# Notes

# Effect of ecotype, sediment composition, and fertility level on productivity of eight Florida ecotypes of American eelgrass (*Vallisneria americana*)

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# INTRODUCTION

Lake habitat enhancement projects are often undertaken to reverse systemic changes caused by altered hydrologic patterns and to restore native littoral vegetation. Revegetation with native aquatic plants improves water quality, reduces wave action and erosion, and restores fish and wildlife habitats. For example, fish populations are healthiest when vegetative cover is between 15 and 85% (Canfield and Hoyer 1992), so submersed aquatic vegetation (SAV) is often planted to improve habitat quality for fish and wildlife (Allen and Tugend 2002). Revegetation efforts at some sites are effective and newly planted SAV thrives, but, in other cases, establishment of self-sustaining populations of SAV is unsuccessful. This suggests that environmental factors play a determinant role in the establishment of SAV. Some workers have developed methods to predict whether a particular site is likely to host self-supporting populations of native plants introduced through restoration and revegetation efforts (e.g., Grodowitz et al. 2009, Mazzotti et al. 2011), but those methods are technical and unlikely to be employed by resource managers.

Revegetation projects rely predominantly on the use of native plants to foster ecological integrity. A primary goal of lake restoration is often to improve habitat structure, so revegetation plans frequently include a mix of emergent, floating-leaved, and submersed aquatic vegetation. For example, restoration plans for El Dorado Lake in Kansas included emergent plants, such as common arrowhead (*Sagittaria latifolia* Willd.) and pickerelweed (*Pontederia cordata* L.), floating-leaved plants, such as fragrant waterlily (*Nymphaea odorata* Ait.) and American lotus (*Nelumbo lutea* Willd.), and submersed plants, such as Illinois pondweed (*Potamogeton illinoensis* Morong) and American eelgrass (*Vallisneria ameri*-

American wild-celery) is a member of the monocotyledonous Hydrocharitaceae family and is native to eastern North America. This entirely submersed species has ribbon-like leaves that emanate from a central rosette, with the meristem and most of the biomass found on, or immediately above, the hydrosoil (Godfrey and Wooten 1979). Sexual reproduction takes place between plants with pistillate or staminate flowers in this dioecious species, but most colonization is the result of vegetative reproduction (i.e., runners and winter buds). Smart et al. (2006) emphasized the importance of using locally grown (or collected) native species in revegetation projects because these regional ecotypes are often adapted to specific geographic regions. Two distinct biotypes of American eelgrass have been identified in North America and differ in their response to winter conditions. Both types function as perennials; however, southern biotypes are evergreen, whereas northern biotypes are deciduous and produce overwintering buds (Smart et al. 2005, 2006). It seems likely that most or all American eelgrass in Florida is the southern biotype of the species; however, it is quite possible that multiple ecotypes have developed because of regional adaptation. American eelgrass is widely adapted and tolerant of

*cana* Michx.) (Dick and Smart 2004). American eelgrass is a highly desired candidate for inclusion in restoration projects

(Jaggers 1994) because the species is a herbaceous perennial

with an open growth habit that provides structure to the

underwater habitat. American eelgrass (also commonly

known as vallisneria, eel-grass, tape-grass, ribbon-grass, or

American eelgrass is widely adapted and tolerant of diverse environmental parameters, including high turbidity (Davis and Brinson 1980), low light levels (Titus and Adams 1979), and various water chemistry regimes (Korschgen and Green, 1988, and references within). A survey of 118 Florida lakes revealed that American eelgrass grows under a wide range of light levels and is present in areas with Secchi depths ranging from 0.4 to 3.6 m (1.3 to 11.8 ft) (Hoyer et al. 1996). Also, Titus and Adams (1979) suggested that American eelgrass is well adapted to low light conditions, and subsequent research supports the hypothesis that this species has a competitive advantage when grown under low-

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light conditions (Harley and Findlay 1994, Rybicki and Carter 2002, Garrison et al. 2005).

In contrast, sediment characteristics strongly influence the growth of American eelgrass because roots of these aquaphytes must act as secure anchors while absorbing nutrients from the sediment. Barko and Smart (1986) stated that sediment density was the most influential factor regulating growth of aquatic plants because extremely high or low sediment densities resulted in multiple nutrient deficiencies. Sediments with high sand fractions (> 75%)and concomitant high bulk densities (0.9 to  $1.3 \text{ g ml}^{-1}$  [7.5 to 10.8 lb gal<sup>-1</sup>]) have poor nutrient holding capacities, whereas sediments with high organic matter fractions (> 20%) and concomitant low bulk densities (< 0.2 g)ml<sup>-1</sup>) are overly porous, resulting in long nutrient diffusion distances (Barko and Smart 1986). Hunt (1963) found that American eelgrass grew best in silty clay but that the species was able to establish in virtually any substrate as long as the substrate was not overly soft and allowed root penetration sufficient to anchor the plants. Most aquatic plants obtain most or all of their required nutrients (i.e., N, P, and K) from the sediment, so nutrients in the water column typically have little or no effect on the growth of SAV (Barko and Smart 1981, Anderson and Kalff 1986). American eelgrass tolerates a wide range of fertility conditions, but Anderson and Kalff (1986) noted that the species attained the greatest biomass when cultured with low levels of N, P, and K.

The ability of American eelgrass to tolerate a wide range of environmental conditions is partially responsible for its frequent inclusion in restoration projects, but it seems likely that establishment is influenced by factors such as sediment density and nutrient levels. Lake restoration plans often specify that plant material used for revegetation be collected from nearby populations. However, "natural" sites often have sediment conditions that differ considerably from those at the site targeted for restoration. For example, many Florida lakes that host naturally occurring populations of American eelgrass have sandy, nutrient-poor sediments, whereas many lakes targeted for restoration have highly organic, flocculent, nutrient-rich sediments (E. Hayes, pers. comm.). As a result, plants that are collected from naturally occurring populations and transplanted to restoration sites may be ill-suited to their new habitat and may perform poorly or fail to establish self-sustaining populations. Therefore, the purpose of these experiments was to measure productivity of different ecotypes of American eelgrass collected in Florida to determine whether they respond differently to changes in artificial sediment organic matter content and fertility levels. Identification of ecotypes of American eelgrass that are more tolerant of highly organic, nutrient-rich sediments could provide restoration managers with an important tool to improve revegetation success.

# MATERIALS AND METHODS

# **Plant material**

Eight ecotypes of American eelgrass were grown in 5 sediment mixtures and 4 nutrient levels. Two phenotypi-

cally distinct ecotypes were obtained from a commercial aquatic plant nursery in north Florida<sup>1</sup> and were designated Narrow and Wide, based on leaf width. These ecotypes were not maintained in culture by the supplying nursery but were collected from proprietary, undisclosed, specific sites in the Santa Fe River. Six other Florida ecotypes of American eelgrass were collected from one site each in Lakes George, Fairview, and Mann, and from 3 geographically discrete sites (Central, East, and North) in Lake Istokpoga. These ecotypes are phenotypically different from one another, and preliminary genetic analysis using intersimple sequence repeat (ISSR) markers by Gettys and Haller (unpub. data) has revealed that they are genotypically different as well.

#### Sediments and fertility

Blends of coarse builder's sand and commercially available peat<sup>2</sup> were used to create a series of 5 artificial sediments with a range of organic matter contents. Sediment blends included 100% sand, 75% : 25% sand : peat, 50% : 50% sand : peat, 25% : 75% sand : peat, and 100% peat. Four nutrient levels were examined in these experiments, with fertility supplied by a controlled-release fertilizer.<sup>3</sup> Nutrient treatments included control (no fertilizer), low (1 g L<sup>-1</sup> [0.133 oz gal<sup>-1</sup>]), medium (2 g L<sup>-1</sup>), and high (4 g L<sup>-1</sup>) rates. Sediment mixtures were thoroughly blended, and fertilizer prills were gently incorporated into sediments before filling containers.

## Experimental conditions and analysis

Each experimental unit consisted of a single nursery container (21 cm diam by 13 cm deep [8.3 in diam by 5.1 in deep]) without holes that was filled to a depth of 7.5 cm (final sediment volume approximately 2.6 L [0.69 gal]) and planted with a single ramet. Four replicate containers were prepared for each ecotype-sediment-nutrient combination. All pots were top-dressed with a 3-cm layer of washed pea gravel to prevent loss of sediment. Experiments were run once and initiated between September and November 2008 and concluded between January and March 2009. All experiments took place in an unheated glasshouse under ambient air temperature conditions at the University of Florida Center for Aquatic and Invasive Plants in Gainesville, FL. Day length was artificially extended to 16 h using high-intensity sodium halide lights, and average water temperature ranged from 17 to 26 C (62.6 to 78.8 F) for the duration of these experiments. Experimental units were placed in 2.5-m-diam (8.2-ft-diam) tanks filled with well water and maintained at a depth of 0.5 m. Eight tanks were used, and each tank held all sediment-fertility combinations and replicates of a single ecotype (i.e., 80 units per tank). Each tank was equipped with a biofilter<sup>4</sup> to reduce waterborne nutrient loads and algae blooms. Plants were cultured for 16 wk; at which time, the number of ramets per container was recorded. A destructive harvest was then conducted to separate aboveground leaves and belowground roots at the sediment line. Plant tissue was washed clean of sediment and other debris and dried in a forced-air oven at 90 C until a constant weight was achieved. Raw data

were subjected to ANOVA and Fisher's Protected LSD test separation using SAS software.<sup>5</sup> The general linear model included ecotype, sediment type, and fertility level as independent variables, both alone and as interactions. Dependent variables were number of ramets, the ratio of root to shoot dry biomass, and total dry biomass.

#### **RESULTS AND DISCUSSION**

#### Ecotype

Plant source, hereafter referred to as ecotype, had a significant effect on productivity-as measured by total dry biomass and ramet production-in these experiments (Figure 1), but root: shoot ratio was not affected by ecotype, sediment composition, or fertility level (P =0.2594). Total dry weights were highest in the Istokpoga East and Narrow ecotypes, which yielded an average dry biomass of 4.25 and 4.10 g container<sup>-1</sup>, respectively (Figure 1A). The Narrow ecotype produced an average of 9.8 ramets container<sup>-1</sup>, which was significantly greater than ramet production by any of the other ecotypes (Figure 1B). The Narrow ecotype yielded both the highest biomass and the greatest number of ramets; therefore, these results suggest that the Narrow ecotype is the most productive-in terms of total dry biomass and ramet production-of the 8 ecotypes of American eelgrass tested in these experiments.

# **Fertility level**

Fertility level also had a significant effect on productivity of these Florida ecotypes of American eelgrass (Figure 2). There was no difference in average biomass per container of plants grown with medium  $(2 \text{ g L}^{-1})$  or low  $(1 \text{ g L}^{-1})$  levels of fertilizer and no difference between biomass of plants grown with low or high (4 g  $L^{-1}$ ) levels of fertilizer, but average biomass per container was lowest in treatments grown with the control (no fertilizer) rate (Figure 2A). There was no difference in ramet production of plants grown with medium or low levels of fertilizer and no difference in ramet production of plants grown with low or high levels of fertilizer, but average ramet production was lowest in treatments grown with the control (no fertilizer) rate (Figure 2B). Plants cultured with any level of fertilizer yielded more biomass and the greatest number of ramets; therefore, it seems likely that the ecotypes of American eelgrass tested in these experiments are most productive when grown under a regime that includes applications of fertilizer.

#### Sediment composition

Similar to ecotype and fertility level, sediment composition had a significant effect on productivity of the Florida ecotypes of American eelgrass examined in these experiments. Total dry biomass of plants grown in sediments with 50% or greater organic matter averaged 2.94 to 3.25 g container<sup>-1</sup>, which was significantly greater than the total dry biomass produced by plants grown in sediments that contained 75% or more sand. Ramet production was also

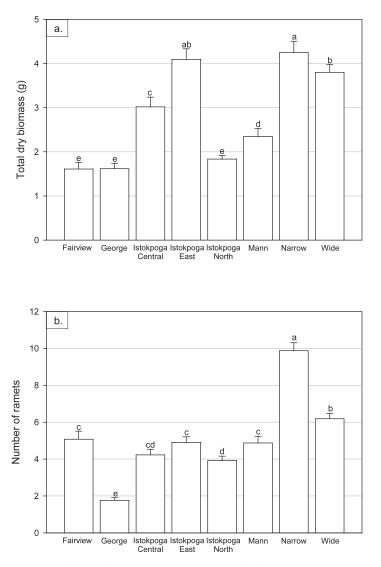


Figure 1. Effect of ecotype on productivity of 8 Florida ecotypes of American eelgrass. Bars represent the mean of 80 replicates, and error bars represent 1 SEM. Treatments coded with the same letter are not significantly different at P = 0.05. (A) Total dry biomass. (B) Ramet production.

highest in plants grown in sediments that included 50% or more organic matter and averaged 5.39 to 5.91 plants container<sup>-1</sup>. These results suggest that the ecotypes of American eelgrass studied in these experiments are most productive when grown in a substrate that includes 50% or more organic matter.

#### Interactions: ecotype and fertility level

The interaction between ecotype and fertility had a significant effect on productivity of the 8 Florida ecotypes of American eelgrass studied in these experiments. Total dry biomass was highest (4.54 to 5.40 g container<sup>-1</sup>) in the Narrow ecotype, fertilized at the low, medium, or high rate; in the Wide ecotype, fertilized at the low rate; and in the Istokpoga East ecotype, fertilized at the low or medium rate. Ramet production was greatest and ranged from 10.15 to

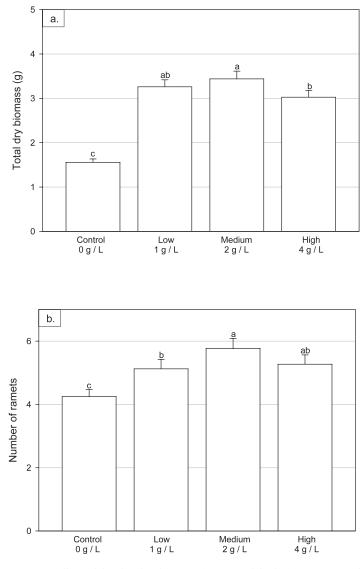


Figure 2. Effect of fertility level on productivity of 8 Florida ecotypes of American eelgrass. Bars represent the mean of 160 replicates, and error bars represent 1 SEM. Treatments coded with the same letter are not significantly different at P = 0.05. (A) Total dry biomass. (B) Ramet production.

11.60 ramets container<sup>-1</sup> in plants of the Narrow ecotype treated with low, medium, or high fertilizer rates.

#### **Other interactions**

The interaction between sediment composition and fertility had no significant effect on productivity of the 8 Florida ecotypes of American eelgrass tested in these experiments. Similar results were noted regarding the interaction between ecotype and sediment composition.

These experiments revealed that ecotype, fertility, and sediment composition are important in productivity—as measured by total dry biomass and ramet production—of American eelgrass. It is clear that the Narrow ecotype was the most productive of the 8 Florida ecotypes of American eelgrass examined in these experiments because total dry biomass and ramet production were highest in the Narrow ecotype. Also, total dry biomass and ramet production were highest in plants cultured with a moderate level of fertilizer in a substrate that contained 50% or more organic matter.

The Narrow ecotype of American eelgrass may be able to tolerate a wider range of sediment and fertility conditions than other ecotypes of American eelgrass because the number of ramets and total dry weight were significantly higher in the Narrow ecotype than in the 7 other ecotypes regardless of sediment composition and fertility level. This may have important implications for restoration ecologists because American eelgrass is anecdotally referred to as a "light feeder" that grows best under a low fertility regime, as noted by Anderson and Kalff (1986). The identification of a ecotype more tolerant of increased fertility levels may allow the use of this species in restoration sites with nutrient levels that are typically considered too high to foster successful establishment of American eelgrass.

Lake restoration programs typically specify the use of locally grown and adapted plant materials in revegetation efforts. However, these materials may not be readily available, or sites located nearby may have environmental conditions that differ significantly from those at the site being considered for restoration. Preliminary field trials by Gettys and Haller (unpub. data) have revealed that different ecotypes of American eelgrass establish with varying degrees of success in Florida lakes targeted for restoration. Because these experiments show that ecotypes of American eelgrass do differ in their response to environmental conditions, prescreening plants for tolerance to the specific conditions present at a restoration site may be a useful tool for restoration ecologists who wish to include this native, submersed species in their restoration and revegetation sites but lack readily available stocks of plants growing in a similar habitat.

#### SOURCE OF MATERIALS

<sup>1</sup>Suwannee Laboratories, 1205 SW King Street, Lake City, FL 32024.

<sup>2</sup>Majestic Earth Lawn and Garden Canadian sphagnum peat moss, Conrad Fafard Inc., 770 Silver Street, Agawam, MA 01001.

<sup>3</sup>Southern Formula Osmocote Plus, 15–9–12 N–P–K, formulated for 5to 6-mo release, The Scotts Co., Inc., 14111 Scottslawn Road, Marysville, OH 43041.

<sup>4</sup>ClearChoice Biofilter PF-1, Tetra/United Pet Group Aquatics, 3001 Commerce Street, Blacksburg, VA 24060.

<sup>5</sup>SAS Version 9.1, SAS Institute, 100 SAS Campus Drive, Cary, NC 27513-2414.

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