12-9-2014

Recruitment Patterns of the Fouling Community on Eelgrass in Long Island Sound

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B.A., Albion College, 2006

A Thesis

Submitted in Partial Fulfillment of the

Requirements for the Degree of

Master of Science

At the

University of Connecticut

2014

APPROVAL PAGE

Master of Science Thesis

Recruitment Patterns of the Fouling Community on Eelgrass

in Long Island Sound

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2014

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Abstract

The seagrass *Zostera marina* (eelgrass) is typically found in shallow protected areas of portions of eastern Long Island Sound (LIS). Although the species and its associated ecosystem have been well studied in many places, work in LIS has been limited. A field study was conducted to assess the fouling communities associated with eelgrass beds around Pine Island, in eastern LIS. Samples were collected from four sites between July and October, 2011. A total of 15 species of fouling organisms were found on eelgrass leaves throughout the study period. Recruitment patterns varied spatially and temporally. Differences in the composition of fouling invertebrates were rarely observed between inner and outer leaves of eelgrass shoots. In general, the abundances of invertebrates increased along the length of leaves, but not significantly. Rafting leaves had slightly higher abundances of fouling species than intact leaves and more species recruited on PVC panels than on eelgrass leaves. While eelgrass leaves are seasonally present in LIS, they are an important substrate for a variety of fouling species. Rafting leaves encrusted with fouling organisms may also be an important dispersal mechanism for these species.

Introduction

The diverse and complex ecosystem created by seagrass beds supports a wide array of animals, such as snails, crabs, fish, birds, turtles and dugongs (Williams & Heck, 2000). Many animals are not permanent residents but spend part of their life cycle in seagrass beds, including commercially important species. Seagrass provides an ideal "nursery ground" to protect eggs and young vertebrates and invertebrates from predation in open water, and an abundance of food (Orth et al., 2006).

The health of seagrass beds can be an indicator of the health of a marine habitat (Gaeckle et al., 2011). Seagrass has been dubbed a "coastal canary" (Orth et al., 2006) due to its rapid and measurable responses to changing environmental conditions. For example, eutrophication can cause algae blooms which in turn reduce light penetration into the water column. This light reduction results in declining health of seagrass and may as such be a proxy for overall water quality. Recently, seagrass has been used as an indicator in regulation of industry and manufacturing. Rather than set regulations on industrial discharge by the amount of pollutant per liter, a particular amount of healthy seagrass beds in the vicinity of discharge can be required and used to indicate good water quality (Orth et al., 2006). These seagrass beds require suitable conditions that depend on physical factors (e.g. water depth, tides, current velocity, and wave height), water quality parameters (e.g., temperature, salinity, dissolved oxygen) and benthic features (sediment type, percentage organics and sulfides) (Vaudrey, 2008). Changes in any of these conditions, due to anthropogenic causes such as industrial waste and farming run-off, affect shallow-water seagrass.

In temperate regions of North America, the most prominent and productive temperate seagrass species is *Zostera marina* (Webster et al., 1998, Williams & Heck, 2001)--commonly called eelgrass. It is a flowering, subtidal, soft-sediment vascular plant found primarily in bays and estuaries (Williams & Heck, 2001) and can grow to a height ranging from a few centimeters to approximately four meters. Eelgrass can co-exist with other marine plant life such as macroalgae and other species of seagrasses, which can contribute significantly to overall primary production. Eelgrass is generally the dominant primary producer of its ecosystem (Williams & Heck, 2001).

Besides providing habitat to organisms that live among the seagrass, these eelgrass beds also provide substrate to a diverse group of organisms that live on the leaves of the grass itself. Photosynthetic organisms that grow directly on the eelgrass leaves are known as epiphytes and include diatoms, dinoflagellates, and microalgae. Epiphytes make up a valuable part of the eelgrass bed ecosystem by contributing to the primary production of the system and providing food for grazers such as snails (Williams & Heck, 2001). In addition, a variety of encrusting invertebrates (e.g. polychaetes, ascidians, bryozoans, sponges), collectively known as fouling organisms, also use eelgrass leaves as substrate for attachment (Williams, 2007).

While contributing to the greater ecosystem productivity, a heavy load of epiphytes can be detrimental to eelgrass. Epiphytes can obstruct the plants' access to sunlight causing atrophy of the encrusted leaves (Reusch & Williams, 1998; Williams, 2007). The availability of adequate quantities of light is often the most important factor in the growth of eelgrass (Dennison, 1987). In situ, ~22% of the surface light measured must penetrate to the eelgrass canopy in order to sustain eelgrass photosynthesis (Vaudrey, 2008). This light level value is a measurement of water clarity but does not take into account factors such as epiphytes that influence the amount of light that reaches the eelgrass leaves. In a healthy eelgrass bed, the biomass of epiphytes in an eelgrass bed can be up to 36% of the total biomass (Vaudrey, 2008). Increased temperature can stimulate the growth of phytoplankton and photosynthetic epiphytes reducing the amount of light reaching the eelgrass. The presence of grazers that feed on the epiphytes, such as herbivorous snails, helps keep down the epiphyte load (Nelson, 1997, Orth & Van Montfrans, 1984). When both water column and leaf surface features are taken into account, the minimum light requirement of eelgrass can be as low as 13-15% (Vaudrey, 2008).

In addition to stimulating photosynthetic epiphytes, temperature can also facilitate invasion of fouling invertebrates (Stachowicz et al., 2002). For example, a tropical anemone (*Bunodeopsis* sp) introduced to temperate California experiences a population boom in the summer, which has been attributed to ocean warming. Native eelgrass in the zone is negatively effected by the massive load of the anemone population settling on the leaves, blocking the light and collapsing the eelgrass canopy (Williams & Heck, 2001).

Recent work by Carman & Grunden (2010) investigated invasive tunicates attached to eelgrass in Lake Tashmoo, Martha's Vineyard, Massachusetts. Native *Molgula* and invasive *Botrylloides, Botryllus, Didemnum, Styela* and *Diplosoma* were present on stalks, leaves and floating leaves. Although the ecological effects of such tunicates have not been assessed, their effects could be more harmful to marine plant communities than other fouling species, such as the lighter encrusting bryozoans, because of the cumulative weight of their biomass and rapid reproduction (Carman & Grunden, 2010).

Extensive work has been done on the LIS fouling communities inhabiting rocks, pilings, PVC panels, and other man-made surfaces (Bullard et al., 2007, Osman et al., 2010, Osman & Whitlatch 1995, 1996, 1998, 2004, 2007, others). This body of work has documented distinct patterns in recruitment of a variety of fouling species at different sites and at different times. Fouling organisms have been observed growing on eelgrass leaves in LIS (Whitlatch, Vaudrey, personal communication), although no previous work has been done to assess the abundance or patterns of recruitment.

LIS is known to have almost 2,000 acres of eelgrass in more than 170 beds (Tiner et al., 2009). Three of the largest beds are found in Quiambog Cove, Little Narragansett Bay, and Fisher's Island (407, 343 and 345 acres, respectively). In the 1930s the majority of the eelgrass beds were wiped out by an "eelgrass wasting disease," since attributed to the slime mold *Labyrinthula zosterae* (Muehstein et al., 1991). Portions of central and eastern LIS have experienced re-growth, but eelgrass beds never recovered in the western end of the sound (Tiner et al., 2009).

A few eelgrass sites in LIS have been extensively studied (Vaudrey, 2008). This work has focused on light, nutrients, sediment type and quality. The goal of these studies has been to identify areas suitable for transplanting eelgrass. The plan is to re-establish disappearing beds and eventually re-introduce *Z. marina* to western LIS. In identifying habitats suitable for the introduction of eelgrass, factors that reduce the amount of light reaching the eelgrass must be addressed, including turbidity, water quality, phytoplankton biomass, and epiphyte load.

Eelgrass has been well studied from a botanical viewpoint. However, relatively little is known about the associated communities of animals. Studies of populations within eelgrass beds tend to focus on a single species, rather than looking at the entire community (Williams & Heck, 2001). Studies on invasive species in eelgrass beds are even more limited. Williams (2007) presented 56 introduced species present in eelgrass habitats including 17 invertebrate fouling species. Few of these species have been further investigated.

Eelgrass leaves that are no longer attached to the shoot float on the surface of the water. These "rafting" eelgrass leaves have been shown to serve as a dispersal mechanism for fouling organisms (Carman & Grunden 2010, Petersen & Svane, 1995, Worcester, 1994). The species observed on eelgrass leaves by Carman & Grunden (2010) are also abundant in eastern LIS (Osman & Whitlatch 1995, 2007). If these fouling organisms are found to be rafting on eelgrass in LIS, it's possible that the eelgrass leaves are being used as a dispersal mechanism in LIS --as was shown to be the case elsewhere (Carman & Grunden 2010, Petersen & Svane, 1995, Worcester, 1994).

Little is known about the invertebrates that inhabit the eelgrass of LIS. Few publications about eelgrass are specific to LIS and none focus on the fauna therein. The aim of this study was to assess the colonization of eelgrass by fouling organisms in eelgrass beds around Pine Island, in eastern LIS.

Present Study

The aim of the study is to assess the invertebrate fouling community in four eelgrass beds in eastern LIS. Five hypotheses guided this testing.

Hypothesis 1: Due to differences in the sampling sites, fouling species' composition and abundances will vary spatially and temporally across sampling sites throughout the recruitment season.

Fouling communities inhabiting eastern LIS have been well documented (Osman et al., 2010, Bullard et al., 2007, Stachowicz et al., 2002, Fell & Lewandrowski, 1981, others). For example, an ongoing study of the fouling community of LIS has been conducted by Whitlatch et al. since 1991(henceforth referred to as LTREB[1]). Weekly or biweekly (depending on the season), four 10cm x 10cm PVC panels are deployed face-down, at several different sites in eastern LIS. The organisms recruited on the panels are counted and identified under dissecting microscopes. Results from this study indicate that the organism abundances and assemblages that settle on the PVC panels vary seasonally and by location (Freestone et al., 2009, Osman & Whitlatch, 1995). The peak larval settling time differs between species causing a continuous shift in dominance.

As shown in figure 1, the four study sites are in close proximity but separated enough to have slightly varying conditions, such as exposure to water flow and waves (personal observations). The spatial variability in recruitment shown by the LTREB data (Osman & Whitlatch, 1995) suggests that variability should also be observed in fouling recruitment on eelgrass leaves.

Table 1 reports the common shallow water invertebrates of the southern New England fouling community identified by Osman et al. (2010). Based on their presence in the local area,

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[1] Formerly funded through the National Science Foundation's LTREB program (Long Term Research in Environmental Biology)

these species were predicted to be found on eelgrass leaves at the four study sites. The present study was conducted from July 5th through October 13th, 2011, covering the peak recruitment times for the identified species (Table 1).

Hypothesis 2: Greater abundances of fouling species will be seen on outer leaves of eelgrass shoots compared to inner leaves because outer leaves have been exposed to recruitment longer than inner leaves.

Eelgrass is a rooted, vascular monocotyledonous, flowering plant. When producing seeds through sexual reproduction, the terminal shoot is reproductive. Eelgrass also reproduces asexually with lateral shoots arising from the rhizome. Individual shoots consist of a sheath bundle containing multiple leaves. The leaves on the outside of the shoot were the first leaves to have grown. Younger leaves grow up from the center of the shoot. This creates a sequence of older to younger leaves from the outside inward. As new leaves grow, the older leaves on the outside are sloughed off, so inner leaves become outer leaves in a continuous progression, as shown in Figure 2. As outer leaves are older, they have been exposed to fouling invertebrate recruitment for a longer period of time than the younger, inner leaves of the shoot. It is therefore hypothesized that the outer leaves will have higher levels of recruitment than the inner leaves which have been exposed to recruitment for a shorter amount of time.

Hypothesis 3: Due to longer time of exposure to recruitment, abundances of fouling organisms will vary along the length of individual leaves.

Eelgrass grows from the base of the leaf. The tip of the leaf is the first part to emerge from the sheath and is the oldest part of the leaf. The base of the leaf is the youngest section, having most recently emerged from the sheath (Figure 2). Individual leaves are increasingly older from base to tip and therefore have been

Figure 1. Study sites A. Beach, B. Bushy C. Jetty and D. Outside (maps.google.com).

Table 1. Common sessile, shallow water, invertebrate fouling species of southern New England (Osman et al., 2010)

Species	Solitary/ colonial	Recruitment Range	Recruitment Peak
Sponges			
Halichondria bowerbanki	C	May–Dec	Jun, Sep
Hydroids			
Tubularia larynx	C	–	–
Obelia sp.	C	May–Oct	May, Sep
Polychaetes			
Spirorbis spp.	S	May–Dec	Sep
Hydroides dianthus	S	May–Oct	Sep
Barnacles			
Balanus spp.	S	May–Nov	Aug
Molluscs			
Mytilus spp.	S	Jun–Sep	Jun
Bryozoans			
Cryptosula pallasiana	C	May–Dec	Jul–Sep
Schizoporella errata	C	Jun–Nov	Jul
Microporella ciliata	C	Jun–Oct	–
Bugula turrita	C	Jun–Nov	Aug–Sep
Bowerbankia gracilis	C	Jun–Oct	Jul–Aug
Membranipora membranacea	C	Jul–Oct	Sep
Ascidians			
Botryllus schlosseri	C	May–Nov	Aug
Botrylloides violaceus	C	Jun–Dec	Jul–Sep
Diplosoma listerianum	C	Jun–Dec	Sep
Molgula manhattensis	S	Jun–Oct	Aug–Sep
Ascidiella aspersa	S	Jun–Dec	Jul–Oct
Ciona intestinalis	S	May–Oct	Jul–Aug
Styela clava	S	Jun–Oct	Jul–Oct

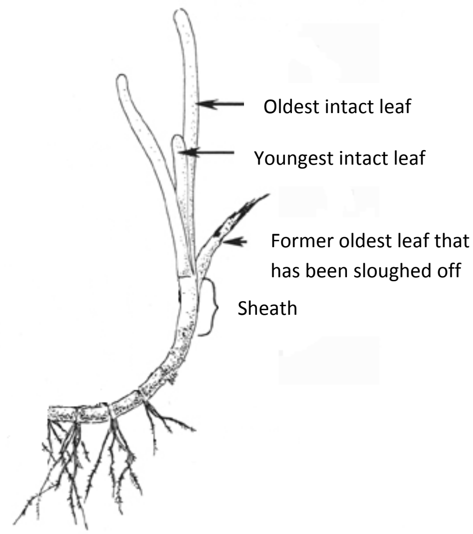

Figure 2. Diagram of an eelgrass shoot. As older, outer leaves are sloughed off, younger, inner leaves become outer leaves. Drawing by Mark Fonseca (Short & Coles, 2001).

exposed longer to fouling species. Having been exposed to recruitment for longer, the tips of individual leaves are predicted to have higher abundances of recruitment than lower sections.

Hypothesis 4: Rafting eelgrass leaves will have greater abundances of fouling invertebrates than intact eelgrass leaves because they have been exposed to recruitment for longer than intact leaves.

Most rafting leaves are those which have been sloughed off from the outside of the shoot (Short & Coles, 2001). These are the oldest leaves which have been exposed to recruitment for the longest periods while intact, followed by more exposure to recruitment while floating at the surface. This increased exposure to recruitment is predicted to result in a greater abundance of fouling invertebrates when compared to intact leaves.

Hypothesis 5: Fouling organism assemblages on eelgrass leaves will be similar to recruitment patterns seen on PVC panels.

Previous work with the fouling community indicates settling organisms do not discriminate between experimental PVC panels and other available hard substrates (Stachowicz, et al., 2002). Colonization abundances and species composition on eelgrass leaves are predicted to be similar to those found on PVC panels.

Materials and Methods

A. <u>Study sites</u>

Eelgrass was sampled from four eelgrass beds found around Pine Island, LIS, off the coast of Avery Point, Groton, CT (Figure 1, Table 2). Site A (hence referred to as "Beach") is located just off a sandy beach on the north side of Pine Island. It contains a patchy eelgrass bed that is found in 1-2 meters of water, depending on the tide. The Beach site is protected from high winds and waves allowing for calmer water than sites located on the eastern side of Pine Island. This site can thus be described as well protected, because the island serves as a barrier from the open water of LIS. Site B (hence referred to as "Bushy") is just of off Bushy Point. Eelgrass at this site is also patchy and is located in 3-4 meters of water, adjacent to Bushy Rock. Site C (further referred to as “Jetty”) is located in a well-protected area adjacent to a stone jetty on the northwestern point of the island. It is a patchy bed 2-3 meters below the surface. Site D (further referred to as “Outside”) is on the outside (south side) of Pine Island. It hosts a denser, less patchy eelgrass bed than the other study sites, located in 3-4 meters of water. It is the most exposed, with little to no protection from prevailing wind and waves.

In general, the Bushy and Outside sites are slightly more exposed to prevailing winds and waves than the Beach and Jetty sites. The Jetty site is the most protected while generally the Outside site is most exposed. Hurricane Irene impacted all four sites (August 28th, 2011), with the most changes seen at the Beach and Bushy sites (personal observations). Eelgrass density was reduced and patchiness increased.

B. Eelgrass and fouling invertebrate sampling

Samples of both intact shoots and floating leaves were collected bi-weekly, from July 5 through October 13, 2011, by snorkeling and SCUBA diving. A minimum of five intact shoots were collected arbitrarily at each site from inside the eelgrass beds (with the exception on the first two collection dates, on which only three shoots were collected from each site). As many floating leaves as were observed were collected from each site, with a maximum of 20 individual leaves processed (Hypothesis 4: recruitment on rafting vs. intact leaves). Samples were transported immediately to the Rankin Seawater Laboratory, Avery Point, CT, and stored in unfiltered flow-through seawater until processing (1-3 days). Settled organisms attached to the leaves were identified using dissecting microscopes. For intact shoots, the position of each leaf in relation to other leaves of the shoot (outer or inner) was observed (Hypothesis 2: recruitment on inner leaves vs. outer leaves). Length of the leaves was measured from the top of the sheath to the tip. Percent of the leaf that was brown (desiccated/wasting disease) was estimated (rounded to 5%, not recorded if less than 5%). The observed number of individuals of each species was recorded for

Table 2. Geographical locations and characteristics of study sites

Site		**Beach**	**Bushy**	**Jetty**	**Outside**
Location		41°18'53"N 72°03'20"W (leeward side of Pine Island)	41°18'56"N 72°03'09"W (windward side of Pine Island)	41°18'47"N 72°03'36"W (leeward side of Pine Island)	41°18'50"N 72°03'16"W (windward side of Pine Island)
Depth (M)		1-2	3-4	2-3	3-4
Sediment size distribution	Silt & clay: very fine sand: fine sand: medium sand: coarse sand: very coarse sand: Granule:	<63µm 1.6% 63-125 µm 2.0% 125-297 µm 2.6% 298-500 µm 4.2% 500-707 µm 5.6% 707-841 µm 1.5% >841 µm 72.5%	<63µm 2.3% 63-125 µm 0.6% 125-297 µm 0.9% 298-500 µm 3.7% 500-707 µm 14.7% 707-841 µm 11.5% >841 µm 66.2%	<63µm 40.8% 63-125 µm 1.8% 125-297 µm 5.4% 298-500 µm 5.4% 500-707 µm 6.8% 707-841 µm 4.1% >841 µm 5.6%	<63µm 0.5% 63-125 µm 0.0% 125-297 µm 0.4% 298-707 µm 18.7% 707-841 µm 11.5% >841 µm 68.8%
Organic content of sediment		1.30%	2.20%	11.60%	40.00%

each eelgrass leaf. Data collected on August 2, 2011 have been excluded from the study due to errors in storage and processing.

Beginning on the August 16, 2011, intact leaves were divided into 20cm segments, beginning at the base of the leaf. The “first section” was defined as the first 20cm above the top of the sheath. Each subsequent 20cm section was a data point included in “middle.” The final section of the leaf was defined as “end.” Only leaves greater than 20cm long were included in divided analysis (Hypothesis 3: variation in recruitment along the length of the leaves). Leaves less than 40cm in length were defined to have a first and end sections. Leaves were pooled by position category (first, middle and end sections) for each site and collection date.

Species were combined into taxonomic groups for analysis: ascidians, barnacles, bryozoans, and polychaetes. Not included in data analysis were eggs and egg casings. Due to extremely low abundances, also excluded were entoprocts, cnidarians, hydroids and sponges. In a few instances, colonial organisms had developed into colonies such that individual recruits could not be determined. In such cases, percent cover was estimated and later converted to individual recruits by estimating that 10 individuals would equal 100% cover per cm of leaf, resulting in the following formula:

$$recruits\ (\#) = \frac{\%\ cover}{100} \cdot leaf\ length\ (cm) \cdot \frac{10\ recruits}{1\ cm\ of\ leaf\ length}$$

Average numbers of individuals were then divided by leaf length for analysis of Hypothesis 1, or leaf section for Hypothesis 2.

In order to test Hypothesis 5 (similarities of assemblages on PVC panels as found on eelgrass), 10cm x10cm PVC panels were deployed at each of the sampling sites twice during the study period. Four panels were attached to a length of PVC pipe that was suspended on a rope between a concrete weight and a subsurface buoy. Two of the panels were oriented with settling surfaces downward facing, similar to other studies (Bullard et al., 2004, Osman & Whitlatch, 1995, Stachowitcz et al., 2002, others) and two panels were oriented sideward facing, to imitate the orientation of intact eelgrass leaves in the water column. The devices were positioned within an eelgrass bed at each of the sampling sites so that the panels were among the eelgrass. The first set of panels were deployed for 23 days, August 16 to September 8, 2011, and the second set of panels were deployed for 21 days, September 22 to October 13, 2011. Panels deployed at the Outside site on August 16, 2011 were lost due to Hurricane Irene, which swept through the area on August 28, 2011. A subsequent set of panels was not deployed at this site. Once retrieved, organisms attached to panels were identified using dissecting microscopes.

C. Data Analysis

Data analysis was performed in Microsoft Excel and Minitab. One-way ANOVAs were performed to determine differences in abundances within and among groups. Significant differences were confirmed using paired t-tests or 2-sample t-tests. Significant difference is defined as p-values of 0.05 or less, and p-values higher than 0.05 are considered not significant.

Results

Table 3 reports that a total of 15 species were observed on collected eelgrass leaves over the course of the study period. The data were dominated by species which fell into four taxonomic groups: ascidians, barnacles, bryozoans, and polychaetes. Species were grouped by these general categories for analysis. Abundance of sponges and hydroids were extremely low and were therefore omitted from all data analysis.

Hypothesis 1

It was hypothesized that recruitment abundances would vary spatially and temporally. Species assemblages varied among sites and within sites (Figure 3). For certain sampling periods the Beach site was dominated by polychaetes, while at other times none of the groups were

Table 3. Species found on collected leaves at each site over the entire sampling period.

Species:	Beach	Bushy	Jetty	Outside
Ascidians				
Botrylloides violaceus	●	●	●	●
Botryllus schlosseri	●		●	
Molgula manhattensis			●	
Bryozoans				
Bowerbankia gracilis	●	●	●	●
Celleporaria hyalina	●	●	●	●
Crisia elongata			●	
Cryptosula pallasiana		●	●	
Electra pilosa	●	●	●	●
Electra crustulenta	●	●	●	●
Membranipora sp.	●		●	●
Microporella ciliata	●	●	●	●
Barnacles				
Balanus sp.	●	●	●	
Hydroids				
Obelia sp	●	●	●	●
Polychaetes				
Spirorbis sp	●	●	●	●
Sponges				
Halichondria sp	●	●	●	●

clearly dominant. The Bushy site had overall low recruitment abundances. The Jetty site was dominated by barnacles and polychaetes. The Outside site was dominated by bryozoans.

Recruitment Comparison by Site

Beach Site Taxonomic Group Comparison

At the Beach site there were significantly more polychaetes than ascidians, barnacles, and bryozoans on all collection dates from July 19, 2011, through September 15, 2011 (Figure 4, Table 4). The different taxonomic groups displayed peaks in recruitment abundances on different dates (Figure 5). The peak in polychaete and ascidian abundance was on August 16, 2011. Barnacles had maximum abundances on July 5, 2011 and September 1, 2011. Bryozoans showed a maximum peak on August 16, 2011 and a subsequent although lower peak on September 1, 2011.

Beach Site Species Comparison

The Beach site was primarily dominated by *Spirorbis* polychaetes throughout the sampling period (Table 5). Beginning on the September 29, 2011 collection date, the bryozoan, *B. gracilis* accounts for approximately 50% of recruitment. On October 13, 2011 *Spirorbis* polychaetes are no longer dominant. Instead, the two bryozoans, *B. Gracilis* and *E. pilosa* account for the majority of recruitment.

Bushy Site Taxonomic Group Comparison

Collections at the Bushy site showed significantly more polychaete and bryozoan recruitment than other taxa on July 5, 2011. Polychaete recruitment was significantly higher than other taxa on August 16, 2011 through September 15, 2011 (Figure 6, Table 6), but not on July 19, 2011. Bryozoan abundance peaked on July 5, 2011 and polychaete abundance peaked on September 1, 2011 (Figure 7). Ascidians and barnacles were in zero or near-zero abundances throughout the sampling period.

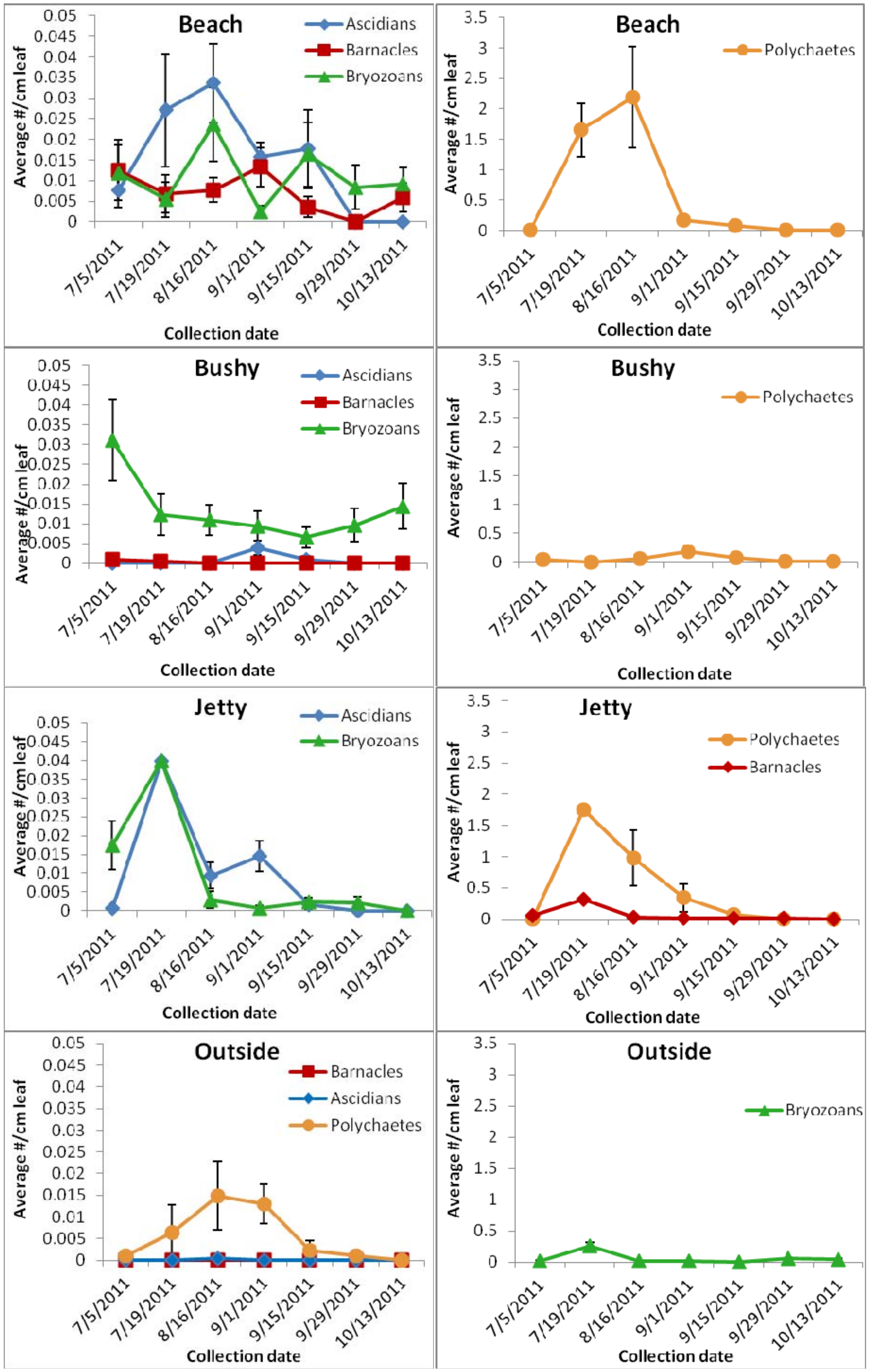

Figure 3. Species assemblages among sites. Left column are species in lower abundance (<0.05/cm leaf). Right column are species in higher abundances (≤2.2/cm leaf). Error bars are ±1 standard error

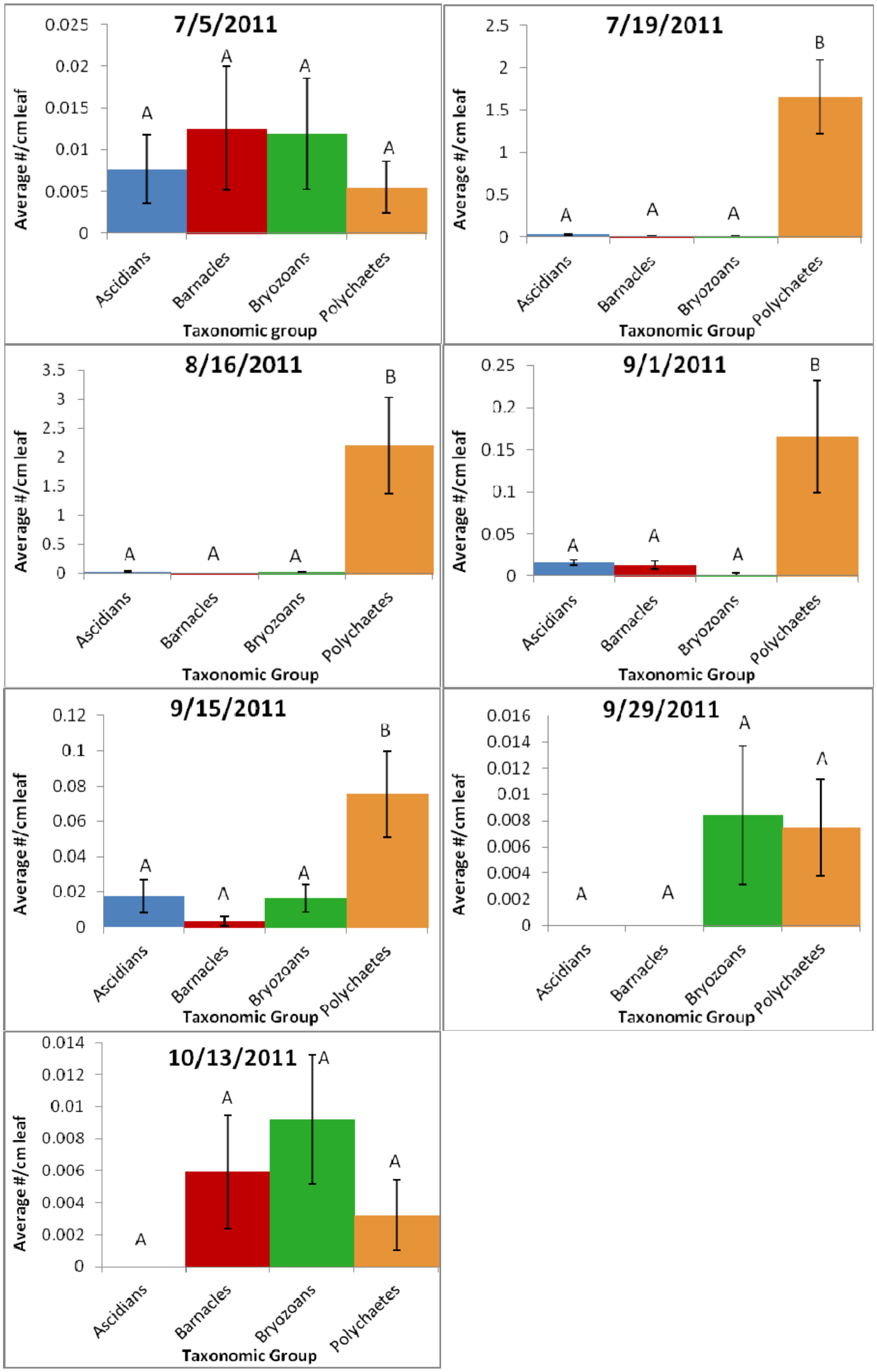

Figure 4. Recruitment abundance for all taxa at the Beach site on each sampling date. Letter indicate significant differences according to Tukey's Analysis of Variance Error bars are ±1 standard error. Note different scales on y-axes.

Table 4. Analysis of variance of ascidian, barnacle, bryozoan and polychaete recruitment abundances at the Beach site across the sampling period.

Taxonomic Group	Tukey's-test Letter	One-way ANOVA p-value
7/5/2011		
Ascidians	A	0.778
Barnacles	A	
Bryozoans	A	
Polychaetes	A	
7/19/2011		
Ascidians	B	<0.001
Barnacles	B	
Bryozoans	B	
Polychaetes	A	
8/16/2011		
Ascidians	B	<0.001
Barnacles	B	
Bryozoans	B	
Polychaetes	A	
9/1/2011		
Ascidians	B	0.002
Barnacles	B	
Bryozoans	B	
Polychaetes	A	
9/15/2011		
Ascidians	B	0.002
Barnacles	B	
Bryozoans	B	
Polychaetes	A	
9/29/2011		
Ascidians	A	0.116
Barnacles	A	
Bryozoans	A	
Polychaetes	A	
10/13/2011		
Ascidians	A	0.153
Barnacles	A	
Bryozoans	A	
Polychaetes	A	

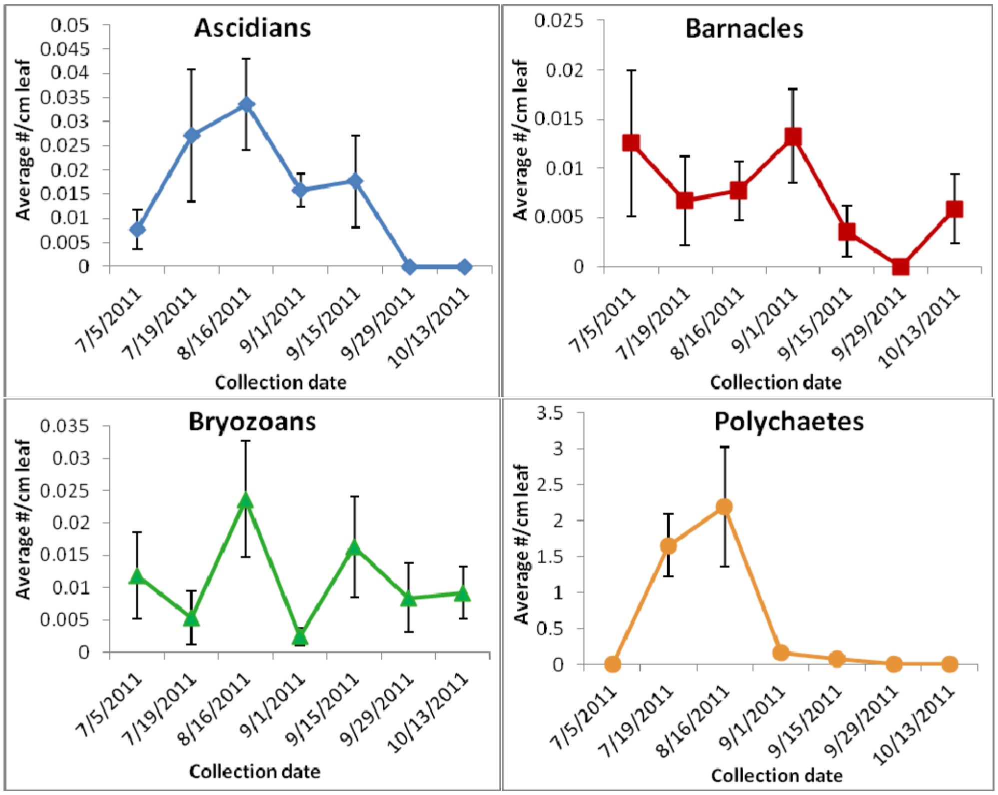

Figure 5. Average abundances of ascidians, barnacles, bryozoans and polychaetes per cm leaf at the Beach site. Error bars are ±1 standard error. Note different scales on y-axes.

Table 5. Fouling invertebrate species observed on eelgrass at the Beach site as fraction of total recruits on each collection date.

Species:	7/5/2011	7/19/2011	8/16/2011	9/1/2011	9/15/2011	9/29/2011	10/13/2011
Ascidians							
Botrylloides violaceus	0.203	0.002	0.012	0.080	0.156		
Botryllus schlosseri		0.014	0.002				
Bryozoans							
Bowerbankia gracilis			0.010	0.013	0.136	0.530	0.443
Celleporaria hyalina		0.002					
Electra pilosa	0.021						0.058
Electra crustulenta	0.182						
Microporella ciliata	0.114						
Barnacles							
Balanus sp.	0.334	0.004	0.003	0.067	0.032		0.322
Polychaetes							
Spirorbis sp.	0.147	0.977	0.971	0.840	0.667	0.470	0.177

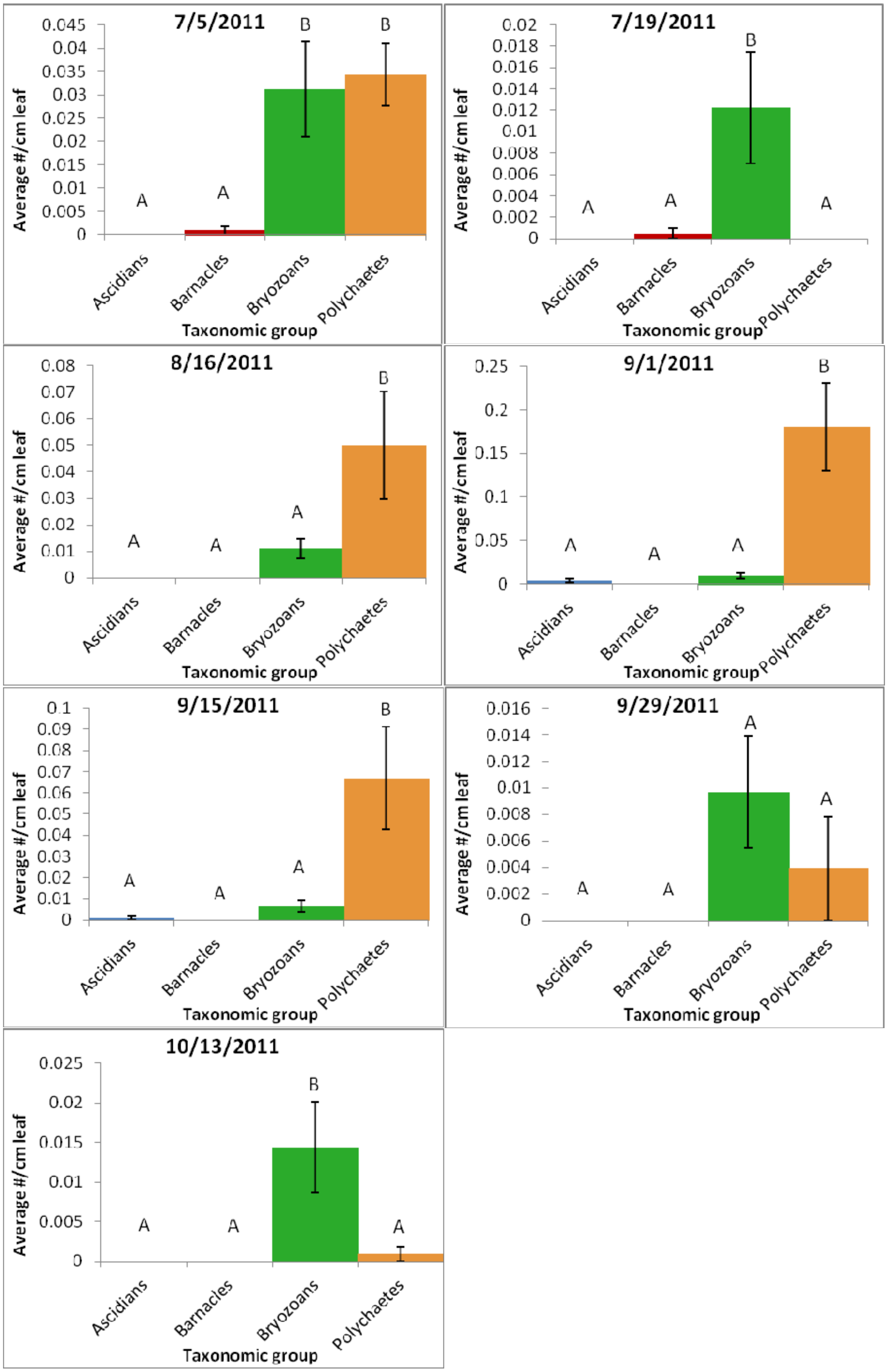

Figure 6. Recruitment abundance for all taxa at the Bushy site on each sampling date. Letter indicate significant differences according to Tukey's Analysis of Variance Error bars are ±1 standard error. Note different scales on y-axes.

Table 6. Analysis of variance of ascidian, barnacle, bryozoan and polychaete recruitment abundances at the Bushy site across the sampling period.

Taxonomic Group	Tukey's-test Letter	One-way ANOVA p-value
7/5/2011		
Ascidians	A	<0.001
Barnacles	A	
Bryozoans	B	
Polychaetes	B	
7/19/2011		
Ascidians	A	0.002
Barnacles	A	
Bryozoans	B	
Polychaetes	A	
8/16/2011		
Ascidians	A	0.003
Barnacles	A	
Bryozoans	A	
Polychaetes	B	
9/1/2011		
Ascidians	A	<0.001
Barnacles	A	
Bryozoans	A	
Polychaetes	B	
9/15/2011		
Ascidians	A	<0.001
Barnacles	A	
Bryozoans	A	
Polychaetes	B	
9/29/2011		
Ascidians	A	0.065
Barnacles	A	
Bryozoans	A	
Polychaetes	A	
10/13/2011		
Ascidians	A	0.001
Barnacles	A	
Bryozoans	B	
Polychaetes	A	

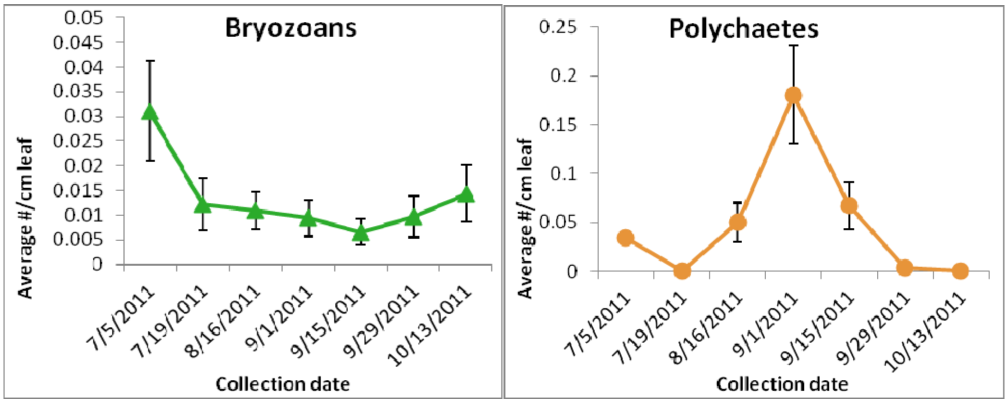

Figure 7. Average abundances of bryozoans and polychaetes per cm leaf at the Bushy site. Error bars are ±1 standard error. Note different scales on y-axes.

Table 7. Fouling invertebrate species observed on eelgrass at the Bushy site as fraction of total recruits on each collection date.

Species:	7/5/2011	7/19/2011	8/16/2011	9/1/2011	9/15/2011	9/29/2011	10/13/2011
Ascidians							
Botrylloides violaceus				0.020	0.012		
Bryozoans							
Bowerbankia gracilis			0.032				
Celleporaria hyalina	0.073	0.127	0.131	0.049	0.089	0.590	0.940
Cryptosula pallasiana						0.055	
Electra pilosa		0.837	0.018				
Electra crustulenta	0.054					0.084	
Microporella ciliata	0.343						
Barnacles							
Balanus sp.	0.013	0.036					
Polychaetes							
Spirorbis sp	0.517		0.820	0.931	0.900	0.273	0.060

Bushy Site Species Comparison

On the initial collection date, July 5, 2011, the Bushy site was dominated by the bryozoan, *M. ciliata* and *Spirorbis* polychaetes (Table 7). The second collection date, July 19, 2011 dominance shifted to the bryozoan, *E. pilosa.* Dominance then shifted again to *Spirorbis* polychaetes until the September 29, 2011, collection date at which point the bryozoan *C. hyalina* was dominant for the rest of the sampling period.

Jetty Site Taxonomic Group Comparison

Barnacles showed significantly higher abundances than other taxa on July 5, 2011 (Figure 8, Table 8) and higher than ascidians and bryozoans, but lower than polychaetes on July 19, 2011. Polychaete abundances were significantly higher than all taxa on starting July 19, 2011, through September 15, 2011. Ascidians, bryozoans, barnacles and polychaetes all showed peak abundances on July 19, 2011 (Figure 9).

Jetty Site Species Comparison

The Jetty site was initially dominated by *Balanus* barnacles, but by the second collection date dominance shifted to *Spirorbis* polychaetes (Table 9). Polychaetes dominated recruitment until the September 29, 2011 collection date, at which point barnacles were again dominant. The final collection date, October 13, 2011, only barnacles were observed to have recruited on collected leaves.

Outside Site Taxonomic Group Comparison

Bryozoans dominated the settlement at Outside (Figure 10, Table 10). There were significantly higher abundances of byrozoans than all other taxa on July 5, July 19, September 29 and October 13, 2011. On August 16, September 1 and September 15, 2011, there were significantly higher abundances of bryozoans than ascidians and barnacles, but not polychaetes. Bryozoan abundance peaked on September 19, 2011 (Figure 11). Polychaete abundance peaked on August 16, 2011, and were significantly higher than ascidians and barnacles on that date. All other taxa were in zero or near-zero abundances throughout the sampling period.

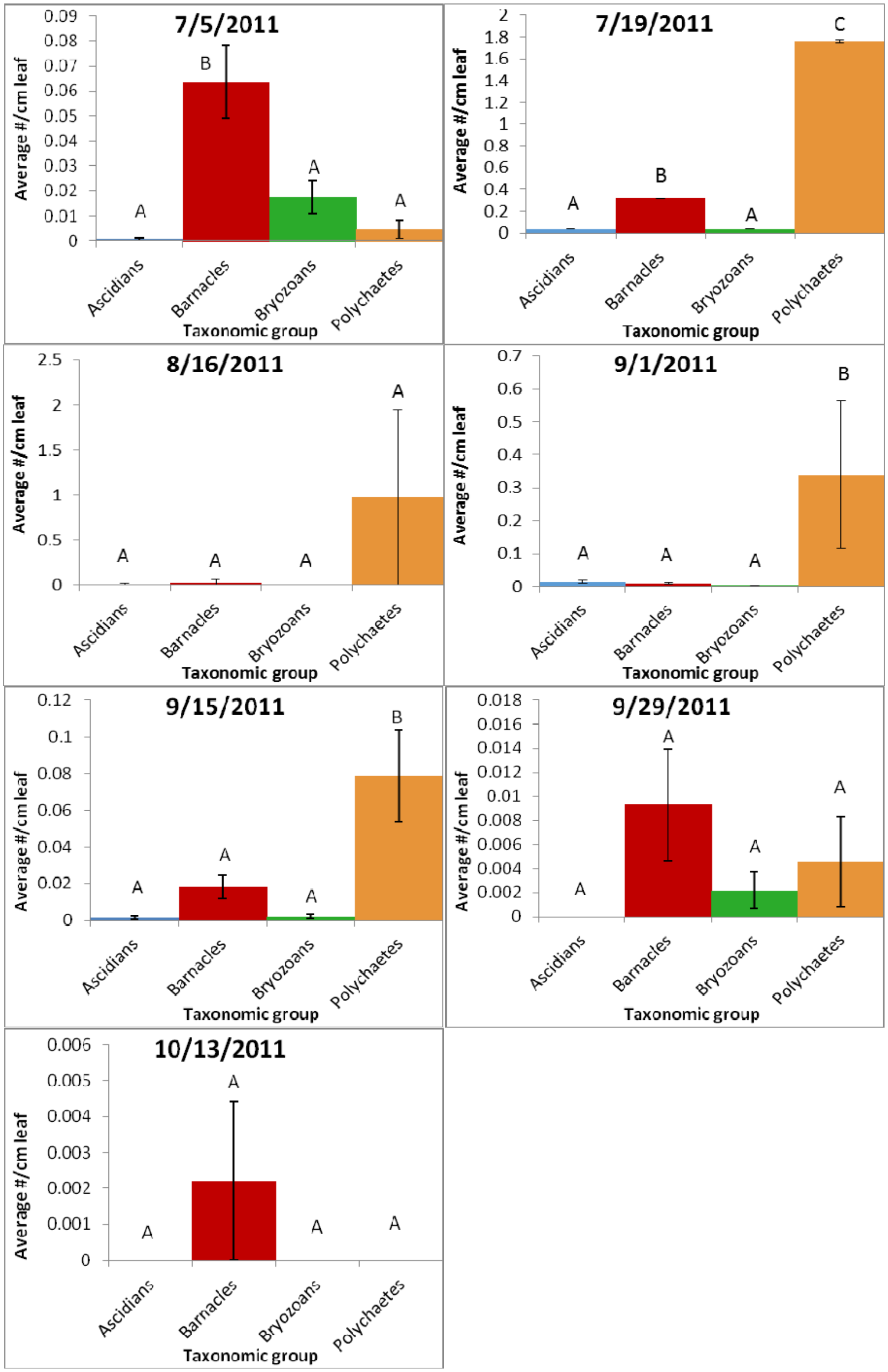

Figure 8. Recruitment abundance for all taxa at the Jetty site on each sampling date. Letter indicate significant differences according to Tukey's Analysis of Variance Error bars are ±1 standard error. Note different scales on y-axes.

Table 8. Analysis of variance of ascidian, barnacle, bryozoan and polychaete recruitment abundances at the Jetty site across the sampling period.

Taxonomic Group	Tukey's-test Letter	One-way ANOVA p-value
7/5/2011		
Ascidians	A	<0.001
Barnacles	B	
Bryozoans	A	
Polychaetes	A	
7/19/2011		
Ascidians	A	0.012
Barnacles	B	
Bryozoans	A	
Polychaetes	C	
8/16/2011		
Ascidians	A	0.004
Barnacles	A	
Bryozoans	A	
Polychaetes	A	
9/1/2011		
Ascidians	A	0.097
Barnacles	A	
Bryozoans	A	
Polychaetes	B	
9/15/2011		
Ascidians	A	<0.001
Barnacles	A	
Bryozoans	A	
Polychaetes	B	
9/29/2011		
Ascidians	A	0.0120
Barnacles	A	
Bryozoans	A	
Polychaetes	A	
10/13/2011		
Ascidians	A	0.399
Barnacles	A	
Bryozoans	A	
Polychaetes	A	

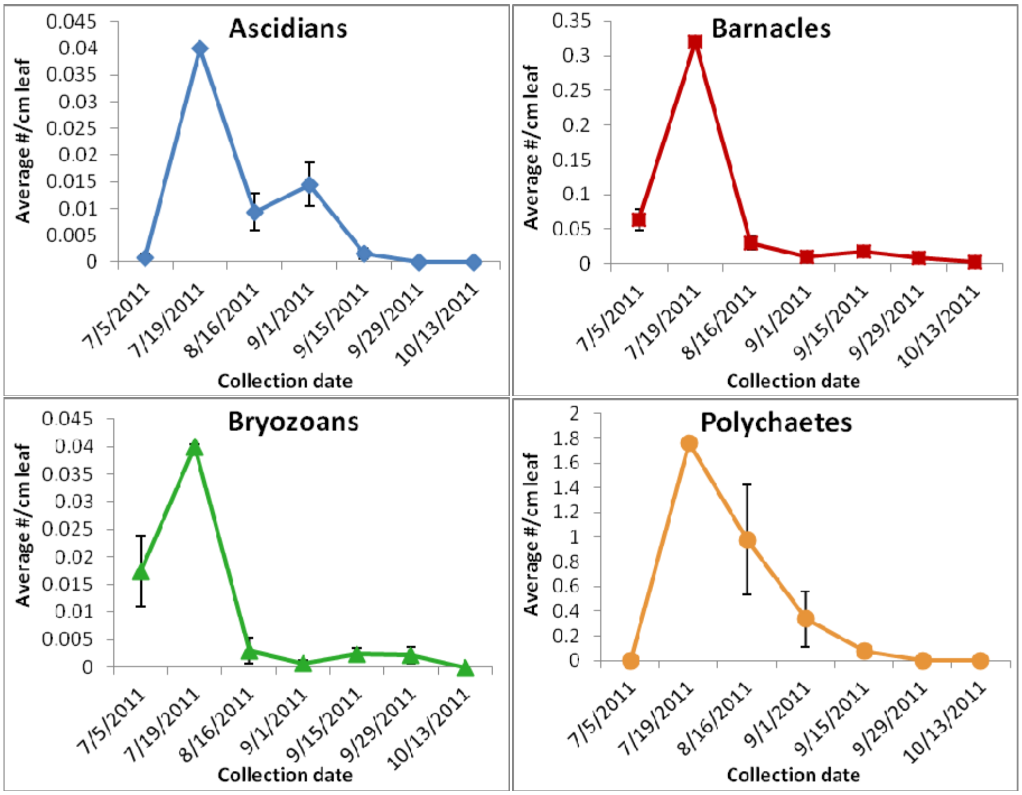

Figure 9. Average abundances of ascidians, barnacles, bryozoans and polychaetes per cm leaf at the Jetty site. Error bars are ±1 standard error. Note different scales on y-axes.

Table 9. Fouling invertebrate species observed on eelgrass at the Jetty site as fraction of total recruits on each collection date.

Species:	7/5/2011	7/19/2011	8/16/2011	9/1/2011	9/15/2011	9/29/2011	10/13/2011
Ascidians							
Botrylloides violaceus	0.008	0.021	0.005	0.041	0.032		
Botryllus schlosseri			0.003				
Molgula manhattensis			0.002				
Bryozoans							
Bowerbankia gracilis			0.002		0.032		
Cryptosula pallasiana						0.081	
Electra pilosa	0.010						
Electra crustulenta	0.105	0.017		0.002			
Membranipora sp.						0.056	
Microporella ciliata	0.006						
Barnacles							
Balanus sp.	0.736	0.151	0.030	0.029	0.010	0.580	1.00
Polychaetes							
Spirorbis sp	0.053	0.811	0.958	0.957	0.925	0.283	

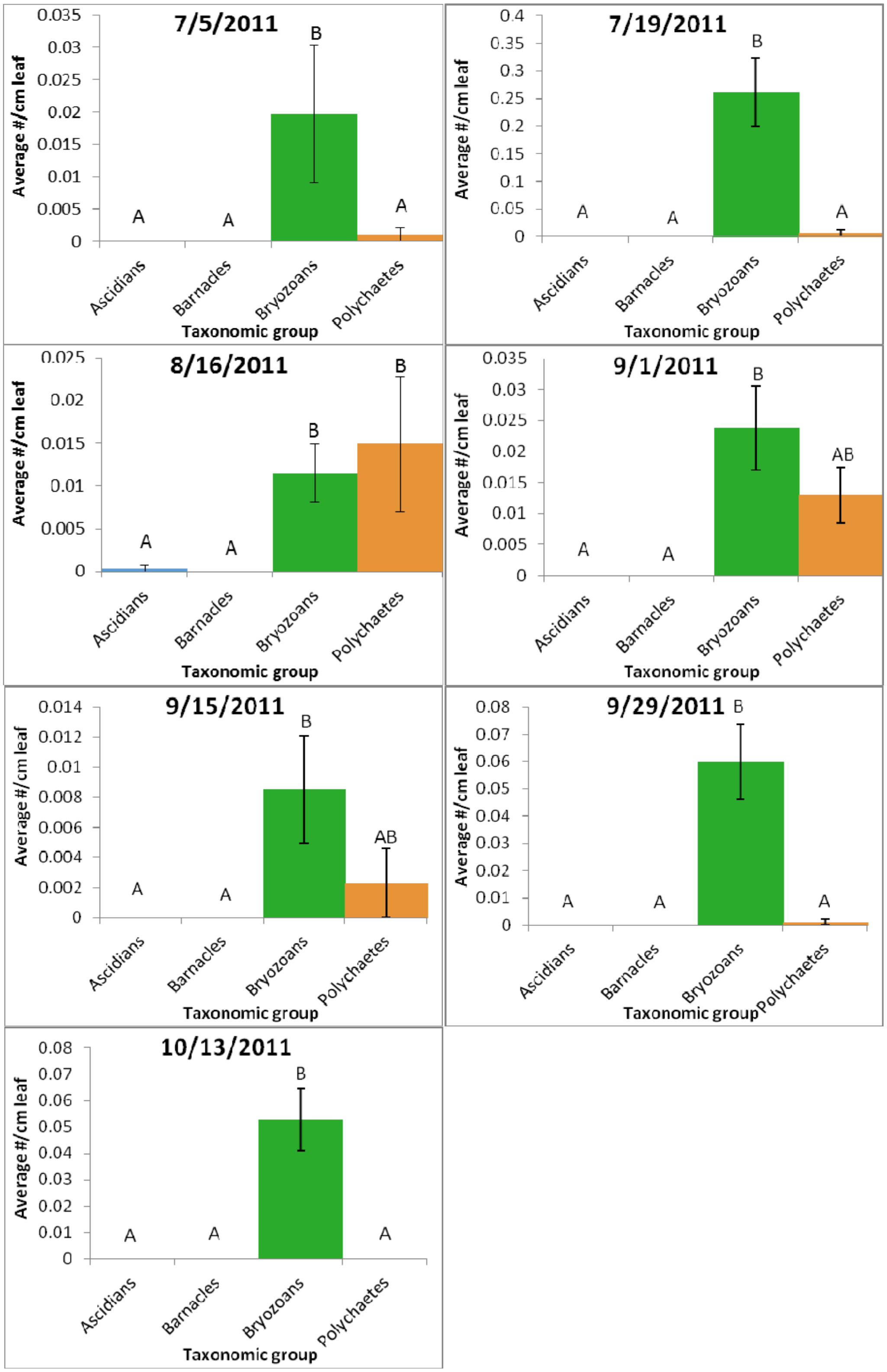

Figure 10. Recruitment abundance for all taxa at the Outside site on each sampling date. Letter indicate significant differences according to Tukey's Analysis of Variance Error bars are ±1 standard error. Note different scales on y-axes.

Table 10. Analysis of variance of ascidian, barnacle, bryozoan and polychaete recruitment abundances at the Outside site across the sampling period.

Taxonomic Group	Tukey's-test Letter	One-way ANOVA p-value
7/5/2011		
Ascidians	A	0.029
Barnacles	A	
Bryozoans	B	
Polychaetes	A	
7/19/2011		
Ascidians	A	<0.001
Barnacles	A	
Bryozoans	B	
Polychaetes	A	
8/16/2011		
Ascidians	A	0.028
Barnacles	A	
Bryozoans	B	
Polychaetes	B	
9/1/2011		
Ascidians	A	<0.001
Barnacles	A	
Bryozoans	B	
Polychaetes	AB	
9/15/2011		
Ascidians	A	0.018
Barnacles	A	
Bryozoans	B	
Polychaetes	AB	
9/29/2011		
Ascidians	A	0.000
Barnacles	A	
Bryozoans	B	
Polychaetes	A	
10/13/2011		
Ascidians	A	<0.001
Barnacles	A	
Bryozoans	B	
Polychaetes	A	

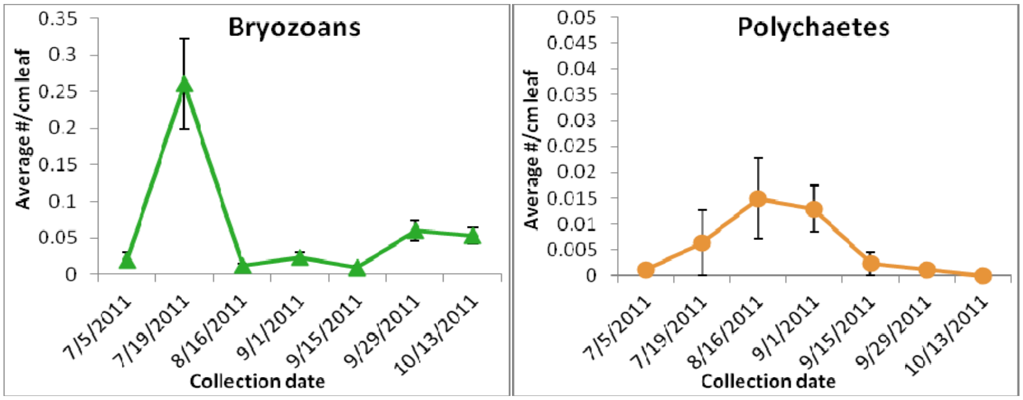

Figure 11. Average abundances of bryozoans and polychaetes per cm leaf at the Outside site. Error bars are ±1 standard error. Note different scales on y-axes.

Outside Site Species Comparison

On the initial collection date the Outside site was dominated by the bryozoan, *M. ciliata* (Table 11). The second date, July 19, 2011, dominance was observed by *E. pilosa,* and the third collection date, August 16, 2011, *Spirorbis* polychaetes were dominant. The bryozoan, *C. hyalina* was then dominant until the final collection date, at which point the bryozoan *B. gracilis* is dominant, and *Balanus* barnacles also substantially contributed to recruitment.

Comparison of Sites by Taxonomic Group

Ascidians

Peaks in ascidian abundance were not seen synchronously among sites (Figure 12). The Jetty site displayed the earliest peak, on July 19, 2011 followed by the Beach site and the Outside site, on August 16, 2011. The Bushy site showed an ascidian peak on September 1, 2011, which was also the date of a second peak at the Jetty site. The Beach site showed a second peak on September 15, 2011, at which time the other sites showed a sharp decline in abundance.

Barnacles

The Jetty site had the most dramatic peak in abundance on July 19, 2011, followed by a sharp decline and consistently low abundances for the remainder of the sampling dates (Figure 13). The Beach site had a great deal of variability in barnacle abundance, although remained consistently low overall. The Bushy site had extremely low barnacle recruitment on July 5 and July 19, 2011, followed by no barnacle recruitment at all for the rest of the sampling period. Barnacles did not recruit at all at the Outside site.

Bryozoans

Bryozoan recruitment was highly variable among sites (Figure 14). The Bushy site had overall low Bryozoan abundances with a peak on July 5, 2011. The Jetty site and the Outside site both had peaks in bryozoan recruitment abundance on July 19, 2011, although the Outside site showed dramatically higher values than the Jetty site. The Jetty site showed near-zero abundances for the rest of the sampling period, while the Outside site showed a later and much lower peak in

Table 11. Fouling invertebrate species observed on eelgrass at the Outside site as fraction of total recruits on each collection date.

Species:	7/5/2011	7/19/2011	8/16/2011	9/1/2011	9/15/2011	9/29/2011	10/13/2011
Ascidians							
Botrylloides violaceus			0.014				
Bryozoans							
Bowerbankia gracilis			0.160	0.038	0.073		0.443
Celleporaria hyalina	0.051	0.018	0.195	0.572	0.715	0.967	
Cryptosula pallasiana						.015	
Electra pilosa		0.955	0.073	0.037			0.058
Electra crustulenta	0.055	0.003					
Microporella ciliata	0.843						
Barnacles							
Balanus sp.							0.322
Polychaetes							
Spirorbis sp	0.051	0.024	0.557	0.352	0.211	0.018	0.177

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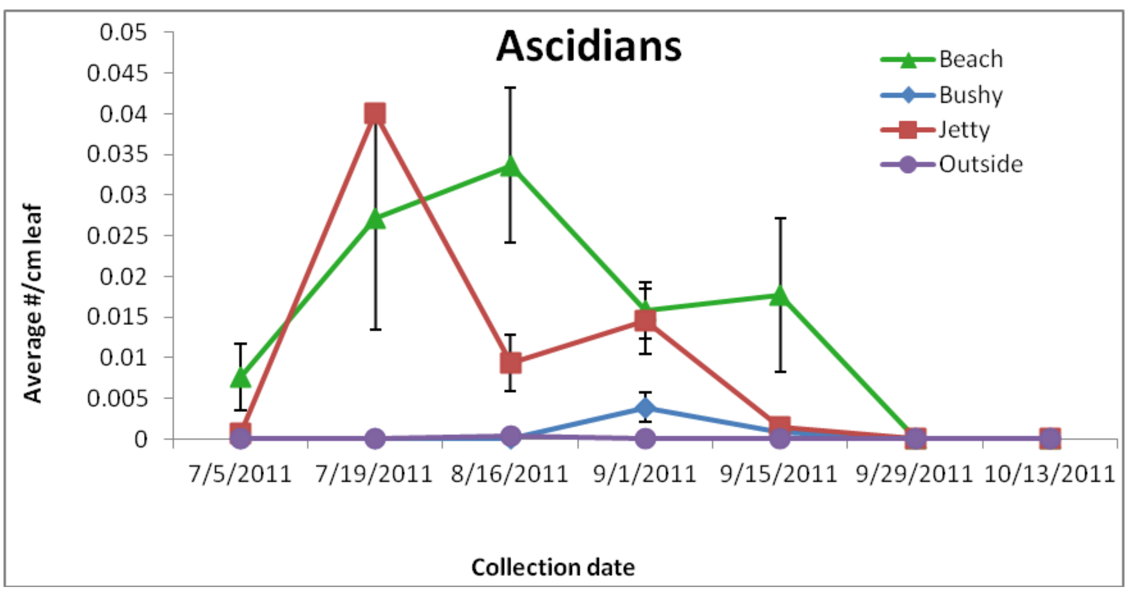

Figure 12. Ascidian abundance among sites

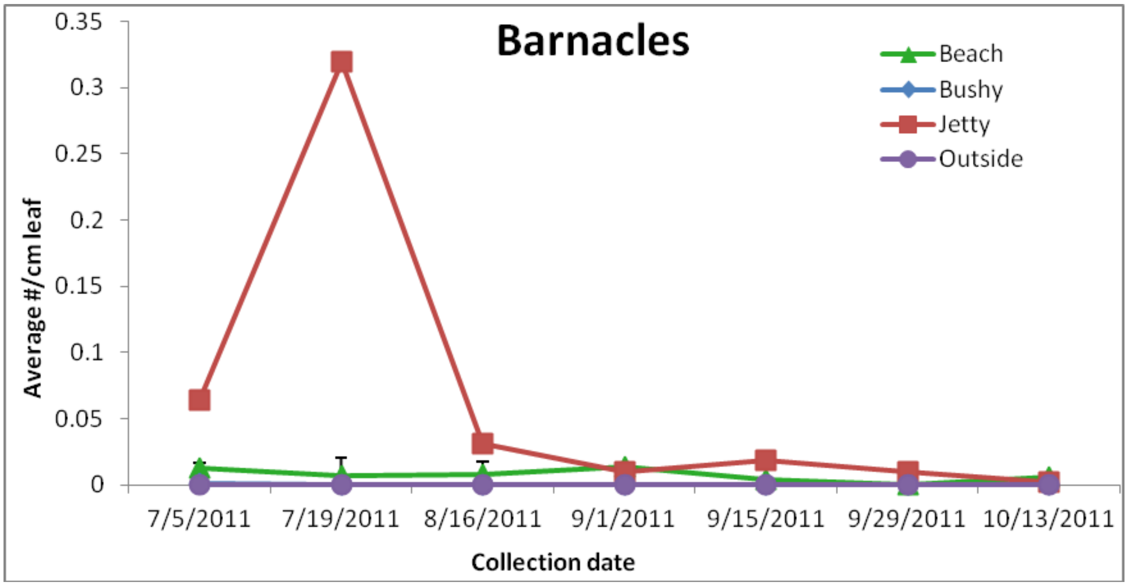

Figure 13. Barnacle abundance among sites

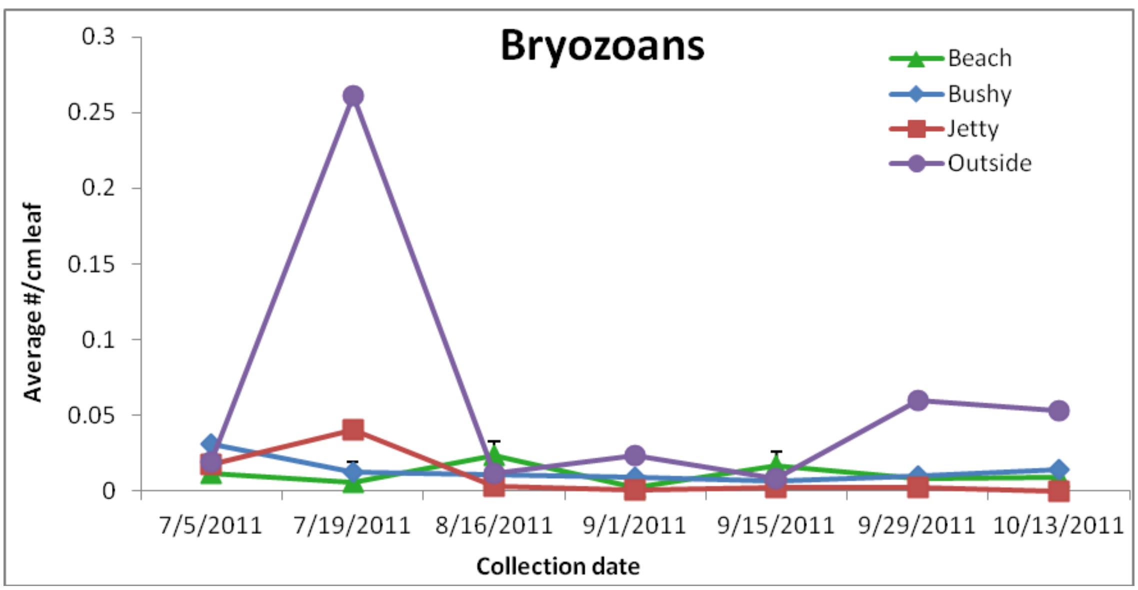

Figure 14. Bryozoan abundance among sites

abundance on September 29, 2011. While recruitment remained low at the Beach site, distinct peaks were displayed on August 16, and September 15, 2014.

Polychaetes

The Jetty site showed a peak in polychaete abundance on July 19, 2011 (Figure 15). The Beach site and the Outside site both had peaks of polychaete abundance on August 16, 2011, although the Beach site was several orders of magnitude greater in abundance. The Bushy site peaked in polychaete abundance on September 1, 2011.

Hypothesis 2

It was hypothesized that outer leaves of collected shoots would show higher abundances of invertebrate fouling recruitment than inner leaves. All outer leaves from the four sites were pooled from all collection dates and all inner leaves pooled the same way, resulting in significant differences in recruitment abundances on outer leaves as compared to inner leaves (Figure 16, Table 12). A series of paired t-tests were then performed comparing outer with inner leaves pooled by date and site (Table 13). This analysis resulted in significant differences between inner and outer leaves for less than 20% of the comparisons. Differences between inner and outer leaves were inconsistent among sampling sites.

Beach site (Figure 17, Table 13): No significant differences in ascidian recruitment abundances were observed between inner and outer leaves for any of the collection dates. On the September 1, 2011 collection date, significantly greater barnacle recruitment was observed on outer leaves relative to inner leaves and on the September 15 and October 13, 2011collection dates, barnacle recruitment was observed on outer leaves but not inner leaves resulting in significantly higher recruitment on the outer leaves. On the July 5, 2011, collection date, bryozoan and polychaete recruitment was observed on inner and not outer leaves, resulting in significantly more of both groups on inner than outer leaves. These are the only cases throughout the study in which the

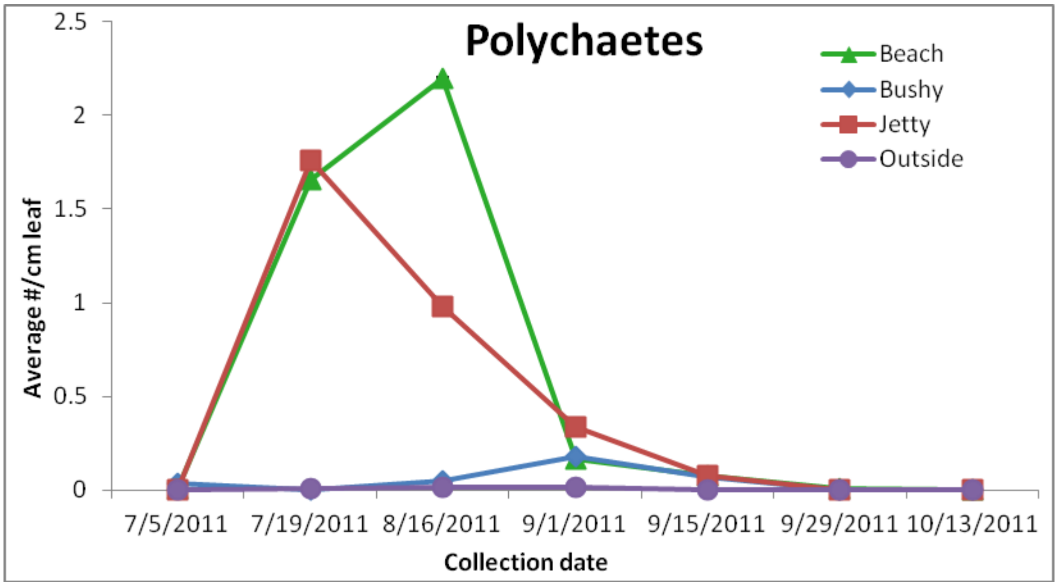

Figure 15. Polychaete abundance among sites

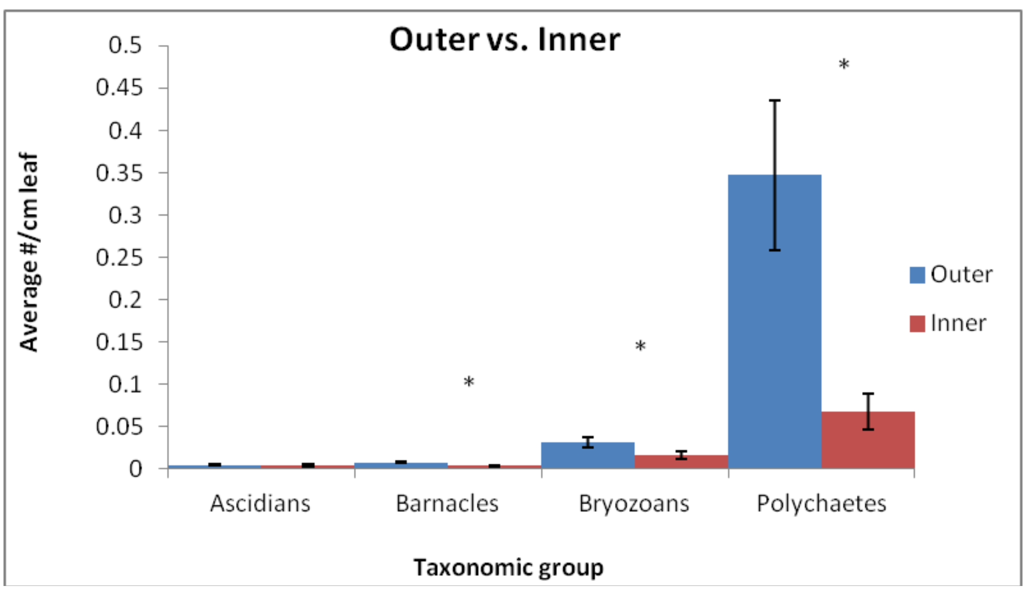

Figure 16. Pooled outer leaves compared with pooled inner leaves from all sites and collection dates. Error bars are ±1 standard error. Significant differences indicated by "*".

Table 12. Recruitment abundances on outer leaves compared to inner leaves with data pooled from all sites and all collection dates.

Taxonomic Group	Tukey's-test Letter	One-way ANOVA p-value	Two-Sample t-test p-value
Ascidians			
Outer	A	0.840	0.841
Inner	A		
Barnacles			
Outer	A	0.006	0.005
Inner	B		
Bryozoans			
Outer	A	0.037	0.034
Inner	B		
Polychaetes			
Outer	A	0.003	0.002
Inner	B		

Table 13. P-values for paired t-tests of pooled recruitment on outer vs. inner leaves. Highlighted values indicate significant differences; yellow indicates greater recruitment on outer than inner leaves. Red indicates greater recruitment on inner than outer leaves. "X" indicates no recruitment was observed on outer or inner leaves.

Ascidians					Bryozoans				
	Beach	Bushy	Jetty	Outside		Beach	Bushy	Jetty	Outside
7/5/2011	>0.05	>0.05	>0.05	>0.05	7/5/2011	<0.001	>0.05	>0.05	>0.05
7/19/2011	>0.05	>0.05	>0.05	>0.05	7/19/2011	>0.05	>0.05	>0.05	>0.05
8/16/2011	>0.05	>0.05	>0.05	>0.05	8/16/2011	>0.05	<0.001	>0.05	0.003
9/1/2011	>0.05	>0.05	0.001	x	9/1/2011	>0.05	>0.05	>0.05	<0.001
9/15/2011	>0.05	>0.05	<0.001	>0.05	9/15/2011	>0.05	>0.05	<0.001	>0.05
9/29/2011	>0.05	>0.05	x	>0.05	9/29/2011	>0.05	>0.05	<0.001	0.040
10/13/2011	>0.05	>0.05	>0.05	>0.05	10/13/2011	>0.05	<0.001	>0.05	0.001
Barnacles					Polychaetes				
	Beach	Bushy	Jetty	Outside		Beach	Bushy	Jetty	Outside
7/5/2011	>0.05	>0.05	>0.05	>0.05	7/5/2011	<0.001	>0.05	>0.05	>0.05
7/19/2011	>0.05	>0.05	>0.05	>0.05	7/19/2011	>0.05	>0.05	>0.05	>0.05
8/16/2011	>0.05	>0.05	>0.05	>0.05	8/16/2011	>0.05	>0.05	0.042	<0.001
9/1/2011	0.016	>0.05	>0.05	x	9/1/2011	>0.05	0.005	>0.05	<0.001
9/15/2011	<0.001	>0.05	>0.05	>0.05	9/15/2011	0.020	0.020	0.014	>0.05
9/29/2011	>0.05	>0.05	<0.001	>0.05	9/29/2011	>0.05	>0.05	<0.001	>0.05
10/13/2011	<0.001	>0.05	>0.05	>0.05	10/13/2011	>0.05	>0.05	>0.05	>0.05

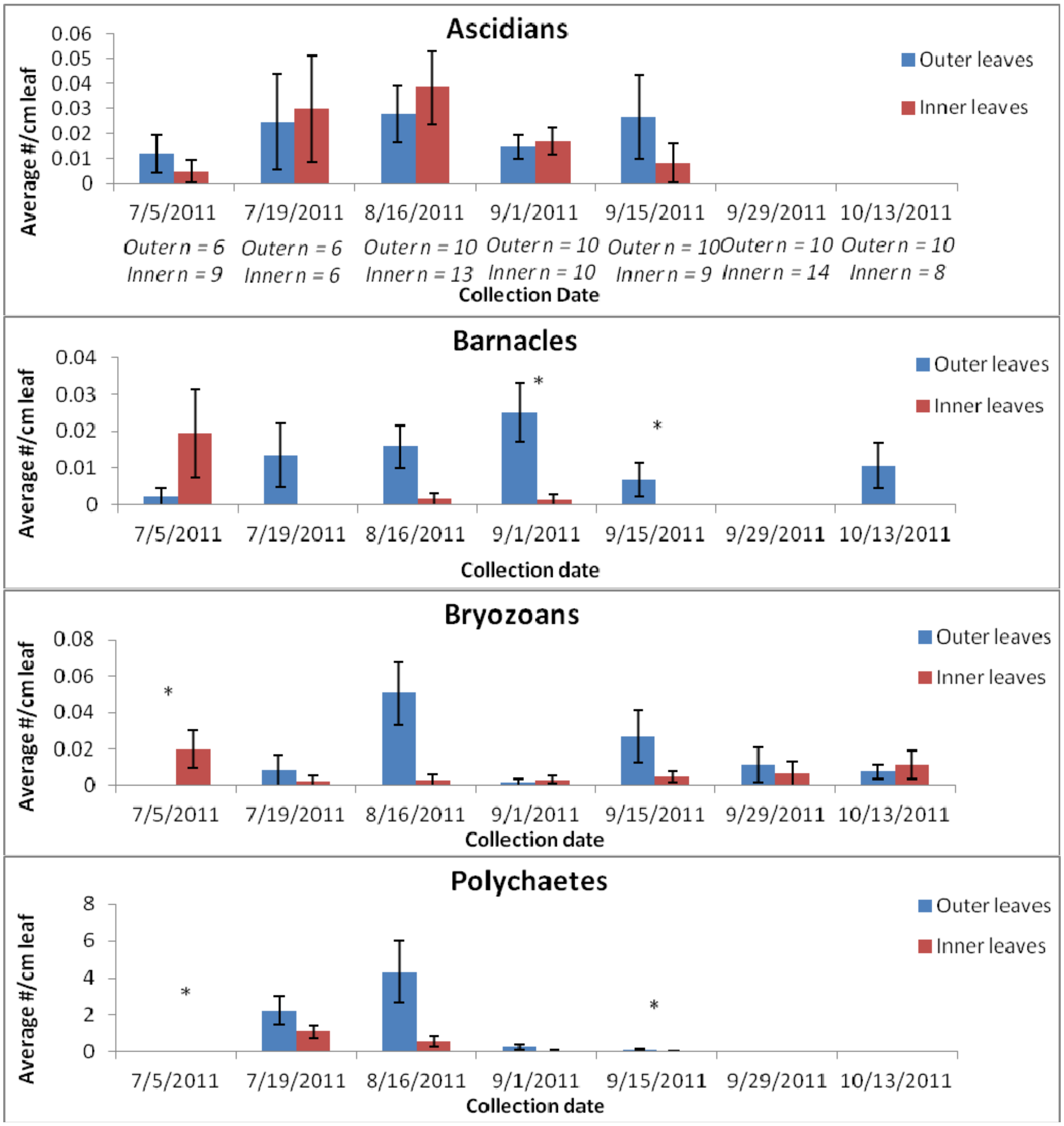

Figure 17. Average number of recruits on outer and inner leaves for Beach site. Error bars are ±1 standard error. Significant differences indicated by *. Note different scales on y-axes.

inner leaves had significantly greater recruitment than outer leaves. The September 15, 2011 collection date also showed significantly greater polychaete recruitment on outer leaves relative to inner leaves.

Bushy site (Figure 18, Table 13): No significant differences in ascidian or barnacle recruitment abundances were observed between inner and outer leaves for any of the collection dates. On the August 16 and October 13, 2011 collection dates, bryozoan recruitment was observed on outer leaves but not inner leaves, resulting in significantly higher recruitment of bryozoans on outer leaves relative to inner leaves. On the September 1 and September 15, 2011 collection dates, significantly greater polychaete recruitment was observed on outer leaves relative to inner leaves.

Jetty site (Figure 19, Table 13)**:** Significantly higher ascidian recruitment was observed on outer leaves relative to inner leaves on the September 1 and September 15, 2011 collection dates. Recruitment of barnacles was only seen on outer leaves on the September 29, 2011, collection date. Recruitment of bryozoans was observed on outer leaves but not inner leaves on the September 15 and September 29, 2011, collection dates. Significantly higher polychaete recruitment was observed on outer leaves relative to inner leaves on the August 16, September 15, and September 29, 2011, collection dates.

Outside site (Figure 20, Table 13): No significant differences in ascidian or barnacle recruitment abundances were observed between inner and outer leaves for any of the collection dates. On the August 16, September 1, September 29, and October 13, 2011 collection dates, significantly higher recruitment of bryozoans was observed on outer leaves relative to inner leaves. On the August 16 and September 1, 2011 collection dates, recruitment of polychaetes was observed on outer but not inner leaves, resulting in significantly higher recruitment on outer leaves

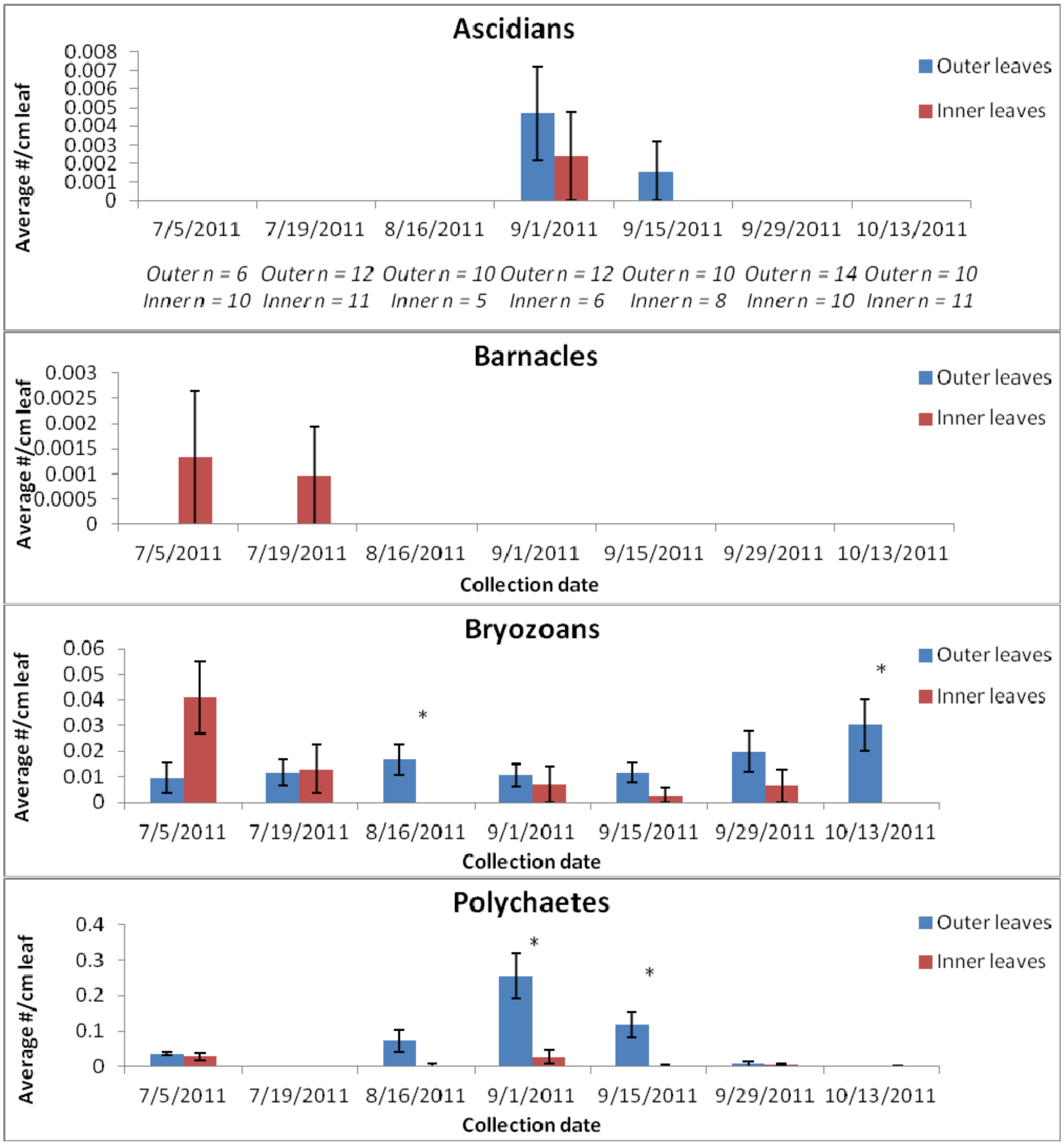

Figure 18. Average number of recruits on outer and inner leaves for Bushy site. Error bars are ±1 standard error. Significant differences indicated by *. Note different scales on y-axes.

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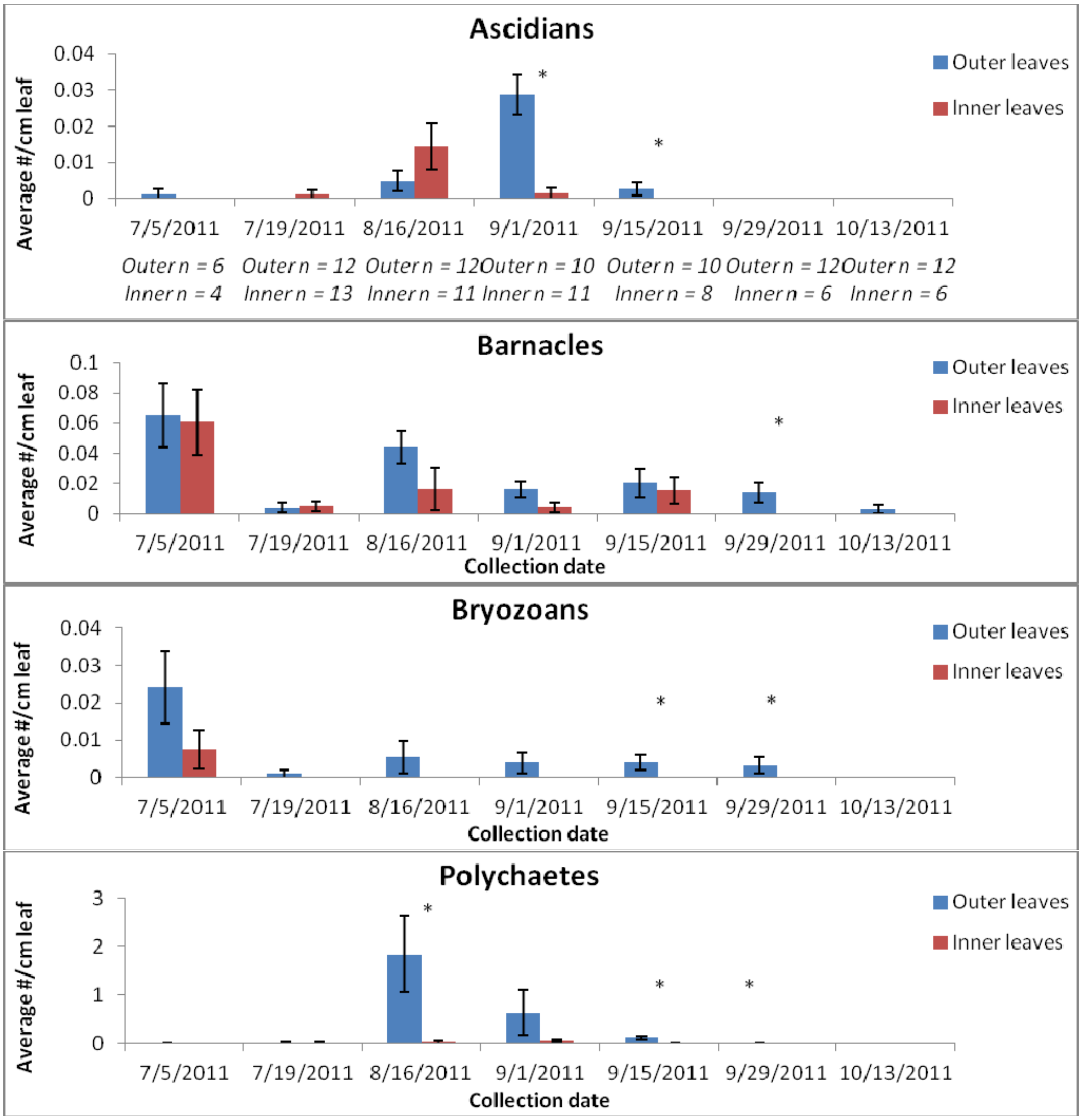

Figure 19. Average number of recruits on outer and inner leaves for Jetty site. Error bars are ±1 standard error. Significant differences indicated by *. Note different scales on y-axes.

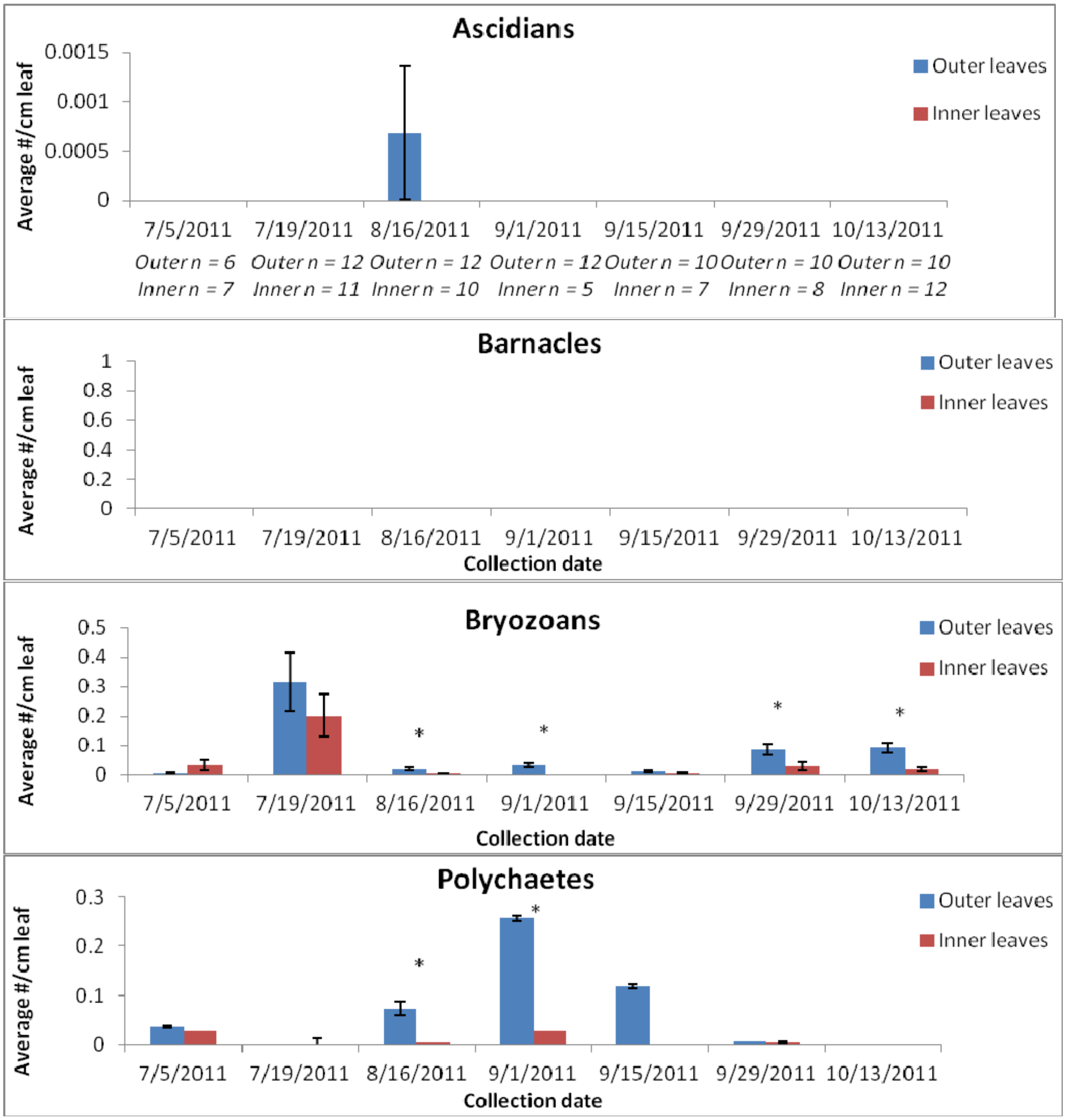

Figure 20. Average number of recruits on outer and inner leaves for Outside site. Error bars are ±1 standard error. Significant differences indicated by *. Note different scales on y-axes.

Hypothesis 3

It was hypothesized that fouling invertebrates would increase in abundance along eelgrass leaves due to the progressively increased amount of time of exposure to recruitment from leaf base to tip. Overall, differences in recruitment were most often found to be not significant between sections of leaf. Significant differences observed were consistent across all sites: both middle and end sections had higher recruitment than first sections and little to no difference in recruitment was observed between middle and end sections.

Beach site (Figure 21, Table 14): There was a general trend toward higher recruitment on middle and end sections than first sections of leaf. The only significant difference was seen for ascidian recruitment between end and middle sections on the August 16, 2011 collection date.

Bushy site (Figure 22, Table 15): There was a general trend toward higher recruitment of bryozoans and polychaetes on middle and end sections than first sections of leaf, with little to no ascidian and barnacle recruitment. Significant differences were seen for polychaete recruitment between the first section and middle section and the first section and end section on the September 1, 2011 collection date and between the first section and middle section on the September 15, 2011 collection date.

Jetty Site (Figure 23, Table 16)**:** There was a general trend toward higher recruitment of fouling organisms on middle and end sections as compared with first sections. Significant differences were seen for polychaete recruitment between the first and middle sections on the August 16, September 1, and September 15, 2011 collection dates. Barnacles showed significantly higher recruitment on end sections as compared with first and middle sections on the September 1, 2011 collection date.

Outside site (Figure 24, Table 17)**:** The Outside side had almost no recruitment of ascidians or barnacles on any leaf sections. Most of the sampling dates showed a trend of middle and end sections experiencing higher recruitment of bryozoans and polychaetes as compared to first

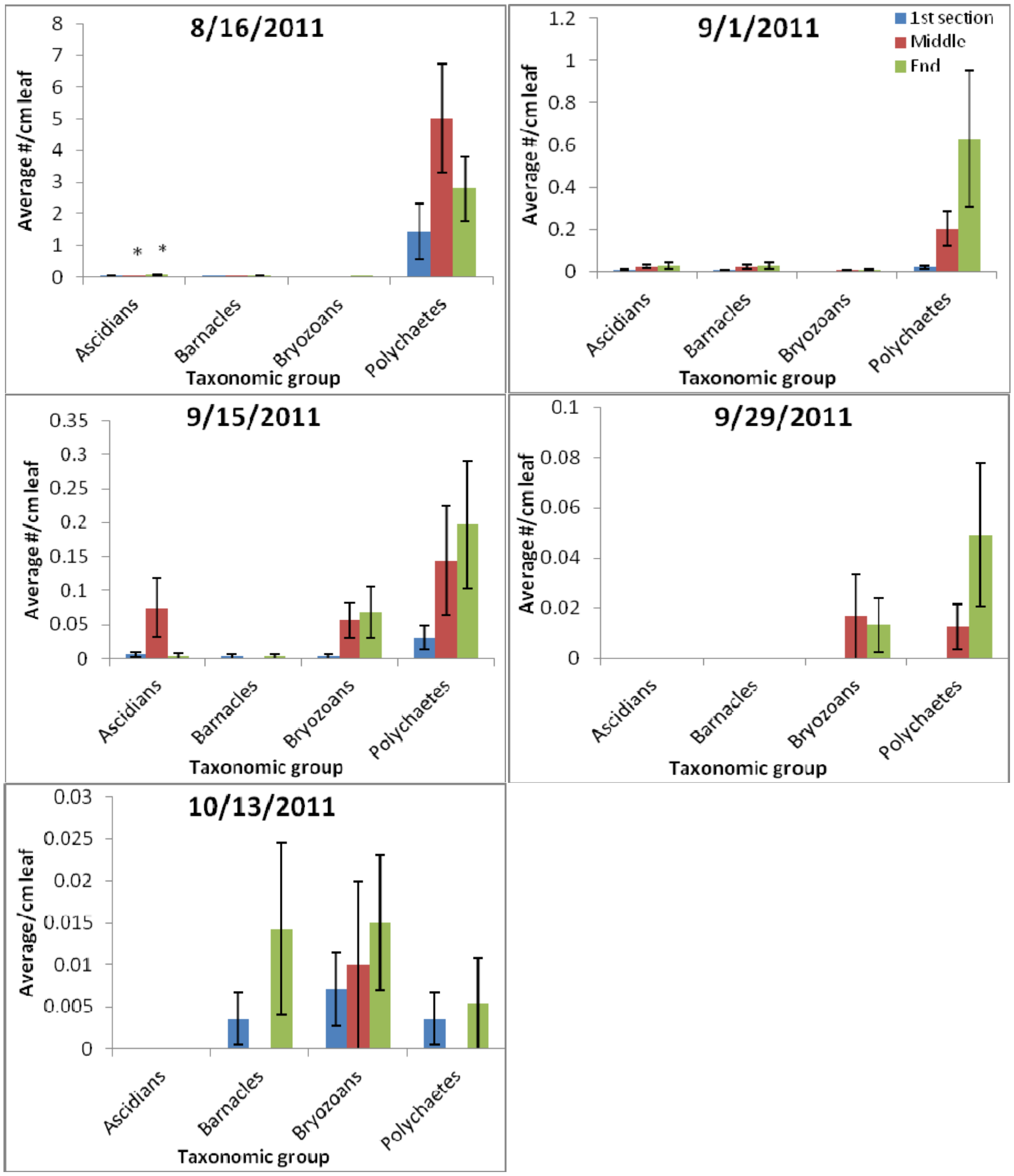

Figure 21. Average recruitment of fouling organisms on first, middle and end sections of leaves for each sampling date at the Beach site. Error bars are ±1 standard error. Sections with significant differences are indicated by *. Note different scales on y-axes.

Table 14. Results of 2-sample t-tests for recruitment abundances on first, middle and end sections of leaves at the Beach site. Section with higher recruitment is listed first in each pair. Highlighting indicates significant differences. Dash in p-value column indicates there was no recruitment in one of the sections compared. Dashes in sections columns indicate no recruitment on either section.

8/16/2011					
Ascidians			**Barnacles**		
Sections		p-value	Sections		p-value
First	Middle	0.200	-	-	-
End	Middle	0.040	-	-	-
End	First	0.197	-	-	-
Bryozoans			**Polychaetes**		
Sections		p-value	Sections		p-value
-	-	-	Middle	First	0.079
-	-	-	Middle	End	0.277
-	-	-	End	First	0.326
9/1/2011					
Ascidians			**Barnacles**		
Sections		p-value	Sections		p-value
Middle	First	0.073	Middle	First	0.103
End	Middle	> 0.25	End	Middle	> 0.25
End	First	0.181	End	First	0.147
Bryozoans			**Polychaetes**		
Sections		p-value	Sections		p-value
Middle	First	-	Middle	First	0.092
End	Middle	> 0.25	End	Middle	0.218
End	First	-	End	First	0.077
9/15/2011					
Ascidians			**Barnacles**		
Sections		p-value	Sections		p-value
Middle	First	0.158	First	Middle	-
Middle	End	0.147	End	Middle	-
First	End	> 0.25	End	First	> 0.25
Bryozoans			**Polychaetes**		
Sections		p-value	Sections		p-value
Middle	First	0.080	Middle	First	0.214
End	Middle	> 0.25	End	Middle	> 0.25
End	First	0.080	End	First	0.106
9/29/2011					
Ascidians			**Barnacles**		
Sections		p-value	Sections		p-value
-	-	-	-	-	-
-	-	-	-	-	-
-	-	-	-	-	-
Bryozoans			**Polychaetes**		
Sections tested		p-value	Sections		p-value
Middle	First	-	Middle	First	-
Middle	End	> 0.25	End	Middle	0.232
End	First	-	End	First	-
10/13/2011					
Ascidians			**Barnacles**		
Sections		p-value	Sections		p-value
-	-	-	First	Middle	-
-	-	-	End	Middle	-
-	-	-	End	First	> 0.25
Bryozoans			**Polychaetes**		
Sections tested		p-value	Sections		p-value
Middle	First	> 0.25	First	Middle	-
End	Middle	> 0.25	End	Middle	-
End	First	> 0.25	End	First	> 0.25

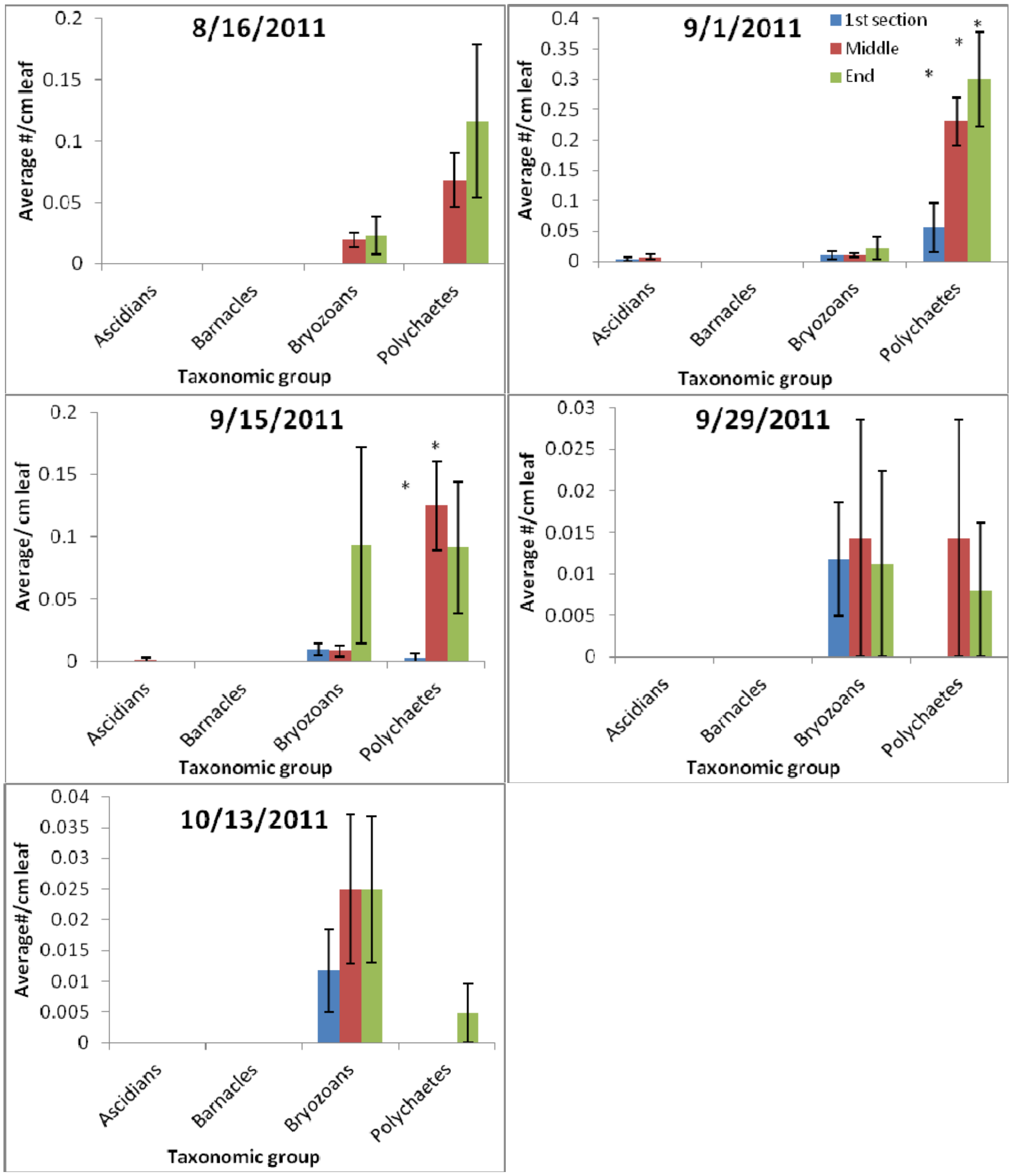

Figure 22. Average recruitment of fouling organisms on first, middle and end sections of leaves for each sampling date at the Bushy site. Error bars are ±1 standard error. Sections with significant differences are indicated by *. Note different scales on y-axes.

Table 15. Results of 2-sample t-tests for recruitment abundances on first, middle and end sections of leaves at the Bushy site. Section with higher recruitment is listed first in each pair. Highlighting indicates significant differences. Dash in p-value column indicates there was no recruitment in one of the sections compared. Dashes in sections columns indicate no recruitment on either section.

8/16/2011					
Ascidians			**Barnacles**		
Sections		p-value	Sections		p-value
-	-	-	-	-	-
-	-	-	-	-	-
-	-	-	-	-	-
Bryozoans			**Polychaetes**		
Sections		p-value	Sections		p-value
Middle	First	-	Middle	First	-
End	Middle	> 0.25	Middle	End	> 0.25
End	First	-	End	First	-
9/1/2011					
Ascidians			**Barnacles**		
Sections		p-value	Sections		p-value
Middle	First	-	-	-	-
End	Middle	> 0.25	-	-	-
End	First	-	-	-	-
Bryozoans			**Polychaetes**		
Sections		p-value	Sections		p-value
Middle	First	> 0.25	Middle	First	0.004
End	Middle	> 0.25	End	Middle	> 0.25
End	First	> 0.25	End	First	0.011
9/15/2011					
Ascidians			**Barnacles**		
Sections		p-value	Sections		p-value
Middle	First	-	-	-	-
Middle	End	-	-	-	-
-	-	-	-	-	-
Bryozoans			**Polychaetes**		
Sections		p-value	Sections		p-value
Middle	First	> 0.25	Middle	First	0.002
End	Middle	> 0.25	End	Middle	> 0.25
End	First	> 0.25	End	First	0.118
9/29/2011					
Ascidians			**Barnacles**		
Sections		p-value	Sections		p-value
-	-	-	-	-	-
-	-	-	-	-	-
-	-	-	-	-	-
Bryozoans			**Polychaetes**		
Sections tested		p-value	Sections		p-value
Middle	First	> 0.25	Middle	First	-
Middle	End	> 0.25	End	Middle	> 0.25
End	First	> 0.25	End	First	-
10/13/2011					
Ascidians			**Barnacles**		
Sections		p-value	Sections		p-value
-	-	-	-	-	-
-	-	-	-	-	-
-	-	-	-	-	-
Bryozoans			**Polychaetes**		
Sections tested		p-value	Sections		p-value
Middle	First	> 0.25	-	-	-
End	Middle	> 0.25	End	Middle	-
End	First	> 0.25	End	First	-

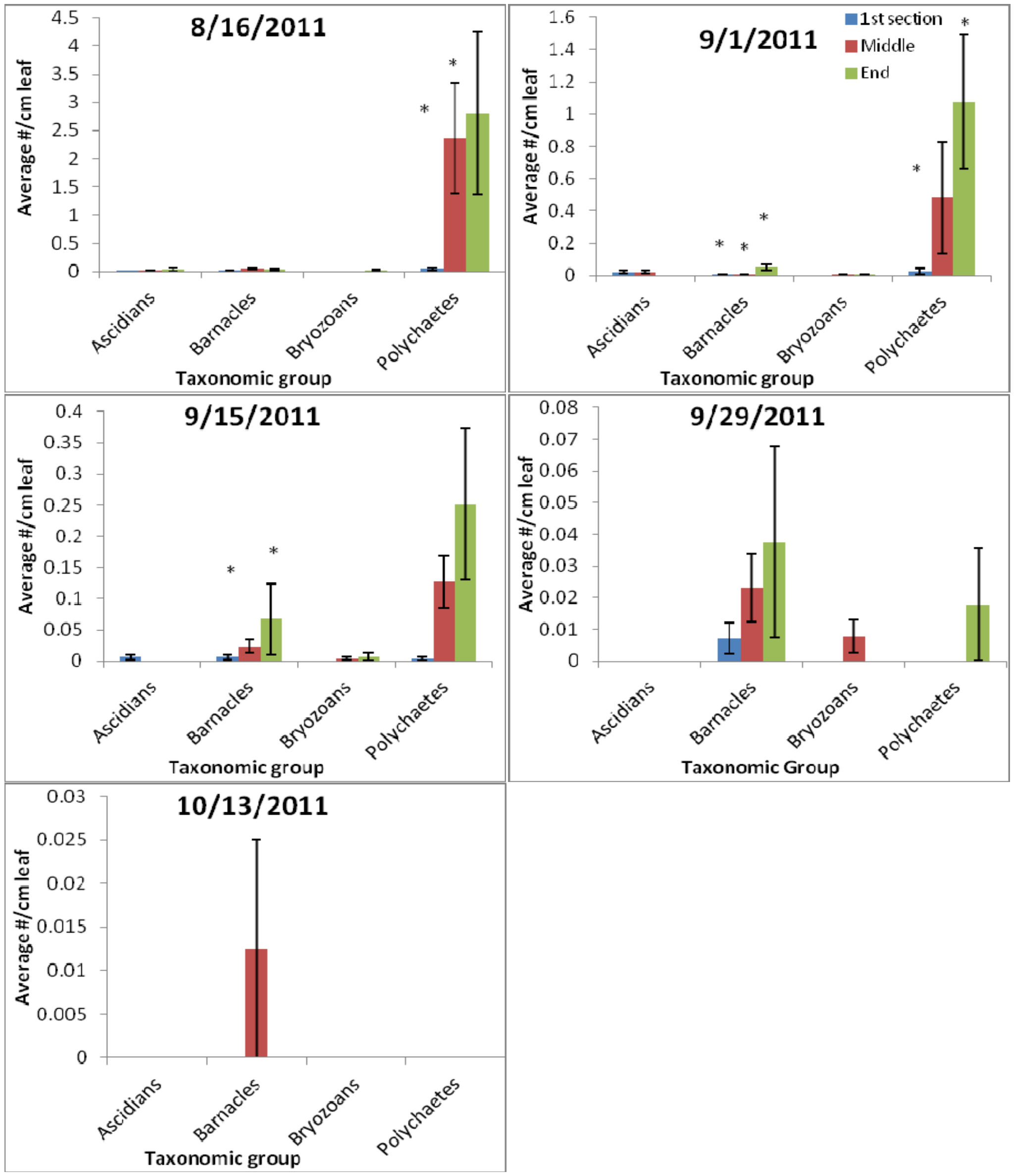

Figure 23. Average recruitment of fouling organisms on first, middle and end sections of leaves for each sampling date at the Jetty site. Error bars are ±1 standard error. Sections with significant differences are indicated by *. Note different scales on y-axes.

Table 16. Results of 2-sample t-tests for recruitment abundances on first, middle and end sections of leaves at the Jetty site. Section with higher recruitment is listed first in each pair. Highlighting indicates significant differences. Dash in p-value column indicates there was no recruitment in one of the sections compared. Dashes in sections columns indicate no recruitment on either section.

8/16/2011					
Ascidians			**Barnacles**		
Sections		p-value	Sections		p-value
Middle	First	> 0.25	Middle	First	0.062
End	Middle	> 0.25	End	Middle	> 0.25
End	First	> 0.25	End	First	0.217
Bryozoans			**Polychaetes**		
Sections		p-value	Sections		p-value
-	-	-	Middle	First	0.028
End	Middle	-	Middle	End	> 0.25
End	First	-	End	First	0.068
9/1/2011					
Ascidians			**Barnacles**		
Sections		p-value	Sections		p-value
Middle	First	> 0.25	Middle	First	> 0.25
End	Middle	-	End	Middle	0.035
End	First	-	End	First	0.032
Bryozoans			**Polychaetes**		
Sections		p-value	Sections		p-value
Middle	First	-	Middle	First	0.197
End	Middle	> 0.25	End	Middle	0.282
End	First	-	End	First	0.022
9/15/2011					
Ascidians			**Barnacles**		
Sections		p-value	Sections		p-value
Middle	First	-	Middle	First	0.149
-	-	-	End	Middle	> 0.25
First	End	-	End	First	> 0.25
Bryozoans			**Polychaetes**		
Sections		p-value	Sections		p-value
Middle	First	-	Middle	First	0.006
End	Middle	> 0.25	End	Middle	> 0.25
End	First	-	End	First	0.058
9/29/2011					
Ascidians			**Barnacles**		
Sections		p-value	Sections		p-value
-	-	-	Middle	First	0.196
-	-	-	End	Middle	> 0.25
-	-	-	End	First	0.339
Bryozoans			**Polychaetes**		
Sections tested		p-value	Sections		p-value
Middle	First	-	-	-	-
Middle	End	-	End	Middle	-
-	-	-	End	First	-
10/13/2011					
Ascidians			**Barnacles**		
Sections		p-value	Sections		p-value
-	-	-	Middle	First	-
-	-	-	End	Middle	-
-	-	-	-	-	-
Bryozoans			**Polychaetes**		
Sections tested		p-value	Sections		p-value
-	-	-	-	-	-
-	-	-	-	-	-
-	-	-	-	-	-

sections, with the exception of the October 13, 2011 sampling date which reversed this trend. There was significantly higher recruitment of bryozoans on first sections compared with end sections on the October 13, 2011 sampling date. The only other significance at the Outside site was on the September 1, 2011 sampling date; middle sections showed significantly higher recruitment of polychaetes as compared with first sections. There were no significant differences between middle and end sections for any of the taxa at the Outside site on any of the collection dates.

Overall, the differences among leaf sections showed very little statistical significance for any of the taxa, study sites or sampling dates.

Hypothesis 4

It was hypothesized that rafting leaves would have higher abundances of fouling invertebrates than intact leaves due to the increased amount of time that rafting leaves have been exposed to recruitment in comparison to intact leaves.

Collection date: July 5, 2011

Rafting leaves (n = 13) were only collected from the Bushy (n = 3) and Outside (n = 10) sites on July 5, 2011, as no rafting leaves were observed at the other sites. Ascidian recruitment was extremely low on both intact and rafting leaves. There was a trend toward slightly higher recruitment of ascidians on rafting as compared to intact leaves from all four of the sites (Figure 25) although the Tukey's test indicated that these differences were not significant. The one-way un-stacked analysis of variance did, however indicate a significant difference (p-value = 0.049) (Table 18). Barnacle recruitment abundances were significantly higher on intact leaves from the Jetty site as compared with rafting leaves. Other sites showed no significant differences in barnacle recruitment abundances. No recruitment of bryozoans was observed on rafting leaves on this collection date. Bryozoan recruitment abundances on intact leaves at all four sites were in such low abundances as to be not significant. Polychaete recruitment abundances showed a trend

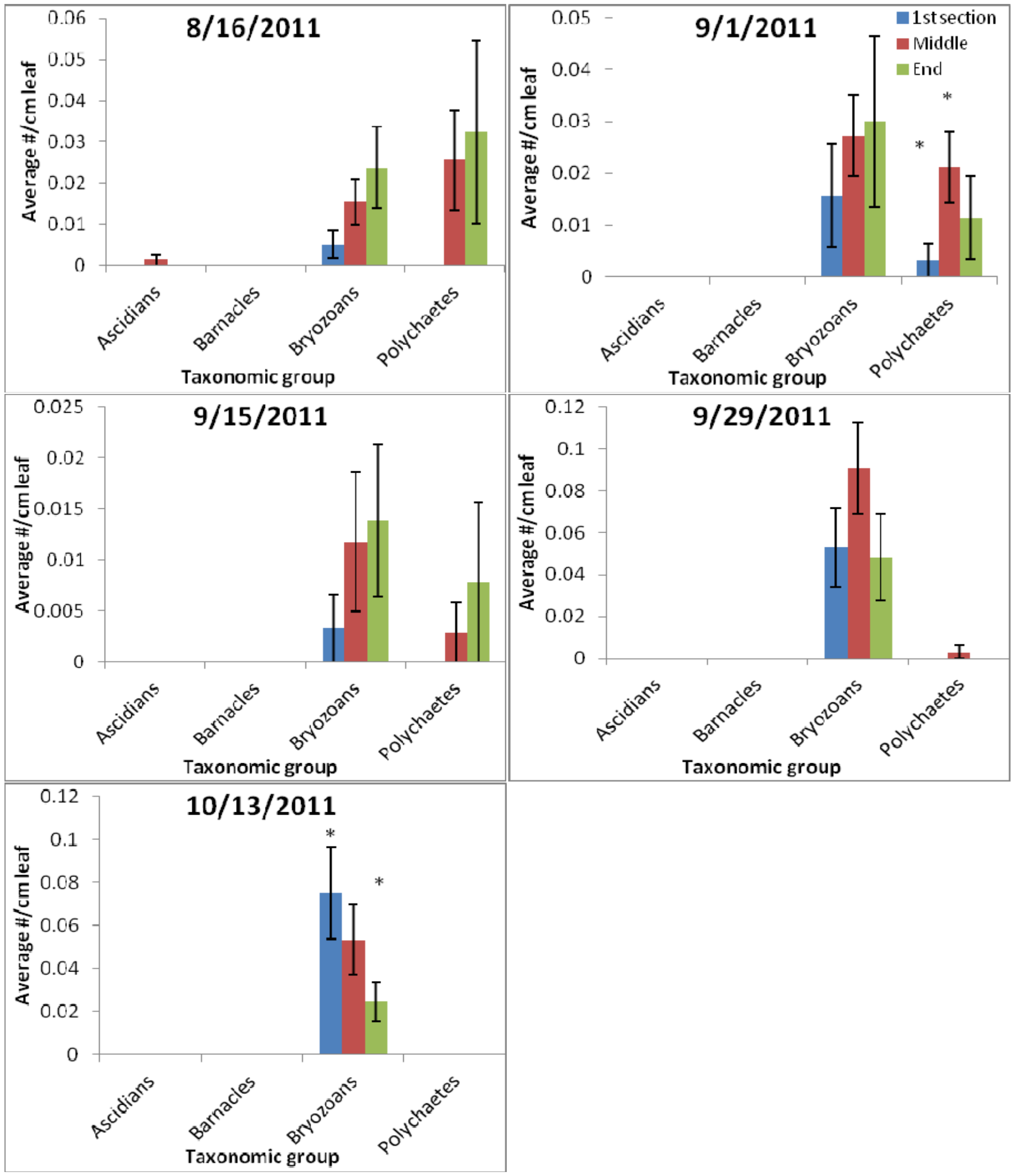

Figure 24. Average recruitment of fouling organisms on first, middle and end sections of leaves for each sampling date at the Outside site. Error bars are ±1 standard error. Sections with significant differences are indicated by *. Note different scales on y-axes.

Table 17. Results of 2-sample t-tests for recruitment abundances on first, middle and end sections of leaves at the Outside site. Section with higher recruitment is listed first in each pair. Highlighting indicates significant differences. Dash in p-value column indicates there was no recruitment in one of the sections compared. Dashes in sections columns indicate no recruitment on either section.

8/16/2011					
Ascidians			**Barnacles**		
Sections		p-value	sections		p-value
Middle	First	-	-	-	-
Middle	End	-	-	-	-
-	-	-	-	-	-
Bryozoans			**Polychaetes**		
Sections tested		p-value	sections		p-value
Middle	First	0.118	Middle	First	-
End	Middle	> 0.25	End	Middle	> 0.25
End	First	0.087	End	First	> 0.25
9/1/2011					
Ascidians			**Barnacles**		
Sections		p-value	Sections		p-value
-	-	-	-	-	-
-	-	-	-	-	-
-	-	-	-	-	-
Bryozoans			**Polychaetes**		
Sections		p-value	Sections		p-value
Middle	First	> 0.25	Middle	First	0.021
End	Middle	> 0.25	Middle	End	> 0.25
End	First	> 0.25	End	First	> 0.25
9/15/2011					
Ascidians			**Barnacles**		
Sections		p-value	Sections		p-value
-	-	-	-	-	-
-	-	-	-	-	-
-	-	-	-	-	-
Bryozoans			**Polychaetes**		
Sections		p-value	Sections		p-value
Middle	First	> 0.25	Middle	First	-
End	Middle	> 0.25	End	Middle	> 0.25
End	First	0.208	End	First	-
9/29/2011					
Ascidians			**Barnacles**		
Sections		p-value	Sections		p-value
-	-	-	-	-	-
-	-	-	-	-	-
-	-	-	-	-	-
Bryozoans			**Polychaetes**		
Sections		p-value	Sections		p-value
Middle	First	0.194	-	-	-
Middle	End	0.167	End	Middle	-
End	First	> 0.25	End	First	-
10/13/2011					
Ascidians			**Barnacles**		
Sections		p-value	Sections		p-value
-	-	-	-	-	-
-	-	-	-	-	-
-	-	-	-	-	-
Bryozoans			**Polychaetes**		
Sections		p-value	Sections		p-value
First	Middle	> 0.25	-	-	-
Middle	End	0.143	-	-	-
First	End	0.040	-	-	-

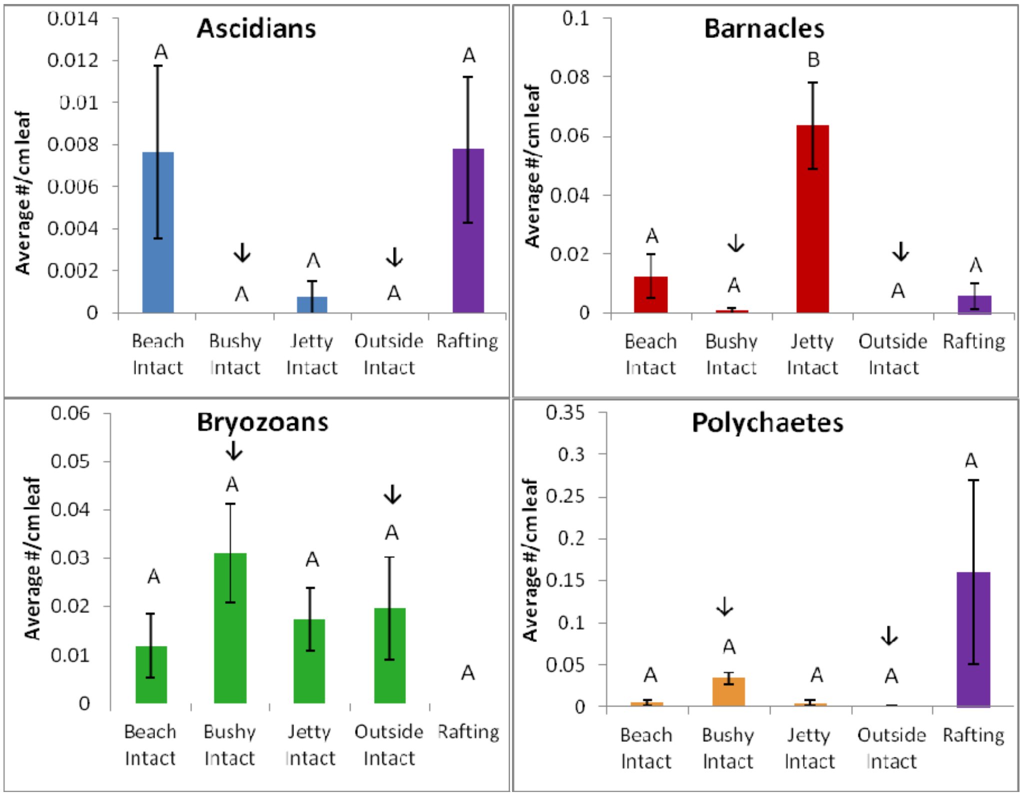

Figure 25. Average recruitment of fouling invertebrates on rafting leaves pooled, and intact leaves from the Beach, Bushy, Jetty and Outside sites on the 7/5/2011 collection date. Arrows indicate sites from which rafting leaves were collected. Error bars are ±1 standard error. Note different scales on y-axes.

Table 18. Results of analysis of variance for intact leaves at the Beach, Bushy, Jetty and Outside sites as compared to rafting leaves on the 7/5/2011 collection date.

Taxonomic Group	Tukey's-test Letter	One-way ANOVA p-value
Ascidians		
Intact Beach	A	0.049
Intact Bushy	A	
Intact Jetty	A	
Intact Outside	A	
Rafting	A	
Barnacles		
Intact Beach	A	<0.001
Intact Bushy	A	
Intact Jetty	B	
Intact Outside	A	
Rafting	A	
Bryozoans		
Intact Beach	A	0.092
Intact Bushy	A	
Intact Jetty	A	
Intact Outside	A	
Rafting	A	
Polychaetes		
Intact Beach	A	0.114
Intact Bushy	A	
Intact Jetty	A	
Intact Outside	A	
Rafting	A	

of being greater on rafting leaves as compared with intact leaves from all four sites, but the difference was not statistically significant.

Collection date: July 19, 2011

Rafting leaves (n = 8) were only collected from the Beach site on July 19, 2011 because no rafting leaves were observed at the other sites on this collection date. Recruitment abundances of ascidians on rafting leaves were significantly greater as compared to on intact leaves from the Bushy, Jetty and Outside sites, but not as compared to on intact leaves at the Beach site (Figure 26, Table 19). Tukey's test showed that barnacle recruitment abundances were not significantly different on rafting leaves as compared with any of the four sites, but the one-way un-stacked analysis of variance did indicate a significant difference (p-value = 0.38). Bryozoan recruitment abundances had significant differences among sites, but none of the four sites showed significant differences in recruitment abundances as compared with rafting leaves. Polychaete recruitment abundances showed clear significant differences between rafting leaves and intact leaves from all four sampling sites.

Collection date: August 16, 2011

Rafting leaves (n = 29) were collected from the Beach (n = 7), Bushy (n = 4) and Jetty (n = 18) sites on August 16, 2011. No rafting leaves were observed at the Outside site on this collection date. While a trend can be seen of greater abundances of ascidian recruitment on rafting leaves as compared with intact leaves from all four sites, the difference is not significant (Figure 27, Table 20). Barnacle recruitment abundances on intact leaves varied among sites, but were not significantly different as compared to rafting leaves. Bryozoan recruitment showed a trend toward greater abundances on rafting leaves as compared with intact leaves, but was also not significantly different. Polychaete recruitment was not significantly different on intact leaves from any of the four sites as compared with rafting leaves.

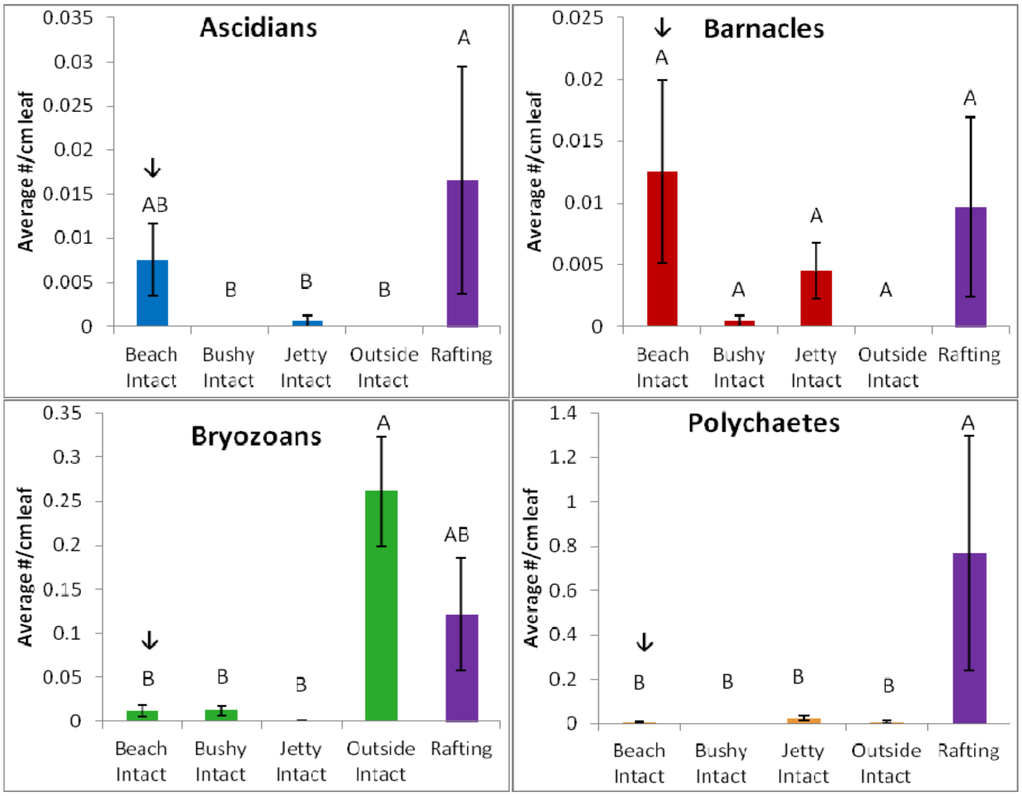

Figure 26. Average recruitment of fouling invertebrates on rafting leaves and intact leaves from the Beach site and pooled rafting leaves from the 7/19/2011 collection date. Arrows indicate sites from which rafting leaves were collected. Error bars are ±1 standard error. Note different scales on y-axes.

Table 19. Results of analysis of variance for intact leaves at the Beach site as compared to rafting leaves from 7/19/2011 collection date.

Taxonomic Group	Tukey's-test Letter	One-way ANOVA p-value
Ascidians		
Intact Beach	AB	0.002
Intact Bushy	B	
Intact Jetty	B	
Intact Outside	B	
Rafting	A	
Barnacles		
Intact Beach	A	0.038
Intact Bushy	A	
Intact Jetty	A	
Intact Outside	A	
Rafting	A	
Bryozoans		
Intact Beach	B	<0.001
Intact Bushy	B	
Intact Jetty	B	
Intact Outside	A	
Rafting	AB	
Polychaetes		
Intact Beach	B	<0.001
Intact Bushy	B	
Intact Jetty	B	
Intact Outside	B	
Rafting	A	

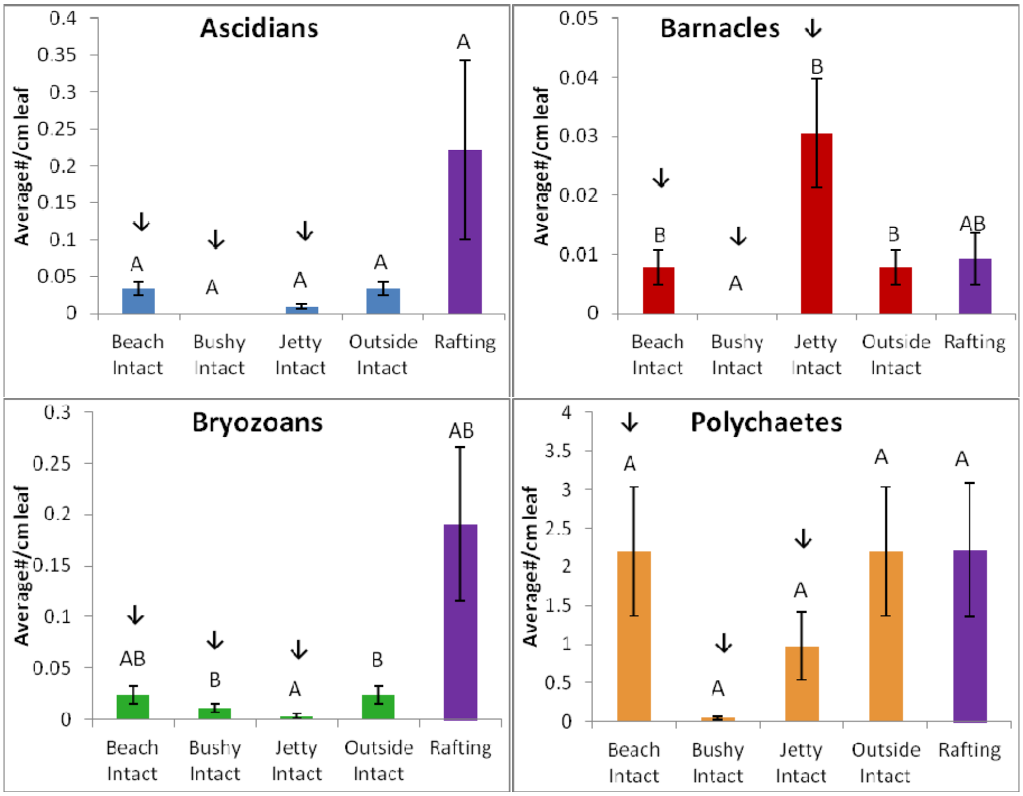

Figure 27. Average recruitment of fouling invertebrates on intact leaves from the Beach, Bushy, Jetty and Outside sites and pooled rafting leaves from the 8/16/2011 collection date. Arrows indicate sites from which rafting leaves were collected. Error bars are ±1 standard error. Note different scales on y-axes.

Table 20. Results of analysis of variance for intact leaves at the Beach, Bushy, Jetty and Outside sites as compared to pooled rafting leaves from the 8/16/2011 collection date.

Taxonomic Group	Tukey's-test Letter	One-way ANOVA p-value
Ascidians		
Intact Beach	A	0.106
Intact Bushy	A	
Intact Jetty	A	
Intact Outside	A	
Rafting	A	
Barnacles		
Intact Beach	B	0.003
Intact Bushy	A	
Intact Jetty	B	
Intact Outside	B	
Rafting	AB	
Bryozoans		
Intact Beach	AB	0.008
Intact Bushy	B	
Intact Jetty	A	
Intact Outside	B	
Rafting	AB	
Polychaetes		
Intact Beach	A	0.163
Intact Bushy	A	
Intact Jetty	A	
Intact Outside	A	
Rafting	A	

Collection date: September 1, 2011

Rafting leaves (n = 60) were collected from all four sites (Beach: n = 20, Bushy: n = 8, Jetty: n = 21, Outside: n = 11) on the September 1, 2011 collection date. For all four taxonomic groups, the Tukey's test indicated no significant differences in recruitment abundances between the intact leaves from any of the four sites and the rafting leaves (Table 21). However, there is a clear trend for all four taxa of greater recruitment on rafting leaves than intact leaves (Figure 28). One-way un-stacked analysis of variance resulted in p-values which indicated significant differences for both ascidians and polychaetes (Table 21).

Collection date: September 15, 2011

Rafting leaves (n = 60) were collected from the Beach (n = 20), Bushy (n = 20) and Jetty (n = 20) sites as there were no rafting leaves observed at the Outside site on the September 15, 2011 collection date. For all four taxonomic groups, the Tukey's test indicates no significant differences in recruitment abundances between the intact leaves from any of the three sites and the rafting leaves (Table 22). However, there is a clear trend for all four taxa of greater recruitment on rafting leaves than intact leaves (Figure 29). One-way un-stacked analysis of variance resulted in a p-value for barnacle recruitment which indicated significant differences (Table 22).

Collection date: September 29, 2011

Rafting leaves (n = 48) were collected from the Beach (n = 20), Bushy (n = 8) and Jetty (n = 20) sites as there were no rafting leaves observed at the Outside site on the September 29, 2011 collection date. For ascidians, barnacles and polychaetes, the Tukey's test indicated no significant differences in recruitment abundances between the intact leaves from any of the sites and the rafting leaves (Table 23). While not statistically significant, there is a clear trend for ascidians and polychaetes of greater recruitment on rafting leaves than intact leaves (Figure 30). Bryozoans showed significantly higher recruitment abundances on intact leaves from the Outside site than on rafting leaves.

Table 21. Results of analysis of variance for intact leaves at the Beach, Bushy, Jetty and Outside sites as compared to pooled rafting leaves from the 9/1/2011 collection date.

Taxonomic Group	Tukey's-test Letter	One-way ANOVA p-value
Ascidians		
Intact Beach	A	0.014
Intact Bushy	A	
Intact Jetty	A	
Intact Outside	A	
Rafting	A	
Barnacles		
Intact Beach	A	0.081
Intact Bushy	A	
Intact Jetty	A	
Intact Outside	A	
Rafting	A	
Bryozoans		
Intact Beach	A	0.514
Intact Bushy	A	
Intact Jetty	A	
Intact Outside	A	
Rafting	A	
Polychaetes		
Intact Beach	A	0.029
Intact Bushy	A	
Intact Jetty	A	
Intact Outside	A	
Rafting	A	

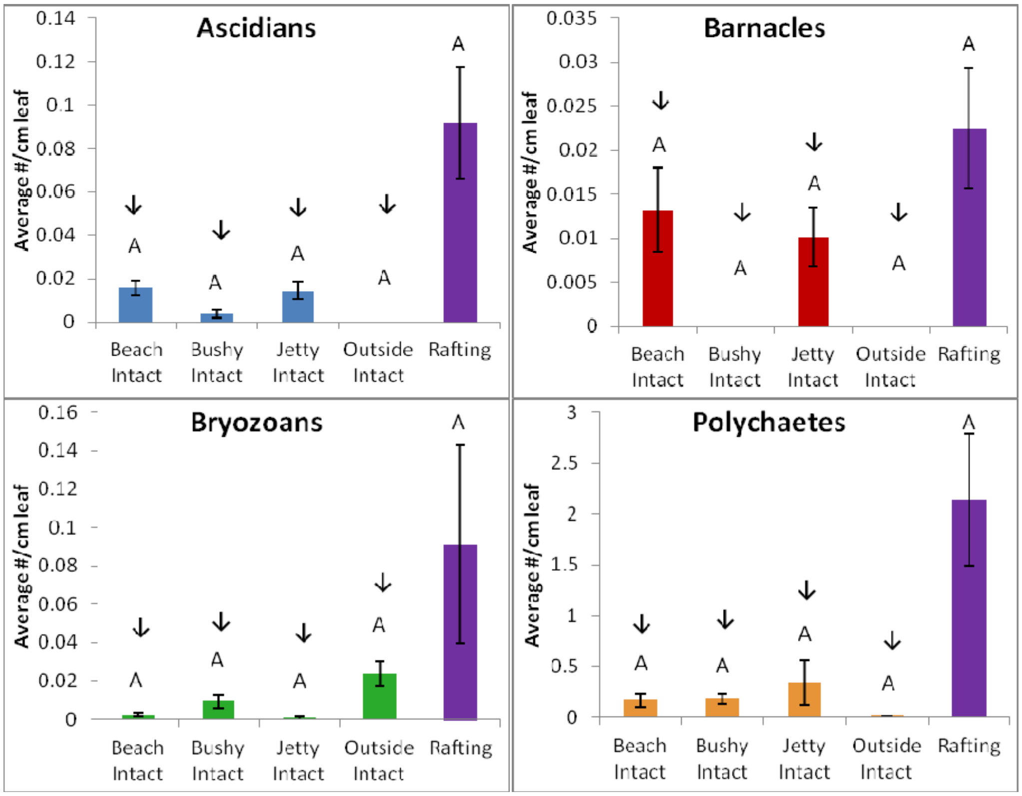

Figure 28. Average recruitment of fouling invertebrates on intact leaves from the Beach, Bushy, Jetty and Outside sites and pooled rafting leaves from all four sites on the 9/1/2011 collection date. Error bars are ±1 standard error. Note different scales on y-axes.

Table 22. Results of analysis of variance for intact leaves at the Beach, Bushy, Jetty and Outside sites as compared to pooled rafting leaves from the 9/15/2011 collection date.

Taxonomic Group	Tukey's-test Letter	One-way ANOVA p-value
Ascidians		
Intact Beach	A	0.060
Intact Bushy	A	
Intact Jetty	A	
Intact Outside	A	
Rafting	A	
Barnacles		
Intact Beach	A	0.011
Intact Bushy	A	
Intact Jetty	A	
Intact Outside	A	
Rafting	A	
Bryozoans		
Intact Beach	A	0.097
Intact Bushy	A	
Intact Jetty	A	
Intact Outside	A	
Rafting	A	
Polychaetes		
Intact Beach	A	0.483
Intact Bushy	A	
Intact Jetty	A	
Intact Outside	A	
Rafting	A	

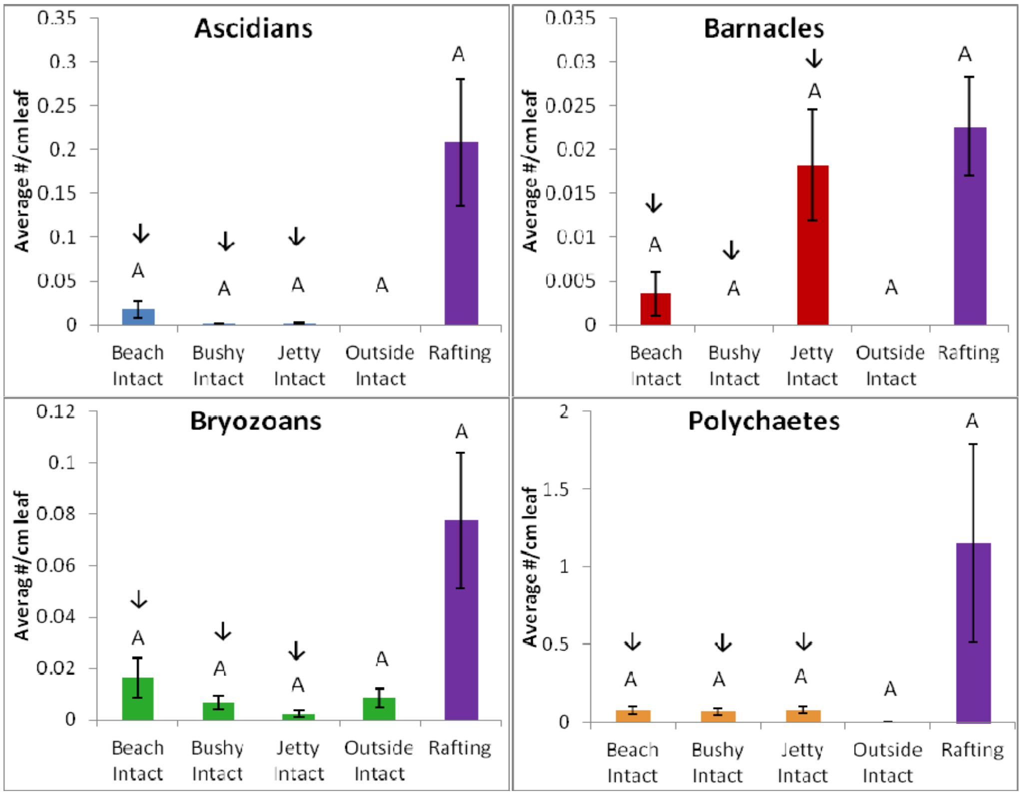

Figure 29. Average recruitment of fouling invertebrates on intact leaves from the Beach, Bushy and Jetty sites and pooled rafting leaves from the 9/15/2011 collection date. Arrows indicate sites from which rafting leaves were collected. Error bars are ±1 standard error. Note different scales on y-axes.

Table 23. Results of analysis of variance for intact leaves at the Beach, Bushy, Jetty and Outside sites as compared to pooled rafting leaves from the 9/29/2011 collection date.

Taxonomic Group	Tukey's-test Letter	One-way ANOVA p-value
Ascidians		
Intact Beach	A	0.106
Intact Bushy	A	
Intact Jetty	A	
Intact Outside	A	
Rafting	A	
Barnacles		
Intact Beach	A	0.615
Intact Bushy	A	
Intact Jetty	A	
Intact Outside	A	
Rafting	A	
Bryozoans		
Intact Beach	B	<0.001
Intact Bushy	B	
Intact Jetty	B	
Intact Outside	A	
Rafting	B	
Polychaetes		
Intact Beach	A	0.427
Intact Bushy	A	
Intact Jetty	A	
Intact Outside	A	
Rafting	A	

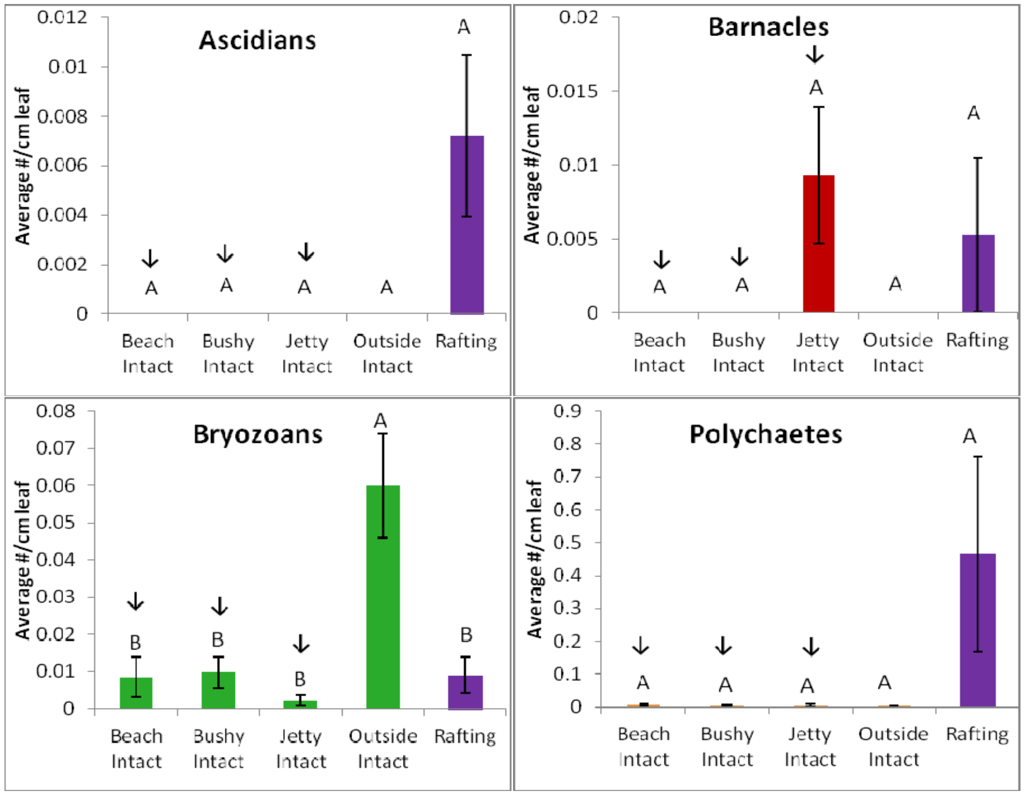

Figure 30. Average recruitment of fouling invertebrates on intact leaves from the Beach, Bushy, Jetty and Outside sites and pooled rafting leaves from the 9/29/2011 collection date. Arrows indicate sites from which rafting leaves were collected. Error bars are ±1 standard error. Note different scales on y-axes.

Collection date: October 13, 2011

Rafting leaves (n = 41) were collected from the Beach (n = 15), Bushy (n = 14) and Jetty (n = 12) sites as there were no rafting leaves observed at the Outside site on the October 13, 2011 collection date. For ascidians, bryozoans and polychaetes, the Tukey's test indicated no significant differences in recruitment abundances between the intact leaves from any of the sites and the rafting leaves (Table 24). While not statistically significant, there is a clear trend for all three taxa of greater recruitment on rafting leaves than intact leaves (Figure 31). Barnacles did not recruit on rafting leaves collected on this date and recruitment abundances on intact leaves from the Beach, Jetty and Outside sites were not significantly high.

Despite few significant differences between rafting leaves and intact leaves, trends toward greater recruitment on rafting leaves as compared with intact leaves can be seen in much of the data. All seven of the collection dates showed a trend toward greater recruitment on rafting leaves as compared with intact leaves for at least two of the taxonomic groups, with most of the collection dates (July 19, September 1, September 15, and October 13, 2011) exhibiting this trend for at least three of the four taxonomic groups. Ascidians showed this trend for all seven collection dates.

Hypothesis 5

It was hypothesized that recruitment of fouling invertebrates on eelgrass leaves would be similar to that on PVC panels, based on previous work with the fouling community that showed similar recruitment on PVC panels as on other hard substrates.

September 8, 2011 Panels

Species of fouling invertebrates were observed to have recruited onto panels that were not seen on eelgrass leaves (Table 25). Five species of ascidians, five species of bryozoans, one species of entoproct, one species of polychaete, and one species of sponge were observed to have

Table 24. Results of analysis of variance for intact leaves at the Beach, Bushy, Jetty and Outside sites as compared to pooled rafting leaves from the 10/13/2011 collection date.

Taxonomic Group	Tukey's-test Letter	One-way ANOVA p-value
Ascidians		
Intact Beach	A	0.778
Intact Bushy	A	
Intact Jetty	A	
Intact Outside	A	
Rafting	A	
Barnacles		
Intact Beach	A	0.052
Intact Bushy	A	
Intact Jetty	A	
Intact Outside	A	
Rafting	A	
Bryozoans		
Intact Beach	A	0.583
Intact Bushy	A	
Intact Jetty	A	
Intact Outside	A	
Rafting	A	
Polychaetes		
Intact Beach	A	0.775
Intact Bushy	A	
Intact Jetty	A	
Intact Outside	A	
Rafting	A	

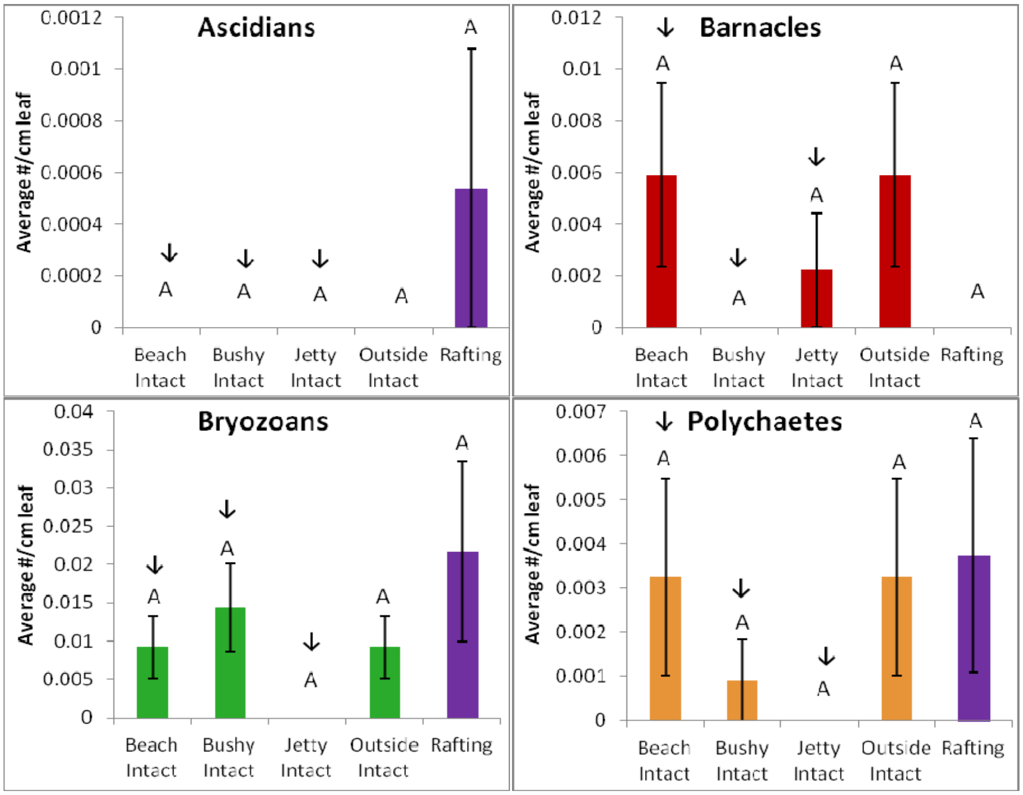

Figure 31. Average recruitment of fouling invertebrates on intact leaves from the Beach, Bushy, Jetty and Outside sites and pooled rafting leaves from the 10/13/2011 collection date. Arrows indicate sites from which rafting leaves were collected. Error bars are ±1 standard error. Note different scales on y-axes.

Table 25. Species found on eelgrass leaves collected 9/1/2011 and 9/15/2011 (pooled) and panels retrieved 9/8/2011, from the Beach, Bushy and Jetty sites. Sideward facing and downward facing panels are pooled.

Species:	Beach		Bushy		Jetty	
Ascidians	Leaves	Panels	leaves	Panels	Leaves	Panels
Aplidium sp.						●
Botrylloides violaceus	●	●	●	●	●	●
Botryllus schlosseri		●				●
Ciona intestinalis				●		
Didemnum vexillum		●		●		●
Diplosoma listerianum		●				●
Molgula manhattensis						●
Bryozoans						
Bowerbankia gracilis	●	●		●	●	●
Bugula neritina		●				●
Bugula turrita		●				●
Celleporaria hyalina			●	●	●	
Cryptosula pallasiana		●		●		●
Electra crustulenta					●	●
Membranipora sp.	●					
Microporella ciliata				●		●
Schizoporella errata						●
Barnacles						
Balanus sp.	●	●		●	●	●
Entoproct						
Barentsia sp.				●		●
Hydroids						
Obelia sp	●					
Polychaetes						
Hydroides dianthus				●		●
Spirorbis sp.	●	●	●	●	●	●
Sponges						
Halichondria sp.	●	●				●
Leucosolenia sp.				●		●

recruited onto panels that did not recruit on the eelgrass from the same site. There was also one species of bryozoan and one species of hydroid that recruited on eelgrass leaves but not on the panels.

Abundances of the four dominant taxonomic groups (ascidians, barnacles, bryozoans, and polychaetes) consistently demonstrated significant differences between collected leaves and PVC panels. There were also significant differences in recruitment abundances between sideward facing and downward facing panels.

Beach site (Figure 32, Table 26): Ascidian recruitment on collected leaves from both the September 1 and September 15, 2011 collection dates were significantly lower than on panels retrieved on September 8, 2011. Sideward facing panels also showed significantly lower recruitment abundances of ascidians as compared to downward facing panels. Barnacle recruitment showed no significant differences between leaves collected on either date or sideward facing panels. Down-ward-facing panels showed significantly greater recruitment abundances of barnacles than both collected leaves and sideward facing panels. Bryozoan recruitment abundances on leaves collected on September 15, 2011 and the side-ward facing panels showed no significant differences as compared to leaves collected on September 1, 2011 or downward facing panels. However, leaves collected on September 1, 2011 showed significantly lower abundances as compared with the downward facing panels. Leaves collected on both dates showed no significant differences in polychaete recruitment abundances. Polychaete recruitment on sideward facing panels was significantly higher than on collected leaves, and recruitment on downward facing panels was significantly higher than on both sideward facing panels and collected leaves.

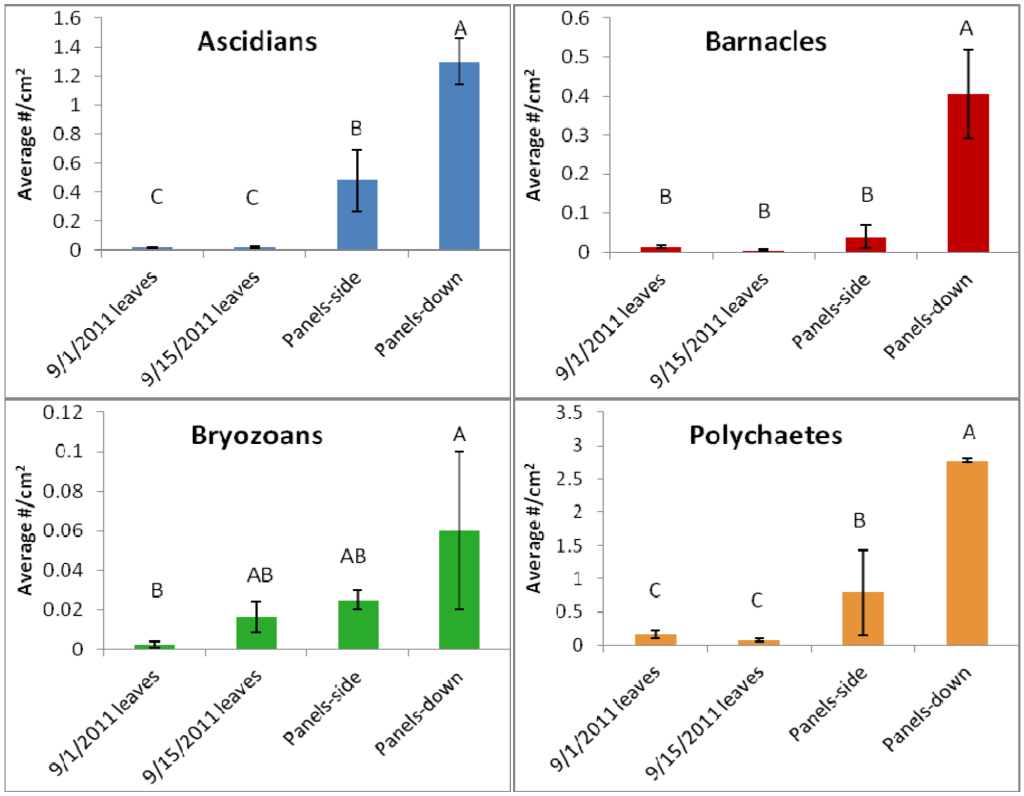

Figure 32. Average recruitment abundances on collected leaves from the 9/1/2011 and 9/15/2011 collection dates, on sideward facing panels and downward facing panels retrieved 9/8/2011, from the Beach site. Letters indicate significant differences. Error bars are ±1 standard error. Note differences in y-axes.

Table 26. Results of analysis of variance for recruitment abundances on leaves from the 9/1/2011 and 9/15/2011 collection dates as compared to sideward facing panels and downward facing panels retrieved on 9/8/2011 from the Beach site.

Taxonomic Group	Tukey's-test Letter	One-way ANOVA p-value
Ascidians		
9/1/2011 leaves	C	<0.001
9/15/2011 leaves	C	
Panels- side	B	
Panels- down	A	
Barnacles		
9/1/2011 leaves	B	<0.001
9/15/2011 leaves	B	
Panels- side	B	
Panels- down	A	
Bryozoans		
9/1/2011 leaves	B	0.019
9/15/2011 leaves	AB	
Panels- side	AB	
Panels- down	A	
Polychaetes		
9/1/2011 leaves	C	<0.001
9/15/2011 leaves	C	
Panels- side	B	
Panels- down	A	

Bushy site (Figure 33, Table 27): Ascidian recruitment abundances showed no differences between leaves collected on either the September 1 or the September 15, 2011 collection dates. There were also no differences in ascidian recruitment abundances between sideward facing and downward facing panels. There was significantly greater recruitment of ascidians on panels as compared with collected leaves. Barnacle recruitment was significantly greater on sideward facing panels as compared with collected leaves and downward facing panels. Barnacles did not recruit on collected leaves from either date or on the downward facing panels. There were no differences in recruitment abundances of bryozoans between collected leaves and sideward facing panels. Downward facing panels had significantly greater bryozoan recruitment as compared with both collected leaves and sideward facing panels. There were no significant differences in polychaete recruitment between collected leaves and panels oriented either way.

Jetty site (Figure 34, Table 28)**:** Ascidian recruitment abundances were significantly greater on panels than on collected leaves. There were also significantly greater abundances on downward facing panels as compared with sideward facing panels. Barnacle recruitment also showed significantly greater abundances on panels as compared with collected leaves but there were no significant differences in barnacle recruitment abundance between the orientations of the panels. Bryozoan recruitment was higher on panels as compared with collected leaves and downward facing panels showed significantly greater abundances as compared with sideward facing panels. Polychaete recruitment showed significantly greater abundances on panels as compared with collected leaves, with significantly higher recruitment on downward facing panels as compared with sideward facing panels.

October 13, 2011 Panels

Similarly to the September 8, 2011 panels, the panels that were retrieved on October 13, 2011 showed recruitment of fouling invertebrates that were not observed on the eelgrass leaves that were collected on the same date (Table 29). Six species of ascidians, seven species of bryozoans, one species of entoproct, one species of polychaete, and one species of sponge

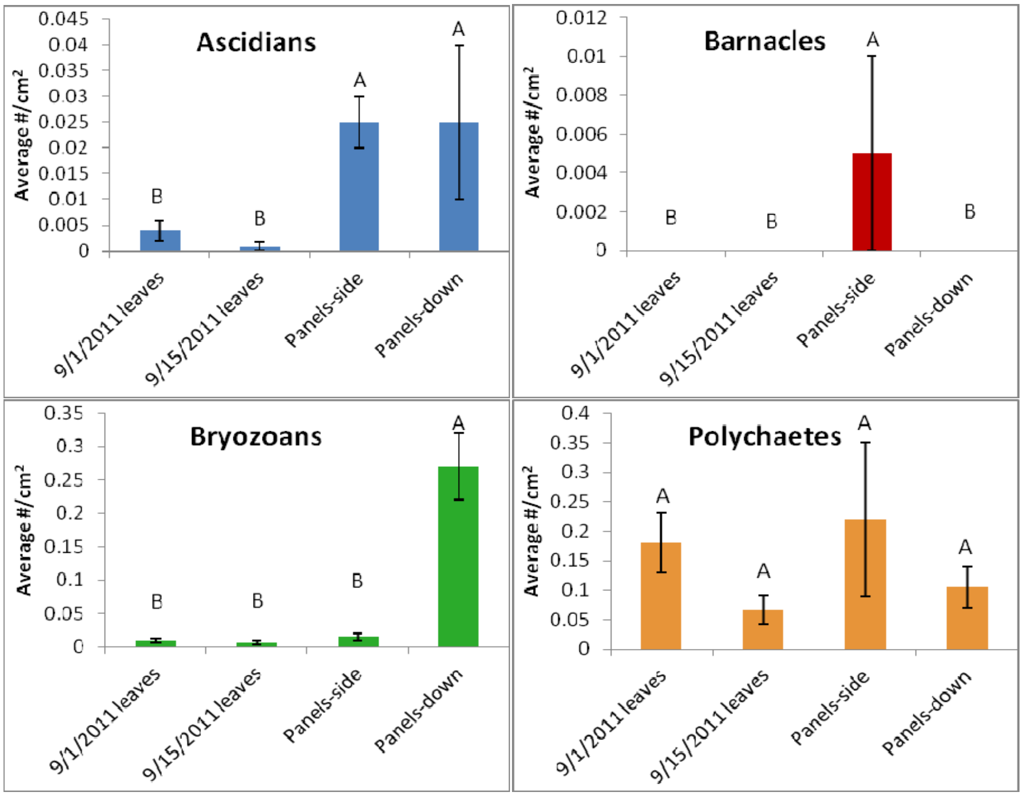

Figure 33. Average recruitment abundances on collected leaves from the 9/1/2011 and 9/15/2011 collection dates, on sideward facing panels and downward facing panels retrieved on 9/8/2011, from the Bushy site. Letters indicate significant differences. Error bars are ±1 standard error. Note differences in y-axes.

Table 27. Results of analysis of variance for recruitment abundances on leaves from the 9/1/2011 and 9/15/2011 collection dates as compared to sideward facing panels and downward facing panels retrieved on 9/8/2011 from the Bushy site.

Taxonomic Group	Tukey's-test Letter	One-way ANOVA p-value
Ascidians		
9/1/2011 leaves	B	<0.001
9/15/2011 leaves	B	
Panels- side	A	
Panels- down	A	
Barnacles		
9/1/2011 leaves	B	<0.001
9/15/2011 leaves	B	
Panels- side	A	
Panels- down	B	
Bryozoans		
9/1/2011 leaves	B	<0.001
9/15/2011 leaves	B	
Panels- side	B	
Panels- down	A	
Polychaetes		
9/1/2011 leaves	A	0.199
9/15/2011 leaves	A	
Panels- side	A	
Panels- down	A	

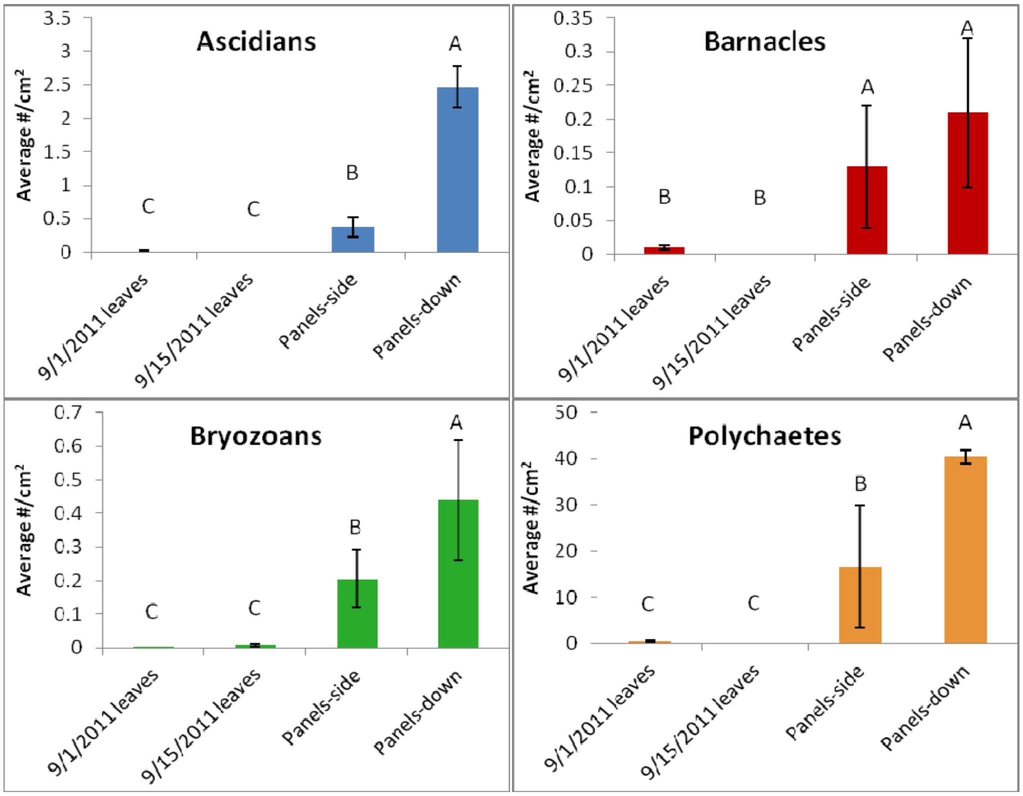

Figure 34. Average recruitment abundances on collected leaves from the 9/1/2011 and 9/15/2011 collection dates, on sideward facing panels and downward facing panels retrieved 9/8/2011, from the Jetty site. Letters indicate significant differences. Error bars are ±1 standard error. Note differences in y-axes.

Table 28. Results of analysis of variance for recruitment abundances on leaves from the 9/1/2011 and 9/15/2011 collection dates as compared to sideward facing panels and downward facing panels retrieved on 9/8/2011 from the Jetty site.

Taxonomic Group	Tukey's-test Letter	One-way ANOVA p-value
Ascidians		
9/1/2011 leaves	C	<0.001
9/15/2011 leaves	C	
Panels- side	B	
Panels- down	A	
Barnacles		
9/1/2011 leaves	B	<0.001
9/15/2011 leaves	B	
Panels- side	A	
Panels- down	A	
Bryozoans		
9/1/2011 leaves	C	<0.001
9/15/2011 leaves	C	
Panels- side	B	
Panels- down	A	
Polychaetes		
9/1/2011 leaves	C	<0.001
9/15/2011 leaves	C	
Panels- side	B	
Panels- down	A	

Table 29. Species found on collected leaves and PVC panels retrieved on 10/13/2011.

Species:	Beach		Bushy		Jetty	
Ascidians	Leaves	Panels	leaves	Panels	Leaves	Panels
Aplidium sp.						●
Botrylloides violaceus				●		
Botryllus schlosseri		●				
Didemnum vexillum		●		●		●
Diplosoma listerianum						●
Molgula manhattensis						●
Bryozoans						
Bowerbankia gracilis	●	●		●		
Bugula neritina						●
Bugula turrita						●
Celleporaria hyalina		●	●	●		
Cryptosula pallasiana		●		●		●
Electra crustulenta						●
Electra pilosa	●					
Schizoporella errata		●		●		●
Barnacles						
Balanus sp.	●	●		●	●	●
Entoproct						
Barentsia sp.				●		●
Hydroids						
Obelia sp	●					
Polychaetes						
Hydroides dianthus		●		●		●
Spirorbis sp.	●	●	●	●		●
Sponges						
Halichondria sp.	●					●

recruited on panels but were not observed on leaves from the same site. Found on leaves but not on panels of the same site, one species of bryozoan, one species of hydroid and one species of sponge were observed.

Beach site (Figure 35, Table 30): No ascidian recruitment was observed on leaves collected on October 13, 2011 and recruitment on sideward facing panels was not significantly different. Significantly greater recruitment of ascidians was observed on downward facing panels as compared with both collected leaves and sideward facing panels. There were significantly greater recruitment abundances of barnacles on panels as compared with collected leaves, and significantly more on downward facing panels as compared with sideward facing panels. Bryozoan recruitment abundances were not significantly different between collected leaves and panels or between the differently oriented panels. Polychaete recruitment was not observed on collected leaves or sideward facing panels, while significant abundances were observed on downward facing panels.

Bushy site (Figure 36, Table 31): No recruitment of ascidians was observed on collected leaves or on sideward facing panels from the Bushy site on October 13, 2011. Ascidians recruitment was observed on downward facing panels in significant abundances. Barnacle recruitment was not observed on collected leaves, but was observed in significant abundances on both orientations of panels, with significantly greater abundances on downward facing as compared with sideward facing panels. Bryozoan recruitment showed significantly greater recruitment abundances on sideward facing panels as compared with collected leaves. Bryozoan recruitment abundances on downward facing panels were not significantly different as compared with collected leaves, or as compared with sideward facing panels. Polychaete recruitment abundances were significantly greater on panels as compared with collected leaves. Downward facing panels showed significantly greater recruitment abundances of polychaetes as compared with sideward facing panels.

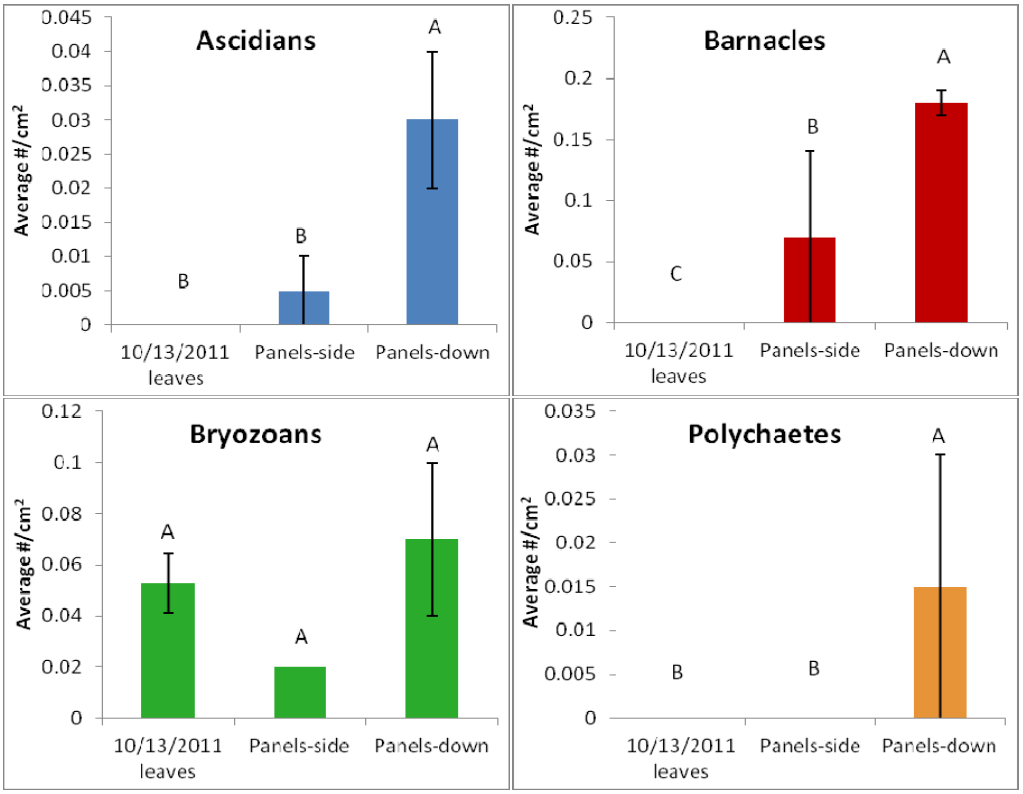

Figure 35. Average recruitment abundances on collected leaves on 10/13/2011, on sideward facing panels and downward facing panels retrieved 10/13/2011, from the Beach site. Letters indicate significant differences. Error bars are ±1 standard error. Note differences in y-axes.

Table 30. Results of analysis of variance for recruitment abundances on leaves from the 10/13/2011 collection date as compared to sideward facing panels and downward facing panels retrieved on 10/13/2011 from the Beach site.

Taxonomic Group	Tukey's-test Letter	One-way ANOVA p-value
Ascidians		
10/13/2011 leaves	B	<0.001
Panels- side	B	
Panels- down	A	
Barnacles		
10/13/2011 leaves	C	<0.001
Panels- side	B	
Panels- down	A	
Bryozoans		
10/13/2011 leaves	A	0.623
Panels- side	A	
Panels- down	A	
Polychaetes		
10/13/2011 leaves	B	0.001
Panels- side	B	
Panels- down	A	

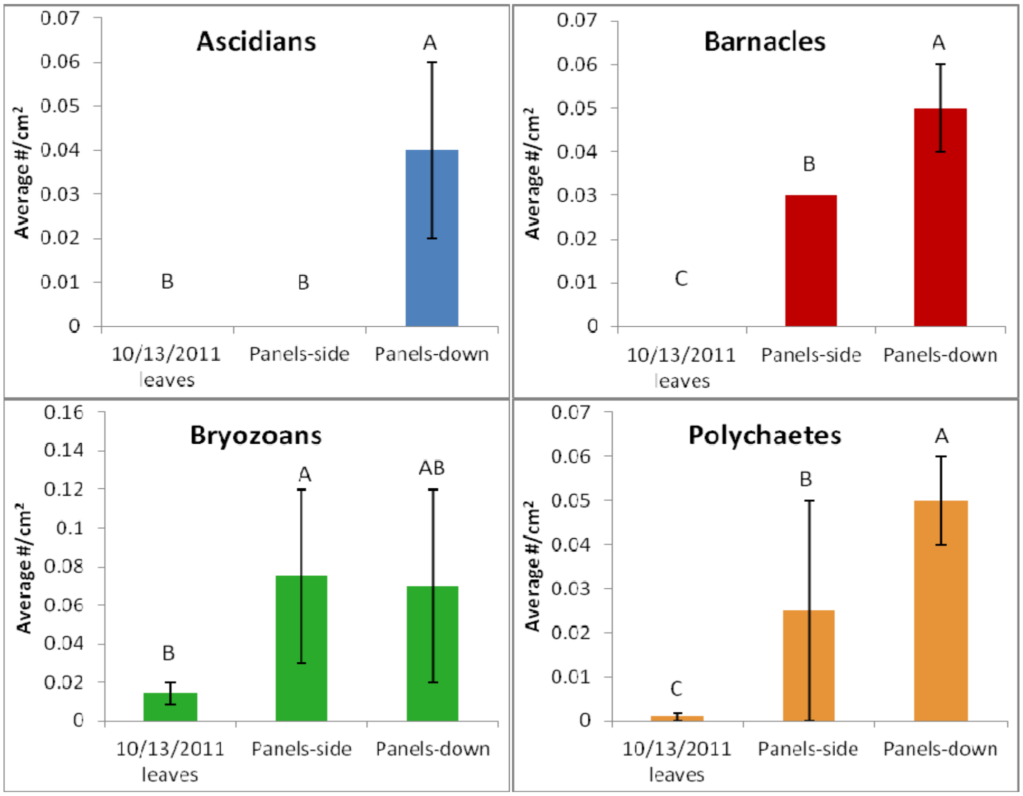

Figure 36. Average recruitment abundances on collected leaves from 10/13/2011, on sideward facing panels and downward facing panels retrieved 10/13/2011, from the Bushy site. Letters indicate significant differences. Error bars are ±1 standard error. Note differences in y-axes.

Table 31. Results of analysis of variance for recruitment abundances on leaves from the 10/13/2011 collection date as compared to sideward facing panels and downward facing panels retrieved on 10/13/2011 from the Bushy site.

Taxonomic Group	Tukey's-test Letter	One-way ANOVA p-value
Ascidians		
10/13/2011 leaves	B	<0.001
Panels- side	B	
Panels- down	A	
Barnacles		
10/13/2011 leaves	C	<0.001
Panels- side	B	
Panels- down	A	
Bryozoans		
10/13/2011 leaves	B	0.012
Panels- side	A	
Panels- down	AB	
Polychaetes		
10/13/2011 leaves	C	<0.001
Panels- side	B	
Panels- down	A	

Jetty site (Figure 37, Table 32): Recruitment of ascidians was not observed on leaves collected at the Jetty site on October 13, 2011. Significant abundances of ascidians recruited on both orientations of panels, with no differences between them. Barnacle recruitment was significantly greater on panels as compared with collected leaves, and significantly greater on downward facing panels as compared with sideward facing panels. Neither bryozoans nor polychaetes were observed to have recruited on collected leaves while significant abundances were observed to have recruited on panels. Significantly greater abundances recruited on downward facing panels as compared with sideward facing panels for both bryozoans and polychaetes.

Overall, greater recruitment abundances were seen on PVC panels as compared with collected eelgrass leaves. Recruitment abundances were consistently greater on downward facing panels as compared with sideward facing panels. More species were also observed to have recruited on panels than were observed on collected eelgrass leaves.

Discussion

Hypothesis 1 was supported by the data: recruitment on eelgrass leaves varied spatially and temporally. Spatially, a number of trends emerged. The two sites located between Pine Island and the mainland, the Beach site and the Jetty site, showed similarities in recruitment of ascidians and polychaetes, with polychaetes being dominant at both of these sites. The ascidian, *B. schlosseri,* was found on the eelgrass leaves at the Beach and Jetty sites but not at the Bushy and Outside sites. Due to the location of the Beach and Jetty sites on the leeward side of the island, these sites are sheltered from prevailing wind and waves by the island itself. The Jetty site is further protected by the presence of the stone jetty. More recruitment of barnacles was seen at the Jetty site than any of the other sites, suggesting that the protected conditions of calmer water promotes recruitment of barnacle larvae. The Jetty site was the only site at which the ascidian, *M. manhattensis* was found on the eelgrass leaves, again suggesting that the conditions at the Jetty site, unlike the other three sites, was conducive the this species' settlement. The Bushy site and

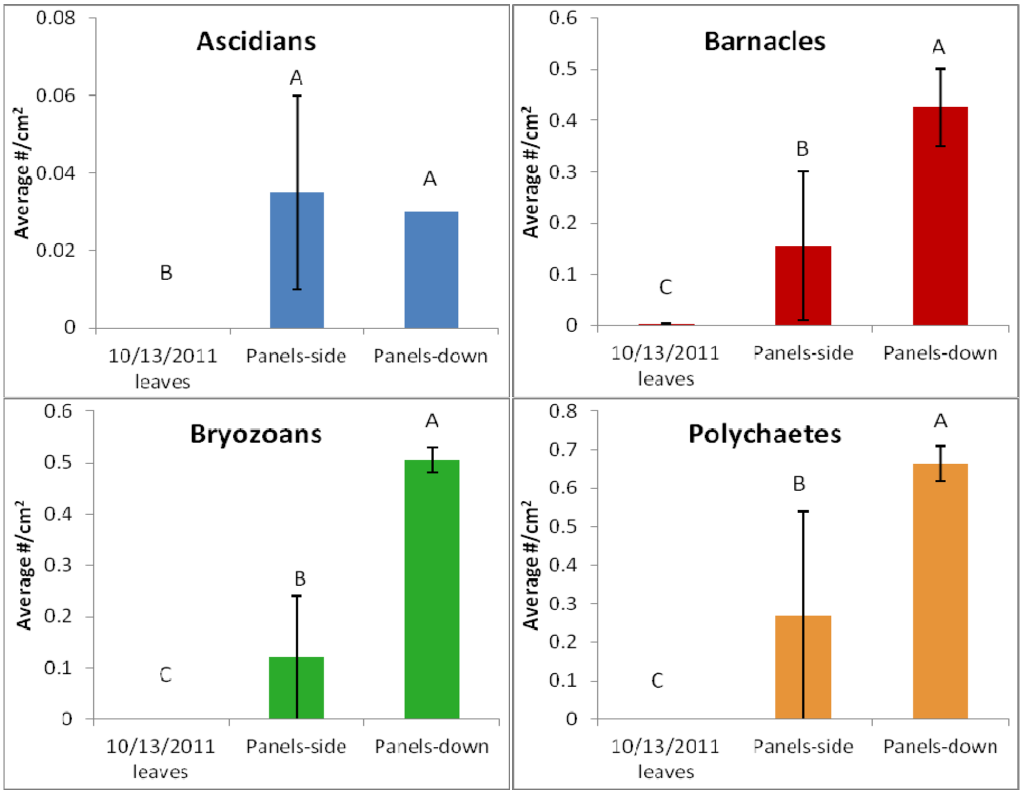

Figure 37. Average recruitment abundances on collected leaves from 10/13/2011, on sideward facing panels and downward facing panels retrieved 10/13/2011, from the Jetty site. Letters indicate significant differences. Error bars are ±1 standard error. Note differences in y-axes.

Table 32. Results of analysis of variance for recruitment abundances on leaves from the 10/13/2011 collection date as compared to sideward facing panels and downward facing panels retrieved on 10/13/2011 from the Jetty site.

Taxonomic Group	Tukey's-test Letter	One-way ANOVA p-value
Ascidians		
10/13/2011 leaves	B	<0.001
Panels- side	A	
Panels- down	A	
Barnacles		
10/13/2011 leaves	C	<0.001
Panels- side	B	
Panels- down	A	
Bryozoans		
10/13/2011 leaves	C	<0.001
Panels- side	B	
Panels- down	A	
Polychaetes		
10/13/2011 leaves	C	<0.001
Panels- side	B	
Panels- down	A	

the Outside site, both located on the windward side of Pine Island, show very low recruitment of ascidians and polychaetes. The Outside site experienced the highest energy of the sampling sites and experienced primarily bryozoan recruitment. Recruitment of all taxa at the Bushy site was relatively lower than the other collection sites

These results are in agreement with Munguia et al. (2011) who also found more protected sites to have higher abundances of ascidians while more exposed sites were dominated by bryozoans. They attributed these differences to the presence of different combinations of both physical and biological stressors (Osman et al. 2010). Such stressors might include contributing physical factors such as water flow and substrate disturbance and biological factors including interspecies competition and predation, combined to produce unique conditions at each site. More work is needed to specifically identify the factors at individual sites that contribute to the dominance of one group over another in these particular systems.

Temporally, different patterns emerged at different sites. All four taxonomic groups showed peaks in recruitment on three different dates among the four sites, with no correlation between them. At the Beach site, ascidians and polychaetes showed clear patterns of recruitment, with a rise, a peak and then decline in recruitment. These results follow a normal pattern in recruitment similar to patterns seen in other studies of fouling community larval settlement (Altman &Whitlatch, 2007, Freestone et al., 2009).

Barnacle and bryozoan recruitment at the Beach site was much more erratic. These two groups show several increases and decreases over the course of the short sampling period. This suggests that there are variable influences affecting these organisms which have less of an impact on ascidians and polychaetes. One example of a variable impact is predation (Osman &Whitlatch 1996, 1998). The presence of predators in the ecosystem can be variable and patchily affect larval settlement. The protected nature of the Beach site allows for the presence of snails and other predators that might be present at lower abundances at the higher energy sites.

While recruitment was overall lowest at the Bushy site, a distinct peak in polychaete recruitment can nonetheless be seen on the September 1, 2011 collection date. This peak is not concurrent with maximum polychaete recruitment at the other sites. The other three collection sites showed peaks in polychaete recruitment on earlier collection dates, although not synchronously-- the Beach and Outside sites showed maximum in polychaete recruitment on August 16, 2011 while Jetty peaked on July 19, 2011. Larval supply is a hypothesized explanation for this variability. If the recruiting larvae at the different sites are being supplied from different sources with asynchronous spawning events, then peaks in recruitment would be staggered, as seen in this study. Again, further work is needed to identify the factors contributing to this variability.

At the Jetty site, all four taxonomic groups showed the same pattern of settlement, albeit in different abundances. A simultaneous peak in recruitment was on the July 19, 2011 collection date, followed by drastically lower abundances on subsequent collection dates. The Jetty site is the most protected of the four sampling sites. There is limited water flow and little to no wave action present at this site. The stillness of the water prevents free swimming larvae from being washed away before settling. As a result, eelgrass leaves collected at this site showed the highest overall settlement.

Just as the sessile invertebrates are protected from water flow at the Jetty site, so are predators. Organisms which prey upon the sessile fouling organisms may have experienced a peak just following the peak in sessile recruitment, due to the relative abundance of prey. An increase in predator abundance would subsequently reduce the numbers of recruits present on the leaves. While predation was not the focus of this study, higher numbers of snails were observed on the eelgrass leaves at the Jetty than any of the other sites, though such observations, including species present, were not recorded. Further study would involve cages with and without predators and their effects on fouling recruitment on eelgrass. Such studies using recruitment on

PVC panels found significant impacts of *Anachis sp.* and *Mitrella lunata* predation (Osman &Whitlatch 1996, 1998).

The Outside site showed little to no recruitment of ascidians or barnacles and was dominated by recruitment of bryozoans and polychaetes. Both groups showed maximum recruitment on the July 19, 2011 collection date, followed by decline. The dominance of these two taxonomic groups over the ascidians and bryozoans is likely due to the high energy nature of the collection site. The Outside site is the most exposed of the four collection sites, and water flow is likely the biggest factor in low recruitment abundances. With high water flow (~60cm/sec, Osman & Whitlatch, 1998), larvae are more likely to be swept away by the current before settling on the eelgrass leaves.

While bryozoans were the dominant taxonomic group at the Outside site, the species which was dominant shifted throughout the study. This suggests asynchronous spawning events of the larval sources for the different species. *M. ciliata, E. pilosa, C. hyalina,* and *B. gracilis* all in turn dominated bryozoan recruitment at the Outside site only to experience little to no recruitment before and after the time period in which they were dominant. For example, at the peak of bryozoan recruitment, the July 19, 2011 collection date, *E. pilosa* accounted for 95.5% of observed recruitment but accounted for 7% or less on all other collection dates.

More bryozoan species were observed on eelgrass leaves than species of other taxonomic groups examined. This is the only taxa then, for which species-level patterns emerged. As discussed at the Outside site, abundances of different species of bryozoans at all four sites shifted throughout the sampling period, with different species being dominant on different sampling dates. This suggests staggered spawning events for the different species observed. For example, at all four sites, *M. ciliata* was observed on eelgrass leaves on the July 5, 2011 collection date, and not on any of the subsequent collection dates, consistent among the sites. This suggests that spawning of this species occurred earlier in the season than other species observed and was no longer recruiting by the second collection date. *B. gracilis* was not observed on the eelgrass leaves until the August 16, 2011 collection date, suggesting that spawning of this species began somewhere between the July 19 and August 16, 2011 collection dates.

Hypothesis 2, that higher recruitment would be seen on outer than inner leaves, was partially supported by the data. When collection dates and sites were pooled, outer leaves had significantly greater recruitment for barnacles, bryozoans and polychaetes. When separated by site and time, however, the overwhelming majority of the data showed no significant differences between recruitment on inner verses outer leaves. While there were no overall patterns with respect to increased recruitment on inner or outer leaves, closer examination of individual taxa allow for possible patterns to emerge. For example, barnacle recruitment at the Jetty site showed even recruitment on inner and outer leaves on all collection dates through September 15, 2011. The following two collection dates, September 29 and October 13, 2011 showed barnacle recruitment on outer but not inner leaves. This pattern lends itself to the idea that barnacle recruitment at the Jetty site was indiscriminant between inner and outer leaves through September 15, 2011 and new recruitment stopped entirely between the September 15 and September 29, 2011 collection dates. Barnacles that were observed on outer leaves on the last two collection dates had already recruited on leaves that were inner leaves at the time of settlement but had become outer leaves by the time of collection through the natural succession of the leaves of the eelgrass shoot (see Figure 2). There is no way of determining the age of any single eelgrass leaf, nor of a post-metamorphosed barnacle. This hypothesis could be tested through careful monitoring of a single eelgrass shoot from time of initial barnacle settlement through the succession of leaves from inner to outer positions.

Similar patterns can be deduced with other taxa and at other sites: during higher settling periods, no significant differences are seen in recruitment between inner and outer leaves implying that larvae are settling indiscriminately with regard to inner or outer leaves. Later collections, after recruitment has dropped, greater recruitment is seen on outer than inner leaves. This pattern supports the idea that post peak settlement, recruits that are seen on outer leaves are those which settled on inner leaves during peak recruitment which subsequently became outer leaves through natural leaf succession. This idea is supported by the rate at which leaf succession occurs, which can range from 7-17 days for eelgrass in New England at this time of year (Gaeckle & Short, 2002). While this recruitment pattern may indeed be the case, the differences between inner and outer leaves are not high enough to influence the pooling of the data that was used in testing of hypotheses 1, 4, and 5.

Hypothesis 3, that recruitment would increase along the length of the leaf, was partially supported. While there was greater recruitment on middle and end than first sections, there was little difference between middle and end sections.

Leaves grow from the base, causing the first sections to be the youngest and end sections of leaves are the oldest. The youngest sections would have been exposed to settlement for the least amount of time. The ages of the respective sections of leaf are relative as leaf age cannot be determined without tracking over time (Gaeckle & Short, 2002). Leaf length and therefore age of sections has to do with nutrients, light availability, current and disturbance (Dennison, 1987, Koch & Beer, 1996).

Middle section of an outer leaf of a shoot could be the same age as the end section of an adjacent leaf on the inside of the same shoot. These sections would be exposed to the same highs and lows of larval supply and recruitment, resulting in the lack of difference seen between middle and end sections.

The first sections consistently had little to no recruitment. These are the youngest sections of the leaves and are somewhat protected by the outer leaves of the shoot. The combination of experiencing some shielding from outer leaves of the shoot and being exposed to settlement for the shortest amount of time accounts for the lowest levels of recruitment seen on the first sections. As the leaf grows, first sections become middle sections as new first sections grow. As leaves grow, enough time allows for recruitment to occur.

While it was not measured or recorded, more photosynthetic epiphytes were observed on middle and end sections of the leaves. This can be accounted for by sheltering of inner leaves by outer leaves and time exposed for epiphytic growth, as with the fouling invertebrates. The coverage by photosynthetic epiphytes and fouling invertebrates in combination has implications for the eelgrass. In combination, the heavier load of organisms growing on the leaves have the potential to reduce light availability for photosynthesis. The older sections of the leaves are higher in the water column and should be experiencing the greatest amount of light. The presence of these organisms on the leaves, however, are reducing the amount of light available to the very portions of the leaf that should be experiencing the greatest amount of light. The younger sections of the leaf that experience the least amount of fouling and epiphytes are also lowest in the water column and experience the lowest levels of light, being shaded by the leaf canopy. Therefore, the sections of the leaf that should be experiencing the greatest light levels are also the most highly impacted by organisms on the leaf surface.

Simultaneously, the longer the leaf grows, and therefore more photosynthetic productivity potential it has the greater the surface it provides for fouling invertebrates. During peak times of settlement, greater surface availability reduces the competition for larval settlement. Greater amount of leaf surface might increase total numbers of settled larvae while reducing the density of recruitment. This is advantageous for both fouling invertebrates and eelgrass in that a smaller percentage of leaf is covered by fouling recruits as well as lower competition for the invertebrate larvae. The peak in larval recruitment coincides with the height of the eelgrass growing season. Whether or not the peak in available substrate on eelgrass leaves contributed to the evolution of local larval settlement peak timing cannot be determined.

Hypothesis 4, that rafting leaves would show higher levels of recruitment than intact leaves, was not supported by the data. In most instances, rafting leaves showed a trend of higher levels of recruitment than intact leaves, although the differences were rarely significant. This result was particularly consistent for ascidians and polychaetes.

The close proximity of the four sites might allow for rafting leaves to move between the sites and other eelgrass beds with currents and the tide. Thus, all rafting leaves were pooled for analysis and compared to all four sites. However, it can be noted that for a number of cases, the site that showed significant differences in recruitment abundances for one or more taxa as compared with abundances on rafting leaves are indeed the sites from which rafting leaves were not collected (July 5, 2011 barnacles, July 19, 2011 ascidians and bryozoans, September 29, 2011 bryozoans). This suggests that rafting leaves were likely from the sites at which they were collected.

Jetty is the most protected site and is least likely to have rafting leaves exported to the other sites, due to the physical barrier caused by the presence of a stone Jetty and the direction of the currents. The Jetty site also had the most rafting leaves. These leaves get caught in the still waters around and on the stone jetty at the site. On most collection dates, few or no rafting leaves were collected from Outside. This has the highest energy of the four sites, and rarely were rafting leaves seen. The higher current of Outside likely carries away rafting leaves quickly.

Rafting leaves are most often the older, outer leaves that have been sloughed off by the plant. In the succession of leaves of an eelgrass shoot, the outer leaves are shed as they are replaced by younger leaves from the inside of the shoot. How long a rafting blade has been floating at the surface cannot be determined. It is possible that some leaves spend a greater amount of time rafting than as part of an intact plant, particularly in protected areas like the Jetty site where they are not being transported by the current. This is supported by the percentages of the rafting leaves that were no longer photosynthetically viable (brown in color-noted in data collection, not presented in results). The rafting leaves collected at the Jetty were generally 28.8-94.5% brown, which suggests a substantial amount of time since detaching from the plant.

Having been exposed to recruitment while part of intact plants and later while rafting was the basis upon which Hypothesis 4 was originated, supposing that this increased exposure to recruitment could contribute to the higher abundances of fouling invertebrates observed on rafting than intact leaves. As this hypothesis was not supported by the data, it must be considered why this was not the case. Rafting leaves perhaps are less likely to be in proximity of sources of larvae. Intact plants adjacent to rocks and other larval sources might be more likely to experience recruitment than rafting leaves that are carried away from such sources by current.

Rafting leaves could also be compared with results of Hypothesis 5 that PVC panels had greater recruitment on downward than sideward facing panels. The orientation of rafting leaves in the water is more similar to the downward facing panels in that rafting leaves have a downward facing surface which would also suggest that they would accrue additional recruits once detached from the plant. Waves and currents, however, can change the orientation of the leaves in the water so that the same face of the leaf is not always facing downward. If direct sunlight or the indirect effects of increased sunlight (such as competition from photosynthetic epiphytes) has a negative effect on the settlement of fouling larvae, then any change in the orientation of the leaf in the water would increase the negative effects of upward orientation on the leaf face while increasing positive effects of downward facing on the opposite leaf face. Further study is needed to clarify this issue.

Rafting leaves have been shown to be a mechanism of dispersal for a number of organisms (Carman & Grunden 2010, Petersen & Svane, 1995). Rafting leaves can transport larvae between sites and over long distances (Worcester, 1994). Carman & Grunden (2010) found ascidians attached to rafting eelgrass leaves in Lake Tashmoo, Martha's Vineyard, Massachusetts. They also found fragments of eelgrass leaves incorporated in colonies growing on manmade substrates, implying that rafting leaves were the means of dispersal. Petersen & Svane conducted a study on the dispersal of *Ciona intestinalis* larvae, and found that eelgrass served as a localized, small-scale means of dispersal for the solitary ascidian. Future study is needed to evaluate the role of rafting eelgrass leaves in the dispersal of the species in this study. No work had been done on the role of rafting eelgrass leaves in LIS.

The present study demonstrates that fouling invertebrates are present on rafting leaves in LIS. This could have profound implications with respect to the dispersal of invasive tunicates. Less than 5km from the study site the invasive ascidian, *Clavelina lepadiformis,* has been found on the pilings of the Custom House Pier (Reinhardt et al., 2010), in the Thames River. The 2009 Eelgrass Survey for Eastern Long Island Sound, Connecticut and New York confirmed the presence of eelgrass beds in New London Harbor, at the mouth of the Thames River (Tiner et al., 2010). While this species was not seen on rafting leaves in this study, it is possible that *C. lepadiformis* could use eelgrass as a means of dispersal to further spread in LIS.

The data did not support Hypothesis 5, that similar recruitment patterns would be seen on PVC panels deployed in the eelgrass beds as on those collected on eelgrass leaves. PVC panels not only had greater abundances in recruitment for all taxa, but also recruitment of species that were not observed on eelgrass leaves at all.

A greater number of overall species recruited on the panels than on eelgrass throughout the sampling period. This result indicates that not all of the larval fouling organisms present in the water column are settling on the eelgrass. Those animals which settled on the panels but not on the eelgrass leaves are likely selectively settling on a more optimal substrate than eelgrass leaves, such as rocks. A better substrate might be a harder, less mobile or a different texture than the eelgrass leaves.

Sideward facing panels consistently had lower recruitment than downward facing panels. This would imply that the downward facing surfaces are a more suitable recruitment surface for the fouling species. This could be due to the lack of direct sunlight hitting the downward surfaces. Less sunlight results in reduced photosynthetic biomass in competition for space on the panels.

The lower levels of recruitment on the sideward facing panels resulted in fewer differences in settlement between the panels and the collected eelgrass leaves. The orientation of eelgrass leaves in the water is more similar to the sideward facing panels. The amount of sunlight and photosynthetic organisms with which the fouling invertebrates must compete for space is thus going to be more like that on the sideward facing panels than the downward facing panels. This has implications for studies of the fouling community in eelgrass ecosystems. PVC panels are often used and deployed in a similar fashion as the downward facing panels of this study (Bullard et al., 2004, Osman & Whitlatch, 1995, Stachowitcz et al., 2002, others). These results indicate that this may not be an accurate way of assessing the fouling organisms that are settling on the eelgrass. The downward facing panels showed recruitment in higher abundances than the collected eelgrass and in fact had recruitment of organisms that were not observed to have recruited on the collected leaves. In future studies that employ PVC panels in order to assess fouling organisms of the eelgrass ecosystem, sideward facing panels ought to be used, if not an entirely different method of assessment such as artificial eelgrass leaves.

These results show that there is a great deal of variability in the recruitment pattern of the invertebrate fouling community on the eelgrass of LIS. In addition to normal physical and biological factors contributing to the patterns observed, a major disturbance swept through the area on August 28, 2011 in the form of Hurricane Irene. The storm did not directly impact the study area but high winds and waves were experienced. Some impacts were seen in the eelgrass beds as loss of plants. Reduction in density of beds and increases in patchiness were seen at the Bushy and Beach sites in particular. As far as larval recruitment patterns, the storm did not appear to have much effect. Some taxa had reductions in abundance before the storm and others after.

A great deal more work is indicated on the fouling community of eelgrass beds. For example, the interaction between photosynthetic epiphytes and fouling invertebrates has not been examined. Previous work has shown that interactions between larvae and established colonies have impact on larval settlement (Osman & Whitlatch 1995, Bullard et al., 2004). Progression of eelgrass leaves does not allow adequate colonial development of sessile invertebrates but the presence of epiphytes on the surface of the leaves could influence larval recruitment.

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