THE EFFECTS OF WATER VELOCITY AND SEDIMENT COMPOSITION ON COMPETITIVE INTERACTIONS BETWEEN NATIVE AND INVASIVE MACROPHYTE SPECIES IN A SPRING FED RIVER

by

Jacob N. Bilbo, B.A.

A thesis submitted to the Graduate Council of
Texas State University in partial fulfillment
of the requirements for the degree of
Master of Science
with a major in Aquatic Resources
August 2015

Committee Members:

Thomas B. Hardy, Chair

Weston H. Nowlin

Paula S. Williamson

FAIR USE AND AUTHOR’S PERMISSION STATEMENT

Fair Use

Duplication Permission

ACKNOWLEDGEMENTS

I would first like to thank my major advisor Dr. Thomas Hardy for giving me the opportunity to work at the Meadows Center for Water and the Environment (MCWE). The time that I have spent conducting habitat restoration on the San Marcos River led me to my interest in invasive species management. I am truly grateful for the experiences and knowledge that I gained as a result of working at the MCWE. I would also like to thank Dr. Weston Nowlin for allowing me to use his laboratory, and Dr. Paula Williamson for her immense help and involvement in this project.

The staff and student workers at the MCWE are a group of highly intelligent people who I have thoroughly enjoyed working with. Special thanks to Kristy Kollaus and Gabrielle Timmons for being so helpful in the development of this project, and for the feedback and insight that they provided me. Large thanks goes out to Tom Heard, Daphne Mitlo, Kristyn Cunningham, John Fletcher, Jessica Frye, Jeremiah Leech, and Colby Maron for all of their assistance in constructing enclosures, setting up and tearing down the field site, and the months of measuring plants in the laboratory. Without their help this project would not have been possible. Thank you all for the fun times, hard work, and unforgettable experiences that we have shared together. Thanks to my friends and family who have supported me during my academic career, both emotionally and financially. Finally, thanks go out to my father Steven Bilbo, and my mother Martha Bilbo. Thank you for all the years of supporting me while I try and achieve my goals, and for helping me build my appreciation for nature and science at a young age.

TABLE OF CONTENTS

Page

CHAPTER

LIST OF TABLES

LIST OF FIGURES

ABSTRACT

Hydrilla (*Hydrilla verticillata*) is an invasive species that is problematic globally and also in the San Marcos River where it competes with native species. Hydrilla has been described as the "perfect aquatic weed" because it is able to propagate under a wide range of environmental conditions including low nutrient and variable light conditions (Langeland 1996). Treatment methods for control of non-native aquatic plants can be restricted due to the co-occurrence of native endangered species, requiring an integrated approach of several methods for restoration, including removal by hand, and manipulating environmental factors to encourage growth of native species. I conducted a competition study to determine if a native species can out-compete non-native species under a set of environmental conditions. The experiment was conducted within Spring Lake at the headwaters of the San Marcos River, Hays Co, Texas between 03/28/2014 and 05/21/2014. I used a three-factor replacement design; (water velocity, substrate type, and competitive pressure) to assess competitive interaction between a native species (Illinois pondweed) and non-native species (hydrilla). Illinois pondweed (*Potamogeton illinoensis*) and hydrilla (*Hydrilla verticillata*) were potted in monoculture (intraspecific competition) and mixtures (interspecific competition) using sand or silt sediment, and high or low velocity for a period of seven weeks. Above- and belowground dry biomass, total stem length, and number of stems were measured. Across all treatments, pondweed demonstrated significantly ($P<0.05$) higher growth rates than hydrilla. Substrate type and monocultures were not statistically significant factors in plant growth. However, growth indices indicated that total dry biomass of both plants was slightly higher in sand substrate and high velocity. I also found intraspecific competition was greater than interspecific competition for both species. Both species produced more biomass when in monoculture and less biomass in mixtures. Therefore, data from this study comparatively better environmental conditions for Illinois pondweed to successfully out-compete hydrilla are in sand substrate and high velocity. These strategies could be used to enhance EAHCP efforts in the San Marcos River where invasive plant management options are limited. Part of the EAHCP plan includes planting native species after removal of non-natives. Illinois pondweed may not be a suitable plant for remedial gardening, not because it is an inferior competitor with hydrilla, but because it may not be competing with it at all. In fact, Illinois pondweed may be aiding hydrilla propagation by slowing water velocity, thus accumulating fine sediments and creating suitable habitat for hydrilla.

CHAPTER 1

THE EFFECTS OF WATER VELOCITY AND SEDIMENT COMPOSITION ON COMPETITIVE INTERACTIONS BETWEEN NATIVE AND INVASIVE MACROPHYTE SPECIES IN A SPRING FED RIVER

Introduction

The aquatic macrophyte hydrilla has become a worldwide problem and exists on every continent except Antarctica (Barnes et al. 2014). Since hydrilla is so widely spread around the world, its original distribution is uncertain (Bowles & Bowles 2001) but, it is thought that hydrilla is native to Asia, Africa, and Australia (Sousa 2011). Hydrilla was introduced into the United States in 1950 or 1951 by an aquarium plant dealer who dumped the species into a canal in Tampa, Florida (Van Driesche et al. 2002). Considered a nuisance invasive macrophyte, infestations of hydrilla have led to marina closures, revenue losses due to decreased recreation by inhibiting boating, swimming, fishing, and blocking of canal and waterways (Van Driesche et al. 2002). Incidental mortality of fish and other species can occur when dense mats of hydrilla are removed. Hydrilla can displace native macrophyte species in lotic environments by forming a dense canopy at the water's surface, blocking light penetration to submerged vegetation ( ). Hydrilla has shown to provide habitat for fish and invertebrates, but once it becomes too dense, it can affect foraging rates for fish, increase fish egg mortality (Haller et al. 1980), and can result in hypoxia leading to fish kills (Colle & Shireman 1980).

Hydrilla was first recorded in the San Marcos River Texas, USA in 1975, but the exact date of its introduction is unknown (Bowles & Bowles 2001). The San Marcos

River is a spring fed river originating in Hays County, Texas from groundwater discharge supplied by the Edwards Aquifer and flows approximately 177 kilometers before it converges with the Guadalupe River near Gonzales, Gonzales County, Texas. The San Marcos River is a diverse ecosystem that supports several threatened and endangered species (Owens et al. 2001), and provides diverse habitats that supports at least 31 species of aquatic plants, 23 of which are native (Saunders et al. 2001).

To date a cost effective and long-term solution for control of hydrilla in rivers such as the San Marcos River that contain listed species has not been found. Methods for removing hydrilla include mechanical, chemical, and biological control. Control of hydrilla requires continually monitoring and removing new growth because hydrilla has the ability to reproduce by fragmentation, auxillary turions, and subterranean tubers that can remain dormant for periods of four years or more (Langeland 1996).Mechanical methods include water level manipulation and removal of the entire plant including the root system by use of a harvester or shovels (Chilton 2011, Owens et al. 2001). Mechanical removal is only effective when care is taken to prevent hydrilla fragments and propagules from becoming established nearby or downstream (Chilton 2001, Owens et al. 2001). Chemical control includes the use of herbicides such as copper, diquat, or endorthall products (Chilton 2011). The San Marcos River supports several endangered species, therefore The Edwards Aquifer Authority Habitat Conservation Plan (EAHCP) was developed. The EAHCP is a fifteen-year plan with long-term goals to enhance or expand habitat (EAHCP 2013). Several species of nonnative vegetation were identified in the EAHCP for targeted removal, one of which is hydrilla. In the San Marcos River, hydrilla competes with native species including the federally endangered Texas wild rice

(*Zizania texana*) (EAHCP 2013), which is endemic to the San Marcos River. The use of mechanical methods and herbicides to remove hydrilla is not feasible in in the San Marcos River because they could harm sensitive ecosystems and native and endangered species endemic to the San Marcos River such as Texas wild rice. An effective documented biological method of controlling hydrilla is with the introduction of triploid grass carp (*Ctenopharyngodon idella*), which consume hydrilla (Cuda & Sutton 1999). However, grass carp cannot be introduced into the San Marcos River because they could consume Texas wild rice and other aquatic vegetation. Because these traditional methods are not viable options to control hydrilla in the San Marcos River, alternative methods currently being employed in the San Marcos River are removal by hand, planting of native vegetation, and remedial gardening (EAHCP 2013).

Another potential method for control of hydrilla is planting a native aquatic species that can out-compete hydrilla. Van et al. (1998) showed that native American eelgrass (*Vallisneria americana*) could be effective for control of hydrilla under low soil fertility conditions. Sago pondweed (*Potamogeton pectinatus*) can also successfully out-compete invasive shortspike watermilfoil (*Myriophyllum exalbescens*) because of its ability to grow throughout the water column and create a canopy at the surface, blocking light to shortspike watermilfoil (Moen & Cohen 1989). In the San Marcos River, one potential candidate plant for restoration is the native Illinois pondweed (*Potamogeton illinoensis*) (Owens et al. 2001). Cursory field observations suggest that Illinois pondweed can effectively colonize an area and out-compete non-native species such as hydrilla after a period of two weeks (Jacob Bilbo, *personal observation*). This may be caused by modification of environmental conditions that would create ideal conditions for the growth of Illinois pondweed.

If native species can outcompete invasive species and lead to exclusion of nonnatives, than it is important to understand the environmental conditions in which hydrilla and Illinois pondweed grow. Varieties of environmental factors affect macrophyte growth and lead to different competitive outcomes (Barko & Smart 1986). Among environmental factors, water velocities and sediment types are among the most critical factors that influence submerged macrophyte communities (Madsen et al. 2001). Generally, macrophyte biomass density declines in current velocity (Chambers et al. 1991) because increased water velocities cause macrophyte damage by fragmentation (Madsen et al. 2001). After removal of non-native vegetation in a flowing river, the resulting increase in water velocity could make it difficult for hydrilla to recolonize that area. The optimal water velocity for hydrilla growth is from 0.15 m/s to 0.70 m/s, above which biomass sharply decreases (Hardy et al. 2010, Saunders et al. 2001). In addition, water velocities greater than 1 m/s begin to limit growth of pondweeds (Howard-Williams & Liptrot 1980). This difference in the velocities suggests that velocities from 0.70 m/s and 1 m/s would lead to a decrease in hydrilla, and thereby potentially allow Illinois pondweed to exhibit greater growth and ability to outcompete hydrilla. Although Illinois pondweed also fragments, its elongated shape and thin leaves may make it more resistant to fragmentation at higher velocity (Chambers et al. 1991).

Sediment type also differentially affects the growth of different macrophyte species (Barko & Smart 1986). Coarse and porous sediments such as sand can typically contain less organic matter and thus less nutrient availability (Barko & Smart 1986).

Coarse sediments make plants more susceptible to uprooting due to stream stress (Barko & Smart 1986). Hydrilla growing in sand may have lower biomass while hydrilla growing in finer less porous soils may produce more biomass (Spencer et al. 1992). Sago pondweed was shown to have a positive correlation with the coarseness of sediment and plant growth (Schmid 1965), suggesting that Illinois pondweed may also have higher growth rates and produce more biomass when grown in sand.

Spatial analysis of vegetation distribution within the San Marcos River has shown that Illinois pondweed and hydrilla occur in both sand and silt, and that the majority of non-native plants growing in the San Marcos River prefer silt (Hardy et al. 2010). If growth of Illinois pondweed is in fact associated with habitats consisting of sand, this could mean that it may be able to suppress hydrilla growing in sandy areas. High water velocity and sandy substrate may be the optimal conditions in which Illinois pondweed would be able to out-compete and suppress hydrilla propagation. In this study, I experimentally evaluated the outcomes of competition between native Illinois pondweed and non-native hydrilla across a gradient of a range of intensities of intraspecific and interspecific competition at two different flow velocities and two different sediment types. I predicted that variation in water velocity and sediment characteristics would alter/affect the outcome of competition between the native macrophyte species (Illinois pondweed) and non-native macrophyte species (hydrilla). I also predicted that Illinois pondweed would exhibit greater growth than hydrilla in high water velocities, sand substrate, and when competitive pressures are equal with or greater than hydrilla. Results of this competition study may determine whether certain environmental conditions allow Illinois pondweed to out-compete hydrilla. And if so may demonstrate that Illinois pondweed is a good candidate to us in restoration to successfully manage areas inundated by hydrilla. Thus identifying these conditions where they naturally occur or altering substrate and velocity to create these conditions could encourage growth of Illinois pondweed and keep hydrilla propagation at manageable levels.

Methods

The experiment was conducted in Spring Lake at the headwaters of the San Marcos River, Hays Co, Texas from 03/28/2014 to 05/21/2014. Healthy plants free of obvious signs of disease, extensive damage from herbivory, and chlorosis were collected from the San Marcos River. Illinois pondweed was collected just below Spring Lake Dam and hydrilla was collected downstream of City Park. Apical tips from each species were cut to 20 cm in length and planted 10 cm into the experimental substrates. Hydrilla tips were stripped of all branches leaving only the main stem with all leaves attached. Hydrilla leaves are small and numerous, thus I did not count all the leaves on each plant, but instead elected to count the number of leaves per whorl, which was consistent among plants at eight leaves per whorl. Illinois pondweed tips were also stripped of all branches but was left with four leaves on each stem. To determine initial total dry biomass for each species eight 20cm apical tips of both hydrilla and Illinois pondweed were dried and weighed. These were then averaged to give a mean initial dry biomass.

Sediment treatments consisted of two sediment types commonly found in the San Marcos River in areas where hydrilla and Illinois pondweed both occur (Hardy et al. 2010). The first sediment type contained a relatively high percentage of sand and a lower percentage of silt and clay (sand 84%, silt 3%, clay 13%; classified as loamy sand by the Soil, Water, and Forage Testing Laboratory at Texas A&M Agrilife Extension Office).

The second type was classified as having greater percentage of silt and clay (sand 52%, silt 27%, clay 21%; classified as sandy clay loam). Hereafter, I will refer to the two sediment types will be referred to as “sand” and “silt” respectively.

Two levels of water velocity were tested. Pots of plants were placed at one site in Spring Lake that had high velocity (0.20 m/s) and another site that had low water velocity (0.00 m/s). The high velocity treatment is moderate in terms of the velocities found in the San Marcos River, but will be referred to as “high velocity”. The high velocity used in this study was chosen because it was the highest velocity found in Spring Lake during site surveys.

The three factor design was arranged as 5x2x2 with five types of competitive pressures, two velocities (high and low), and two substrate types (sand and silt). To test the effects of intra- and interspecific competition between Illinois pondweed and hydrilla, I used a “replacement design” (Kelty & Cameron 1995), which involves growing two species in different proportions (Vanclay 2006). In this experimental design the varying competitive pressures were species exposed to only intraspecific competition, species exposed to equal levels of interspecific competition, and species exposed to moderate levels of interspecific competition. The intraspecific competition treatment consisted of monocultures of two stems of each species (hydrilla:pondweed in a 2:0 or 0:2 ratio). Equal interspecific competition consisted of planting two apical tips of hydrilla with two apical tips of Illinois pondweed (2:2 ratio). Moderate interspecific competition between the two species consisted of one apical tip of hydrilla planted with three apical tips of Illinois pondweed and the reverse situation of one apical tip of Illinois pondweed planted with three apical tips of hydrilla (1:2 or 2:1). Each treatment combination of competitive pressure was duplicated and subjected to moderate (0.20 m/s) and low (0.00 m/s) velocity conditions with either sand or silt sediment, and was replicated 12 times for a total of 240 pots.

Two 11m^2 experimental plots were established within Spring Lake (Figure 1). Both experimental plots had a uniform depth across the plot of 0.75 meters. Two plots were constructed consisting of four 15cm x 1.5m x 1.8m treated wood frames in which the pots were placed. One was placed at the low velocity site and the other plot was placed at the high velocity site. Low velocity and high velocity experimental plots were enclosed in chicken wire (2 cm aperture) to exclude materials from drifting in and out of the plots. The low velocity plot was 0.5 meters below the surface resting on the lake bottom. The high velocity site was elevated on metal t-posts so that the plant beds were at a depth of 0.5 meters. Each enclosure was monitored weekly for damage to plants or presence of foreign plants. Foreign plants found growing in the pots were manually removed. Water velocity was monitored and recorded weekly using a flow meter 0.75 meters below the surface at the leading edge of each holding structure. The experiment ran for twenty three days: Plants were placed into frames on 03/28/2014 and were harvested on 05/21/2014.

Growth Data Collection

Following harvest, aboveground and belowground plant parts were separated in the lab and stored at -80°C. The numbers of stems were counted and total lengths of branches were measured to the nearest centimeter. Plants were then subsequently washed over a 0.2 mm sieve to remove sediment and debris. Plants were dried at 60°C for 24 h and the aboveground and belowground dry weight in grams was measured.

Linear Mixed-Effects Models

Growth data for each species were analyzed using linear mixed effect models (Table 1) and then incorporated into an Akaike Information Criterion (AIC) model selection table created for the response variables total length, aboveground dry biomass, belowground dry biomass, and number of stems (Table 2). The purpose for model selection is to estimate model parameters that are of particular interest or to identify a model that can be used for predicting a response variable. When there was clear support for one model, maximum likelihood parameter estimates or predictions of the response variable from that model were used. Competing model parameters were averaged if they were within 2 AIC units of one another (Posada & Buckley 2004). One model may have parameters which are not included in another competing model. In this case, only the parameters included in both models are averaged together, but the additional parameter is included in the final averaged model (Burnham & Anderson 2002, Posada & Buckley 2004). In the example below the parameter $\beta_3\chi_3$ is not included in model two; therefore it is not averaged but is included in the final averaged model.

$$\text{Model 1: } Y=\beta_o + \beta_1\chi_i + \beta_2\chi_2 + \beta_3\chi_3 + \epsilon$$

$$\text{Model 2: } Y=\beta_o + \beta_1\chi_i + \beta_2\chi_2 + \epsilon$$

$$\text{Averaged Model: } Y=\beta_o + \beta_1\chi_i + \beta_2\chi_2 + \beta_3\chi_3 + \epsilon$$

Data violated the assumption of homoscedasticity and were $\log_{10}$-transformed before analysis. All analysis and modeling were conducted using the statistical program RStudio (Version 0.98.1049). The LmerTest package was used in “R” to calculate p-values of the linear model.

Growth Indices

In order to analyze the yield of mixtures compared to the yield in monocultures I calculated indices commonly used in replacement design experiments using formulas from Weigelt and Jolliffe (2003) to quantify the outcome of competition. The indices used were (1) yield, (2) relative yield, and (3) change in contribution (Williams & McCarthy 2001). Yield is defined as the mean of the entire replicate. For this analysis I measured total biomass (above- and belowground) in grams of all pots in a given treatment. Yield was calculated as the mean of all plants in monoculture treatments as Y_H or Y_P (Y_X, where x is the species of interest).

As plants are often grown at different densities the superscript after the Y is used to denote the density at which all plants are grown. In this case density was held constant for each replicate. Y_H^D = yield of hydrilla at an overall density of D, and Y_P^D = yield of Illinois pondweed at an overall density of D. Y_{HP} or Y_{PH} is the total yield of one species in the presence of another in mixture. This is not a combined value of both species but, is instead separate for both species. The mean of each treatment was used to calculate total yield in mixture.Y_{HP} = yield of hydrilla in the presence of Illinois pondweed, and Y_{PH} = yield of Illinois pondweed in the presence of hydrilla.

Proportions were varied in each replicate and were used to calculate both yield and change in contribution. The proportion at which each plant was grown is represented by p_H and p_p with values always equal to 1. For example: mixtures grown at a ratio of 3:1 would have a proportion of 75% H and 25% P, so that p_H= 0.75 and p_p = 0.25. Mixtures of equal competitive pressure (2:2 ratio) had p_H = 0.5 and p_p = 0.5. Mixtures at moderate competitive pressures` had p_H = 0.75 and p_p = 0.25 (3:1 ratio), or p_H = 0.25 and p_p = 0.75 (1:3 ratio).

p_H = proportion at which hydrilla was sown

p_p = proportion at which Illinois pondweed was sown

Relative yield (RY) measures the yield in a mixture divided by the yield in a monoculture taking into account proportions at which the species are grown (Williams & McCarthy 2001). RY is expressed as a proportion that species performed in a mixture compared to a monoculture where $RY^{D}_{H}=Y^{D}_{HP}/(p_{H}Y^{D}_{H})$ equals relative yield of hydrilla, and $RY^{D}_{P}=Y^{D}_{PH}/(p_{P}Y^{D}_{P})$ equals relative yield of Illinois pondweed. When the RY value equals 1.0 both species grew equally well with each other in mixture as they did in monoculture, implying equal effects of intra- and interspecific competition. In contrast, RY values greater than 1.0 indicate a species did better in mixture than it did in monoculture (i.e. effects of intraspecific competition was greater than interspecific competition). Values less than 1.0 indicate that a species did better in monoculture than it did in mixture, (i.e. effects of interspecific competition was greater than intraspecific competition).

Change in contribution (CC) is a percentage calculated separately for each species which gives the proportional change of biomass in a mixture compared to a monoculture (Williams and McCarthy 2001). A CC value of zero means that there was no change in proportion in a mixture compared to what was expected in a monoculture. A value greater than zero indicates a proportional increase in biomass, while a value less than zero indicates a loss in biomass. For example: if CC_H= -0.18 and CC_P=0.05 this means that species H had an 18% decrease in biomass while species P had a 5% increase in biomass. Values were calculated by dividing the proportion of biomass a species attained in a mixture by the expected proportion from a monoculture (Williams & McCarthy 2001).

$CC^{D}_{H}=(Y^{D}_{HP}/(Y^{D}_{HP} + Y^{D}_{PH}))/((p_{H}Y^{D}_{H})/(p_{H}Y^{D}_{H} + p_{P}Y^{D}_{P}) -1$ = change in contribution of hydrilla

$CC^{D}_{P}=(Y^{D}_{PH}/(Y^{D}_{HP} + Y^{D}_{PH}))/((p_{P}Y^{D}_{P})/(p_{H}Y^{D}_{H} + p_{P}Y^{D}_{P}) -1$ = change in contribution of Illinois pondweed

Results

In linear mixed-effects model analysis, AIC model parameters for total stem length, number of stems, and aboveground dry mass (Table 3) were averaged because they were not significantly different (i.e., within two AIC units). Total length and aboveground dry mass were not significant in low velocity. Number of stems and belowground dry biomass at low velocity were significant. Illinois pondweed had significantly greater total length, aboveground dry mass, number of stems, and belowground dry mass than hydrilla in both monoculture and mixtures. Total length, aboveground dry mass, and number of stems were significant in monocultures of both species. Mixtures of equal or moderate interspecific competitive pressure were not significant response variables in the AIC models. Total length ($P=0.043$) and number of stems ($P=0.043$) were significant in both species in both low velocity and silt treatments.

The various growth indices exhibited some variation (Table 4). The yield of total biomass of Illinois pondweed (Y_{PH}) was greater than hydrilla (Y_{HP}) at every level of competitive pressure, velocity, and sediment type. Both Illinois pondweed and hydrilla performed better in mixtures than in monoculture (RY values >1.0). At equal competitive pressures (2:2 ratios) Illinois pondweed (RY_{P}) had higher RY values than hydrilla (RY_{H}) in both velocities and both substrate types. However, when Illinois pondweed was planted at a higher competitive pressure than hydrilla (Illinois pondweed >hydrilla, or 3:1), hydrilla had greater RY values than Illinois pondweed. This result was the same in both velocities and substrate types. When hydrilla had greater competitive pressure than Illinois pondweed (Illinois pondweed< hydrilla, or 1:3), Illinois pondweed had greater RY values than hydrilla in both velocities and both substrate types. At equal competitive pressures (2:2 ratio), hydrilla (CC_H) had proportional losses in total biomass (CC values less than zero) in both velocities and substrate types, while Illinois Pondweed (CC_P) had proportional increases in both velocities and substrate types. When Illinois pondweed was planted at a higher competitive pressure than hydrilla (Illinois pondweed >hydrilla, or 3:1), hydrilla had greater proportional increases in total biomass than Illinois pondweed. This was true in both velocities and substrate types. When Illinois pondweed was planted at lower competitive pressures (Illinois pondweed< hydrilla, or 1:3), Illinois pondweed had greater proportional increases in biomass than hydrilla in both velocities and substrate types.

The yield of hydrilla in low velocity (Y_{HP}) was slightly higher than the yield in high velocity. The opposite was observed for Illinois pondweed (Y_{PH}) which had slightly higher yields in higher velocity than in low velocity. A similar trend was found when plotting the mean and standard error of total length, number of stems, belowground dry mass, and aboveground dry mass (Figures 3a-3h). There was higher total length, number of stems, belowground dry biomass, and aboveground dry biomass for hydrilla at lower velocity and for Illinois pondweed at higher velocities.

Yield for both Illinois pondweed and hydrilla was lower in silt sediment than it was in sand sediment. Relative yield for both species was too close between sand and silt sediments to draw a definitive conclusion as to which sediment had more relative yield. Boxplots showed differences in the values of all response variables between sand and silt sediment (Figures 2a-d and 2a-d). Hydrilla had higher total length and number of stems in silt substrate than in sandy substrate. Illinois pondweed had lower total length and number of stems in silt than in sand.

Discussion

Overall Illinois pondweed had higher number of stems, greater above- and belowground dry biomass, and greater total stem length than hydrilla. Spencer and Rejmanek (1989) showed that initial propagule size can influence the competition between macrophyte species, and that hydrilla grown from fragments may take longer to propagate compared to fragments of other macrophyte species. A faster initial growth of Illinois pondweed from fragments compared to the growth of hydrilla from fragments may explain the overall greater growth of Illinois pondweed observed in monoculture and mixtures.

I predicted that Illinois pondweed would exhibit greater growth than hydrilla in high water velocity. Although the statistical analysis did not show any significant difference in growth of Illinois pondweed or hydrilla in high water velocities, indices suggested that higher water velocities may be affecting macrophyte growth. There was a higher yield of total biomass of Illinois pondweed than hydrilla in both high and low velocities. Illinois pondweed biomass decreased slightly in low velocity while hydrilla biomass increased in low velocity. This suggests that higher water velocity was suppressing the growth of hydrilla, while creating favorable growing conditions for Illinois pondweed.

Sediment characteristics may affect the outcome of competition between Illinois pondweed and hydrilla. Sand has less organic material and thus less nutrient availability (Barko & Smart 1896). Sago pondweed (*Stuckenia pectinata*) exhibits a positive correlation between the coarseness of sediment and plant biomass (Barko & Smart 1986), whereas hydrilla grown in finer sediment produces greater biomass (Spencer et al. 1992). I predicted that Illinois pondweed would have higher growth in sand sediment than hydrilla, and that hydrilla would have higher growth rates in silt sediment than Illinois pondweed. In fact the yield of both Illinois pondweed and hydrilla was lower in silt sediment than in sand. Illinois pondweed and hydrilla probably both had higher yields of biomass in sand because the sand used in the experiment had higher nutrients levels and thus more nutrient availability. The sand had nitrate and phosphorus levels of 27 ppm and 5 ppm respectively, while the silt had nitrate and phosphorus levels of 4ppm and 3 ppm respectively. Although the differences in yield of Illinois pondweed and hydrilla between sand and silt sediments were not statistically significant, the slightly higher total biomass of Illinois pondweed in sand suggests that sediments with increasing coarseness may be more conducive to Illinois pondweed growth.

Out of the three factors tested, competitive pressure was the factor that influenced growth the greatest. Prior research has shown that it is possible for native macrophytes to outcompete a non-native species (Moen & Cohen 1989, Van et al. 1998). Spencer and Ksander (2000) found that in some instances an established stand of American pondweed (*Potamogeton nodosus*) might reduce the propagules or biomass of invading hydrilla. Spencer and Ksander (2000) also found that interspecific competition between hydrilla and American pondweed grown in mixtures was greater than intraspecific competition between hydrilla. I predicted that Illinois pondweed would exhibit greater growth when competitive pressures were equal with or greater than hydrilla. Whereas Illinois pondweed did produce more biomass than hydrilla in equal mixtures, it produced less biomass when it was planted at higher ratios than hydrilla. The same can be said for hydrilla which produced less biomass when planted at higher densities. In this experiment relative yield and change in contribution indicated the intraspecific competition of both species was greater than the interspecific competition of both species, so both species produced more biomass in mixture than in monoculture. These higher levels of intraspecific competition explain why Illinois pondweed and hydrilla were more productive when they were planted at a lower ratio than the competitor.

An explanation for the higher levels of intraspecific competition observed in each species is that they are not competing with one another. Classic competition theory suggests that two species are not competing when intraspecific competition is greater than interspecific competition, particularly when aboveground and belowground competition is equally important (Aguiar et al. 2001). Furthermore, both species may have different niche requirements and thus may not be interacting with one another. In the San Marcos River, Illinois pondweed and hydrilla both occur in similar sediments and velocities; however as discussed earlier, this study suggests that each species may produce more biomass in different sediment types and velocities.

The results of this study suggest that sand and high velocities are two environmental conditions which give Illinois pondweed a competitive advantage against hydrilla. Although the velocities which were tested in this experiment were not high compared to usual flows in the San Marcos River, the slight variation in yield and change in contribution between both velocities and substrate types suggests that high velocity and sand sediment may have an effect on the growth of Illinois pondweed and hydrilla. These findings might be useful in management strategies, particularly when other means of vegetation removal are not available, as in the case in the San Marcos River. Identifying areas in water bodies where abiotic and biotic environmental conditions provide the resources necessary for native species to out-compete non-natives may be an effective strategy in habitat restoration. Planting native plant species in such areas may require minimal effort to continually manage after they become established. These strategies could be used to enhance EAHCP efforts in the San Marcos River where invasive plant management options are limited. Part of the EAHCP plan includes planting native species after removal of non-natives. Illinois pondweed may not be a suitable plant for remedial gardening, not because it is an inferior competitor with hydrilla, but because it may not be competing with it at all. In fact, Illinois pondweed may be aiding hydrilla propagation by slowing water velocity, thus accumulating fine sediments and creating suitable habitat for hydrilla.

This research must be considered a preliminary step in researching the effects of environmental conditions on competitive interactions of macrophytes in a lotic system. Natural communities are much more diverse with a multitude of factors influencing a wider range of interacting species. Further research is needed understand the competitive interactions between native and invasive species in riverine ecosystems. Understanding the interactions between macrophyte species and specific environmental conditions which influence them can be used to successfully manage an area inundated by an invasive species and is critical for future habitat restoration efforts and long-term management of invasive species.

Table 1: Model statements for linear mixed-effect models.

Model	**Model Statement**
1	$Y= \beta_o + \beta_{velocity} * \beta_{substrate} + \beta_{plant\ type} + \beta_{competitive\ pressure} + \epsilon$
2	$Y= \beta_o + \beta_{velocity} + \beta_{substrate} + \beta_{plant\ type} + \beta_{competitive\ pressure} + \epsilon$
3	$Y= \beta_o + \beta_{velocity} + \beta_{substrate} + \beta_{plant\ type} + \epsilon$
4	$Y= \beta_o + \beta_{substrate} + \beta_{competitive\ pressure} + \epsilon$
5	$Y= \beta_o + \beta_{substrate} + \epsilon$
6	$Y= \beta_o + \beta_{competitive\ pressure} + \epsilon$
7	$Y= \beta_o + \beta_{plant\ type} + \epsilon$
8	$Y= \beta_o + \beta_{velocity} + \epsilon$
9	$Y= \beta_o + \beta_{plant\ type} + \beta_{competitive\ pressure} + \epsilon$

The same models were used for each response variable (total stem length, number of stems, aboveground dry biomass, and belowground dry biomass(Y) = response variable; (β_o) = conditional mean when x = 0; ($\beta_{velocity,}$ $\beta_{substrate}$, etc.) = slope, change in mean of Y per 1 unit change x; (ϵ) = residual error.

Table 2: AIC Model selection tables for each response variable.

Total Stem Length

Model	df	logLik	AICc	delta	weight
1*	9.000	-525.887	1070.306	0.000	0.403
9*	6.000	-529.459	1071.164	0.859	0.263
2*	8.000	-527.914	1072.252	1.947	0.152
7*	4.000	-532.329	1072.775	2.470	0.117
3	6.000	-530.870	1073.987	3.681	0.064
6	5.000	-555.016	1120.206	49.901	0.000
4	6.000	-554.435	1121.115	50.810	0.000
8	4.000	-556.586	1121.288	50.983	0.000
5	4.000	-556.771	1121.659	51.353	0.000

Number of Stems

Model	df	logLik	AICc	delta	weight
1*	9.000	-350.147	718.826	0.000	0.657
2*	8.000	-352.199	720.822	1.996	0.242
9	6.000	-355.287	722.820	3.994	0.089
3	6.000	-357.642	727.529	8.703	0.008
7	4.000	-360.706	729.528	10.702	0.003
6	5.000	-377.796	765.767	46.941	0.000
8	4.000	-379.628	767.372	48.546	0.000
4	6.000	-377.795	767.837	49.011	0.000
5	4.000	-382.319	772.755	53.929	0.000

Above-ground Dry Biomass

Model	df	logLik	AICc	delta	weight
2*	8.000	-399.766	815.957	0.000	0.440
1*	9.000	-398.760	816.053	0.096	0.420
9	6.000	-403.048	818.342	2.385	0.134
3	6.000	-406.349	824.945	8.988	0.005
7	4.000	-409.591	827.299	11.342	0.002
4	6.000	-475.699	963.645	147.688	0.000
6	5.000	-477.034	964.243	148.287	0.000
5	4.000	-479.470	967.056	151.099	0.000
8	4.000	-479.802	967.720	151.763	0.000

Below-ground Dry Biomass

Model	df	logLik	AICc	delta	weight
1	9.000	-454.717	927.967	0.000	0.582
2	8.000	-456.508	929.441	1.473	0.279
9	6.000	-459.616	931.479	3.512	0.101
3	6.000	-460.929	934.105	6.138	0.027
7	4.000	-463.836	935.789	7.822	0.012
8	4.000	-575.518	1159.152	231.185	0.000
6	5.000	-575.214	1160.604	232.636	0.000
5	4.000	-576.687	1161.491	233.523	0.000
4	6.000	-575.023	1162.293	234.325	0.000

(df) =degrees of freedom; (logLik) =natural logarithm of the maximum likelihood for the model; (AIC) =rounded Akaike weights; (delta) =measure of model relative to all other models. Models with a delta of 0-2 are highly supported; (weight) =intuitive measure of how much better the model may fit the data. Weights with values closer to 1.0 have a better fit.

Table 3: Summaries of linear mixed-effect models for each response variable.

Total Stem Length				
Coefficients	Estimate	Std. Error	t value	P Value
Intercept	229.210	1.181	5.611	<0.001*
Low Velocity	1.023	1.208	1.739	0.903
Silt	1.386	1.215	0.740	0.095
Illinois pondweed	2.411	1.125	13.654	<0.001*
Moderate interspecific competition	1.072	1.156	0.017	0.635
Intraspecific competition	1.449	1.179	3.004	0.025*
Silt:Low Velocity	-0.620	1.266	1.415	0.043*

Number of Stems				
Coefficients	Estimate	Std. Error	t value	P Value
Intercept	9.927	1.105	22.923	< 0.001*
Low Velocity	1.384	1.106	3.210	0.001*
Silt	1.177	1.109	1.578	0.115
Illinois pondweed	1.647	1.074	7.030	0.000
Moderate interspecific competition	1.071	1.091	0.792	0.429
Intraspecific competition	1.364	1.104	3.143	0.002*
Silt:Low Velocity	-0.749	1.153	-2.032	0.043*

Aboveground Dry Biomass				
Coefficients	Estimate	Std. Error	t value	P Value
Intercept	1.913	1.122	5.611	< 0.001*
Low Velocity	1.225	1.123	1.739	0.082
Silt	-0.920	1.126	0.740	0.459
Illinois pondweed	3.065	1.085	13.654	< 0.001*
Moderate interspecific competition	-1.000	1.105	0.017	0.986
Intraspecific competition	1.409	1.120	3.004	0.003*
Silt:Low Velocity	-0.890	1.177	0.695	0.487

Belowground Dry Biomass				
Coefficients	Estimate	Std. Error	t value	P Value
Intercept	-0.601	1.145	-3.765	0.000*
Low Velocity	1.519	1.146	3.059	0.002*
Silt	1.148	1.150	0.992	0.322
Illinois pondweed	5.812	1.101	18.360	< 0.001*
Moderate interspecific competition	-1.194	1.124	-1.517	0.130
Intraspecific competition	1.194	1.143	1.328	0.185
Silt:Low Velocity	-1.440	1.212	-1.897	0.059

α=0.05; Significant P-values indicated by (*). High velocity, sand, hydrilla, and equal interspecific competition were coded as reference categories and are represented by the intercept.

Table 4. Competition growth indices of hydrilla and Illinois pondweed grown under different competitive pressures

	High Velocity			**Low Velocity**			**Sand**			**Silt**		
	H=	H<	H>	H=	H<	H>	H=	H<	H>	H=	H<	H>
	P=	P>	P<	P=	P>	P<	P=	P>	P<	P=	P>	P<
Y_{HP}	2.96	1.90	5.68	3.83	2.24	6.35	3.59	2.10	5.89	2.79	2.18	5.85
Y_{PH}	13.41	14.26	7.88	11.30	16.31	7.95	13.21	18.23	8.52	11.60	12.46	7.86
RY_H	1.49	1.92	1.91	1.35	1.59	1.50	1.43	1.67	1.56	1.22	1.91	1.71
RY_P	1.91	1.35	2.24	1.40	1.35	1.98	1.49	1.37	1.92	1.85	1.32	2.50
CC_H	-0.18	0.37	-0.09	-0.03	0.15	-0.13	-0.03	0.19	-0.11	-0.27	0.38	-0.18
CC_P	0.05	-0.03	0.07	0.01	-0.02	0.14	0.01	-0.02	0.09	0.10	-0.05	0.20
	$Y_H = 3.96$			$Y_H = 5.64$			$Y_H = 5.03$			$Y_H = 4.55$		
	$Y_P = 14.04$			$Y_P = 16.1$			$Y_P = 17.76$			$Y_P = 12.56$		

Values were calculated using the total dry biomass of hydrilla (H) and Illinois pondweed (P). (=), Both species were grown at equal interspecific competitive pressure (2:2); (>) species grown at 75% of mixture; (<), species grown at 25% of mixture. Y_{HP} and Y_{PH} = yield of mixture calculated as the mean of the entire treatment (grams). RY= relative yield (percentage); CC= change in contribution (percentage).

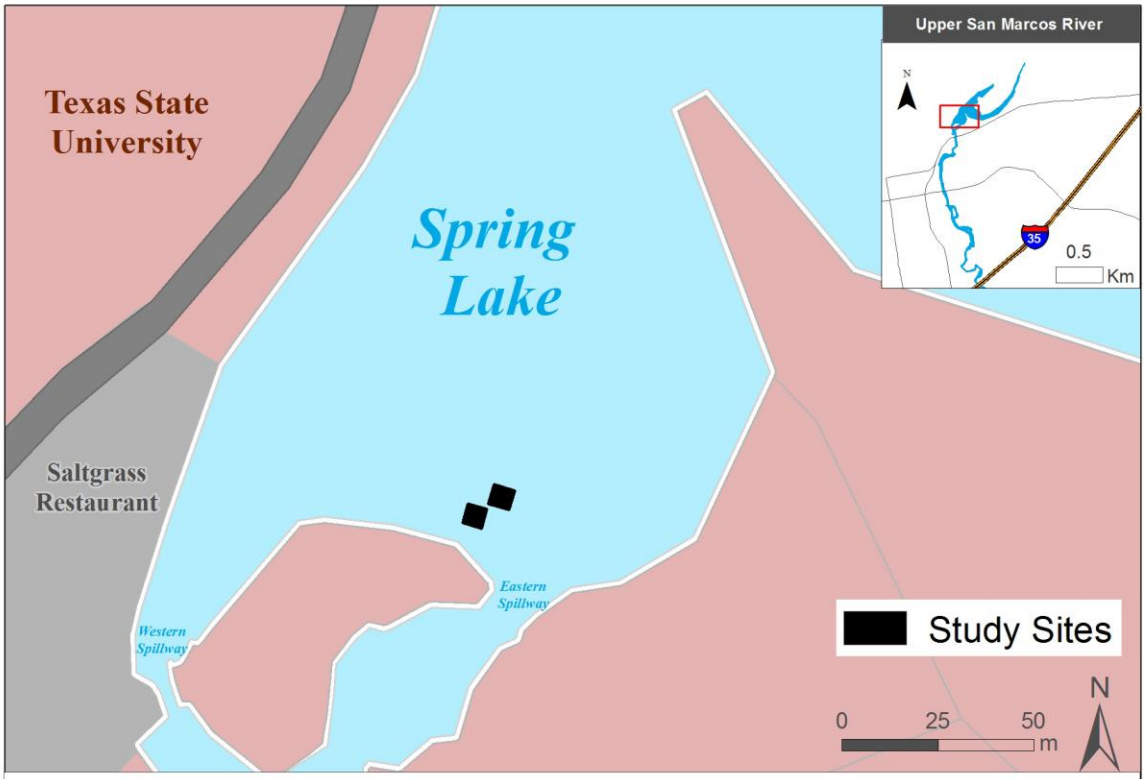

Figure 1: Locations of experimental plots in Spring Lake.

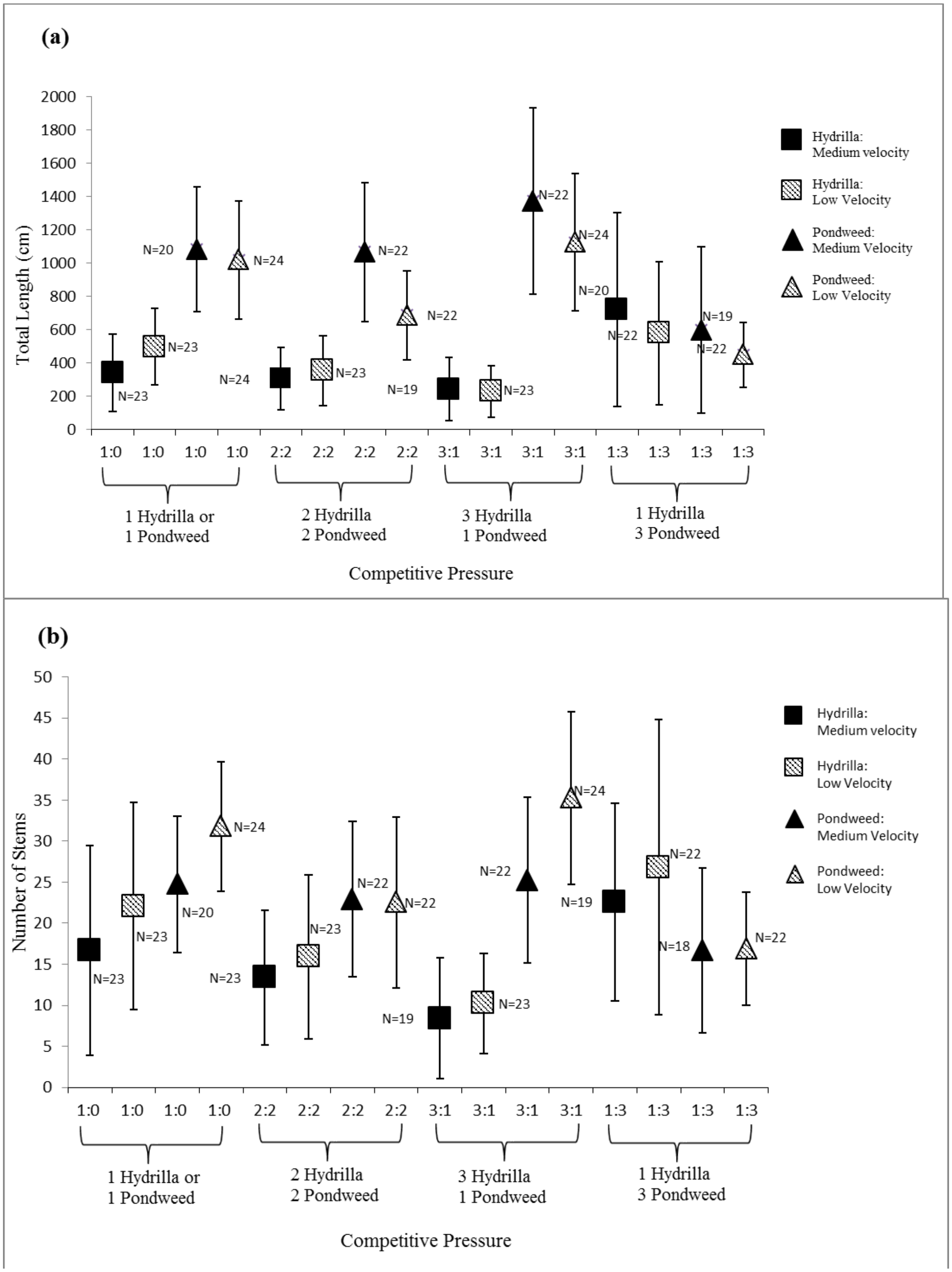

Figure 2a-d: Mean and standard error of (a) total stem length (cm), (b) number of stems, (c) belowground dry biomass, and (d) aboveground dry biomass of hydrilla and Illinois pondweed at high and low current velocities. Sediment type is held constant. Sample size is given next to each data point.

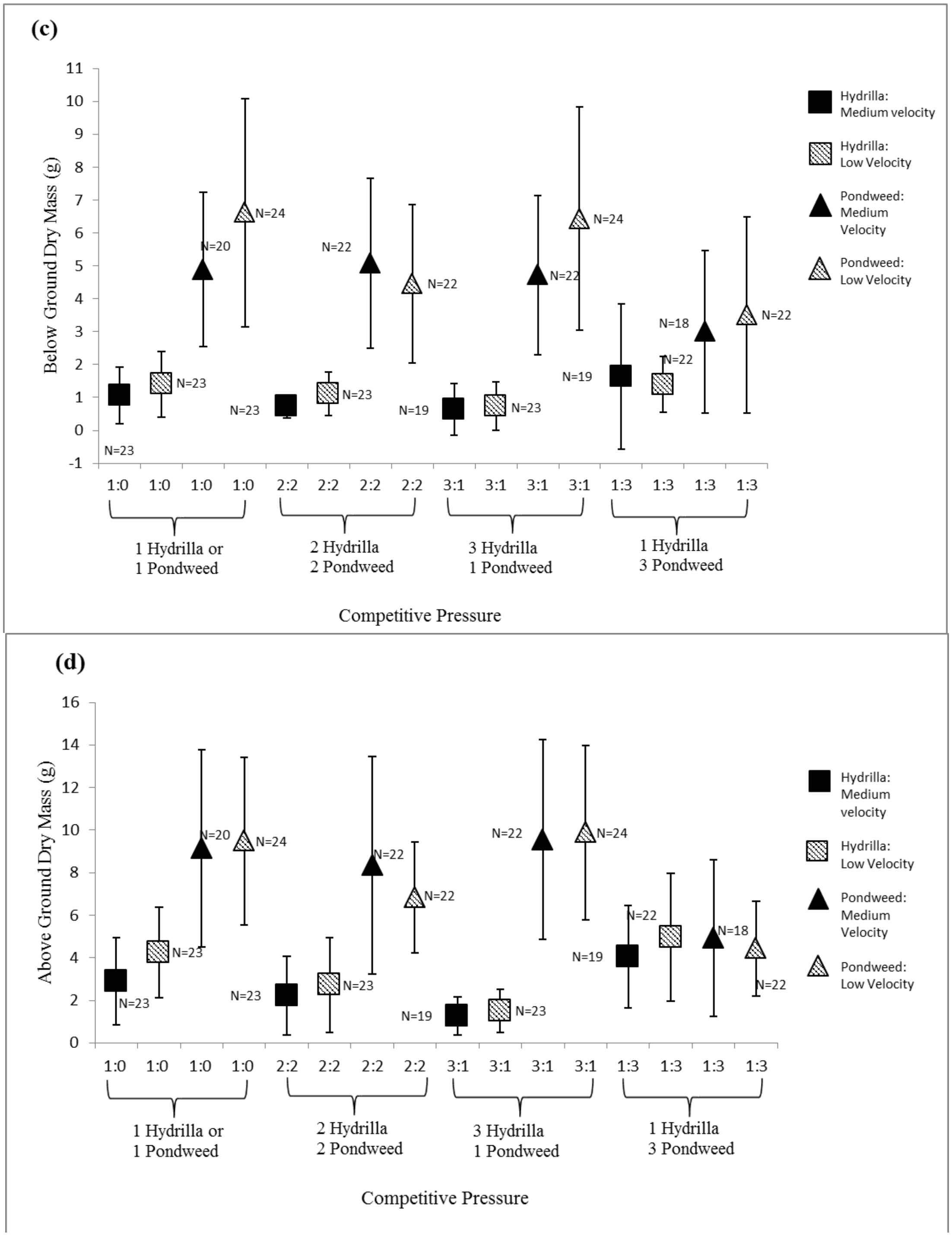

Figure 2, continued.

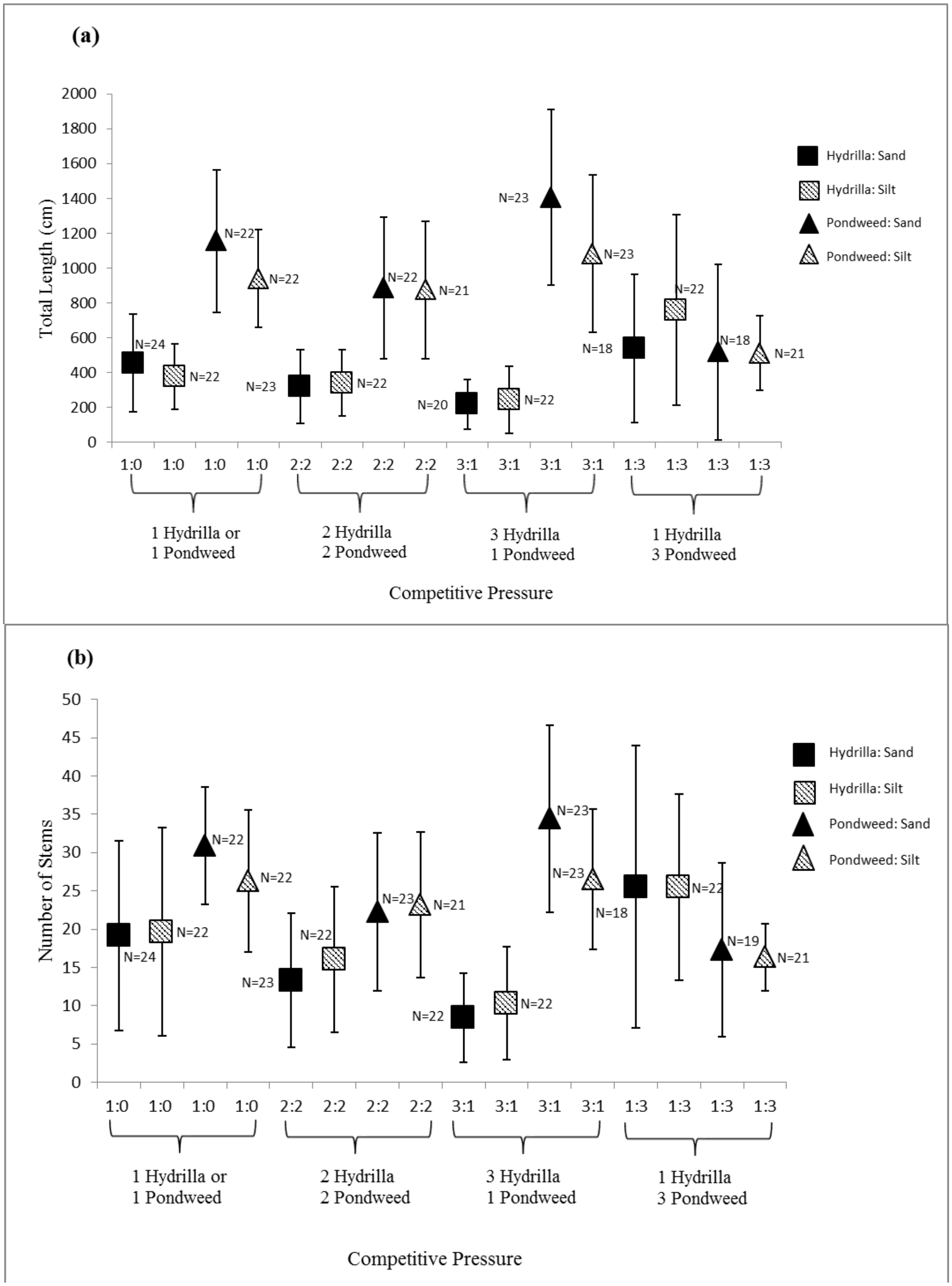

Figure 3a-d: Mean and standard error of (a) total stem length (cm), (b) number of stems, (c) belowground dry biomass, and (d) aboveground dry biomass of hydrilla and Illinois pondweed with different sediment types. Water velocity is held constant. Sample size is given next to each data point.

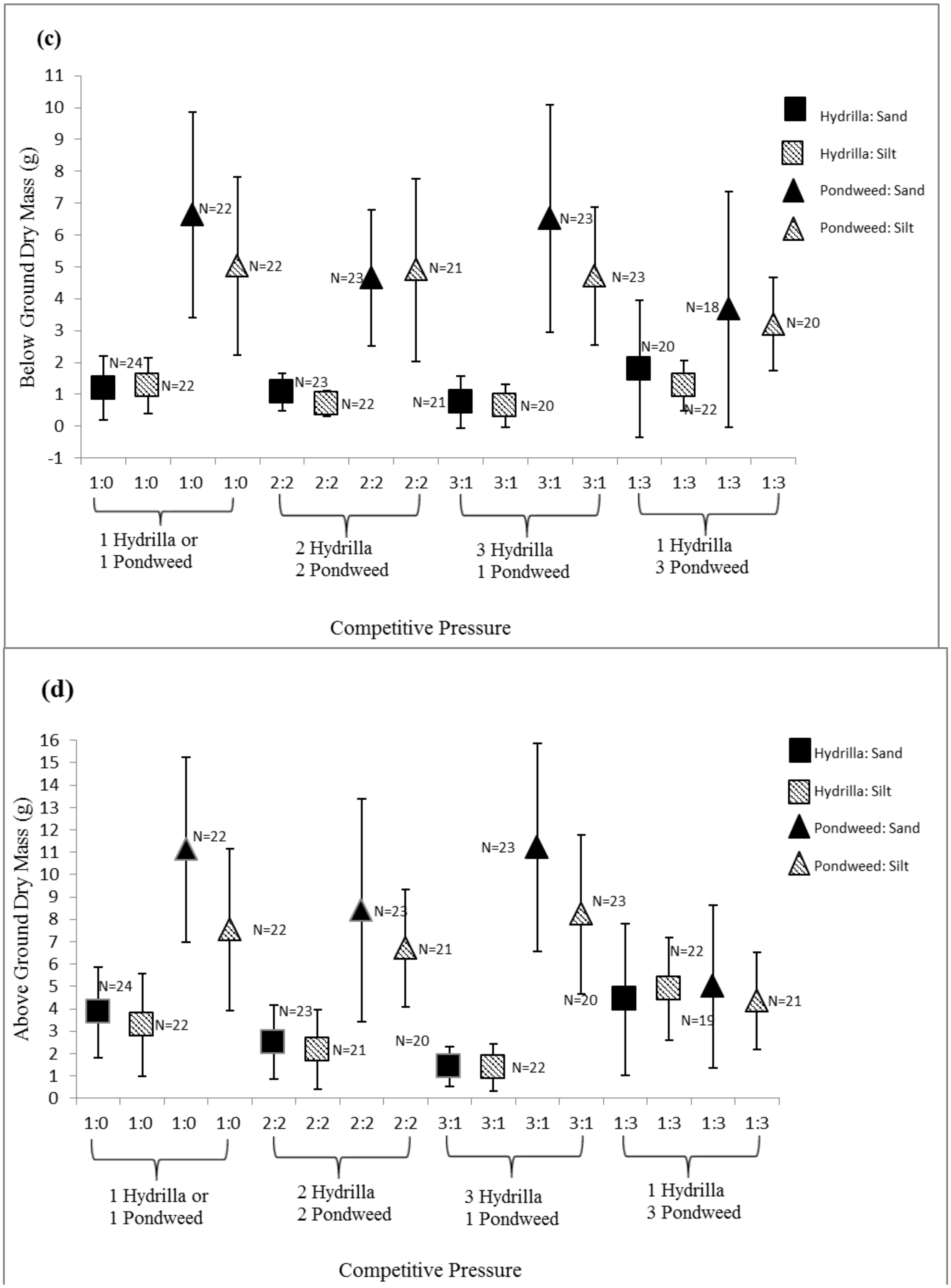

Figure 3, continued.

REFERENCES

Aquiar MR, Lauenroth WK, & Peters DP. 2001. Intensity of intra- and interspecific competition in coexisting shortgrass species. Journal of Ecology 89:40-47

Amundsen PA, Knudsen R, & Klemetsen A. (2007). Intraspecific competition and density dependence of food consumption and growth in arctic charr. Journal of Animal Ecology 76: 149–158

Barko JW, Smart M. 1986. Sediment-related mechanisms of growth limitations in submersed macrophytes. Ecology 67:1328-1340

Barnes MA, Jerde CL, Wittmann ME, Lindsay C, Ding J, Zhang J, Purcell M, Budhathoki M, & Lodge D. 2014. Geographic selection bias occurrence data influences transferability of invasive Hydrilla verticillata distribution models. Ecology and Evolution 4:2584–2593

Bowles DE, Bowles, BD. 2001. A review of the exotic species inhabiting the upper San Marcos River, Texas, U.S.A. Texas Parks and Wildlife Department, Austin, TX. pp.30

Burnham KP, Anderson DR. 2002. Model selection and multimodel inference: a practical information-theoretic approach. Springer Science & Business Media. pp. 481

Chambers PA, Prepas EE, Hamilton HR, & Bothwell ML. 1991. Current velocity and its effect on aquatic macrophytes in flowing waters. Ecological Applications 1:249-257

Chilton, EW. 2011. Aquatic Vegetation Management in Texas: A Guidance Document. Texas Parks and Wildlife Department Inland Fisheries 13:1-55

Colle DE, Shireman JV. 1980. Coefficients of condition for largemouth bass, bluegill, and redear sunfish in hydrilla-infested lakes. Transactions of the American Fisheries Society 109:521-531

Cuda JP, Sutton DL. 1999. Is the aquatic weed Hygrophila, *Hygrophila polysperma* (Polemoniales: Acanthaceae), a suitable target for classical biological control? Proceedings of the X International Symposium on Biological Control of Weeds 337-348

EAHCP. 2013. Edwards aquifer habitat conservation plan. Edwards Aquifer Authority. Available online: http://eahcp.org/ (February 20, 2014)

Haller WT, Shireman JV, & Durant DF. 1980. Fish harvest resulting from mechanical control of hydrilla. Transactions of the American Fisheries Society 109:517-520

Hardy TB, Kollaus K, & Tower K. 2010. Evaluation of the proposed Edwards Aquifer recovery implementation program drought of record minimum flow regimes in the Comal and San Marcos River systems. The Edwards Aquifer Recovery Implementation Program pp.22

Howard-Williams C, Liptrot MRM. 1980. Submerged macrophyte communities in a brackish South African estuarine-lake system. Aquatic Botany 9:101-116

Kelty MJ, Cameron IR. 1995. Plot designs for the analysis of species interactions in mixed stands. Commonwealth Forestry Review 74: 322-332

Lemke DE. 1989. Aquatic macrophytes of the upper San Marcos River, Hays Co., Texas. The Southwestern Naturalist 34:289-291

Langeland KA. 1996. *Hydrilla verticillata* (L.F.) Royle (Hydrocharitaceae), "The perfect aquatic weed". Castanea 61:293-304

Madsen JD, Chambers PA, James WF, Koch EW & Westlake DF. 2001. The interaction between water movement, sediment dynamics and submersed macrophytes. Hydrobiologia 444:71-84

Moen RA, Cohen Y. 1989. Growth and competition between *Potamogeton pectinatus* L. and *Myriophyllum exalbescens* Fern in experimental ecosystems. Aquatic Botany 33:257-270

Owens C, Madsen J, Smart RM, & Stewart, RM. 2001. Dispersal of native and nonnative aquatic plant species in the San Marcos River, Texas. Journal of Aquatic Plant Management 39:75-79

Posada D, Buckley T. 2004. Model selection and model averaging in phylogenetics: advantages of Akaike information criterion and Bayesian approaches over likelihood ratio tests. Systematic Biology 5315: 793-808

Saunders KS, Mayes KB, Jurgensen TA, Trungale JF, Kleinsasser LJ, Aziz K, Fields JR, & Moss RE. 2001. An evaluation of spring flows to support the upper San Marcos River spring ecosystem, Hays County, Texas. Texas Parks and Wildlife Department River Studies Report 16:1-33

Schmid, WD. 1965. Distribution of aquatic vegetation as measured by line intercept with scuba. Ecology 46:816-823

Sousa, WTZ. 2011. *Hydrilla verticillata* (Hydrocharitaceae), a recent invader threatening Brazil’s freshwater environments: a review of the extent of the problem. Hydrobiologia 669:1-20

Spencer DF, Ksander GG, & Bissell SR. 1992. Growth of monoecious hydrilla on different soils amended with peat or barley straw. Journal of Aquatic Plant Management 30:9-15

Spencer DF, Ksander GG. 2000. Interactions between American pondweed and monoecious hydrilla grown in mixtures. Journal of Aquatic Plant Management 38: 5-13

Spencer DF, Rejmanek M. 1989. Propagule type influences competition between two submersed aquatic macrophytes. Oecologia 81:132-137

Van T, Wheeler G, & Center TD. 1998. Competition between *Hydrilla verticillata* and *Vallisneria americana* as influenced by soil fertility. Aquatic Botany 62:225-233

Vanclay JK. 2006. Experimental designs to evaluate inter- and intra-specific interactions in mixed plantings of forest trees. Forest Ecology and Management 233:336-374

Van Driesche R, Blossey B, Hoddle M, Lyon S, & Reardon R. 2002. Biological Control of Invasive Plants in the Eastern United States. USDA Forest Service Publication FHTET-2002-04 1-413

Weigelt A, Jolliffe P. 2003. Indices of plant competition. Journal of Ecology 91:707-720

Williams CA, McCarthy BC. 2001. A new index of interspecific competition for replacement and additive designs. Ecological Research 16:29-40