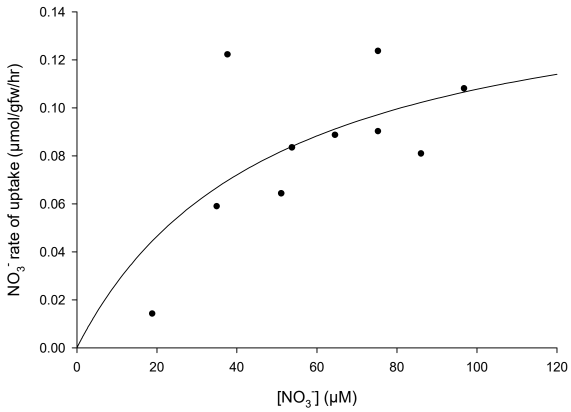
Eriophorum angustifolium



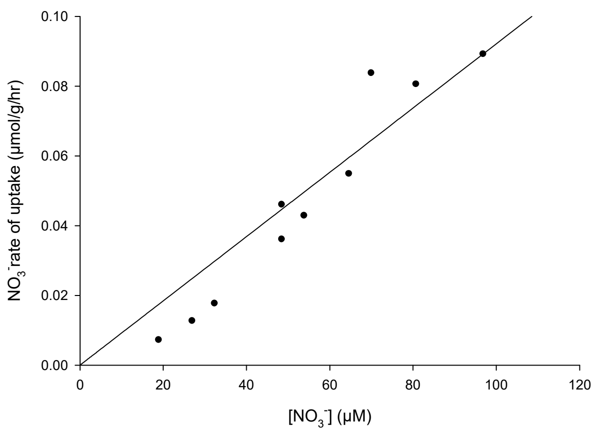
K. virginica NO_3 Means



S. cyperinus NO_3^- Means

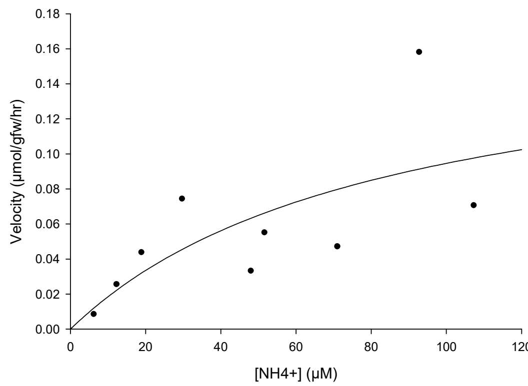


Solidago sempervirens

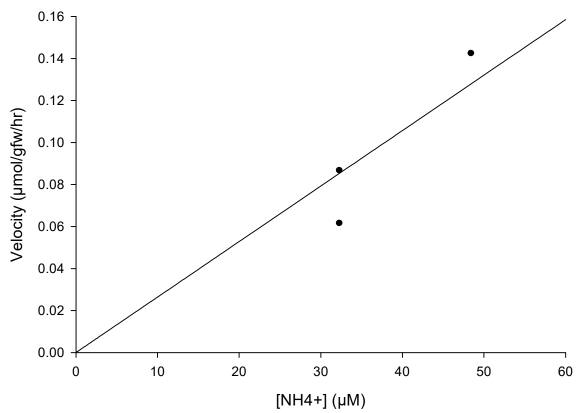


Year 2009:

P. virgatum - NO_3^- mean



S. americanus - NO_3^- mean.



Kinetics Parameters:

Plant species	NH_4^+		NO_3^-	
	V_{max} (µmol·gfw^{-1}·hr^{-1})	K_m	V_{max} (µmol·gfw^{-1}·hr^{-1})	K_m
C. vulpinoidea	0.52 ± 0.24	86.8 ± 70.9	9.60 ± 1290.30	8148.70 ± 1.12×10^6
D arundinaceum	0.21 ± 3.15×10^{-2}	8.8 ± 7.41	0.32 ± 0.70	784.80 ± 1948.76
S. cernuus	0.82 ± 0.17	28.9 ± 17.64	1947.90 ± 5.06×10^6	2.45×10^6 ± 6.37×10^9
A.calamus	0.80 ± 8.12×10^{-2}	10.40 ± 4.32	0.061 ± 4.85×10^{-2}	25.9 ± 60.44
D. verticillatus	0.56 ± 0.24	10.30 ± 21.83	1616.00 ± 9.25×10^6	2.90×10^6 ± 1.66×10^{10}
E. angustifolium	0.97 ± 0.68	47.20 ± 75.10	0.09 ± 0.20	250.30 ±

				737.90
S. cyperinus	0.22 ± 0.11	2.65×10^{-7} ± 20.89	0.16 ± 7.61×10^{-2}	49.20 ± 52.60
S. sempervirens	1.00 ± 0.51	17.00 ± 33.04	1.67×10^{4} ± 1.74×10^{8}	1.77×10^{8} ± 1.85×10^{11}
P. virgatum	0.81 ± 0.26	19.51 ± 24.87	0.1742 ± 0.1625	84.12 ± 141.43
S. alternifolia	0.48 ± 0.29	108.73 ± 110.81	** ongoing **	** ongoing **
S. americanus	0.23 ± 6.74x10-2	2.40 ± 13.59	31,454,5690 ± 1.180×10^{9}	1.191×10^{7} ± 4.47×10^{11}
K. virginica	2.50 ± 2.36	115.00 ± 172.70	4431.70 ± 3.34×10^{7}	9.54×10^{6} ± 7.20×10^{10}

Performance summary:

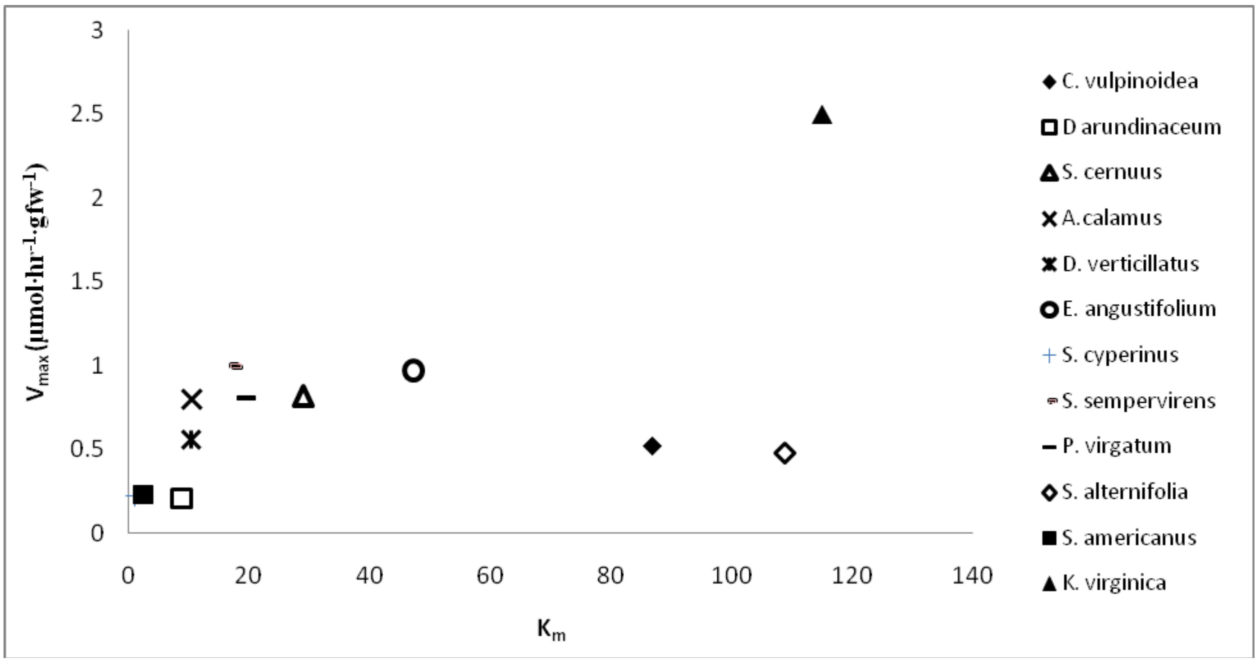
	NH_4^+		NO_3^-	
	Affinity (K_m)	**Velocity (V_{max})**	**Affinity (K_m)**	**Velocity (V_{max})**
Highest	*S. cyperinus*	*K. virginica*	*A. calamus*	*S. americanus*
	S. americanus	*S. sempervirens*	*S. cyperinus*	*S. sempervirens*
	D. arundinaceum	*E. angustifolium*	*P. virgatum*	*K. virginica*
	D. verticillatus	*S. cernuus*	*E. angustifolium*	*S. cernuus*
	A. calamus	*P. virgatum*	*D. arundinaceum*	*D. verticillatus*
	S. sempervirens	*A. calamus*	*C. vulpinoidea*	*C. vulpinoidea*
	P. virgatum	*D. verticillatus*	*S. cernuus*	*D. arundinaceum*
	S. cernuus	*C. vulpinoidea*	*D. verticillatus*	*P. virgatum*
	E. angustifolium	*S. alternifolia*	*K. virginica*	*S. cyperinus*
	C. vulpinoidea	*S. americanus*	*S. americanus*	*E. angustifolium*
	S. altermifolia	*S. cyperinus*	*S. sempervirens*	*A. calamus*
Lowest	*K. virginica*	*D. arundinaceum*		

- Of all twelve species examined for ammonia uptake, *S. cyperinus* was found to have the highest affinity for ammonia, while *K. virginica* has the lowest affinity.
- *K. virginica* was found to be the fastest taker-upper of ammonia of all species examined and *D. arundinaceum* is the slowest.
- Our results show that *C. vulpinoidea* does not use NO_3^- and that *S. cyperinus* does not use NH_4^+.
- In nitrate experiments, *A. calamus* was found to have the highest affinity for NO_3^- while *S. sempervirens* has the lowest.

- *S. americanus* takes up NO_3^- at a higher velocity when compared to the other species and *A. calamus* is the slowest species to take up NO_3^-.
- No nitrate data is available for *S. alternifolia* because experiments are in progress.
- *P. virgatum, S. cyperinus* and *A. calamus* are the only species to show evidence of saturation in NO_3^- uptake experiments. All other species show evidence of NO_3^- use (with the exception of *C. vulpinoidea*), but no saturation; therefore, the Michaelis-Menten kinetics do not apply.

Year 2010

V_{max} vs. K_m for NH_4^+:



Statistical Analysis:

One-way ANOVA NH_4^+ V_{max}

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	386.8552	1	386.8552	69.71213	1.59E-11	4.006873
Within Groups	321.8608	58	5.549325			
Total	708.7161	59				

One-way ANOVA NH_4^+ K_m

ANOVA

Source of Variation	SS	df	MS	F	P-value	F crit
Between Groups	17426.3	1	17426.3	13.80324	0.000458	4.006873
Within Groups	73223.75	58	1262.478			
Total	90650.05	59				

One-way ANOVA NO_3^- V_{max}

ANOVA

Source of Variation	SS	df	MS	F	P-value	F crit
Between Groups	148.4195	1	148.4195	16.6445	0.000703	4.413873
Within Groups	160.5065	18	8.917029			
Total	308.926	19				

One-way ANOVA NO_3^- K_m

ANOVA

Source of Variation	SS	df	MS	F	P-value	F crit
Between Groups	127836.1	1	127836.1	5.089017	0.036758	4.413873
Within Groups	452160.1	18	25120.01			
Total	579996.3	19				

Notes:

Our overall results show that there is a significant difference among V_{max} and K_m values between species in both Ammonia and Nitrate nutrient solutions.

Phytotron evaluation of plant species nutrient uptake

This objective was initiated in early 2008 and had to be repeated during the late spring due to mechanical failure of the phytotrons (special plant growth chambers) Twelve species were cultured using replicated shallow water culture tanks in two phytotrons. Plant species tested were:

Genus	Common name	Climate adaptability
Acorus calamus	Sweet flag	Mid Atlantic to Maine
Carex vulpinoidae	Fox sedge	Mid Atlantic to Maine
Decadon verticillatus	Swamp loosestrife	Mid Atlantic to Maine
Dulichium aruninaceum	Three-sided sedge	Mid Atlantic to Maine
Eriopherum angustifolium	Cotton grass	Mid Atlantic to Maine
Kolestkia virginica	Seashore mallow	Mid Atlantic

Panicum hemitomon	maidencane	Mid Atlantic to SE
Saururus cernuus	Lizards tail	Mid Atlantic to Maine
Scirpus olneyi or Schoenoplectic americanus	Olney's bulrush or Olney three square	Mid Atlantic to Maine
Scirpus cypernus	Wool grass	Mid Atlantic to Maine
Spartina alternifola	Smooth cord grass	Mid Atlantic to Maine
Solidago sempervirons	Seaside goldenrod	Mid Atlantic

Table 2 Plant species average growth in UMD phytotron chambers.

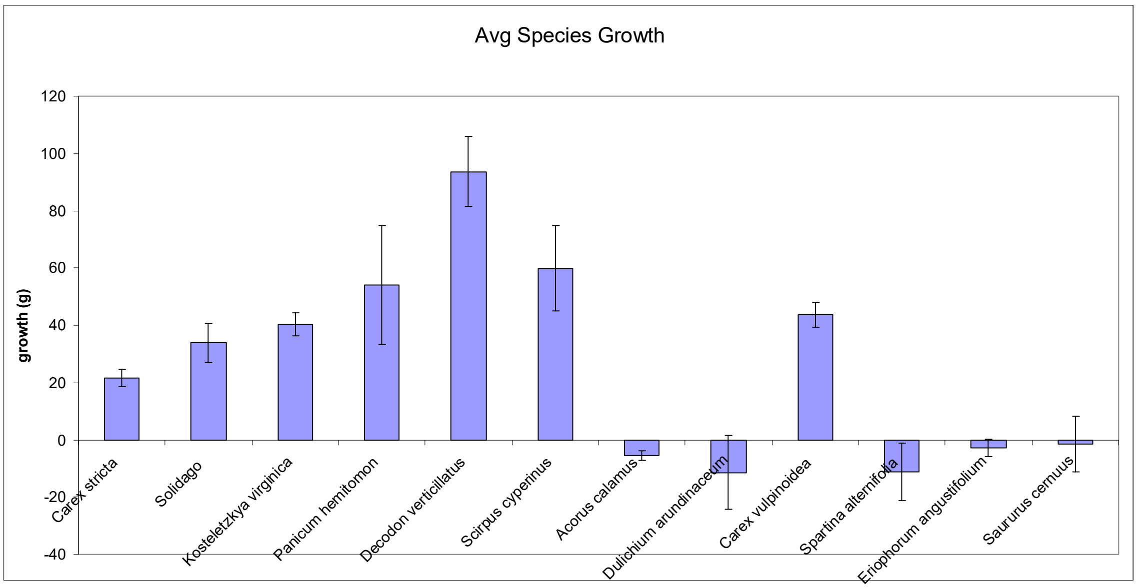
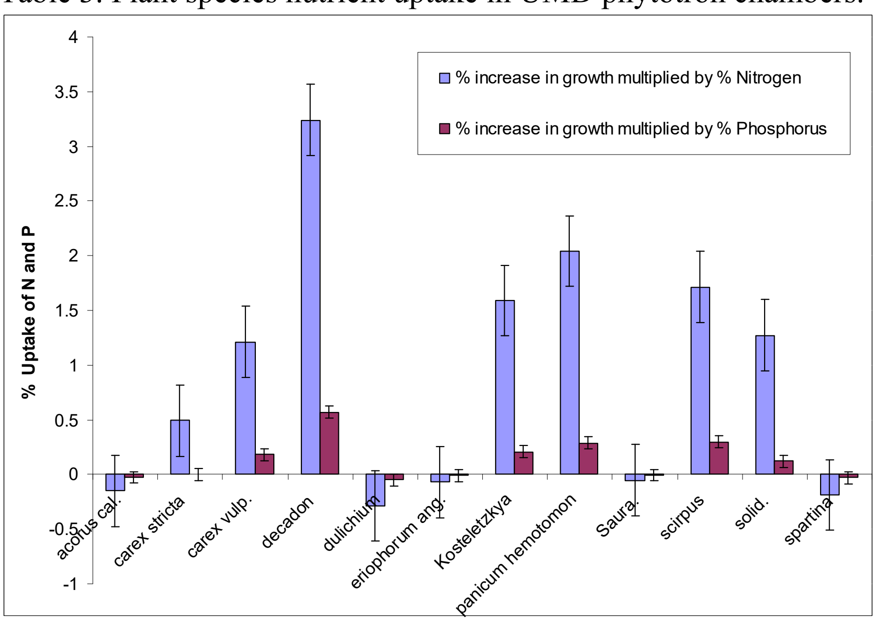
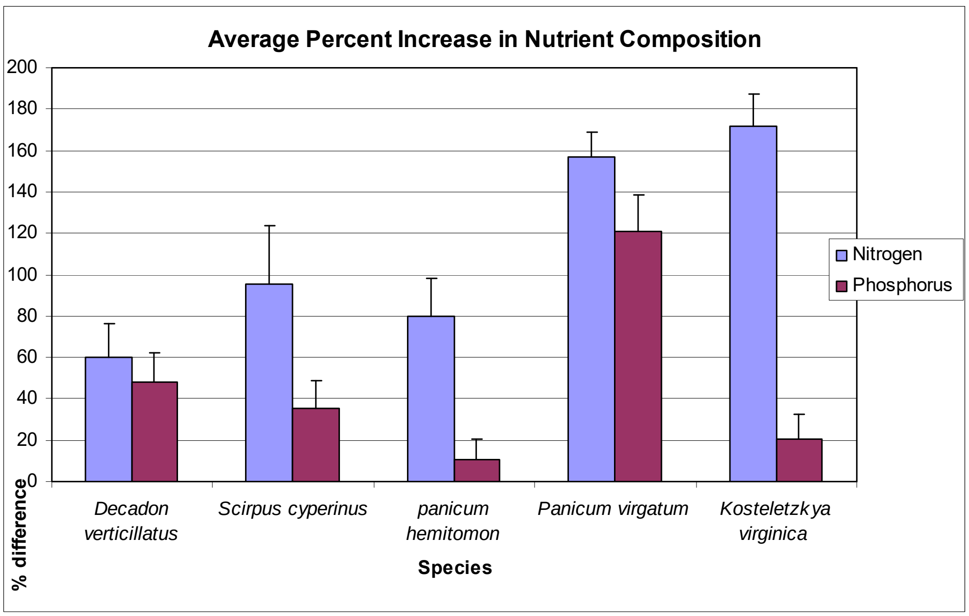


Table 3. Plant species nutrient uptake in UMD phytotron chambers.



Based on phytron studies of nutrient uptake and growth of the twelve plant species, four were selected for scaling up into outdoor mesocosm studies as follows: *Decadon verticullatus*, swamp willow; *Kosteletzkya*, swamp mallow; *Panicum hemitomon*, maidencane; and *Scirpus cypernus*, woolgrass. In addition, *Panicum viragatum,* switchgrass was tested in the mesocosm study due to its potential value as biofuel and similarity to *P. hemitomon*. The outdoor mesocosm studies with the five plant species was initiated in May 2008 and a total of five plant tissue analysis samples are expected. Table 4 presents a comparison of nitrogen and phosphorus uptake for the five species. Two additional samples were yet to be analyzed, therefore final conclusion of plant nutrient uptake is not expected until late 2008.

Table 4. Initial nutrient increase of five aquatic plant species.



Year 2 Phytotron nutrient uptake studies:

Table 1. Plant growth at lower temperature (20^0C)

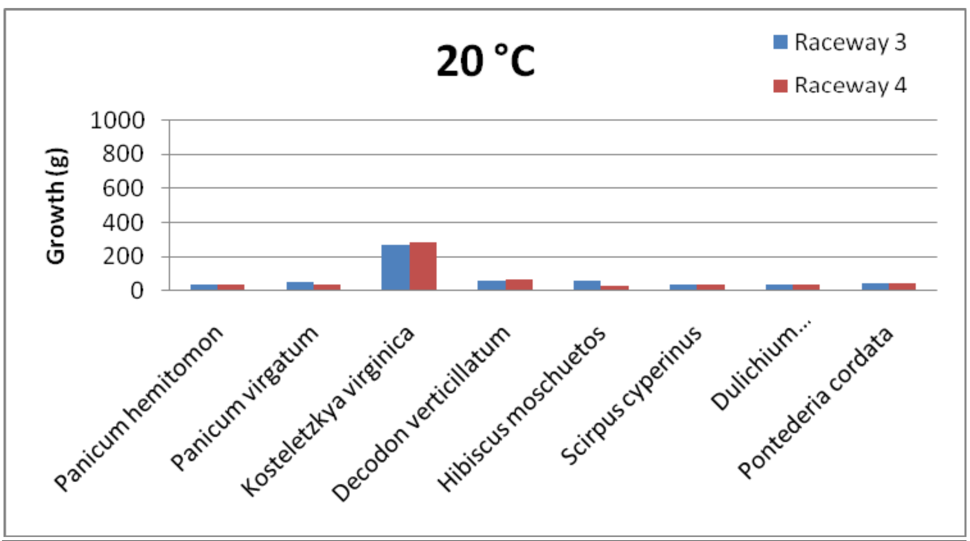


Table 2. Plant growth at higher temperature (30^0C).

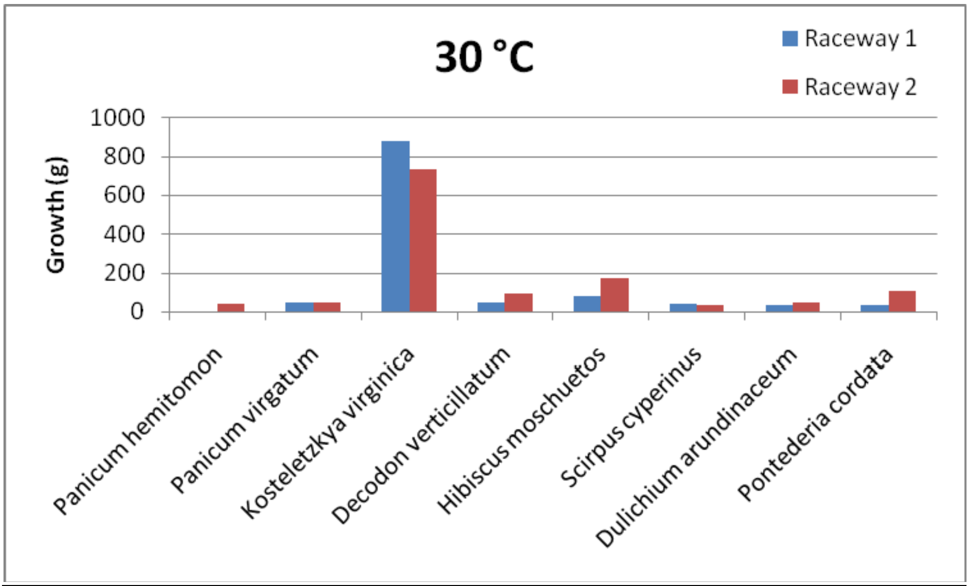


Table 3. Macronutrient content of varied plant species at lower temperature at harvest.

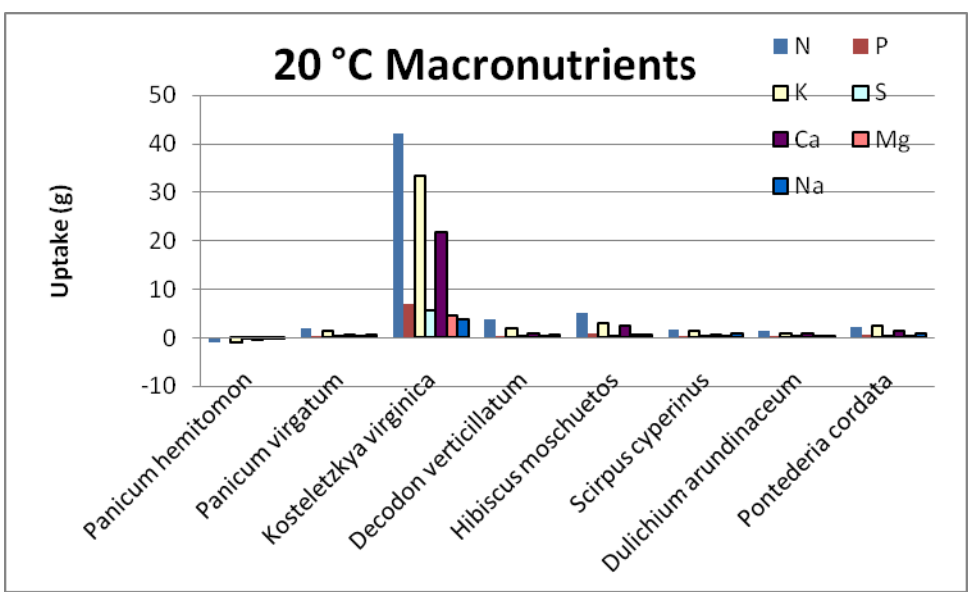
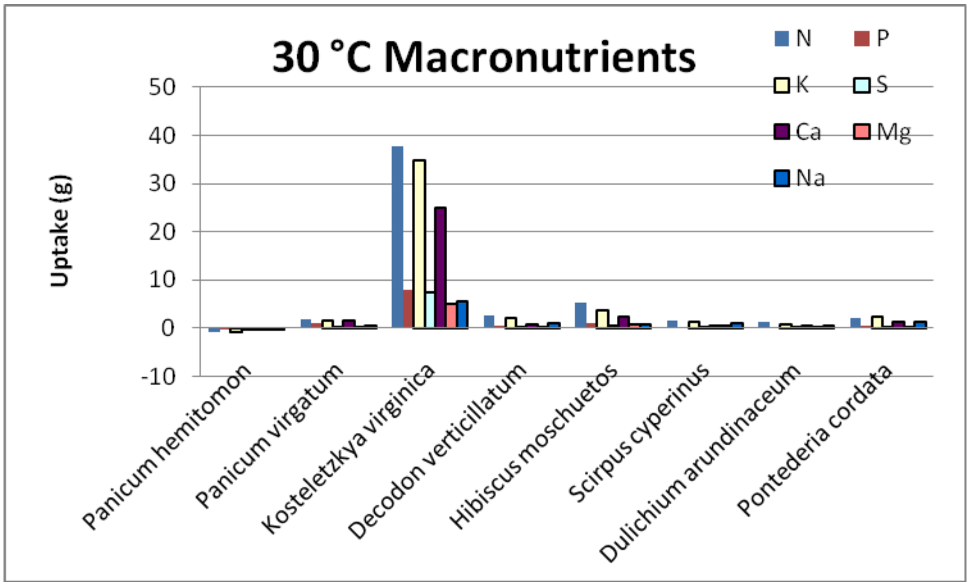


Table 4. Harvest macronutrient content of plants grown under higher temperatures.



Results show that the greatest plant growth and nitrogen and phosphorus uptake was by swamp mallow, *Kosteletzkya virginica*, followed by swamp hibiscus, *Hybiscus* m*ochuetos*.

<u>Demonstration of Commercial Production Methods</u>

6. Develop and demonstrate technology/procedures and conduct individual plant growth and nutrient uptake performance selection trials for several plant species under field conditions;

Outdoor Mesocosm Study

<u>Methods</u>

The study was conducted at the Horn Point Laboratory on the eastern shore of the Chesapeake Bay in Maryland. There were four outdoor raceway tanks used, approximately 6m long, 3 m wide, and 0.6 m deep. The water level was approximately 0.45m deep and was maintained by a stand pipe with needle valves which is connected to a pump from a holding tank. A standing pipe on the opposite end of the raceway drains out the used water.
Four of the plant species used were be based off a previous experiment conducted in Horn Point Lab. In an ongoing project, Northeastern Regional Aquaculture Center (NRAC), at the Horn Point Laboratory twelve species were initially selected based upon the commercial potential to sell and the expected ability to take up nutrients namely nitrogen and phosphorus. This previous portion of the experiment was conducted in a phytotron using hydroponics. The species selected for this experiment are the ones that removed the most nutrients. These species were *Decadon verticillatus, Scirpus cyperinus, Panicum hemitomon, and Carex vulpinoidea*. The species *Panicum virgatum*

was not used in the previous experiment but was selected based on its hardy characteristics as well as its potential for use as a biofuel. In a study conducted by the U.S. Department of Agriculture in ten farms in the Great Plains P*anicum vergatum,* or switchgrass, was found to have potentially more than 500% more renewable energy than the energy used in its growth and production. It also has significant soil conservation benefits as well as low green house gas emissions (Schmer et al. 2008). If this species can take in nutrients it would then be more efficient to use as biofuel rather than compost piles which put nitrogen and phosphorus back into the environment.

Most of these species used in the experiment grow in swamps, marshes, and other shallow bodies of water between Maine and Maryland and West Virginia with prime growing seasons between June and September. *Decadon verticillatus* or Swamp Loosestrife can grow to about 0.6m to 2.7m. *Scirpus cyperinus* or woolgrass is between 1.2- 1.5m tall. It grows in dense clumps and has many inconspicuous flowers (Tiner 1987). *Panicum hemitomon,* also referred to as maidencane, reaches heights of about 0.6 to 0.8m with long leaf blades. *Panicum virgatum* or switchgrass is specifically found in the upland regions of salt marshes or savannahs. It has many branches and spikelets and has been known to be used as ornamental. It has can grow to be 1.9-2.0m. *Kosteletzkya virginica* or Virginia saltmarsh mallow is a hearty plant with round rough and hairy stem. It can grow to 2-4 feet tall and generally flowers August through September with Pink five petaled flowers.

There were 480 plants total, 120 plants in each raceway. This allowed for twenty- four replicates of each respective species. The plants were all purchased from Maryland Aquatic Nurseries. This ensured that each plant plug was grown under the same conditions with the same soil nutrient composition. Plants were weighed before implanting them into the raceways. Initial samples were taken the week of the planting for tissue analysis. Samples were taken every three weeks throughout the experimental period. Samples were dried in a forced air convection oven at 60°C then ground and were sent to A&L Eastern Agricultural Laboratories in Richmond, Virginia. The plant's roots were wound in black mesh and placed inside plastic pots. These pots fit into floating black rafts that will covered the surface of the raceways. This ensured that no light reaches the water and no algal growth occurred. The hydroponic solution used was a 5% Hoagland's Solution which includes potassium nitrate, calcium nitrate, phosphate, magnesium sulfide, and iron. A portion of the solution also contained micronutrients which are KCl, H_3BO_3, $MnSO_4$, $ZnSO_4$, $CuSO_4$, and H_2MoO_4. The solution gave the plants a concentration of about 10mg/L of Nitrogen.

<u>Results and Discussion</u>

Plant growth:

The plants were set in the raceways three and four on June 27, 2008. The remaining raceways were completed on June 29, 2008. Initial weights were taken on these dates. On July 8, 2008 new *Kosteletzkya virginica* was put in place in the raceways due to the poor appearance the original plants were in. After just over three weeks final weights were taken. The percent increase in growth was calculated for each individual sampled. Then averages were taken and are shown in Figure 1. The greatest growth was seen in the species *Kosteletzkya virginica* with an increase of 89.7%. The second greatest growth was exhibited by *Decadon verticillatus* with an estimated growth of 47.9%.

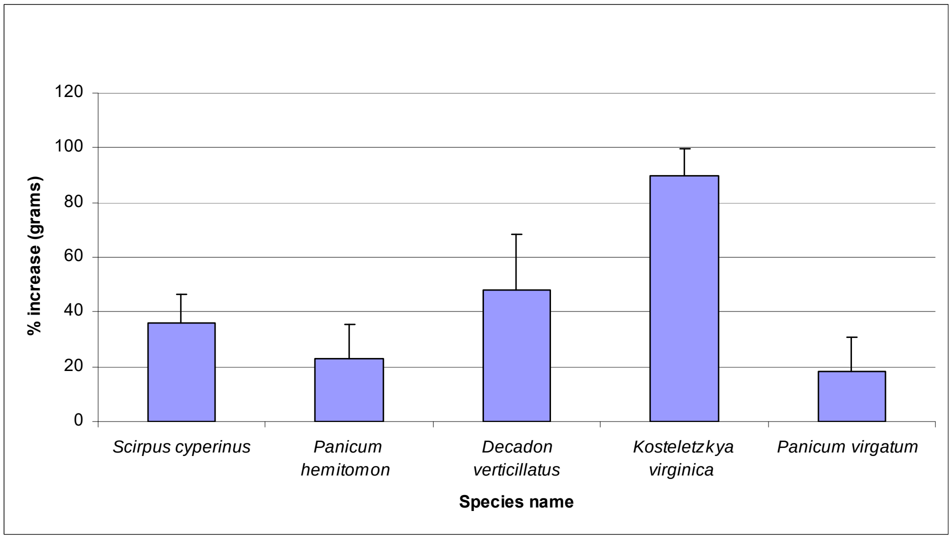


Figure 1. Graph to show the average % increase in growth from the implementation of the plants at the end of June and beginning of July until the first sampling three weeks.

Uptake results:

The tissue nutrient concentration measured by A&L were compared to assess uptake among the species. The difference of the final and initial percent composition of Nitrogen and Phosphorus plant tissue was calculated. It was then divided by the initial composition and multiplied by 100. This was done for each of the six replicates for all five species. The mean differences of nitrogen and phosphorus uptake between the final and initial plants are shown in the Figure 2 below. As seen in the Figure 2 the greatest uptake of nitrogen is represented by *Kosteletzkya virginica*. *Panicum vergatum* has the second highest percent composition of nitrogen. *Panicum vergatum* also took in the most phosphorus. The worst performer of all the species was *Decadon verticillatus.*

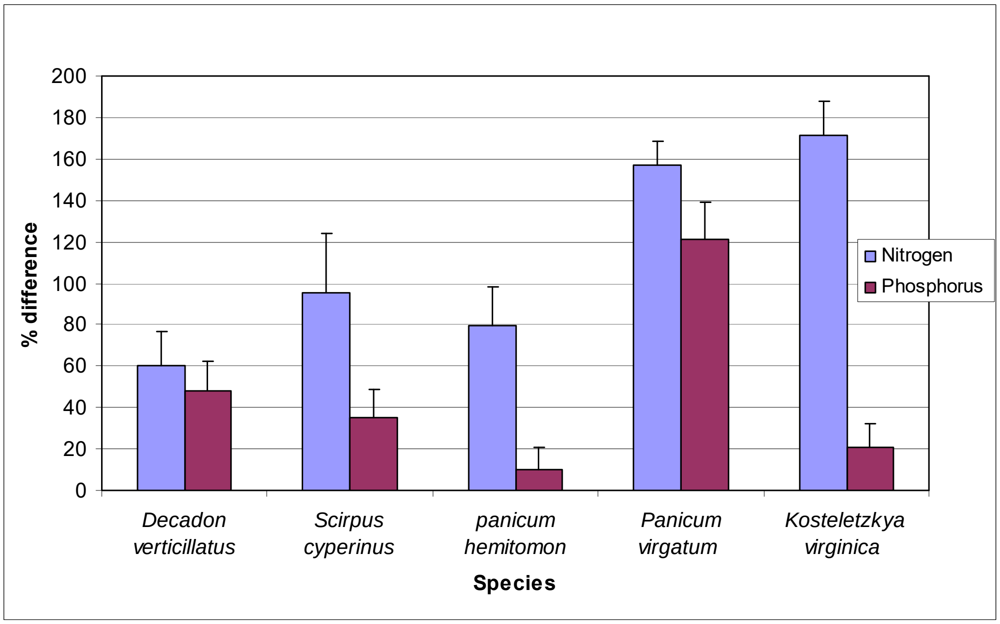


Figure 2. Graph to show the average percent increase in the nitrogen and phosphorus composition of the five plant species over a growing period of just over three weeks.

Throughout the course of the experiment the plants showed signs of nitrogen deficientcy. They exhibited all of the classic symptoms including chlorotic yellowed leaves. In some of the *Decadon verticillatus* a reddish purple appeared on the underside of the leaves as a sign of nitrogen deficiency. Water quality assessments were done on each of the four raceways and the head tank. The results of the water quality tests are shown in table 1. The nitrate/nitrite levels look normal. The anticipated range was between 500-700uM. However, the levels of ammonium in each of the raceways were close to zero, a stark contrast from the level in the head tank which was 93.4uM. One possible explanation for this is that the marsh plants may be selective for ammonium because of their natural habitats. In one study done on the marsh species *Typha latifolia* the uptake kinetics of ammonium and nitrate were compared. The results showed that when grown with ammonium the plant had higher growth, higher nutrient tissue concentrations, and higher contents of adenine nucleotides (Brix 2002). No research has been done on the specific species used in this study. Normally, plants more readily take up nitrate (Reece et al. 2005). In freshwater marsh systems the soil is rich in ammonium. This could be one explanation for the nitrogen deficiency found in the plants. It is possible that they require large amounts of ammonium to sustain them. These plants may also use nitrate as a source of nitrogen, however, it is costly in terms of energy to produce nitrate reductase to transdorm nitrate into ammonium. It is possible that the results for plant growth and uptake are skewed due to the nitrogen deficiency which could possibly slow or stunt the growth of certain species of these plants. *Decadon verticillatus*, for instance, fared the best in the previous phytotron experiment but was one of the species hit hardest by nitrogen deficiency. Adjustments were made in the fourth week of the experiment, after the first sampling to increase the ammonium phosphate by 50% while decreasing the calcium nitrate by 25% to maintain the nitrogen concentration of 10g/L. It was anticipated that this adjustment would allow the plants to recover from their nitrogen deficiency. After this adjustment water quality tests were taken to monitor for any signs of improvement of ammonium levels. To our dismay the numbers came back even lower. It is possible that the ammonium can be disappearing due to the presence of nitrifying bacteria. These bacteria reduce the ammonium and convert it to nitrate and nitrite which eventually could be denitrified under anaerobic conditions. It is not clear however, where these might exist in the system in which the head tanks are continuously bubbled with air.

Upon completion of this experiment a more detailed knowledge of which specific species have the greatest potential to uptake nutrients in a natural setting. There still remains three rounds of plant sampling and nutrient analysis in the experiment. These results may reveal more about the uptake potential of each species over an entire growing season. After the first sampling it can be estimated that P*anicum vergatum* and *Kosteletzkya virginica* took in the most nitrogen and *Panicum vergatum* did the best with Phosphorus uptake. It is possible that in the future these species will be marketable for this ability to take in nutrients from the water. The mitigation of all of these species is vital if the future for waste water management lies in aquatic plants. As the Chesapeake watershed becomes more and more developed it becomes increasingly important to stress the environmental impact that each of us has. Residents may be persuaded to grow their own plants in stormwater ponds that will be suitable for their landscaping needs and that benefit their water quality. This type of maintenance of urban properties may in the long run increase the value of the home or business. To put the uptake observed in this study into perspective on a larger scale, and using our observed uptake of 13.6g.ft^2 and 1.08g/ft^2 of nitrogen and phosphorus respectively; and assuming a 1% coverage rate of the floating wetland per pond acre; 13.05 pounds of nitrogen and 1.03 pounds of phosphorus per acre would be taken up. Expanding this figure to the 200,000 acres of stormwater ponds in Maryland; an estimated 2.61 million pounds of nitrogen and 206,000 pounds of phosphorus would be taken up and removed from stormwater, provided the vegetation is removed from the pond annually. This research also benefits local fisheries including a commercial finfish hatchery in Maine, pond aquaculture at Delaware State University, and a trout system in Virginia. The implications of this experiment may provide valuable information for the future of water treatment and commercial plant sales.

7. Verify the growth performance, nutrient remediation, and aesthetic value of several aquatic plant species in various field applications including stormwater management, hatchery and raceway effluent treatment and pond production; and

Salmon Hatchery System

The Maine Aquaculture Association continued work on its portion of this project. Over the course of the project so far work has progressed steadily with all of our milestones being achieved for the project. The collaboration with two local high school classes that began in year one of the project was continued in year two of the project. We also added collaboration with a chemistry professor at the University of Maine in Machias (UMM).

We continued to weigh the plants from the second field season in July, August and September of 2009 (Fig 2). The plants were then harvested in October of 2009. Randomly selected plants were dried and sent out for plant nutrient analysis.

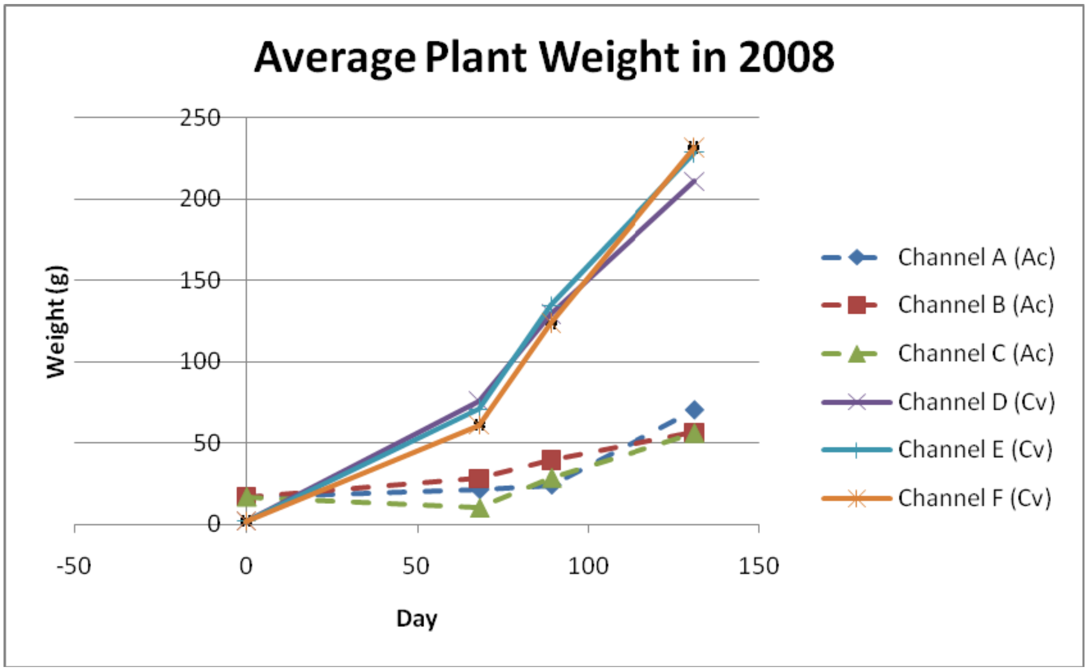


Figure 1: Average plant wet weight in each plant channel for the *Carex vulpinoidea* (Cv) and *Acorus calamus* (Ac) grown in 2009.

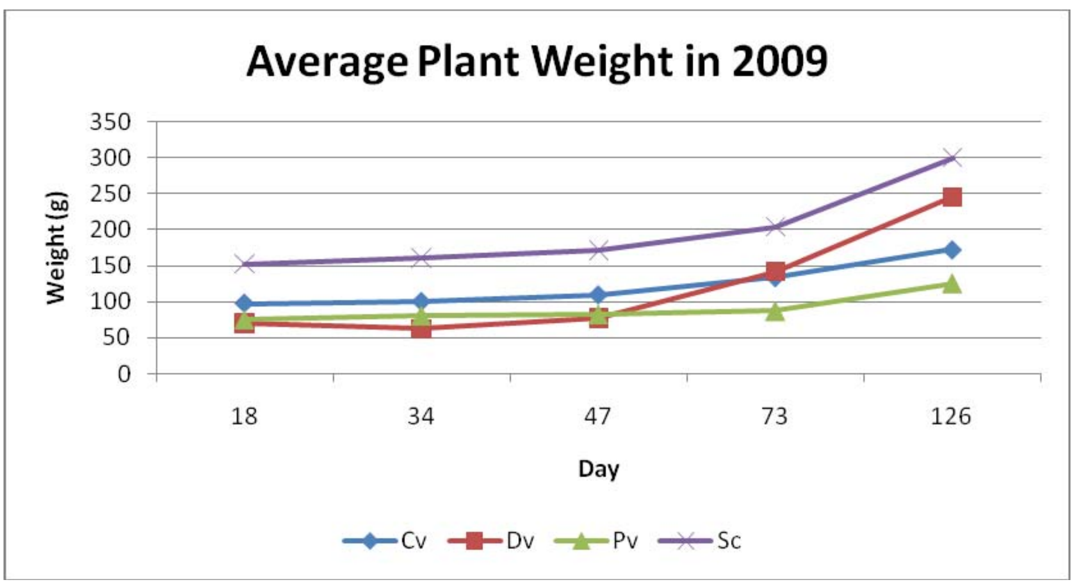


Figure 2: Average plant wet weight across all plant channels for the four species, *Carex vulpinoidea* (Cv), *Decodon verticillatus* (Dv), *Panicum virgatum* (Pv), and *Scirpus cyperinus* (Sc) grown in 2009.

As mentioned above we formed a new collaboration with a Chemistry professor at UMM. This came about as a way to get improved nutrient removal information. The established water quality monitoring program was not being followed by hatchery staff, and was not sufficient to detect changes in the water quality by the plants. Instead, a new trial was designed in the spring of 2009 to establish 4 small plant channels with and equal volume of water per plant as the large plant channels. The water chemistry was then monitored on a 24 hour basis by Dr. Otto who collected water samples to measure the orthophosphate in each small chamber at time 0 and 23 hours after the water change. Sixteen plants of the same species were all randomly assigned to one of the four plant channels after being weighed. At the end of the week the plants were reweighed.

This trial faced several challenges as it was being conducted. The largest of which was the in ability to detect a significant change in the phosphorous level over 24 hours or 7 days. This failure to be able to detect a difference is due to two reasons. First, the amount of phosphorous removed by the plants in the trial periods was calculated to be very small. The second reason was that the high levels of organic enrichment in the water was able to mask our ability to detect the change in phosphorous.

On May 29, 2009 six plant channels and five wetland plots each 4 feet by 8 feet were planted with 72 plants. Each grow out system was planted with 18 plants of each of the following species: fox sedge (*Carex vulpinoidae*), wool grass (*Scirpus cypernus*), switch grass (*Panicum virgatum*), and swamp willow (*Decodon verticillatus*). The plants were planted by the two high school classes, Jill Weber and Mike Pietrak. At the time of planting 5 samples of each species were collected for nutrient analysis. These plants had any potting media washed off of their root stock, were wet weighed and then dried to a constant weight in a 78 C oven. They were then sent to A&L Eastern Labs for composition analysis. At 18 days post planting a randomly selected group of ten plants of each species in each plant channel was marked and weighed. These plants were then reweighed on days 34, 47, 73 and 126 post planting. The trial was terminated on October 2, 2009 when final weights were taken, and a random selection of 4 plants of each species was chosen from the groups being monitored. These were dried and sent for composition analysis.

The dry weight growth of the sampled plants is presented in figure 3. Dry bio mass growth was calculated by subtracting the maximum wet weight of each plant multiplied by the percent water for that species in that plant channel from the maximum wet weight. Figure 4 is the amount of nitrogen removed by each plant over the course of the trial. This was calculated by multiplying the calculated dry biomass of each plant by the average percent nitrogen for that species in that plant channel. The average amount of phosphorous removed by each plant was also calculated in the same manner as Nitrogen.

Utilizing the calculated average amount of phosphorous and nitrogen taken up by an individual plant in the trial, and the average dry weight of each species, the amount of phosphorous and nitrogen taken up on a m^2 basis for our system was calculated. The amount of phosphorous removed (figure 5) was highest for *Decodon*> *Scirpus*> *Carex*>

Panicum. The same pattern was true for the nitrogen removed. Given the very small amounts of phosphorous removed per square meter, 3.7g-1.0g, of plants it was clearly evident that the amount of space needed to remove a sufficient amount of phosphorous from a typical flow through fish hatchery was prohibitively large. For this reason the use of plants to primary remove nutrients from discharge water is not practical for flow through facilities. They may however be potentially useful in a recirculation application prior to a biofilter or some secondary water quality polishing may be achieved if the plants are being cultured for other reasons such as economic diversification of the farm.

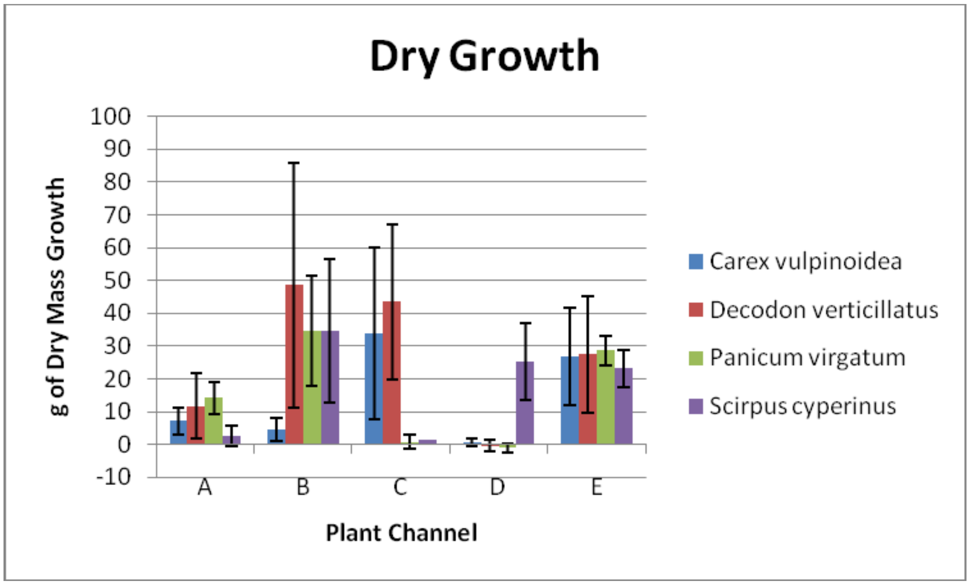


Figure 3: Amount of dry biomass accrued over the growing season in 2009 for each plant species in each plant channel (n=4).

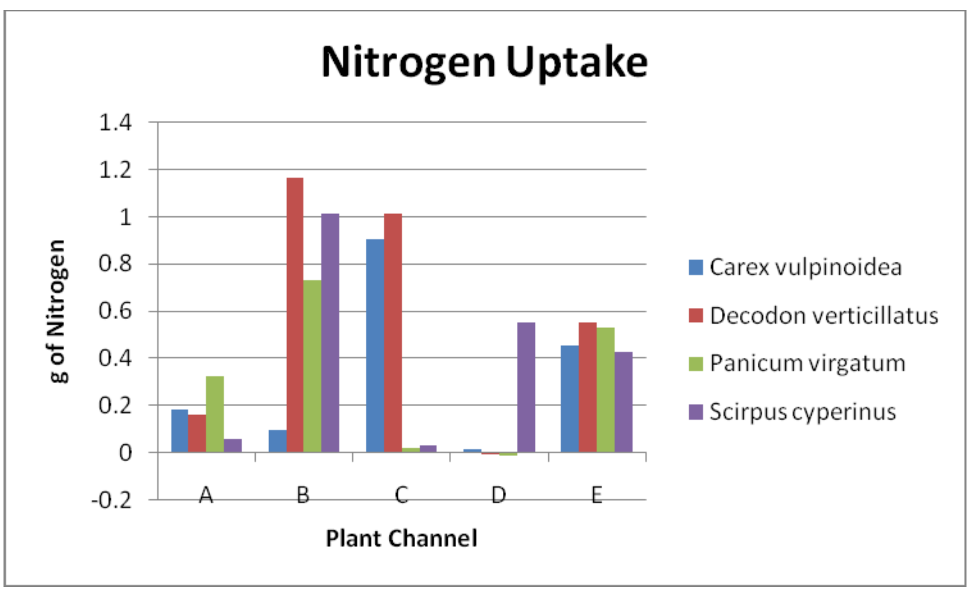


Figure 4: Average amount of Nitrogen removed by each plant.

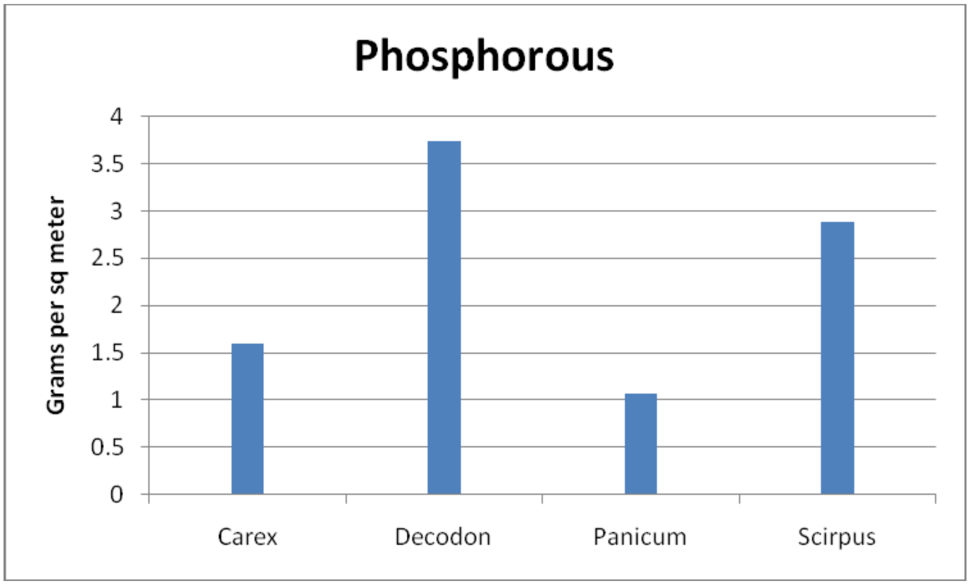


Figure 5: The calculated grams of phosphorous removed per square meter of plant channel. Plants in our plant channel were planted at a denisty of ~ 24 per square meter.

Aquatic Plant and Stormwater Pond Management

Floating aquatic plant rafts or 'floating wetlands" were employed in two stormwater ponds in a 350 home community in Easton, MD in collaboration with project partner, Maryland Aquatic Nurseries. Plant growth for 2008 was monitored, and final samples were collected in October and were not analyzed in time for this report. In addition, 2008 was a dry year with only one large rain event that provided pond outflow data, and therefore, assessment of nutrient uptake of ponds. For Pond 1 described as an old farm pond converted to a stormwater pond, average nitrogen uptake over a 4 day rain event was 26.9% and 38.5 % for phosphorus. Nitrogen and phosphorus reduction s for the Pond 2 (a new two stage stormwater pond) was 8.8% and 15.8% respectively. In both ponds, it was noted that nutrient uptake peaked at second day of rain event. Outflows of nutrients exceeded inflow during third day for Pond 1. In this case, Pond 1 receives runoff from several other ponds within the community (except Pond 2), which may suggest that flow rates during major storm events, exceed the capacity of plants (especially at current stocking rate of five- 48 square feet raft units) to mitigate continuous nutrient uptake. Pond 2, the two stage system observed lower nutrient outflows compared to inflow four all four days of the rain event. The forebay (stage one) of Pond 2 treatment may have provided for nutrient capture and reduced water velocities.

View of floating wetland raft system exposing root growth.

View of floating wetlands deployed in Farm Pond (3 months of growth)

Nutrient uptake by plants in floating wetlands:

The following table shows results of nitorgen and phosphorus uptake by the plants in the floating wetlands (4 months growth).

No. of rafts	Plant biomass production (lbs)	Biomass (dry wt. lbs.)	Total Nitrogen Uptake (lbs)	Total Phosphorus Uptake (lbs)
8	1020	268.7	9.04	0.71

Overall nutirent uptake by plants in floating wetlands showed nutrient uptake of 146.3 g/m^2 and 11.7g/m^2 for nitrogen and phosphorus respectively. Having this information will enable estimation of plant and floating wetland stocking density in ponds of varied nutrient concentrations.

Trout Raceway Integrated System

The aquaculture system in WV is a cold water flow-through raceway system that raises trout. The water source is an artesian spring which supplies 400 gpm. Due to the spring water source, water temperatures are very constant, averaging 12.5°C and varying less than 2°C throughout the year. The combination of cold spring water and a flow-through rearing system that contributes low nutrient inputs yields water that has characteristics quite unlike that which is typically used for either hydroponic or aquaponic plant production. Therefore, there was little guidance as to which plants would perform well within this system. A previous experiment under the USDA-SARE program demonstrated that ornamental plants that are classified as warm season crops do poorly within this system despite a high tolerance for cold temperatures while plants that are considered cool season crops and thrive under cold conditions do well in this system.

At WV, the first year experiment was designed to identify native ornamental species that performed well within a cold water flow-through raceway system. Thirty-three native plant species were selected from a list of plants that may potentially be used in restoration and remediation. The plants were placed in an 8' long plant growing channel constructed of plywood lined with EPDM. Water depth, controlled with a standpipe at the end of the channel, was set at 6". Four replicates of each species were used in the experiment. Three replicates were placed in the channel for the growing season while the fourth was destructively sampled to determine initial biomass (dry weight). Dry weights were determined at the end of the growing season from the three random samples. Mean growth ratio (final wt./initial wt.) was used to evaluate plant performance (Table 1.). This experiment was repeated in yr 2 to ensure results reflect plant performance despite year-to-year variation in growing conditions.

Table 1. Native Plant Trial Mean Growth Ratios

Scientific Name	Common Name	Initial Weight	Mean Final Weight	Mean Growth Ratio
Carex vulpinoidea	Fox Sedge	12.09	325.29	26.91
Myriophyllum aquaticum	Parrot Feather	2.53	121.83	48.15
Sisyrinchium montanum	Blue-Eyed Grass	2.83	116.13	41.03
Typha latifolia	Braod Leaved Cattail	24.17	106.24	4.40
Decodon verticillatus	Swamp Water Willow	1.37	60.80	44.38
Mimulus ringens	Lavender Musk	4.78	59.14	12.37
Typha angustifolia	Narrow Leaved Cattail	6.98	50.77	7.27
Myosotis scirpoides	Forget-Me-Not	1.99	45.49	22.86
Myriophyllum spp.	Red Stemmed Parrot Feather	2.59	44.04	17.01
Dulichium arundinaceum	Dwarf Bamboo	7.91	39.53	5.00
Typha latifolia 'variegata'	Variegated Cattail	6.94	34.42	4.96
Pontederia cordata	Pickerel Rush	3.86	30.65	7.94
Acorus americana	Sweet Flag	2.61	29.59	11.34
Taxodium distichum	Bald Cypress	9.49	26.28	2.77
Kostelekya virginica	Seashore Mallow	4.98	25.52	5.13
Lysimachia nummularia	Creeping Jenny	0.93	24.04	25.85
Justica americana	Water Willow	1.82	23.23	12.76

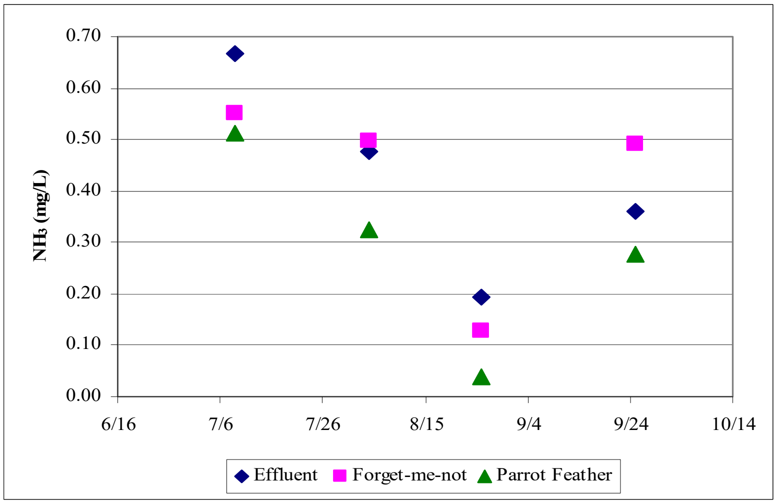
Eriophorum angustifolium	Cotton Grass	6.07	19.89	3.28
Lobelia cardinalis	Cardinal Flower	0.24	17.08	71.17
Acorus calmus 'Variegatus'	Sweet Flag, Variegated	3.42	15.95	4.66
Typha minima	Dwarf Cattail	1.36	11.55	8.49
Alisma plantago-aquatica	Water Plantain	1.94	9.35	4.82
Menyanthes trifolia	Bog Bean	4.48	8.61	1.92
Saururus cernuus	Lizard's Tail (Plug)	1.12	8.43	7.52
Equisetum hyemale	Horsetail	0.4	7.99	19.98
Aponogeton distachyus	Water Hawthorne	2.93	6.55	2.24
Ceratophyllum demersum	Coontail	5.53	5.84	1.06
Juncus efusus 'Spiralis'	Corkscrew Rush	1.6	5.56	3.48
Lobelia fulgens	Queen Victoria Lobelia	1.51	4.07	2.70
Orontium aquaticum	Golden Club	1.55	4.04	2.60
Sagittaria latifolia	Arrowhead	0.88	3.46	3.93
Egeria densa	Anachris	3.56	3.42	0.96
Peltandra virginica	Arrow Arum	0.45	3.42	7.59

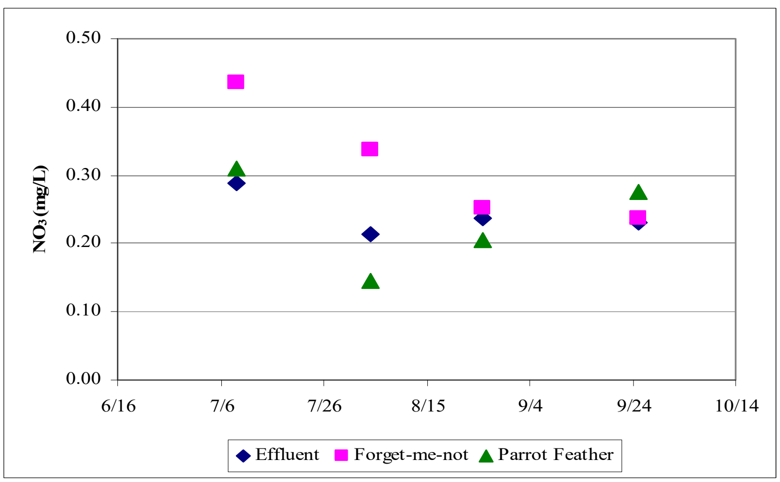
In years 2 & 3, Forget-me-not (*Myosotis scirpoides*) and Parrot feather (*Myriophyllum aquaticum*), two of the top biomass producing species identified in plant growth trials, were selected for in-depth study of growth and nutrient removal over the growing season. In early summer plants potted in vermiculite were placed in a 2' x 18' channel that received effluent from the flow-through trout raceway. Each channel was planted with 144 plants of a single species with 3 replicate channels for each species. Biomass and nutrient samples were taken periodically through the summer.

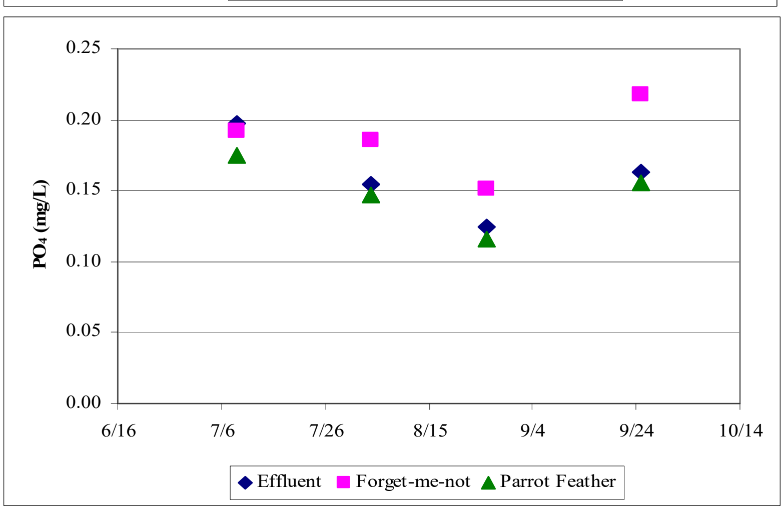
Forget-me-not and Parrot Feather were evaluated in both 2008 and 2009. These two plant species were repeated in 2009 to be able to collect two growing seasons of data but also to be able to gain data on parrot feather since it performed very poorly in the 2008 trial. During the course of the 12-week evaluation, Parrot Feather and Forget-me-not were sampled every 3 weeks. Parrot Feather performed better than the Forget-me-not with respect to sampled biomass taken every 3 weeks and the total crop biomass evaluated at week 12. Forget-me-not and Parrot Feather produced a total crop biomass respectively of 35 and 297 kg. Parrot Feather did exceptionally well in 2009. This plant would have been an excellent commercial source for mitigation plant contracts as well as useful by commercial aquaculture producers as a nutrient and fish solids filter. Forget-me-not did not perform as well in 2009 as compared to 2008. Forget-me-not has the potential to also be an excellent commercial source for mitigation plant contracts. It has good biomass production as well as ease of production in commercial pots for transportation and transplanting. Plant nutrient analysis is ongoing.

Over the summer, effluent nutrient concentrations averaged 0.42 mg/L NH_3, 0.24 mg/L NO_3 and 0.16 mg/L PO_4 (Figure 1.). Parrot feather was more effective at nutrient removal than forget-me-not. Over the growing season parrot feather removed 39% of the NH_3, 22% of NO_3 and 20% of PO_4 while forget-me-not removed only 3% of the NH_3 and there was no reduction in NO_3 or PO_4 concentrations. Lack of effective removal by forget-me-not was due in some part to overtopping by the rampant growth of parrot feather.

Figure 1. Soluble nutrient concentrations of the raceway effluent entering the plant channels and concentrations at the end of the plant channel after removal by forget-me-not or parrot feather.







In year 3, Cardinal Flower (*Lobelia cardinalis*) and Fox sedge (*Carex vulpinoidea*) were selected for in-depth study of growth and nutrient removal over the 2010 growing season based on mean growth rate ratios determined from yr 1. Plant evaluation and plant/water nutrient analysis is ongoing.

The WV portion of the study demonstrated that with proper plant choice, aquaponics could be integrated into a flow-through raceway system despite low nutrient concentrations and low water temperatures. A wide variety of plant species may be grown in this system and provide additional products for fish producers. Some species, such as parrot feather, have the added benefit of improving water quality through nutrient removal. These plants may be sold or composted thereby permanently removing nutrients from the aquatic system.

Objective; Grower Outreach and Education

8. Plan and conduct an Extension program that will disseminate research-based information from this project to the plant industry on the economics of the project, provide information on plant species in a performance database and demonstrate culture technology to industry participants and other stakeholders, including terrestrial and aquatic nurseries, restoration/mitigation contractors, natural resource management agencies, contractors/developers, and research and Extension faculty involved in aquatic environment education.

Objective: Grower Outreach and Education

Outreach activities of this project included technical support to interested parties primarily via one on one discussions and tours of the various integrated systems, workshops and presentations.   Fifteen presentations have been made relating information on integrating aquatic plants in various settings for nutrient remediation. Over 740 people were educated. Tours of the three integrated systems (DSU, ME and WV) and the stormwater ponds in MD, provided technical information and support to over 650 people. Nine publications have been produced with an estimated circulation of over 10,000 people. Fifteen publications have resulted by project end with two manuscripts for journals under development. Web sites at the three universities also include information about this project and its results.

PROJECT COMPLETION REPORT
SIGNATURE PAGE

PROJECT CODE: **SUBCONTRACT NO.:**

PROJECT TITLE: Evaluating Restoration and Mitigation Aquatic Plant Species and Markets to Advance the Commercialization of the Industry

PREPARED BY:

*Andrew M. Lazur*_________________
Project Coordinator

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Date