Nutrient competition between algae and *Juncus effusus* in the Lake Fayetteville Artificial Wetland

An Undergraduate Honors Thesis

in the

Department of Crop, Soil, and Environmental Science

Submitted in partial fulfillment of the requirements for the
University of Arkansas
Dale Bumpers College of Agricultural, Food and Life Sciences
Honors Program

by

Toryn Jones

April 2015

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J. Thad Scott, Ph.D., Chair

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Lisa Wood, Ph.D.

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Curt Rom, Ph.D.
Summary

The effect of N and P availability on macrophyte and algal growth has been extensively documented. The same is true for research regarding competition between wetland macrophytes. However, there is considerably less research focusing exclusively on how nutrient competition between algae and wetland macrophytes affects the growth of these plants. This study examined the relationship between nutrient concentrations (N and P), algal concentrations, and the growth of *Juncus effusus*, or soft rush. *Juncus effusus* growth in the Lake Fayetteville Spiral Wetland was monitored over a four month period during the prime growing season. Towards the end of the growing season, 18 plants were taken from the wetland and replanted in 1 of 6 treatments: ‘plant-only’, ‘algae-only’, ‘combined’, ‘plant-only +supplement’, ‘algae-only +supplement’, or ‘combined +supplement’. The algae and combined environments received an inoculation of algae, and the +supplement treatments received an infusion of an N and a P supplement. An ANOVA test was conducted using SAS to determine the presence of a significant relationship between *Juncus effusus* growth, nutrient concentrations, and/or algal growth. No significant relationship existed between *Juncus effusus* and nutrient concentrations or between *Juncus effusus* and algal concentrations. There was a significant relationship between algal growth and the presence of *Juncus effusus*, which produced an additive effect causing the greatest algal growth in the ‘combined +supplement’ treatment. Results indicate that nutrient competition between *Juncus effusus* and algae in the Lake Fayetteville Spiral Wetland is not the limiting factor in *Juncus effusus* growth in the wetland. Algae in the lake may be able to photosynthesize longer into the year due to light limitations imposed by the Spiral Wetland delaying the rate of non-photochemical quenching in algae.
I. Introduction

Over the past few decades, rising social, scientific, and political anxiety regarding subjects such as global warming, desertification, eutrophication, and numerous other environmental concerns has resulted in the creation of a new, educational art style. Ecological art, or eco-art for short, is an increasingly popular form of art that uses various media to raise public awareness of several environmental issues (Sanders, 1992). Examples of eco-art cover a broad range of possibilities. Some eco-artists create simple picture portfolios to portray the progression of deleterious environmental process; some create sculptures out of refuse, and still others create functioning wildlife habitats and/or habitat improvements from various materials (Sanders, 1992).

The Lake Fayetteville Spiral Wetland was an artificial wetland covering approximately 1000 ft$^2$ near the dam of Lake Fayetteville, Arkansas. The floating wetland was designed and built by Stacy Levy, an artist who has created multiple eco-art installations (http://www.stacylevy.com/). Construction of the Spiral Wetland project began in the spring of 2013, and was completed in July 2013 when plants were put into the structure. The artificial wetland was decommissioned in October of 2014. The purpose behind the Lake Fayetteville Spiral Wetland was to educate the public on the effects of eutrophication and to help reduce the impact of nitrogen and phosphorus enrichment on the lake. Also, the artificial wetland provided an aesthetically pleasing view from the lake’s park trail, and served as a habitat for many insects, fish, and birds. However, an unanticipated issue arose in that the plant installed in the wetland, *Juncus effuses*, did not appear to grow after being established in Lake Fayetteville. This was surprising to the artist and other project participants because they assumed Lake Fayetteville would support substantial plant growth since it is a hypereutrophic lake (Scott and Grantz 2013).
Figure 1 shows the spiral wetland and information which was displayed on a plaque near the wetland for interested persons.

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### a. *Eutrophication: cause and effect*

Eutrophication can be defined as the excessive nutrient enrichment of a water body (Smith, et. al, 1999). The two most common nutrients involved in eutrophication are nitrogen and phosphorus. Nitrogen and phosphorus are the two limiting agents in plant growth, and, when an excess is introduced into the environment, plants in that environment can begin to grow at a consistent rate until they either reach critical mass or exhaust the excess nutrients (Koottatep and Changrak, 1997). Unfortunately, nitrogen and phosphorus are highly concentrated in common run-off contaminants such as animal feces, leaf litter, food, and nutrient fertilizers making eutrophication of local water bodies a fairly common occurrence, especially near agricultural areas (Hammer and Knight, 1994).

Algae and/or cyanobacteria in the water body are able to use a combination of the excess nutrients and energy from sunlight to photosynthesize, fueling growth and reproduction at overwhelming levels (Smith, 1982). Algae and cyanobacteria continue to grow and thrive until they deplete the excess nutrients. Even while they are still growing and reproducing, large quantities of algae and cyanobacteria are dying off and collecting at the bed of the water body. Many detritivore macro-invertebrates and heterotrophic bacteria feed on the decaying algae and cyanobacteria. When these species feed, they respire at great rates, using large quantities of dissolved oxygen (Stevenson, et. al, 1996). This process quickly depletes the dissolved oxygen stored in the water body by the algae and cyanobacteria. Eutrophic zones are areas bound by
extremes. These areas alternate between dangerously high and equally dangerously low concentrations of dissolved oxygen. Many ecological consequences stem from eutrophication.

The most notable effects of eutrophication are algal blooms and hypoxic zones. With the possible exception of a runoff channel visibly contaminated by fertilizer and/or excrement, algal blooms are one of the first definitive signs of eutrophication. Algal blooms are the emergence of extremely high concentrations of algae and cyanobacteria (Stevenson, et. al. 1996). These blooms are characterized by their stench, deep red and green colorings, and their high rates of photosynthesis. Hypoxic zones are areas where dissolved oxygen is so low that very few aquatic lifeforms can survive. Hypoxic zones are created once algal blooms die off and detritivores deplete the dissolved oxygen through respiration, while breaking down the algae for chemical energy (Smith, et. al, 1999).

Each of these results has a deleterious effect leading to the slow breakdown of the aquatic ecosystem. Loss of aquatic species and resources can lead to further damage by negatively impacting the surrounding terrestrial species and environment. For humans, eutrophication can lead to the loss of recreation waters, drinking water, food sources, and a host of other benefits drawn from neighboring water bodies and related terrestrial environments (Smith, et. al, 1999).

b. Artificial Wetlands: agents of eutrophic remediation

Under section 404 of the Clean Water Act of 1972, wetlands are defined as, “areas that are inundated or saturated by surface or ground water at a frequency and duration sufficient to support, and that under normal circumstances do support, a prevalence of vegetation typically adapted for life in saturated soil conditions” (EPA, 2012). An artificial wetland is a manmade reproduction of a wetland ecosystem that typically serves the purpose of either replacing a
damaged wetland or assisting in the remediation of a polluted waterbody (Kadlec and Wallace, 2009). Artificial wetlands have become increasingly abundant since the amendment of section 404 of the Clean Water Act of 1972. This particular act requires any company, developer, and/or government body that destroys a wetland area to mitigate said destruction by constructing an artificial wetland of equal or greater size in a neighboring area (EPA, 2012). This act was established following the discovery of hazardous ecological imbalances stemming from mass drainage of the Florida everglades and much of Louisiana’s marshes. Despite the vast environmental destruction caused by the regulated draining of wetlands, this practice did produce one good consequence in that we now understand, to a much greater degree, the ecological functions and importance of wetlands. Thus, it is with this understanding that we have begun to construct artificial wetlands to assist in the remediation of aquatic bodies heavily polluted by such things as fertilizers, sewage and other contaminants with relatively high nutrient concentrations (Shutes, 2001).

There are three primary functions by which artificial wetlands are able to prevent, manage, and, in some cases, reverse eutrophication. These functions are nutrient competition (Reinhardt, et. al, 2006), light attenuation (Kadlec and Wallace, 2009), and BOD reduction (Karathanasis, et. al, 2003). Hydrophytic vegetation, like most plants, is limited in growth primarily by its access to nutrients and sunlight. It can generally be assumed that, in the presence of excess nutrients and ample sunlight, hydrophytes will reach their maximum growth potential at a relatively fast pace (Gusewella and Koerselman, 2002). In a water body experiencing eutrophication, algae and cyanobacteria are often the only organisms capable of using the nitrogen and phosphorus suspended away from the shore (Stevenson et. al, 1996). However, when an artificial wetland is constructed and placed over the waterbody, it introduces a number
of new hydrophytes that are able to utilize large quantities of nitrogen and phosphorus for
growth, thus reducing the amount of nutrients available to algae and cyanobacteria (Vymazal et.
al, 2007). The competition for nutrients brought on by the introduction of an artificial wetland
puts a stress on algae and cyanobacteria which helps to regulate growth and prevent algal blooms
(Crumpton, 1989). In situations involving point source pollution, wetlands can be constructed
near the point of contamination. Wetlands have a tendency to increase residence time of
contained water and contaminants (Saunders and Kalf, 2001). In point source pollution, this
feature allows certain wetland hydrophytes to filter out a high concentration of various
contaminants before they reach the waterbody being protected.

A somewhat similar effect happens with sunlight. Because algae and cyanobacteria use
photosynthesis as a means of energy production, sunlight is crucial in their growth process.
Studies involving the planting of trees near stream and river banks have shown that tree lines
serve as a runoff barrier, as well as creating intricate root systems which compact the soil and
limit erosion of nutrient enriched soil into the water (Dwyer, et. al, 1992). Theoretically
speaking, many of these trees could also shade the streams and rivers, thus limiting the amount
of consistent sunlight reaching algae as they flow downstream. However, such actions are not as
viable in lentic systems such as lakes or even large ponds where shady trees cannot cover a bulk
of the water’s surface. Hydrophytes, like most other plants, absorb sunlight energy through
photosynthesis. And, while placing an artificial wetland in a lake would technically create a
competition for sunlight energy, algae and cyanobacteria require so little energy relative to what
is available that the environmental stress created by the competition would be insignificant
(Kadlec and Wallace, 2009). While competition is an unlikely factor in algal light absorbance,
there is still a way for wetlands to reduce photosynthetically available light to algae. By creating
a wetland with a solid, opaque layer, light in unable to reach the algae and cyanobacteria under the constructed wetland. In this way, wetlands are able to partially manage the amount of energy available to algae and cyanobacteria (Kadlec and Wallace, 2009).

Wetlands also have an ability to reduce BOD, or biological oxygen demand (Karathanasis, et. al, 1997). This feature, however, is predominantly prominent in shore based artificial wetlands. BOD is the measure of dissolved oxygen (DO) needed to completely dissolve organic matter in a water body (Karathanasis, et. al, 1997). Much like in the aforementioned point source situation, high residence time and retention rate in a wetland allows for microbial organisms to break down organic matter from runoff before it enters the primary waterbody (Karathanasis, et. al, 1997). This helps to slow the depletion of dissolved oxygen within the waterbody. While the reduction of BOD by controlling the amount of incoming organic matter does not directly remEDIATE eutrophication, it does slow the rate of dissolved oxygen depletion which is a destructive result of eutrophication.

There are several benefits and drawback to constructing an artificial wetland as a remediation strategy. The key factors are time, space, money, and personnel. Wetland remediation can take several decades to have a significant effect (Turner, et. al, 2000). Depending on the waterbody being remediated and the wetland design, remediation by wetlands can also be costly and require heavy maintenance and monitoring for the first several years (Turner, et. al, 2000). Yet, due to their resiliency and flexibility, artificial wetlands are an effective long-term and supplemental remediation strategy (Barbier, 1993). This research project seeks to examine the effectiveness of the Lake Fayetteville artificial wetland at reducing the rate of eutrophication in Lake Fayetteville and to propose design modifications which could improve future remediation strategies.
II. Research Hypotheses

There were three hypotheses for this study:

Hypothesis 1. Juncus effusus biomass and algal biomass will be greater where they are grown independently than where they are grown together.

Hypothesis 2. Juncus effusus biomass and algal biomass will increase with increasing nitrogen and phosphorus concentrations in Lake Fayetteville water.

Hypothesis 3. Algae and Juncus effusus grown independently from the other and with added nitrogen and phosphorus will display a synergistic effect rather than an additive effect.

III. Materials and Methods

a. Observation Site and Sampling Methods

The Lake Fayetteville Spiral Wetland covered approximately 1000 square feet and held 3,000 Juncus effusus samples with 500 replacement samples throughout its existence. Change in plant biomass and plant carbon to nitrogen ratios (C:N) were measured over the 2014 growing season. Starting April 19th, 2014, three plants were collected from the wetland at three week intervals. For each set of samples, one plant was taken from the base of the spiral, one plant was taken from the elongated “neck” of the spiral, and one plant was taken from the outermost layer of the spiral. Other than their general location on the spiral, the plants were selected at random; neither root nor shoot length were taken into account. Seven total sample sets were taken. Sampling ended on August 3rd, 2014.
b. Nutrient Competition Experiment

To test the effects of nutrient competition on the growth of *Juncus Effusus*, a two-variable experiment was conducted that compared six varying growth conditions for algae and *Juncus Effusus*. The variables being considered were (1) the presence of a competitive organism (algae and/or *Juncus effusus*) in the growing environment and (2) availability of nutrients, specifically nitrogen and phosphorus, in the growing environment. Fifteen planters with *Juncus effusus* roughly similar in root and shoot length were selected from various parts of the wetland and taken back to the University of Arkansas Experiment Station in Fayetteville where the experiment was conducted. The roots and shoots of these plants were cleaned to minimize unwanted contamination later in the design. Eighteen five-gallon buckets were laid out in three, 2x3 sets. Each set was a replicate. Each bucket was filled with five-gallons of freshwater from a pump at the experimental station. Afterwards, twelve of the buckets (four in each set) had a planter harnessed in using bailing wire. These planters were harnessed just above the water so the plant roots were submersed in the water. The remaining six buckets (two per set) received a 75ml inoculation of concentrated algal biomass (algae was obtained from a separate experiment occurring near Lake Fayetteville). In each set, two buckets already containing a planter were also given a 75ml inoculation of concentrated algae biomass. This resulted in each set of replicates having two buckets with only algae, two buckets with only *Juncus effusus*, and two buckets with algae and *Juncus effusus* combined. In each set, one bucket from each of the previously mentioned pairs was also given a 30ml infusion of trisodium phosphate and a 50ml infusion of potassium nitrate. These dosages were calculated using the maximum concentrations found in Lake Fayetteville. Thus, each set contained one “algae-only” bucket, one “algae-only
+supplement” bucket, one “plant-only” bucket, one “plant-only +supplement” bucket, one “combined” (algae and plant) bucket, and one “combined +supplement” bucket. Each bucket was labelled accordingly and numbered 1, 2 or 3 depending on the set to which it belonged. To further decrease bias, the position of each bucket in a set was chosen at random using a grid and random number generator. Figure 2 displays a basic schematic of the experimental design. The buckets were filled weekly and supplied with aerators. This set up was monitored and allowed to run for approximately two months, at which time it was disassembled and the plants were harvested to measure biomass and C:N. The 3 plants (of the 15 taken from the lake) not accounted for in this process were taken to the lab shortly after the assembly of this experiment and measured for biomass and C:N. This was done to compare the average starting mass and nutrient concentration with the post-experiment measurements.

c. Plant Biomass

To measure the differences in plant biomass, each plant sample taken from the artificial wetland and from the aforementioned experiment was removed from its planter. Due to extended time in water, most of the soil had washed out of the planters. Instead, the planters contained thick masses of roots that appeared roughly identical in size; therefore it was assumed that differences in plant mass within the planters were negligible (assuming plants were of similar size and density). To measure the relative weights of the plant roots and the plant shoots over time, the roots of each plant were removed beginning at the bottommost part of the soil conglomerate and the shoots of the plants were removed beginning at the uppermost part of the soil conglomerate. The roots and shoots were carefully gathered and put into separate bags. This was done for each plant with each plant having its own set of bags. Due to the mass of roots
within the soil being considered negligible, the mid-section of each plant was discarded. After
the roots and shoots of each sample set were divided and bagged, the bags for that set were
placed in a drying oven for no less than one week and up to three weeks. At the end of the drying
period, the contents of each bag were measured using a Mettler Toledo Xs104 balance. This
process provided the root weight, shoot weight, and total weight of each plant sample. Following
this process, plants were stored until they could be ground for C:N analysis.

d. Plant C:N

    Dried and weighed plant samples were ground into a powder using a plant mill at the
University of Arkansas Experiment Station. The roots and shoots of each plant were ground
individually and stored in plastic scintillation viles. To ensure data integrity, the grinder was
opened and cleaned between each run. The 66 samples (33 roots and 33 shoots) were ground
over a two day period. Due to limitations of the C:N analyzer, the initial grinding of the samples
did not provide fine enough plant material; thus, each sample was also run through a Wig-L-Bug
grinder, which is a modified ball mill that produces a fine powder in plant samples. The finely
ground contents were placed back into their respective viles. This process was completed for
each sample, with each sample being ground for 45-seconds. Samples were analyzed for carbon
and nitrogen content using a Thermo Flash 2000-C:N analyzer. Multiplying the dry biomass by
the relative proportions of carbon and nitrogen provided the weight of carbon and weight of
nitrogen for each plant, shoot, and root.
e. Chlorophyll-A Concentrations

During the course of the experimentation, eight sets of water samples were taken at irregular intervals. To do this, the water in each of the 18 buckets was stirred and a 300ml, brown, plastic bottle was submerged in the water. The water in these bottles was vacuum-filtered and filters were retained for chlorophyll-a analysis. The amount of water filtered for each sample was recorded. The filters were stored in a lab freezer until all water samples were collected. When all samples were collected, the filters were placed in individual test tubes which were filled with seven milliliters of 90% acetone. These test tubes were then capped and stored in the freezer overnight. The following day, three milliliters of each sample was placed into smaller test tubes. These test tubes were analyzed on a pre-calibrated Turner Fluorometer which provided the chlorophyll concentration of each sample. As stated, initially there were eight sets of water samples taken throughout the experimental portion of this project. However, due to human error involving the fluorimeter, half of these samples were rendered useless. Thus, only four sets of chlorophyll data are reported.

f. Statistical Methods

Data was compiled using Microsoft Excel. All statistics were conducted using SAS. A two-way analysis of variance (ANOVA) was run on the chlorophyll-a concentrations to test the significance of algae biomass concentrations across the six treatments. A one-way analysis of variance (ANOVA) was run on the carbon weights, nitrogen weights, and the CN ratios of each root sample, shoot sample, and whole plant harvested from the experimental phase. The reason
that a two-way analysis was run on the chlorophyll-a concentrations but not the rest of the data is because the chlorophyll-a measurements represented temporal data.

**IV. Results**

a. *Lake Fayetteville Plant Mass*

Data gathered during monitoring of wetland plant growth on the lake showed an average increase in biomass of about 569% with average plant weights starting at 2.6 grams and peaking at 14.8 grams. Root biomass reached an average increase in weight of approximately 271% with an average starting weight of 1.7 grams and an average peak weight of 4.6 grams. Shoot biomass reached an average increase in biomass of approximately 1133%, with an average starting weight of 0.9 grams and an average peak weight of 10.2 grams. All starting weights were averaged from data gathered on April 19\textsuperscript{th}, 2014, and all peak weights were reached on August 3\textsuperscript{rd}, 2014. Subsequent samples were taken on August 11\textsuperscript{th}, 2014 to compare the average growth of the experimental plants conditions. Data from this set showed a slight decrease in biomass weight with total plant biomass decreasing by an average of 1.1 grams. Figure 3 shows the increase in total plant weight and the changes in percent shoot and root mass over time.

The University of Arkansas water quality lab provided temperature, dissolved oxygen, chlorophyll-A and nitrate concentration data for Lake Fayetteville during that summer. From April 4\textsuperscript{th} to May 16\textsuperscript{th}, surface water temperature fluctuated from 14.06 degrees Celsius to 19.54 degrees Celsius. From June 3\textsuperscript{rd} to July 31\textsuperscript{st}, surface water temperature stayed relatively steady with fluctuations between 25.43 degrees Celsius and 26.91 degrees Celsius. The final temperature measurement was taken on August 18\textsuperscript{th}, and showed a 3 degree increase from 25.82
to 28.99 degrees Celsius. Dissolved oxygen measurements taken throughout the summer fluctuated primarily between 6.37 mg/L and 12.11 mg/L. Nitrate concentrations dropped by 97% from 0.86 mg/L in mid-March to 0.02 mg/L in mid-July, with several fluctuations between those times. Figure 4 shows the changes in nitrate compared to temperature during the research period. It should be noted that no statistical analysis was run on any of the aforementioned data. This is due to the nature of the data. All measurements were taken over time by two separate research entities and dates of measurements, though over the same general time period, do not coincide. Therefore, statistical analysis could not be run.

b. Experiment: Plants

There were no statistically significant differences between the plant (total, root, or shoot) masses or plant carbon to nitrogen ratios across the various competition and/or fertilizer treatments. However, the general patterns observed in the data were biologically meaningful and worth examination. In general, plant weight was greatest in plant-only environments with nutrient supplements added (hereafter referred to as ‘plant-only +supplement’). Conversely, plant weight was least in combined (plant and algae) environments receiving supplements (hereafter referred to as ‘combined +supplement’). Plant-only and combined environments without supplements were roughly equal in total weight and measured weights between the two supplement-receiving plant sets. Figures 5, 6, and 7 show the average final weights of the roots, shoots, and total plants across the four plant growing treatments.

Carbon and nitrogen measurements for the plants were erratic. In plant roots, ‘plant-only +supplement’ displayed the greatest concentrations of both carbon and nitrogen with the other three treatments being roughly similar in both aspects. Plant shoot nitrogen was roughly
equivalent for all four treatments. Measured plant root nitrogen only ranged from 109.9 mg to 110.2 mg. Plant shoot carbon was similar to the roots in that the ‘plant-only + supplement’ had the greatest concentration. Plants in treatments not receiving nutrients had roughly similar carbon concentrations in their shoots, while ‘combined + supplement’ had a relatively low average concentration of carbon in its shoot. Plant nitrogen and plant carbon levels essentially mirrored the pattern explained for plant shoot carbon. Total plant C:N was greatest in the ‘plant-only’ treatment, however, all treatments were roughly similar with a plant C:N range between approximately 36.26 and 40.60. Figures 8a through 10b compare the carbon and nitrogen concentrations for the roots, shoots, and total plant across the four plant growing treatments. Figure 11 shows the C:N ratio of the total plants across the four plant growing treatments. It should be restated that none of the data mentioned in this section were found to be significant. All analysis of variance tests conducted on the above data returned a p-value greater than the alpha-value of 0.05.

c.  *Experiment: Algal Growth*

Algal growth was the only variable measured in this experiment that was found to be statistically significant. With an alpha-value of 0.05, the p-value returned by the analysis of variance test was 0.0347. Source data from SAS showed that the p-value for chlorophyll-a across growing conditions (algae, plant, or combined) was the most significant factor with a p-value of 0.0154. The p-value for chlorophyll-a across supplement treatments (supplement or no supplement) was 0.0729. Thus, the effect of plant presence was significant, while the presence of a nutrient supplement was not-significant.
Algal concentrations displayed a clear pattern when averaged together. In all growing conditions (algae, plant, combined), algal concentrations were greater when a nutrient supplement was added. Thus, concentrations in ‘algae-only + supplement’ were greater than in ‘algae-only’, and the same is true for plant-only and combined environments. In regards to growing conditions, algal concentrations were greatest in the combined environments with ‘combined + supplement’ treatments reaching 800 mg of chlorophyll-a per liter, and ‘combined’ treatments reaching 345.6 mg of chlorophyll-a per liter. Algae-only environments displayed the lowest concentrations of algae at 94.5 (with supplement) and 9.7 (without supplement) micrograms of chlorophyll-a per liter. The plant-only environments were the relative medium values at 415.7 (with supplement) and 232.3 (without supplement) micrograms of chlorophyll-a per liter. Figure 12 compares the concentrations of algae across all six treatments.

d. Summation of Treatments:

The following section is meant to serve as a summary of the final conditions for the six treatments. The ‘algae-only’ treatment had the lowest measured concentration of chlorophyll-a. The ‘algae-only + supplement’ treatment has the second lowest concentration of chlorophyll-a, but was still approximately 10 times greater than the concentration found in ‘algae-only’. No other data was collected for these treatments.

The ‘plant-only + supplement’ and ‘plant-only’ treatments displayed medium algae concentrations relative to the other two growing conditions with the ‘plant-only + supplement’ treatment having algae concentrations approximately 1.8 times greater than the ‘plant-only’ treatment. The ‘plant-only + supplement’ treatment produced the plant with the largest mass at
13.9 grams and the second lowest C:N ratio at 37.13. The ‘plant-only’ produced the plant with the second largest mass at 11.4 grams and the highest C:N ratio at 40.60.

The combined (plants and algae) environments produced the highest amount of algae with the ‘combined + supplement’ treatment reaching concentrations approximately 2.3 times greater than concentrations found in the ‘combined’ treatment. Plant mass in the combined environments was the inverse of the plant growth in the plant-only environments. The ‘combined + supplement’ produced the plant with the lowest mass at 9.5 grams and the second highest C:N at 38.62. The ‘combined’ treatment produced a plant that was a close third in mass at 11.3 grams (0.1 gram less than ‘plant-only’) and has the lowest C:N at 36.26.

V. Discussion

a. Analysis of Juncus effusus Growth Limitations

Data collected for plant growth in the various treatments leads us to reject the hypotheses that Juncus effusus growth in the Lake Fayetteville Spiral Wetland is being limited by a) insufficient nutrient (nitrogen and phosphorus) concentrations and/or b) competition caused by the presence of algae. However, the patterns in the data support the general idea that algae may outcompete emergent plants for nutrients, probably due to the rapid relative growth rate of algae and their ability to move with water currents. The lack of statistical significance could be a function of my limited replication in the two-way experiment.

The effect of nutrient concentration on Juncus effusus growth was isolated by comparing the mass of Juncus effusus produced in the ‘plant-only’ treatment with the mass produced in the ‘plant-only + supplement’ treatment. Similarly, the mass of Juncus effusus in the ‘combined’ treatment was compared to the mass in the ‘combined + supplement’ treatment. Doing this
allowed for nutrient concentrations to be the only variable influencing plant growth differences. It was hypothesized that *Juncus effusus* placed in an environment with a higher nutrient concentration would experience accelerated growth in comparison to the plant without a supplement. This hypothesis was based on data by Gusewella and Koerselman (2002). However, analysis of variance conducted on the data from the two treatments determined that there was no significant relationship between mass of *Juncus effusus* and nutrient concentration. Therefore, nutrient availability alone is not a limiting factor for plant growth in the Lake Fayetteville Spiral Wetland.

A similar process was applied to determine if competition caused by algae alone was a significantly limiting factor in *Juncus effusus* growth. To isolate the effects of algal concentrations on plant growth, the mass of *Juncus effusus* from the ‘plant-only’ treatment was compared to the mass from the ‘combined’ treatment. Likewise, the plant mass from the ‘plant-only + supplement’ treatment was compared to the plant mass acquired from the ‘combined + supplement’ treatment. This made variation in algal concentrations the only variable affecting plant growth differences. It was hypothesized that *Juncus effusus* placed in an environment with higher algal concentrations would experience more stress as a result of greater competition, which would be evidenced by differences in plant mass and nutrient contents. This hypothesis is in accordance with data from Engelhardt and Ritchie (2002). Yet, like nutrient concentrations, analysis of variance conducted on the treatments showed that there was no significant relationship between algal concentrations and mass of *Juncus effusus*. Thus, algal concentration alone is not a significantly limiting factor for plant growth in the Lake Fayetteville Spiral Wetland.
The last variable tested as a limiting factor for *Juncus effusus* growth was a possible synergistic effect forged by the combination of algal and nutrient concentrations. For this additive effect to be tested, the masses of *Juncus effusus* from the ‘plant-only’ treatment and the ‘combined + supplement’ treatment were compared. The idea behind this comparison was that the presence of excess nutrients could accelerate the growth of algae which are generally capable of absorbing nutrients at greater rates than macrophytes. This acceleration in growth would cause rapid eutrophication in such a small environment and exert the greatest amount of stress of any treatment. Despite the ‘combined-only’ treatment having the highest concentration of algae, lowest mass of *Juncus effusus*, and lowest whole-plant carbon to nitrogen ratio, the analysis of variance results were the same in that no significant relationship was apparent. Hence, no additive effect of nutrient and algal concentrations is present which effects the growth of *Juncus effusus* in the Lake Fayetteville Spiral Wetland.

*b. Analysis of Algal Growth*

While no relationship was established between the concentration of algae and *Juncus effusus* growth, analysis of variance tests conducted on algae samples taken from each treatment revealed a significant relationship between the presence of *Juncus effusus* and algal growth. Essentially, *Juncus effusus* growth was not affected by the presence of algae, but algal concentrations were greatest when *Juncus effusus* was present. Furthermore, given that the presence of a nutrient supplement could be construed as significant depending on the chosen alpha value, it is possible that the presence of *Juncus effusus* and a nutrient supplement in the ‘combined + supplement’ treatment produced a synergistic effect on the algae which caused the ‘combined + supplement’ treatment to have the highest concentration of algae.
There are essentially five factors which regulate the growth of algae: light availability, nutrient availability, space, predation, and competition (Stevenson, et. al, 1996). As long as an algal colony has access to light and nutrients, it will continue to grow until it experiences an environmental stress from space limitation, predation, or competition. The buckets in which the algae were growing were free of predation, received ample light, and provided plenty of space for the two months the algae were growing. Thus, the only factors that were influential were competition (with *Juncus effusus*) and nutrient availability (presence of supplement).

Considering algal concentrations were significantly greater in treatments involving *Juncus effusus*, and, algal concentrations had no significant impact on *Juncus effusus* growth, it can be inferred that there is no substantial, if any, competitive stress being inflicted upon the algae by the *Juncus effusus*.

All this, however, fails to explain why algal concentrations were greater when *Juncus effusus* was present. Assuming *Juncus effusus* failed to exert a significant competitive stress on algae, the concentrations in the ‘combined + supplement’ replicates should, at best, be roughly equal to the concentrations in the ‘algae-only + supplement’ replicates. For algal concentrations to be greatest when *Juncus effusus* is present, *Juncus effusus* would have to provide some benefit to the algae. One possible explanation is that in adding the *Juncus effusus* plants to the experimental treatments also transferred algae attached to these plants. In other words, the plants effectively seeded the experimental units with algae.

c. *Implications for the Lake Fayetteville Wetland*

Data gathered from Lake Fayetteville from May 2014 to August 2014 displays the general trends that are expected of a eutrophic lake in the summer. As temperatures steadily
increase throughout the summer, there is a drastic decrease in nitrate concentrations and an increase in DO. This is because, as temperature increases, available light energy also increases. The increase in available light energy allows an increase in photosynthetically-active algae. The photosynthetically-active algae are then able to consume large amounts of nitrate to grow and reproduce, which leads to a rapid reduction in nitrate concentrations (Lee, et. al, 2009).

Conversely, while the algae are consuming nitrate and light energy, the chemical reaction used during photosynthesis is producing DO (Crumpton, 1989). If monitoring had continued into late August and September, based on previous findings, it is likely that there would be a sudden decline in photosynthesis leading to a steady reduction in measured CHL-A (Smith, et. al, 1999). Nitrate concentrations would steadily increase and DO would suddenly decrease rapidly. This is because light intensity would reach levels that cause non-photochemical quenching in algae, which prevents photosynthesis and ultimately results in the mass death of algal communities (Muller, et. al, 2001). Without photosynthetically-active algae present, nitrate levels would build. And, in the presence of mass of amount of dead organic matter (the algae), aquatic detritivores would consume large quantities of DO in a chemical process used to break up the organic matter for a food source (Smith et. al, 1999). Around mid-March, the process would begin again. This cycle is natural; however, in the presence of excess nutrients, nitrate, DO, and CHL-A concentrations begin to fluctuate at dangerous levels.

The Lake Fayetteville Spiral Wetland was designed to raise awareness for eutrophication and how nutrient enrichment in aquatic systems can be disastrous. The designers also expressed a desire for the wetland to serve an ecological purpose and benefit the lake by reducing the rate of eutrophication by some degree. However, in the wetland’s first year, there was minimal plant growth and the Juncus effusus samples which were expected to grow to their full 4’, scarcely
reached 6”. The initial concern was that *Juncus effusus* was not sufficiently capable of competing with algae and cyanobacteria for nutrients needed for growth (N and P). However, after monitoring the wetland during its second summer and conducting this study, it is apparent that competition with aquatic vegetation was not a limiting factor in plant growth. Alternative limitations which could have hindered *Juncus effusus* growth are: growing season, plant anatomy, and physical disturbance/stress.

Construction began on the Spiral Wetland in the late spring of 2013; however, it is entirely likely that, when the wetland was finished and seeds fully planted, *Juncus effusus* had already missed part of its growing season and was not able to reach expected heights in the remaining time. While this theory explains the limited growth in the first growing season, it does not necessarily explain why plants grew in weight by approximately 570% during the second growing season.

*Juncus effusus* anatomy could also have an impact on why the plants grew poorly in the first growing season. According to a *Juncus effusus* fact sheet provided by the Natural Resource Conservation Service (NRCS, a branch of the USDA), *Juncus effusus* seeds need to over-winter near the surface of the soil in order to grow properly (2002). Furthermore, in an average growth cycle, initial shoots tend to emerge in late summer and reach 4’ the following spring (NRCS, 2002). Lastly, *Juncus effusus* grows best in environments with a pH between 4.0 and 6.0, salinity < 14ppt, and water no deeper than 6”. Therefore, it is entirely possible that the “limited” growth observed during the first growing season was simply the emergence of new shoots. However, these shoots did not reach 4’ by the following spring. In fact, by the end of the second growing season, most of the *Juncus effusus* still appeared to be under 2’. This may be due to poor growing conditions based on *Juncus effusus* physiology. While no pH data were gathered for the planters
in which the *Juncus effusus* were grown, pH data were provided for the rest of the lake. The surface water for Lake Fayetteville was consistently greater than 6 (ranged 7.85 to 10.7 from April to August, 2014). Similarly, *Juncus effusus* grows naturally in moist soils 6” or less below water (NRCS, 2002). The *Juncus effusus* growing in the Spiral Wetland were growing in moist, shallow planters sitting above the lake. This means, rather than the plants being rooted in the soil and growing up through the water as they do naturally, they were grown in soil and their roots extended down, well past the planters, into the open water. Both pH and the design of the planters in the wetland could have had significant impacts which limited the growth of *Juncus effusus*.

The last credible option for limiting *Juncus effusus* growth is the constant physical disturbance suffered by the *Juncus effusus* during the course of the Spiral Wetland. While situated in Lake Fayetteville, the spiral wetland it suffered ice, wind, and even hail damage. Due to lack of a skeletal structure, high-winds easily flipped the wetland over on itself leaving dozens of plants at a time completely submerged underwater with their roots in the air. The wetland displayed damage caused by large birds roosting on the soft, foam body and even some tears and litter left by boaters. Constant physical disturbance at that level could also explain the stunted growth exhibited by the *Juncus effusus*. Due to research rejecting the hypothesis that nutrient competition was a primary factor in limiting plant growth, it is likely that one (or a combination) of the aforementioned factors are responsible for limiting *Juncus effusus* growth.

d. Improvements for Future Studies

Due to space and resource limitations, there were a few design characteristics that need to be revised in future research. First, there were no steps taken to measure or catalog the starting
weights of the plants in the experimental portion of the project. Thus, design error could exist in that an abnormally light or heavy plant could greatly offset the final average weight. It is recommended that future studies should diminish this potential error by either a) taking a rough measurement of plant weight before setting up the experiment, b) using seeds instead of partially grown plants to remove previous growth bias, or c) make enough replicates to isolate the effects of an outlier.

Another factor that could greatly influence the outcome of plant growth is the time of year the experiment is conducted. Based on measured plant growth from the artificial wetland, it is likely that the experiment in question was conducted towards the end of the growing season. This timeframe could limit growth in plants as the temperature increases past the optimal range for plant growth. Therefore, it is suggested that future studies be conducted towards the beginning of selected plant’s growing season to ensure continuous growth throughout the experimental period.

Lastly, there were certain issues involving the algae measurements. As the title suggests, the ‘plant-only’ treatments (with and without supplements) were not meant to register any algal concentrations when measured. No algae supplements were added to these treatments. Therefore, it is likely that preliminary cleaning of the plant roots failed to remove algae that were clinging to the roots from the lake. It could then be assumed that the same is true for the plants in the ‘combined’ treatment. Remnant algae from the lake could skew the algae concentration measurements. This is another example of why using seeds instead of partially grown plants could be beneficial.

Due to remnant algae from the lake, there were likely two separate species of algae present in the experiment. The algae used to inoculate the replicates were cultivated by scraping
rocks near the edge of the lake. It is probable that the algae used to inoculate the replicates and
the algae suspended in the lake were different species. It is unclear what effects this had on the
measured algal concentrations, nonetheless, it would be ideal to avoid multiple algae types in the
future studies, unless it were a key study point.

e. Suggested Modifications to Artificial Wetland Design

The Spiral Wetland was removed from Lake Fayetteville on October 20th, 2014 after only
a year and a half of residency. Interest has been expressed in a more permanent artificial wetland
design with aesthetic value, less maintenance requirements, and a more measurable effect in the
environment. With these criteria in mind, and considering the data obtained in this study, I have
developed a wetland design that should provide aesthetic value, require only minimal
maintenance, and provides easily measured data.

Based of algal concentration data in this experiment, the most important design
adjustment for any future artificial wetland is for it to be a solid figure. Light penetration allowed
by the spiral shape likely had the same effect on algal growth as the combined treatments, in that
enough light was blocked to prevent non-photochemical quenching, but not enough to prevent
algal photosynthesis. Therefore, I propose that any future wetland that is to be placed over Lake
Fayetteville be a solid shape. This will greatly increase the opacity of the wetland and limit the
photosynthetic ability of the algae beneath. The design I am proposing is an octagon.

One of the primary nuisances resulting from the Spiral Wetland was the constant need for
repairs and upkeep. The spiral wetland was constructed using foam mats similar to those found
in many gyms and kindergarten classrooms. These mats are very buoyant which made them ideal
for the body of the wetland. However, due to the wetlands spiral shape and lack of support
material, sections were easily flipped and/or damaged due to human and environmental factors. Thus, the outer edge of the octagonal wetland should be constructed using interconnected, sealed pvc pipe. Pvc pipe is buoyant, light, and sturdy. By using pvc pipe to outline the octagonal wetland, the wetland will gain protections from factors such as high winds and boaters. Maneuverability was another problem for the spiral wetland. But, by installing lag eye screws in the pcv pipe at every 90°, an octagonal wetland could easily be moved for maintenance. Another benefit of using pvc pipe as an exoskeletal structure is that, in the event of multiple, “free-floating” wetlands, the pvc pipe can be painted to identify the different wetlands.

Due to its effectiveness as a body material, the foam mats should remain the load bearing part of the wetland. These mats can easily be connected to the pvc pipe using zip-ties, chains, rope, etc. Figure 13 shows a rough draft of what an octagonal wetland could look like. The design presented in Figure 13 holds 100 planters. In the center of the octagonal wetland is a protective case for monitoring equipment used to continuously gather data. A rough estimate puts the wetland at 6 feet in diameter with a total area of approximately 30 square feet. This design covers significantly less area than the spiral wetland, but the smaller, octagonal design allows for multiple wetlands to be placed on the lake and used as monitoring sites.

In order to improve monitoring data quality, the octagonal design includes a space for long term water monitoring equipment. By using a system such as the Horiba W-20XD Series Water Quality Monitoring System in conjunction with a chlorophyll meter, a month’s worth of data could be acquired and stored in the wetland. To collect data, researchers would only need to visit the wetland(s) at the end of each month, upload the data, and then reset the monitoring systems for the next month. The octagonal wetland(s) could be allowed to float freely and collect lake and GPS data at different points of the lake, thus providing a look at nutrient, chlorophyll,
salinity, temperature, and D.O. levels across the lake. Alternatively, the octagonal wetland(s) could be held at certain points on the lake by attaching anchors to the lag eye screws. This would allow for continuous monitoring of a single point on the lake and would require minimal maintenance outside of the monthly data collection. In general, the octagonal wetland design serves as an aesthetically pleasing and ecologically beneficial monitoring system that could be used in a variety of studies or for routine monitoring.

f. Conclusion

Results from this study revealed that *Juncus effusus* in the Lake Fayetteville Spiral Wetland was not limited in growth by nutrient availability or competition with algae. The masses of plants grown in treatments containing algal inoculations and/or nutrient supplements were not significantly different from plant masses produced in the control treatments (plant-only). Similarly, there was no significant variation in the C:N ratios or the nutrient weights across the various treatments. Ample research has been conducted which shows the effects of wetland ecosystems on algae (Hermond and Benoit, 1988) and vice versa (Crumpton, 1989). There are also numerous studies regarding competition among wetland plant species (Englehardt and Ritchie, 2002). Little research exists to evaluate the direct competition between wetland plants and algae. Although the results of this study were not statistically significant and could therefore not fully explain the patterns observed, this could have been the result of poor replication. A more comprehensive study with greater replication could show that the biological patterns observed in this study were meaningful, which could inform the future construction and maintenance of floating wetlands for aesthetic and water quality improvement purposes.
VI. References


VII. Appendix

Figure 1: An image of the spiral wetland, and information explaining the background of the wetland. Both have been provided by Stacy Levy.

Figure 2: Visual representation of experimental design

Figure 3: *Juncus effusus* root, shoot, and total weight over time

Figure 4: Lake Fayetteville surface water nitrate and temperature over time

Figure 5: Average final weights of experimental plant roots across various treatments

Figure 6: Average final weights of experimental plant shoots across various treatments

Figure 7: Average final, total weights of experimental plants across various treatments

Figure 8:

A. Average carbon concentrations for experimental plant roots across various treatments
B. Average nitrogen concentrations for experimental plant roots across various treatments

Figure 9:

A. Average carbon concentrations for experimental plant shoots across various treatments
B. Average nitrogen concentrations for experimental plant shoots across various treatments

Figure 10:

A. Average carbon concentrations for whole experimental plants across various treatments
B. Average nitrogen concentrations for whole experimental plants across various treatments

Figure 11: Average carbon-nitrogen ratio of whole plants across various treatments

Figure 12: Average, final algal concentrations across various treatments

Figure 13: Basic diagram of octagonal wetland design
Stacy Levy  
Born 1960

Spiral Wetland, May 2013  
Closed-cell foam floating wetland mats, soft rush (juncus effuses), and rope

Spiral Wetland is an outdoor eco-art project inspired by Spiral Jetty (1970), Robert Smithson’s famous earthwork sited in the Great Salt Lake, Utah. Celebrating the beauty of the spiral in nature, Stacy Levy uses both art and science as a vehicle for translating the process of patterns in the natural world into the language of human understanding.

Stacy Levy designed Spiral Wetland with an ecological goal: to improve the water quality of Lake Fayetteville. Spiral Wetland uses the native plant, soft rush, growing in a floating sculpture. The plant helps remove excess nutrients like nitrogen and phosphorus from the lake water, and adds shade for fish habitat. When the installation is taken down (estimated date Summer 2014), sections of the wetland will be adopted and transplanted into other wetlands and retention basins in the region, so their benefits can continue in new waters.

Levy graduated from Yale University with a B.A. in Sculpture and a minor in Forestry. She earned an M.F.A. from Tyler School of Art, Temple University, Philadelphia. Her numerous awards include the Pew Fellowship in the Arts and an Excellence in Estuary Award from the Partnership for the Delaware River Estuary.

Figure 1
Figure 2

- **C** = Combined
- **P** = Plant only
- **A** = Algae only
- **S** = Nutrient Supplement Added

= 1 Bucket
Figure 3
Figure 5

- p = 0.4780
- f = 0.91
- α = 0.05

Comparison of root weight (g) between Plant and Combined Competition Treatment groups with and without supplements.
Figure 6

![Graph showing shoot weight comparison between treatments.](image)

- p = 0.1295
- f = 2.54
- a = 0.05

Legend:
- Black: No Supplement
- Gray: With Supplement

Comparison Treatments:
- Plant
- Combined
Figure 7

- **Total Plant Weight (g)**

- **Competition Treatment**
  - **Plant**
  - **Combined**

- **Legend**:
  - Black: No Supplement
  - Grey: With Supplement

- **Statistical Information**:
  - p = 0.1273
  - f = 2.75
  - a = 0.05
Figure 8

- **Part A**: Comparison of root carbon weight under different competition treatments.
  - No Supplement: Black bars
  - With Supplement: Gray bars
  - Statistical details:
    - \( p = 0.4905 \)
    - \( f = 0.88 \)
    - \( a = 0.05 \)

- **Part B**: Comparison of root nitrogen weight under different competition treatments.
  - No Supplement: Black bars
  - With Supplement: Gray bars
  - Statistical details:
    - \( p = 0.6146 \)
    - \( f = 0.63 \)
    - \( a = 0.05 \)
Figure 9
Figure 10

Graph A: Comparison of Total Carbon Weight (g) under different competition treatments.
- Plant: No Supplement, With Supplement
- Combined: No Supplement, With Supplement

Graph B: Comparison of Total Nitrogen Weight (g) under different competition treatments.
- Plant: No Supplement, With Supplement
- Combined: No Supplement, With Supplement

P-values:
- Graph A: p = 0.1091
- Graph B: p = 0.3782
Figure 11
Figure 12
Figure 13

Hard, Skeletal Structure

Planter

Protective Monitor Case

Lag-Eye Connector