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**SHORT COMMUNICATION****GROWTH PATTERNS OF SHOAL GRASS *HALODULE WRIGHTII* AND MANATEE GRASS *SYRINGODIUM FILIFORME* IN THE WESTERN GULF OF MEXICO**

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**INTRODUCTION**

Seagrass beds are a valuable resource because of the numerous roles they play in coastal systems. Seagrass blades provide habitat for abundant micro- and macro-algal communities, which in conjunction with the seagrass, contribute substantially to primary productivity in estuarine systems (Heck and Valentine 2006). The blades and roots stabilize sediments, which improves water clarity and increases light penetration, further increasing primary production (Zieman 1982, Gacia and Duarte 2001). Seagrass beds may also provide increased growth rates, critical habitat, and predation refuges for a variety of organisms (e.g., Irlandi and Peterson 1991, Hemminga and Duarte 2000) and have an increased abundance and diversity of juvenile and adult fish and other epibenthic organisms (Summerson and Peterson 1984, Heck et al. 1995).

In Texas, like most of the western Gulf of Mexico (GOM), shoal grass (*Halodule wrightii*), manatee grass (*Syringodium filiforme*), and turtle grass (*Thalassia testudinum*) are the most common seagrasses (Zieman 1982, Quammen and Onuf 1993, Withers 2002). Eighty percent of seagrass beds in Texas currently occur in the Laguna Madre (hereafter LM), a hypersaline lagoon that separates a coastal barrier island (Padre Island) from the Texas mainland (Pulich 1998, Tunnell and Judd 2002). Seagrasses were once common in many Texas bays, but have disappeared or declined in coverage in many areas due to anthropogenic causes (Pulich and Onuf 2007). Fortunately, the loss of seagrasses in Texas bays was offset by the increase in seagrass abundance in the LM due to the moderation of salinity after dredging of the Gulf Intracoastal Waterway (Quammen and Onuf 1993). Shoal grass can survive and grow in salinities from 5–80 (McMillan and Moseley 1967) and this seagrass dominated the LM for decades because of its ability to withstand these extreme salinities (Withers 2002).

Shoal grass harbors a diverse resident fauna (Tolan et al. 1997) and the migratory redhead duck (*Aythya americana*) population depends on shoal grass in the LM for food during the winter season (Cornelius 1977). Shoal and manatee grass are currently present in the upper LM, with shoal grass historically being dominant (Quammen and Onuf 1993) and extensively studied (e.g., Dunton 1994, 1996). However, manatee grass is increasing in coverage

in much of the LM and is steadily replacing shoal grass in this system (Quammen and Onuf 1993, Pulich and Onuf 2007). In other areas such as Corpus Christi Bay, shoal, manatee, and turtle grass have coexisted for the past 20 yrs (Czerny and Dunton 1995, K. Dunton pers. comm.).

Although manatee grass is becoming increasingly abundant in Texas bays, its growth characteristics have not been measured in the western GOM. Changes in seagrass species composition can have significant community effects (Micheli et al. 2008), but the effects of a transition from shoal to manatee grass in the LM have not been extensively studied (but see Tolan et al. 1997). The goals of our study were to measure growth patterns of these two seagrass species in two locations in the western GOM that vary in salinity, epiphyte loads, and nutrient inputs.

**MATERIALS AND METHODS**

We selected two locations (bays) for this study: the East Flats section of Corpus Christi Bay (CCB) and another in the upper LM. Corpus Christi Bay is an urban estuary that receives substantial nutrient inputs, which are much higher than in the LM. Water exchange occurs more readily in CCB, giving this location lower, albeit more variable, salinity. By utilizing these locations, we were able to measure growth characteristics of shoal and manatee grass under different abiotic conditions. In both CCB and LM, we sampled monospecific stands of manatee grass that were adjacent to monospecific stands of shoal grass. Seagrass beds were separated by ~50 m in LM and ~500 m in CCB, and all were located in about 1.3 m of water (referenced to MLLW). GPS coordinates were 27°24.793'N, 97°21.224'W (shoal grass) and 27°24.805'N, 97°21.214'W (manatee grass) in the LM and 27°48.581'N, 97°07.323'W (shoal grass) and 27°48.758'N, 97°07.195'W (manatee grass) in CCB. We placed a PVC pole near the center of each seagrass bed and all growth characteristics were made within 10 m of these poles for each species.

Hydrolab minisondes were deployed in both locations and set to measure salinity and water temperature over a 60 s period daily for 10–14 d. We averaged the salinity and temperature for each 60 s measurement and then averaged these values for a grand mean of

temperature and salinity for each deployment period

We quantified epiphyte load on shoal and manatee grass in both locations in June 2007 when epiphytes were abundant, as epiphytes can strongly influence seagrass growth and mortality (Burd and Dunton 2001, Duarte 2002, Lirman and Cropper 2003). Our methods consisted of taking 5 randomly sampled, 10 cm diameter core samples using a polyvinyl chloride (PVC) corer (Johnson and Heck 2006) from each seagrass type. Within each core sample, 3 seagrass shoots were randomly selected for epiphyte quantification. The blade surface area was standardized by only using the oldest 10 cm of growth (top of the grass) with no obvious signs of grazing or other damage. We then carefully scraped off the epiphytes with a scalpel and transferred them to pre-weighed (0.001 g) Whatman GF/C filter paper. The filter paper and epiphytes were dried in a convection oven and weighed (0.001 g), and the total weight was subtracted from the original filter paper weight to quantify the epiphyte load. The epiphyte weight of the 3 blades was averaged to produce one epiphyte value per core sample. This produced 5 samples of epiphyte weight per location for both shoal and manatee grass.

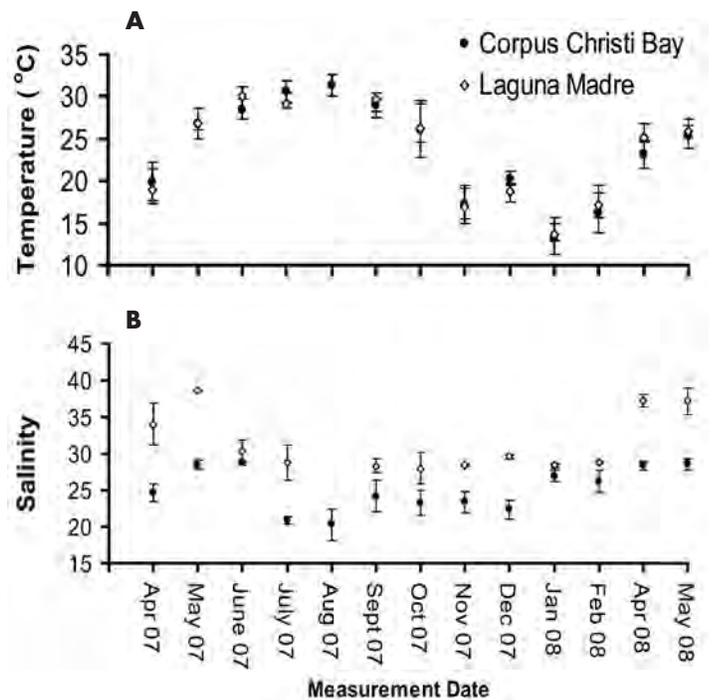
We sampled seagrass growth characteristics during consecutive peak growing seasons from March 2007 through June 2008. Sampling was conducted every 10–28 d ( $n = 27$  dates) depending on the season and weather conditions. We measured shoot density, root:shoot ratio (RSR), and primary growth rate in each location to determine the annual mean primary productivity patterns for each seagrass species. We also measured the density of reproductive shoots produced by manatee grass in each location. Daily variation in temperature and salinity are less likely to influence seagrass growth patterns than longer term differences (Dunton 1990, 1994), thus we pooled our data over date to focus specifically on comparing only seagrass growth patterns.

To measure primary growth, we used the clipping technique of Virnstein (1982); however, due to turbidity, we “harvested” the samples rather than photographing growth (Dunton 1990). This technique consisted of haphazardly selecting and trimming 0.25 m<sup>2</sup> plots ( $n = 3$ ) of shoal grass and 0.25 m<sup>2</sup> plots ( $n = 3$ ) of manatee grass in each location on each sampling date. Cuts were made 1.0 cm above the basal sheath for shoal grass, which allowed us to sample continuous growth (Dunton 1994). For manatee grass, cuts were made 6.0–7.0 cm above the basal sheath as preliminary results indicate that clipping below this height resulted in blade death. On each sampling date, a 10.0 cm diameter core sample, ~10.0 cm deep, was taken from each newly clipped plot as well as from each plot that had been clipped on the previous sampling date. Ten blades from the sample of the newly clipped plot were measured and averaged to determine the mean cut length at time 0 (i.e., mean blade length above substrate after clipping). The length of every blade was measured from the second core, which was taken 10–28 d after clipping. The mean cut length calculated im-

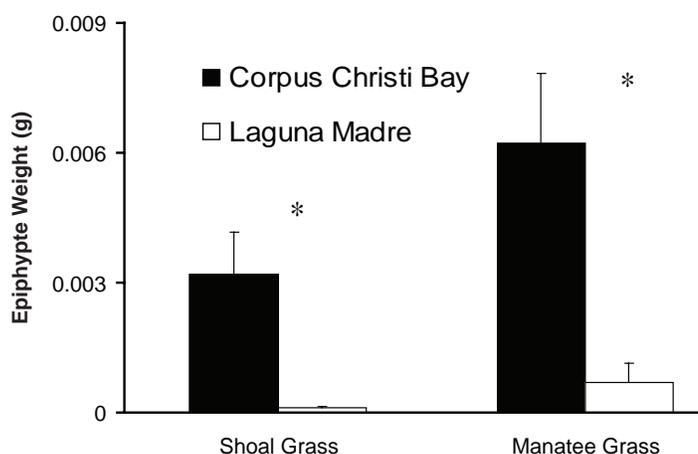
mediately after clipping was subtracted from the mean total length in the second core to determine the amount of growth in each of the 3 plots sampled. Growth rate (cm/d) was calculated by dividing the mean growth rate by the number of days between clipping and harvesting. The growth rates calculated for each core sample were averaged to calculate the grand mean growth rates for each grass by location and date.

We also measured shoot density (#/m<sup>2</sup>) by collecting 78.5 cm<sup>2</sup> core samples ( $n = 3$ ) from an area outside our clipped plot, counting the shoots in each core, and multiplying by 127.4 to convert the value to m<sup>2</sup>. We pooled our density measurements from each core sample to calculate a mean density for each species by location and then averaged these mean density measures by date ( $n = 27$ ) to calculate a grand mean for shoal and manatee grass in each location

To determine RSR, aboveground biomass (blades, g) and underground biomass (roots and rhizomes, g) were measured from shoal grass and manatee grass beds in each location during each sampling date ( $n = 27$ ). Three random core samples (78.5 cm<sup>2</sup>, 10.0 cm diameter) were taken to a depth of ~15.0 cm to ensure the collection of all root and rhizome structures. We haphazardly removed 10 blades from each core sample that had rhizomes attached, divided them into above and below ground sections, and scraped off any epiphytic material from the above ground portion. Above and below ground sections were dried separately in a convection oven at 60°C for 96 h and individually weighed from each



**Figure 1.** Monthly water quality measurements in each location (bay) during the study. A. Temperature (mean  $\pm$  sd). B. Salinity (mean  $\pm$  sd). The variability within each measurement period was low and using se resulted in the error bars being obscured by the data points.



**Figure 2.** Plot of epiphyte weight (g; mean + se) on shoal and manatee grass ( $n = 5$ ) from both locations (bays). \* - significant difference between locations.

core sample to calculate the RSR for each blade. Mean RSRs were calculated by species in each location by date ( $n = 27$ ). A grand mean RSR was calculated for each seagrass species in each bay by averaging the RSR values from all sampling dates.

We also counted the number of reproductive shoots in the RSR core samples collected in manatee grass beds on 6 sampling dates between March and May in 2007 and 5 sampling dates between March and May 2008. This was done because a high number of reproductive shoots may suggest lateral growth that would not be apparent from a 'clip and harvest' measurement. Shoal grass reproductive shoots were not observed during the study. As with shoot density, we multiplied the number of reproductive shoots by 127.4 to convert this value to number per  $m^2$ . Since we took 3 core samples on each sampling date, the number of reproductive shoots in each core sample was averaged.

#### Data analysis

We compared the grand mean of temperature and salinity between CCB and LM with a Student  $t$ -test (Sokal and Rohlf 1995). We then compared grand mean density, grand mean growth rates and grand mean RSR for each seagrass species between locations with a Student  $t$ -test (Sokal and Rohlf 1995). We compared the mean number of shoots pooled by date ( $n = 6$  in 2007 and  $n = 5$  in 2008) between locations by year with separate Mann-Whitney U tests because our data did not meet  $t$ -test assumptions (Sokal and Rohlf 1995).

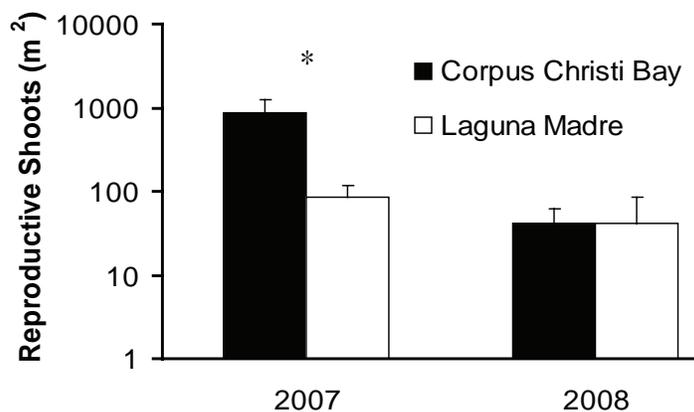
## RESULTS AND DISCUSSION

Temporal measurements of water temperature and salinity are presented in Figure 1 to illustrate seasonal trends. However, statistical analysis was performed only on the grand mean values ( $n = 13$ ) between locations. Water temperature ranged from 13.7°–29.9°C and was not statistically different between the LM and CCB ( $t = 0.18$ ,  $p = 0.85$ , Figure 1A). Salinity was significantly higher in LM ( $t = 4.46$ ,  $p < 0.01$ , Figure 1B). The mean salinity in the

LM was 34.0 and ranged between 28.0–38.5 as compared to CCB with a mean salinity of 24.7 (range 20.3–28.8).

Epiphyte weight on shoal grass was significantly (32x) greater in CCB than in LM ( $t = 3.18$ ,  $p < 0.05$ ,  $n = 5$ , Figure 2). Similarly, the epiphyte weight recovered from manatee grass was significantly (9x) greater in CCB than in LM ( $t = 3.30$ ,  $p < 0.05$ ,  $n = 5$ , Figure 2). Previous research revealed that nutrient inputs are much greater in CCB than in LM (Quammen and Onuf 1993, Lee and Dunton 2000) and higher ambient nutrient levels are most likely responsible for the greater epiphyte weight measured in this study.

Shoal grass shoot density was significantly higher and more variable in the LM ( $t = 2.94$ ,  $p < 0.01$ , Table 1). Shoal grass shoot density peaked in LM in March 2008, reaching a density of ~7000 shoots/ $m^2$ . In CCB, shoal grass density peaked at ~3600 shoots/ $m^2$  in May of 2008. Shoot density was lowest in LM with ~470 shoots/ $m^2$  in October 2007 and CCB had its lowest density of ~850 shoots/ $m^2$  in November of 2007. Manatee grass shoot density was also greater and more variable in LM (Table 1). A  $t$ -test revealed statistical differences between the shoot densities of manatee grass between locations ( $t = 3.18$ ,  $p < 0.01$ , Table 1). The LM reached its peak density of ~4000 shoots/ $m^2$  in May 2007 and CCB density peaked at ~2100 shoots/ $m^2$  in June 2007. The lowest density observed was ~420 shoots/ $m^2$  in CCB in October 2007 and ~540 shoots/ $m^2$  in LM in November 2007.



**Figure 3.** Plot of number of reproductive shoots (mean + se) produced by manatee grass from March to May 2007 ( $n = 6$ ) and March to May 2008 ( $n = 5$ ) in Corpus Christi Bay and the Laguna Madre. \* - significant difference between locations.

Root:shoot ratios determine seasonal differences between the aboveground and belowground biomass fractions of seagrass, reflecting seagrass energy allocation (Dunton 1994, 1996). Higher ratios occur during the winter season when plants are dormant and are allocating more energy into roots and other below ground structures, but ratios decrease when energy is allocated toward above ground growth in the spring and summer. The RSR ratios ranged from 1.05–4.9 in shoal grass and 0.44–2.56 in manatee

**TABLE 1.** Grand mean growth characteristics  $\pm$  se (range of values in parentheses) of shoal and manatee grass in two Texas bays. Grand means were calculated from the means of 27 sampling events of monospecific stands of shoal grass and manatee grass in Corpus Christi Bay, TX and the Laguna Madre Texas. \* - significant difference between locations.

Growth Parameter	Shoal Grass		Manatee Grass	
	Corpus Christi Bay	Laguna Madre	Corpus Christi Bay	Laguna Madre
Shoot Density (number/m <sup>2</sup> )	2274 $\pm$ 439 (854-3605)	3347 $\pm$ 555 (471-6969)*	1254 $\pm$ 281(420-2128)	1774 $\pm$ (548-3965)*
Root:Shoot Ratio (RSR)	2.25 $\pm$ 0.37 (1.05-4.90)	2.13 $\pm$ 0.23 (1.08-4.2)	1.17 $\pm$ 0.19 (0.44-2.56)	1.28 $\pm$ 0.21 (0.49-2.13)
Growth Rate (cm/day)	0.41 $\pm$ 0.06 (0.07-0.80)*	0.27 $\pm$ 0.05 (0.07-0.87)	0.52 $\pm$ 0.14 (0.060-01.2)	0.41 $\pm$ 0.11 (0.04-1.1)

grass (Table 1). Ratios were not significantly different between CCB and LM for either shoal grass ( $t = 0.05$ ,  $p = 0.61$ , Table 1) or manatee grass ( $t = 0.93$ ,  $p = 0.35$ , Table 1).

Shoal grass grew significantly faster in CCB ( $t = 2.68$ ,  $p < 0.05$ , Table 1). Annual mean growth rates for shoal grass were 0.41 cm/d and 0.27 cm/d in CCB and LM, respectively. In both locations, the period of slowest growth occurred in January 2008 and was calculated in both at 0.07 cm/d. Shoal grass peak growth of 0.87 cm/d occurred in LM in September 2007, while growth peaked at 0.81 cm/d in CCB in July 2007.

Manatee grass grew faster in CCB with an annual mean rate of 0.52 cm/d as compared to 0.43 cm/d in LM, but these rates were not statistically different ( $t = 1.13$ ,  $p = 0.23$ , Table 1). Peak growth rate of 1.11 cm/d occurred in LM in June 2007, while the least growth of 0.05 cm/d occurred in March 2007. Peak growth rates of 1.20 cm/d occurred in CCB in August 2007, while the slowest growth in CCB was 0.06 cm/d in January 2008.

Shoal grass reproductive shoots were not observed during the study. Manatee grass produced a significantly higher number of reproductive shoots in LM ( $\sim 875$  reproductive shoots/m<sup>2</sup>) as compared to CCB ( $\sim 85$  reproductive shoots/m<sup>2</sup>,  $z = 2.16$ ,  $p < 0.05$ , Figure 3) from March to May 2007. Numbers of reproductive shoots were not significantly different from March to May 2008 ( $z = 0.63$ ,  $p = 0.73$ , Figure 4) with  $\sim 41$  shoots/m<sup>2</sup> at both locations.

Because manatee grass has historically been much less common than other seagrasses in Texas (Quammen and Onuf 1993), its seasonal growth patterns in the field have not been carefully studied in this region. Our study provides the first documentation of manatee grass growth and energy allocation patterns in the western GOM. Our estimates of shoal grass growth and RSR ratios are consistent with earlier measurements made by Dunton (1990, 1994, 1996), suggesting that our technique provided an appro-

priate assessment of primary production of both species.

Our study locations are exhibiting different patterns of seagrass succession. Seagrass succession in the LM is following the traditional model proposed by Zieman (1982) where shoal grass, the pioneer species, is replaced by manatee grass and finally by turtle grass, the climax community (Quammen and Onuf 1993, Pulich and Onuf 2007). In contrast, all three seagrasses have coexisted for the past 20 yr in CCB without an obvious loss in overall coverage of any one species (Czerny and Dunton 1995, K. Dunton pers. comm.).

Fluctuations in salinity can be stressful to seagrasses and slow or stop succession so that multiple species coexist (Montague and Ley 1993). Salinity fluctuations in CCB, coupled with higher epiphyte loads, may act like moderate disturbances, thus stalling seagrass succession and promoting coexistence of these species. Conversely, the more constant salinity levels and lower epiphyte loads in LM are allowing succession to proceed with manatee grass slowly replacing shoal grass. Both seagrasses grew faster in CCB and had higher epiphyte loads, which were likely caused by greater nutrient inputs at this location.

We observed higher shoot density in both seagrasses as well as greater energy allocation to lateral growth and reproductive structures by manatee grass in LM, suggesting that conditions in this location are more favorable for seagrasses than in CCB. Should seagrass succession proceed in LM, a significant change in species composition in this important and unique ecosystem will likely occur. We must continue to monitor changes in seagrass composition in Texas to better understand potential consequences of species replacement. Since seagrass composition strongly influences community structure (Tolan et al. 1997, Micheli et al. 2008), it is necessary to understand the mechanisms driving seagrass change especially with the current decline in seagrasses worldwide. This study provides important baseline information to begin this process.

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