INTERACTIONS BETWEEN AN INVASIVE EPIPHYTIC BRYOZOAN AND
SPECIES OF ROCKY SUBTIDAL HABITATS OF NOVA SCOTIA

by

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DEPARTMENT OF BIOLOGY

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For Ian

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Abstract

In Nova Scotia subtidal habitats, the invasive bryozoan *Membranipora membranacea* interacts with native bryozoan *Electra pilosa* on kelps, which offer high space availability but are highly dynamic, and on non-kelp algae, which provide low space but high stability. Settlers and colony cover of *M. membranacea* at various stages critical to its population dynamics, as well as relative abundance and encounter outcomes of *M. membranacea* and *E. pilosa*, were quantified on these substrates. I also examined the effects of various factors on growth rates of *E. pilosa*. For *M. membranacea* populations, the roles of kelp and non-kelp substrates varied intra- and inter-annually, as well as spatially. *Membranipora membranacea* was relatively more abundant on kelps than on *Fucus*, likely due to large colony size, faster growth, and strong overgrowth abilities. While kelps provide spatial resources for seasonal peaks in abundance of *M. membranacea*, non-kelp refuges can preserve local populations in time.

List of Abbreviations and Symbols Used

Abbreviation/ Symbol	Definition
α	Significance level
ANCOVA	Analysis of covariance
ANOVA	Analysis of variance
BH	Birchy Head
χ^2	Chi-square statistic
CS	Colony size
df	Degrees of freedom
F	*F*-value
Fs	*Fucus* species
FS	Feltzen South
GR	Growth rate
HSD	Honestly significant difference
Ld	*Laminaria digitata*
MS	Mean squares
N	Sample size
NS	Nova Scotia
p	*p*-value
PH	Paddy's Head
R^2	Coefficient of determination
SC	Sandy Cove
SD	Standard deviation of the mean
SE	Standard error of the mean
Sl	*Saccharina longicruris*
SMB	St. Margarets Bay
sp.	Species (singular)
spp.	Species (plural)

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Chapter 1

Introduction

Alterations to native communities by introduced species have led to substantial environmental and economic impacts, and have evoked negative reaction from humans (Larson 2007; Mack et al. 2008). The origins of invasion biology were influenced by reactionism and political militarism (see Elton 1958), and the use of emotive language persists in this field (Colautti and MacIsaac 2004; Larson 2008). There remains an intensive demand for first-response invasion research, which typically focuses on impacts of invasive species on native communities and human interests. As a consequence, often little is known about the abundance, interactions, and impacts of invasive species in their native ranges (Hierro et al. 2005). Within introduced ranges, sampling of all potential invaders is biased towards cases where invasive species have established successfully, with greater abundance, size, or fecundity than that exhibited in their native ranges (Simons 2003). A call for stronger standards of objectivity by Harding (1992), which includes the critical examination of beliefs and interests forming a scientific project, has led to important paradigm shifts in science, including biology (Schiebinger 1997; Harding 1998). A balanced approach in invasion biology, which considers both native and introduced ranges, as well as contexts of varying levels of invasion success within introduced ranges, may reveal new directions critical to broadening our understanding of interactions between invasive species and their non-native communities.

Several introduced species have become established in Nova Scotia rocky subtidal habitats since the late 19th century (e.g. *Fucus serratus*, *Carcinus maenas*, *Codium fragile*, *Membranipora membranacea*) (Audet et al. 2003; Bird et al. 1993; Coyer et al. 2003; Scheibling et al. 1999), and some have altered community dynamics substantially (Saunders and Metaxas 2008; Scheibling and Gagnon 2006). Prior to recent species introductions, the oscillation of rocky subtidal communities between 2 dominant states, kelp beds with high productivity and barrens with low productivity, has been controlled by the green sea urchin *Strongylocentrotus droebachiensis* (Scheibling et al. 1999). Populations of this dominant grazer cycle between high densities, during which they form destructive feeding aggregations, and low densities, caused by disease (Scheibling 1984). Recent species introductions include an epiphytic bryozoan, *Membranipora*

membranacea, which encrusts kelps, causing defoliation of kelp beds in storms (Saunders and Metaxas 2008), and a green alga, *Codium fragile*, that colonizes spaces opened by defoliation and prevents recruitment of native kelps (Levin et al. 2002; Scheibling and Gagnon 2006).

The impacts of *M. membranacea* on rocky subtidal communities have been studied extensively (e.g. Berman et al. 1992; Lambert et al. 1992; Levin et al. 2002; Saunders and Metaxas 2008; Scheibling et al. 1999), but our understanding of the population dynamics of this bryozoan in its non-native range is limited to kelps. Additionally, no limitations to its dominance as an invasive epiphyte (such as through competition or predation) have been identified in the rocky subtidal communities of Nova Scotia. Kelp blades provide large space for colonization, but these substrates are highly dynamic and unstable due to rapid growth and erosion (Mann 1972; Saunders and Metaxas 2009; Krumhansl and Scheibling in press). In its native range in the northeast Atlantic, *M. membranacea* also occurs on non-kelp substrates, including *Fucus* species, which are smaller than kelps but more stable due to slow, seasonal growth (Mann 1973; O'Connor et al. 1979). *Membranipora membranacea* does not dominate epiphytic communities in its native range, and populations are likely regulated by more complete resource use and increased competition (O'Connor et al. 1980; Stebbing 1973). The epiphyte assemblage on *Fucus* substrates in the northeast Atlantic includes *Electra pilosa*, an encrusting bryozoan that is also native to subtidal habitats of the northwest Atlantic.

The ecology of *M. membranacea* in its native range suggests that it may occur on non-kelp substrates in Nova Scotia rocky subtidal communities, a setting where its presence has not been studied to date. Research on its interactions with alternative non-kelp substrates, as well as other epiphytes, may reveal mechanisms behind its success and persistence and identify limitations to its dominance in its introduced range. The overall objective of this thesis is to address this gap. In Chapter 2, I examine the roles of kelp and non-kelp algal substrates in the population dynamics of *M. membranacea*. To determine how settlement varies between kelp and non-kelp species, I quantified settlers, both early in the settlement period and near the peak of settlement, on *Saccharina longicruris*, *Laminaria digitata*, *Fucus evanescens*, and *F. serratus*, at four sites that

differ in kelp abundance and distribution. To compare extent and survival of colonies of *M. membranacea* between these kelp and non-kelp substrates, I measured colony cover during the period of peak abundance in autumn and at the end of winter.

In Chapter 3, I focus on interactions between *M. membranacea* and the native bryozoan *Electra pilosa* on kelp and non-kelp substrates. I quantified the relative abundance of *M. membranacea* and *E. pilosa* and evaluated outcomes of their encounters on *S. longicruris*, *L. digitata*, and *Fucus* spp. at four sites in Nova Scotia. I also examined the effects of substrate, temperature, and food on growth rates of *E. pilosa* in the laboratory, and the effect of temperature on growth of *E. pilosa* in the field. I compare my findings on factors affecting growth of the native bryozoan to those affecting growth of *M. membranacea*. Chapters 2 and 3 are intended as standalone manuscripts for publication in the primary literature. As a result there is necessarily some repetition among chapters.

Lastly, in Chapter 4, I combine the findings of Chapters 2 and 3 to summarize factors involved in the success, expansion, and persistence of the invasive bryozoan, as well as mechanisms that can limit its dominance in non-native habitats. I also discuss the relevance of these findings to our understanding of the population dynamics of *M. membranacea* which are, in turn, directly connected to the dynamics of kelp beds in Nova Scotia.

Chapter 2

Roles of Kelp and Fucoid Substrates in the Population Dynamics of the Invasive Bryozoan *Membranipora membranacea* in Nova Scotia, Canada

2.1 Abstract

The invasive epiphyte *Membranipora membranacea* occurs in high abundance on kelps which offer high space availability in non-native habitats, but are highly dynamic; however, this bryozoan also occurs on algae other than kelps in its native range, including *Fucus* species, which provide low space availability but high stability. We quantified settlers and colony cover of *M. membranacea* on the kelps *Saccharina longicruris* and *Laminaria digitata* (both native), and on *Fucus evanescens* (native) and *F. serratus* (introduced), at four sites in Nova Scotia at various stages, critical to the population dynamics of the bryozoan. The relative importance of kelp and fucoid substrates varied both intra- and inter-annually, as well as spatially. Settlement was higher on kelps than on *Fucus* at sites where kelps were abundant; however, the abundance of settlers on *Fucus* was similar to or greater than that on kelps at sites where kelps were sparse or spatially separated from *Fucus*. During the period of high colony cover in late autumn, cover was highest on *L. digitata* and lowest on *Fucus* across all sites. Following winter, *M. membranacea* cover decreased by an order of magnitude on kelps, but remained stable on *Fucus*, suggesting high overwintering survival on fucoid algae. While kelps provide spatial resources for seasonal peaks in abundance of the invasive bryozoan, refuges can preserve local populations in time. *Fucus* provides an important refuge for overwintering colonies, particularly where defoliation of kelps has been extensive, and characteristics of this substrate likely facilitate early reproduction and local spread.

2.2 Introduction

Invasive species possess a number of attributes that can enhance their spread, establishment, and persistence in a variety of geographical and ecological settings. Adaptations for long-distance dispersal enable species to reach new regions at broad spatial scales, and breadth of ecological tolerance allows them to succeed in different habitats (Ricciardi and Rasmussen 1998). Within non-native communities, the ability to rapidly produce propagules enables local spread (Sakai et al. 2001), and species that can

expand by asexual or clonal growth can quickly use available resources to increase their abundance (Wright and Davis 2006). For an introduced marine epiphyte, such attributes may enable colonization of both stable and dynamic host substrates, facilitating temporal persistence as well as spatial dominance in non-native habitats.

Membranipora membranacea is an invasive colonial encrusting bryozoan that has caused considerable changes to the dynamics of rocky subtidal habitats of the Northwest Atlantic. With native ranges in the Pacific and Northeast Atlantic Oceans, *M. membranacea* was first observed in the Northwest Atlantic in New Hampshire in 1987, where it established itself as a dominant epiphyte on kelp within two years (Berman et al. 1992). Likely introduced from Europe, *M. membranacea* has since spread to the Atlantic coast of Nova Scotia, Canada, where it was first observed in Mahone Bay and St. Margarets Bay in 1992 (Scheibling et al. 1999; Schwaninger 1999). In its introduced range, colonies of *M. membranacea* encrust the blades of native kelps, making them brittle and susceptible to breakage, and in years of population outbreaks of *M. membranacea*, kelp beds can be severely defoliated in autumn storms (Berman et al. 1992; Lambert et al. 1992; Saunders and Metaxas 2008; Scheibling et al. 1999; Scheibling and Gagnon 2009). Furthermore, an invasive green alga, *Codium fragile fragile*, colonizes spaces opened by defoliation and, once established, can prevent recolonization by kelps (Levin et al. 2002; Scheibling and Gagnon 2006).

Outbreaks of *M. membranacea* have been partially linked to early timing and high rates of settlement following warm winters in combination with high colony growth in warm autumns (Saunders et al. 2010). Larvae of *M. membranacea* can be induced to settle by chemical cues, and are reported to exhibit preference for kelp species, such as *Saccharina longicruris* (Seed 1976; Stricker 1989), although the mechanisms behind this preference have not been demonstrated. However, the physical structure of kelp bed canopies may also influence patterns of settlement. For example, larvae can settle in high abundance on kelp fronds upon encounter, never reaching understory species of algae beneath the kelp canopy (Duggins et al. 1990).

Kelps are highly dynamic substrates for epiphytic organisms. In Nova Scotia, kelps can sustain significant losses of biomass in autumn and winter storms; additionally, they reach maximum growth rates in winter and early spring, during which fronds are

completely replaced through rapid basal growth and distal erosion (Mann 1972; Mann 1973). Populations of *M. membranacea* are greatly affected by kelp dynamics, and bryozoan colony cover on these algae peaks in autumn, following the period of settlement and growth, and preceding the period of kelp erosion and breakage (Saunders and Metaxas 2009b). Early research in this region suggested that *M. membranacea* may contribute to its own decline through reduction in kelp abundance by defoliation (Levin et al. 2002); however, nearly two decades after its recorded arrival, *M. membranacea* remains in these habitats as an abundant epiphyte.

Previous research in the Northwest Atlantic has focussed on the occurrence of *M. membranacea* on kelps, specifically *Saccharina longicruris* and *Laminaria digitata* (Lambert et al. 1992; Levin et al. 2002; Saunders and Metaxas 2007, 2008, 2009a, 2009b; Scheibling and Gagnon 2006, 2009). However, *M. membranacea* occurs on various substrates throughout its range and, in addition to various kelp species, it also occurs on other algae. In its native range (Northwest Europe), *M. membranacea* is a component of the native bryozoan assemblage on *Fucus serratus* (O'Connor et al. 1979; Ryland and Stebbing 1971; Walters and Wethey 1986), and has been reported on *Ascophyllum nodosum* and *Halidrys siliquosa* (Ryland 1959; Ryland 1962). The ecology of *M. membranacea* on *Fucus* species in its introduced range, however, has not been examined, and records of occurrence of *M. membranacea* on species other than kelp in the Northwest Atlantic are limited. Its occurrence has been qualitatively reported on *Fucus evanescens* and *Chondrus crispus* in northern New England (Walters and Wethey 1986), on *C. fragile* and *Desmarestia aculeata* in the Gulf of Maine (Harris and Tyrrell 2001), and on *F. evanescens*, *F. serratus*, *A. nodosum*, *C. crispus*, and *C. fragile* in Nova Scotia (Saier and Chapman 2004; Watanabe et al. 2009; Yorke and Metaxas submitted).

Of the fucoid substrates, *Fucus evanescens* is distributed widely throughout Nova Scotia in shallow subtidal habitats, and *F. serratus* is dominant in localized areas of the region, having been introduced to the province in the late 19th century (Edelstein et al. 1971). These algae likely provide a more stable substrate for epiphytes than kelp, as they have much lower intrannual variation in biomass and slower growth rates (Mann 1973; McCook and Chapman 1991). Additionally, these species may serve as vectors for local-scale alongshore spread of *M. membranacea* in areas where kelps are absent or sparse

(Watanabe et al. 2009). Defoliation of colonized kelp blades potentially affects the populations of *M. membranacea*, both through reduction of the adult population, and through removal of preferred settlement substrates. However, if *M. membranacea* can successfully settle, grow, and overwinter on *Fucus* species in subtidal communities in Nova Scotia, a reduction in kelp abundance will not necessarily result in bryozoan decline.

Our understanding of the seasonal population cycle of *M. membranacea* on kelps suggests that alternative algal substrates may contribute to the success of this invasive bryozoan at various stages. Settlers of *M. membranacea* increase in abundance throughout summer and autumn, reaching high rates of settlement over a short period (weeks), after which settlement decreases (Saunders and Metaxas 2007). New recruits mature and colonies grow throughout late summer and autumn, and colony cover reaches its peak in late autumn, prior to mortality from kelp breakage and colony senescence in late autumn and winter (Saunders and Metaxas 2008, 2009b). Some colonies persist through winter when growth is insignificant due to low temperatures, and growth and reproduction resume with warming water temperatures in early summer (Saunders and Metaxas 2009b; Saunders et al. 2010). Fucoid algae may provide alternative substrates for *M. membranacea* settlers when kelps are sparse, particularly when rates of settlement are high and space on kelps may become limiting. Where recruitment and growth occur on species other than kelp, colony cover will likely persist on these stable substrates throughout late autumn and winter, providing adult populations in the beginning of the following season. Post-winter cover on fucoid algae may be relatively high compared to kelps, particularly where substantial defoliation of kelps has occurred.

In this study, we examined the relative importance of two abundant kelp species and two *Fucus* species at various stages critical to the dynamics of populations of the introduced epiphytic bryozoan *Membranipora membranacea* in Nova Scotia, Canada. To determine how settlement varies between kelp and fucoid species, we quantified settlers, both early in the settlement period and near the peak of settlement, on *Saccharina longicruris*, *Laminaria digitata*, *Fucus evanescens*, and *F. serratus*. We sampled four sites that differ in kelp abundance and distribution. To compare extent and survival of colonies of *M. membranacea* between these kelp and fucoid species, we measured colony

cover during the period of peak abundance in autumn and at the end of winter. This study will allow us to qualify the contribution of fucoid algae to settlement, growth, and overwintering of colonies of *M. membranacea*. The association between *Fucus* and *M. membranacea* that has been recorded in the native range of this invasive bryozoan may also provide an important refuge in its introduced range, particularly where this epiphyte has caused defoliation of kelps.

2.3 Materials and Methods

2.3.1 Study Sites

We sampled 4 sites in and near St. Margarets Bay, Nova Scotia, Canada (Fig. 2.1), where the population dynamics of *M. membranacea* have been previously studied on kelps (Saunders and Metaxas 2007, 2008, 2009a, 2009b; Scheibling and Gagnon 2009). Feltzen South (44º19.9'N, 64º16.9'W) is near the mouth of Lunenburg Bay, 30 km southwest of St. Margarets Bay. The site has a gently sloping bedrock substratum with a steep bedrock ridge offshore. Birchy Head (44º34.5'N, 64º02.5'W) is on the western shore of St. Margarets Bay, and consists of a moderately sloping granite substratum with boulders and cobbles. Paddy's Head (44º31.6'N, 63º57.2'W), on the eastern shore of St. Margarets Bay, includes a steeply sloping exposed outer wall, as well as a more sheltered shoreward cove. Sandy Cove (44º27.8'N, 63º42.5'W) is 20 km east of St. Margarets Bay at the mouth of Terence Bay in Pennant Bay. The site includes an outer slope with bedrock ridges, as well as a gently sloping sheltered inner cove with a cobble bottom. All sites are characterized by mixed kelp beds, dominated by *Saccharina longicruris* and *Laminaria digitata*. Species of *Fucus* are also present throughout the study area, with *F. serratus* occurring at Feltzen South and *F. evanescens* at Birchy Head, Paddy's Head, and Sandy Cove.

Access of settling larvae to algae other than kelp may be affected by kelp abundance and distribution if kelp act as "filters" of settlers; consequently, sites were selected to include variation in the openness of the kelp canopy. Feltzen South and Birchy Head were chosen to represent open canopies: kelps are spatially separated from *Fucus* at Feltzen South and they were sparse at Birchy Head during sampling. Paddy's Head and Sandy Cove were selected as sites with abundant kelp and closed canopies.

Since Birchy Head and Paddy's Head are both in St. Margarets Bay, we assumed they share the same larval source and timing of settlement.

2.3.2 Sample Collection

Sampling was designed to capture different stages in the annual population cycle of *M. membranacea*, based on the known population dynamics of the species in the region. We measured: early settlers (Aug 2008), peak settlers (Oct 2008), pre-winter cover (Nov - Dec 2008), and post-winter cover (Jul 2008 and 2009). We describe early settlers as those that occur prior to the major settlement event, when settlers are sparse but detectable, and increasing in abundance. Peak settlers are those that occur during the period of highest settlement. Pre-winter cover describes the spatial extent of colonies at the end of the periods of settlement and growth and prior to the onset of slow growth and mortality due to senescence. Post-winter cover describes the spatial extent of colonies that have overwintered successfully, censused prior to the accelerated growth associated with increasing temperature.

At each site and each stage, abundance of *S. longicruris*, *L. digitata*, and *Fucus* spp. (*F. serratus* at Feltzen South; *F. evanescens* at all other sites) were sampled in 8 - 10 randomly selected quadrats along transects using SCUBA. Transects were positioned within the zone of highest abundance for each algal taxon at each site, and were orientated to include the range of depths over which each taxon occurred, to a maximum depth of 14 m (Table 2.1). If the alga of interest was absent from a quadrat, contiguous quadrats were sampled until the alga was found. The sampled area was then pooled across all contiguous quadrats. For post-winter cover, we sampled only algae that were encrusted by *M. membranacea*, whereas for all other periods, all thalli of *S. longicruris*, *L. digitata*, and *Fucus* spp. were collected from each quadrat for quantification of kelp abundance and algal biomass (see below). Algae were transferred to dry tubs at the surface, and transported to the laboratory for storage in aquaria with ambient running seawater until processing was completed.

2.3.3 Quantification of M. membranacea

For early settlers, all settlers [defined as in Saunders and Metaxas (2007) as any colony with < 2 zooid rows] were counted on a subsample of 1 - 5 kelp thalli, or ~0.5 m^2 surface area for *Fucus* spp., from each quadrat. For peak settlers (Oct 2008), we only counted settlers on 1 randomly chosen quarter of each kelp blade (as in Saunders and Metaxas 2007) and 1 side of *Fucus* thalli of subsampled algae. For pre-winter and post-winter cover, all mature colonies of *M. membranacea* on each thallus were traced, photographed with a Nikon Coolpix 995 digital camera, and surface area was measured with SigmaScan Pro Image Analysis 5.0 (SPSS). Processing generally occurred within 1 to 8 days of sampling, with the exception of peak settlers on *Fucus* which were quantified within 20 to 26 d. Settlers and colonies did not grow while in aquaria, indicating that the length of time prior to processing did not affect results.

The surface area of algal thalli was also measured using image analysis of photographs, as for tracings of *M. membranacea* colonies. Uncolonized algal surface area was calculated in Oct 2008 from measurements of thallus surface area and area of mature colonies.

For *S. longicruris*, surface area cannot be determined from a single view in two dimensions because of blade crenulations. For this species, thalli were cut up and the fragments laid flat for photography and analysis, as above. Several (6 - 13) thalli from a range of conditions (2 sites for each of Aug, Oct, and Nov - Dec 2008) were used to calculate the ratio of fragment surface area to intact surface area for *S. longicruris*. Analysis of the effects of site and period on these ratios revealed a significant effect of period (2-way ANOVA; Site: $F_{(2, 50)} = 2.061$, $p = 0.138$; Period: $F_{(2, 50)} = 5.772$, $p = 0.006$; Site x Period: $F_{(1, 50)} = 0.119$, $p = 0.732$). Simple linear regressions were therefore generated for each period to correct intact surface area measurements for *S. longicruris*.

To obtain total surface area of algae per unit area substratum for early and peak settlers and pre-winter cover, we developed indices of surface area to biomass. Up to 5 thalli (or ~0.5 m^2 surface area, ~140 g wet weight, for *Fucus* spp.) from each quadrat at each of these sampling periods were weighed in the laboratory using an electronic balance (accuracy 0.001 g) and their surface area measured. From these measures, simple linear regressions of surface area with biomass were generated for each algal

taxon at each sampling stage. During sampling, most quadrats contained 1 - 22, and 1 - 10 thalli, for *S. longicruris* and *L. digitata*, respectively. For quadrats with <5 thalli (or <140 g *Fucus* spp.), total surface area per quadrat was measured in the laboratory, as described above. For quadrats with >5 thalli (or >140 g *Fucus* spp.), total algal mass in each quadrat was measured in the field using a spring balance (accuracy 25 g), and the linear relationships obtained in the laboratory were then used to calculate algal surface area per unit area substratum.

2.3.4 Statistical Analyses

The effects of site (fixed factor, 4 levels: Feltzen South, Birchy Head, Paddy's Head, and Sandy Cove) and period (fixed factor, 3 levels: Aug, Oct, and Nov - Dec 2008) on the abundance of kelp (thalli m^{-2} substratum) were examined for each of *S. longicruris* and *L. digitata*, using 2-way ANOVA. Site was a fixed factor because sites were selected for their locations (inside and outside St. Margarets Bay) and for characteristics of kelp canopies. Period was also a fixed factor because sampling corresponded with specific events in the annual population cycle of *M. membranacea*.

The effects of algal taxon (fixed factor, 3 levels: *S. longicruris*, *L. digitata*, and *Fucus* spp.) and site were examined by 2-way ANOVA for each of the following variables: abundance of early settlers m^{-2} algae (Aug 2008) and abundance of peak settlers m^{-2} algae (Oct 2008); abundance of peak settlers m^{-2} uncolonized algae (Oct 2008); abundance of early settlers m^{-2} substratum (Aug 2008) and abundance of peak settlers m^{-2} substratum (Oct 2008); cm^2 pre-winter cover m^{-2} algae and cm^2 pre-winter cover m^{-2} substratum (Nov - Dec 2008). The effects of algal taxon, site, and year (random factor, 2 levels: 2008 and 2009) on cm^2 post-winter cover m^{-2} substratum (Jul 2008 and 2009) were examined by 3-way ANOVA.

For these analyses, the following transformations were applied to reduce heterogeneity of variances: log(x) for kelp abundance; log(x+0.01) for early settler abundance, pre-winter cover, and post-winter cover; and log(x+1) for peak settler abundance. Homogeneity of variance (Levene's test, $p < 0.05$) was not achieved through transformation for the following: *L. digitata* abundance m^{-2} substratum, early settler abundance m^{-2} algae and m^{-2} substratum, pre-winter cover m^{-2} algae and m^{-2} substratum,

and post-winter cover m^{-2} substratum (2008 and 2009); therefore a more conservative α_{crit} = 0.01 was used for these analyses. According to the Shapiro-Wilk test ($p < 0.05$), early settler abundance m^{-2} algae and m^{-2} substratum, pre-winter cover m^{-2} algae and m^{-2} substratum, and post-winter cover (2008 and 2009) were not distributed normally; however, ANOVA is robust to deviations from normality (Zar 1999). Statistical analyses were conducted using SPSS 15.0. Where appropriate, homogeneous subsets were identified using Tukey's HSD tests.
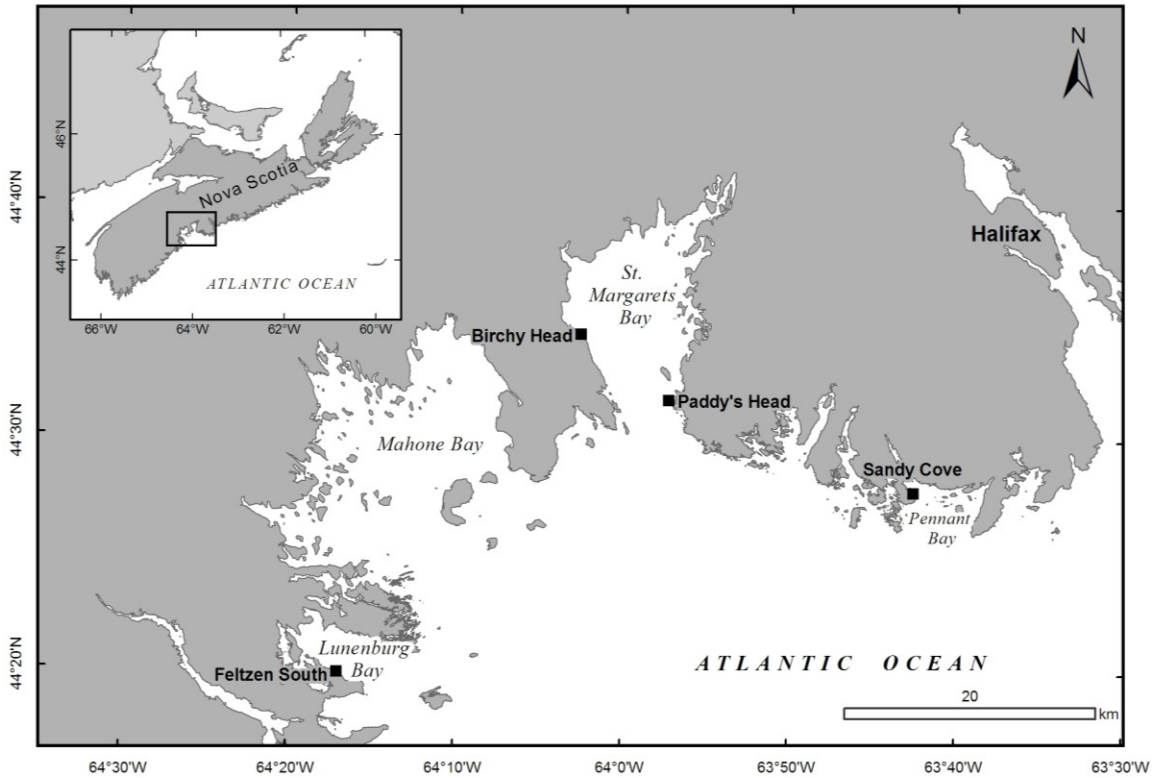
Fig. 2.1 Study area on the southern shore of Nova Scotia, Canada showing sampling sites in Lunenburg Bay (Feltzen South), St. Margarets Bay (Birchy Head and Paddy's Head), and Pennant Bay (Sandy Cove). (© Department of Natural Resources Canada. All rights reserved.)

Table 2.1 Details of sampling distributions of algal taxa at each site. Orientation of transect is relative to the shoreline.

Site	Algal species	Depth range (m)	Orientation of transect
Feltzen South	*Fucus serratus*	2.8 - 6.2	perpendicular
	Laminaria digitata	5.0 - 7.8	parallel
	Saccharina longicruris	5.9 - 8.0	parallel
Birchy Head	*Fucus evanescens*	0.1 - 0.8	parallel
	Laminaria digitata	0.1 - 1.5	parallel
	Saccharina longicruris	1.1 - 8.2	perpendicular
Paddy's Head	*Fucus evanescens*	0.1 - 3.1	parallel
	Laminaria digitata	2.5 - 14	perpendicular
	Saccharina longicruris	2.3 - 14	perpendicular
Sandy Cove	*Fucus evanescens*	0.2 - 3.7	parallel
	Laminaria digitata	0.9 - 7.1	perpendicular
	Saccharina longicruris	0.6 - 7.1	perpendicular

2.4 Results

2.4.1 Abundance of Kelps and Distribution of Algae

At Feltzen South, kelps and *Fucus* are spatially separated; the site includes an extensive, dense meadow of *F. serratus* at ~3 - 6 m depth, and both kelp species occur primarily on a steep bedrock ridge offshore from the *Fucus* meadow at ~5 - 10 m depth. At Birchy Head, kelps occur sparsely at <11 m depth, with *Laminaria digitata* occurring primarily at <2 m depth, and *F. evanescens* occurring in shallower areas (<1 m) with a patchy distribution. The outer wall at Paddy's Head is dominated by abundant *Saccharina longicruris* and *L. digitata* at depths >2 m, and *F. evanescens* occurs sparsely shoreward at depths <3 m. At Sandy Cove, the outer slope has luxuriant kelp beds of *S. longicruris* and *L. digitata* at depths ~1 - 10 m, and *F. evanescens* occurs nearby in the inner cove at <4 m.

The measured patterns in abundance of kelp and algal distribution in 2008 indicated that Birchy Head and Feltzen South were sites with canopies that were more open than those at Paddy's Head and Sandy Cove. Abundance of kelp ranged from 3.8 ± 1.8 thalli m^{-2} substratum (mean ± SD, N = 7 - 11) at Birchy Head (Aug) to 29.4 ± 33.8 at Sandy Cove (Dec) for *S. longicruris* (Fig. 2.2A), and from 3.1 ± 2.1 at Birchy Head (Aug) to 11.6 ± 4.9 at Feltzen South (Oct) for *L. digitata* (Fig. 2.2B). Abundance of *S. longicruris* was significantly lower at both Birchy Head and Feltzen South than at Paddy's Head and Sandy Cove, and abundance of *L. digitata* was significantly lower at Birchy Head than at Feltzen South and Paddy's Head (Table 2.2). Abundance of *S. longicruris* was significantly greater in Oct than in Aug, with no significant differences between Nov-Dec and either of the other periods (Table 2.2). For *L. digitata*, abundance was significantly greater in Oct than in both Aug and Nov-Dec.

Although we interpret the interaction between site and period on the abundance of *L. digitata* to be marginally non-significant, the *p*-value suggests that there may indeed be an interaction between these factors. Post-hoc tests (Tukey's HSD tests, *a* = 0.05) based on a significant interaction reveal lower abundance of *L. digitata* at Birchy Head than at Feltzen South and Paddy's Head, and lower abundance at Sandy Cove than at Paddy's Head, in Aug, with no significant differences among sites in Oct or Nov-Dec. Additionally, abundance was greater in Oct and Nov-Dec than in Aug at Birchy Head,

and greater in Oct than in Aug at Sandy Cove, with no differences among periods at Feltzen South and Paddy's Head.

2.4.2 Settlement of M. membranacea

Abundance of peak settlers was an order of magnitude greater than that of early settlers, at 19 ± 14 (mean $\pm$ SD, N = 8 - 11) to 1177 ± 867 peak settlers m^{-2} algae (Fig. 2.3B), compared to 0 ± 0 to 82 ± 66 early settlers m^{-2} algae (Fig. 2.3A). There were significant interactions between algal taxon and site for both early and peak settlers (Table 2.3).

Settlers were generally more abundant on kelps than on *Fucus* spp. both during early and peak settlement stages at all sites except at Birchy Head, with no significant differences between *S. longicruris* and *L. digitata*, except at Sandy Cove (Table 2.3). At Birchy Head, abundance of both early and peak settlers was similar on kelp and fucoid algae. Also, at Feltzen South, early settlers did not differ between *Fucus* and *L. digitata*. Whereas settlement per unit area of algae was similar across sites on *S. longicruris* (except at Sandy Cove), abundance of early and peak settlers on *Fucus* was greatest at Birchy Head. Patterns in abundance among sites and algal taxa were generally consistent between early and peak settlers. Patterns in the abundance of peak settlers per unit area of uncolonized algae did not differ from those found for peak settlers per unit area of algae; however, taking into account the amount of uncolonized area changed the relationships among sites. Abundance per unit area of uncolonized algae was significantly greatest at Birchy Head for all algal taxa (Fig. 2.3C; Table 2.3).

Patterns of settler abundance per unit area substratum among algal taxa were similar to those detected per unit area of algae in most cases, for both early and peak settlers (Fig. 2.4; Table 2.4). At sites where settlement occurred on *Fucus* spp. (Birchy Head and Feltzen South), although mean abundance of early settlers per unit area substratum was higher on *Fucus* than on kelps, differences were not statistically significant. At the other sites, however, significant differences in abundance of early settlers were detected. The abundance of peak settlers was significantly lower on *S. longicruris* than on both *L. digitata* and *F. evanescens* at Birchy Head, likely due to low abundance of this kelp species at that site. Abundance of early settlers on *S. longicruris*

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per unit area substratum was generally lower at Sandy Cove than at other sites, and abundance of peak settlers was significantly greater at Paddy's Head than at Birchy Head. Patterns among sites in abundance of early and peak settlers per unit area substratum on *Fucus* spp. were similar to those observed for abundance per unit area of algae, with highest abundance occurring at Birchy Head.

2.4.3 Pre-winter Cover of M. membranacea *Colonies*

Measures of area covered by *M. membranacea* colonies per unit area of algae and per unit area substratum in Nov-Dec 2008, the period that precedes winter mortality, spanned orders of magnitude across sites (Fig. 2.5). Minimum cover of *M. membranacea* occurred on *F. evanescens* at Sandy Cove, and maximum cover occurred on *L. digitata* at Paddy's Head. Area of *M. membranacea* colonies was consistent among algal taxa across sites, when quantified both per unit area of algae and per unit area substratum. Colony cover was significantly greatest on *L. digitata* and lowest on *Fucus* spp. Pre-winter cover was generally greater at Birchy Head than at the other sites (Table 2.5).

2.4.4 Post-winter cover of M. membranacea *colonies*

Following winter mortality, patterns of *M. membranacea* cover also varied among sites and algal taxa, and between years. Post-winter cover, measured in Jul 2008 and 2009, ranged from 0.0 ± 0.0 cm^2 *M. membranacea* m^{-2} substratum (mean $\pm$ SD, N = 7 - 11) on *S. longicruris* and *Fucus* spp. at various sites in both years, to 899.8 ± 1173.2 cm^2 *M. membranacea* m^{-2} substratum on *F. serratus* at Feltzen South in 2008 (Fig. 2.6). There was a significant interaction among algal taxon, site, and year on colony cover per unit area substratum (Table 2.6). In 2008, post-winter cover was significantly greater on *Fucus* spp. than on *S. longicruris* at both Feltzen South and Birchy Head (Table 2.6). At Paddy's Head and Sandy Cove, colony cover did not vary significantly among algal taxa. In 2009, post-winter cover was significantly greater on *Fucus* spp. than on both kelp species at Feltzen South and Birchy Head, and greater on both *Fucus* and *L. digitata* than on *S. longicruris* at Paddy's Head. As in 2008, there were no differences among taxa at Sandy Cove. In both years, post-winter colony cover on *S. longicruris* did not vary

across sites, whereas on *Fucus* spp. it was consistently greatest at Feltzen South. On *L. digitata*, patterns among sites differed between years.
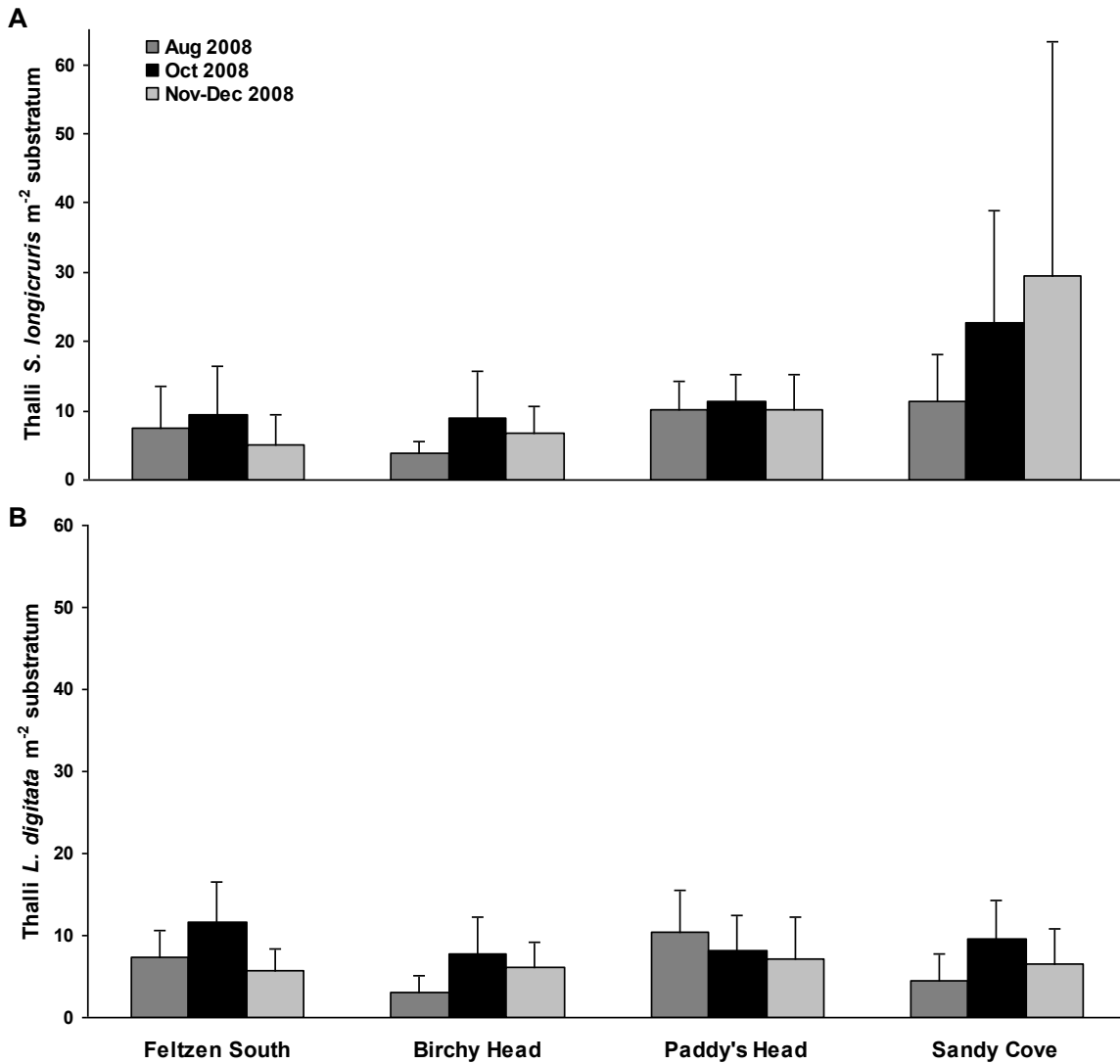
Fig. 2.2 Abundance of two species of kelps at each of four sites on the southern shore of Nova Scotia in Aug 2008, Oct 2008, and Nov-Dec 2008, showing: (A) thalli of *Saccharina longicruris* per m^{-2} substratum (mean + SD, N = 7 - 11), and (B) thalli of *Laminaria digitata* per m^{-2} substratum (mean + SD, N = 8 - 11).

Table 2.2 Results of 2-way ANOVA examining the effects of site (fixed factor, 4 levels: Feltzen South, Birchy Head, Paddy's Head, Sandy Cove) and period (fixed factor, 3 levels: Aug 08, Oct 08, Nov-Dec 08) on the abundance of kelp thalli m^{-2} substratum. FS = Feltzen South; BH = Birchy Head; PH = Paddy's Head; SC = Sandy Cove. Bold font indicates significant *p*-values (*Saccharina longicruris*, $\alpha = 0.05$; *Laminaria digitata*, $\alpha < 0.01$, see Methods). Only significant differences in post hoc tests are shown (at $p < 0.05$).

Effect	MS	$F_{(df)}$	*p*	Tukey's HSD
S. longicruris				
Site	1.369	$17.202_{(3, 99)}$	**< 0.001**	SC > PH > FS, BH
Period	0.302	$3.801_{(2, 99)}$	**0.026**	Oct > Aug
Site x Period	0.141	$1.767_{(6, 99)}$	0.114	
Error	0.080			
L. digitata				
Site	0.440	$4.920_{(3, 99)}$	**0.003**	FS, PH > BH
Period	0.544	$6.082_{(2, 99)}$	**0.003**	Oct > Nov-Dec, Aug
Site x Period	0.268	$2.994_{(6, 99)}$	0.010	
Error	0.089			
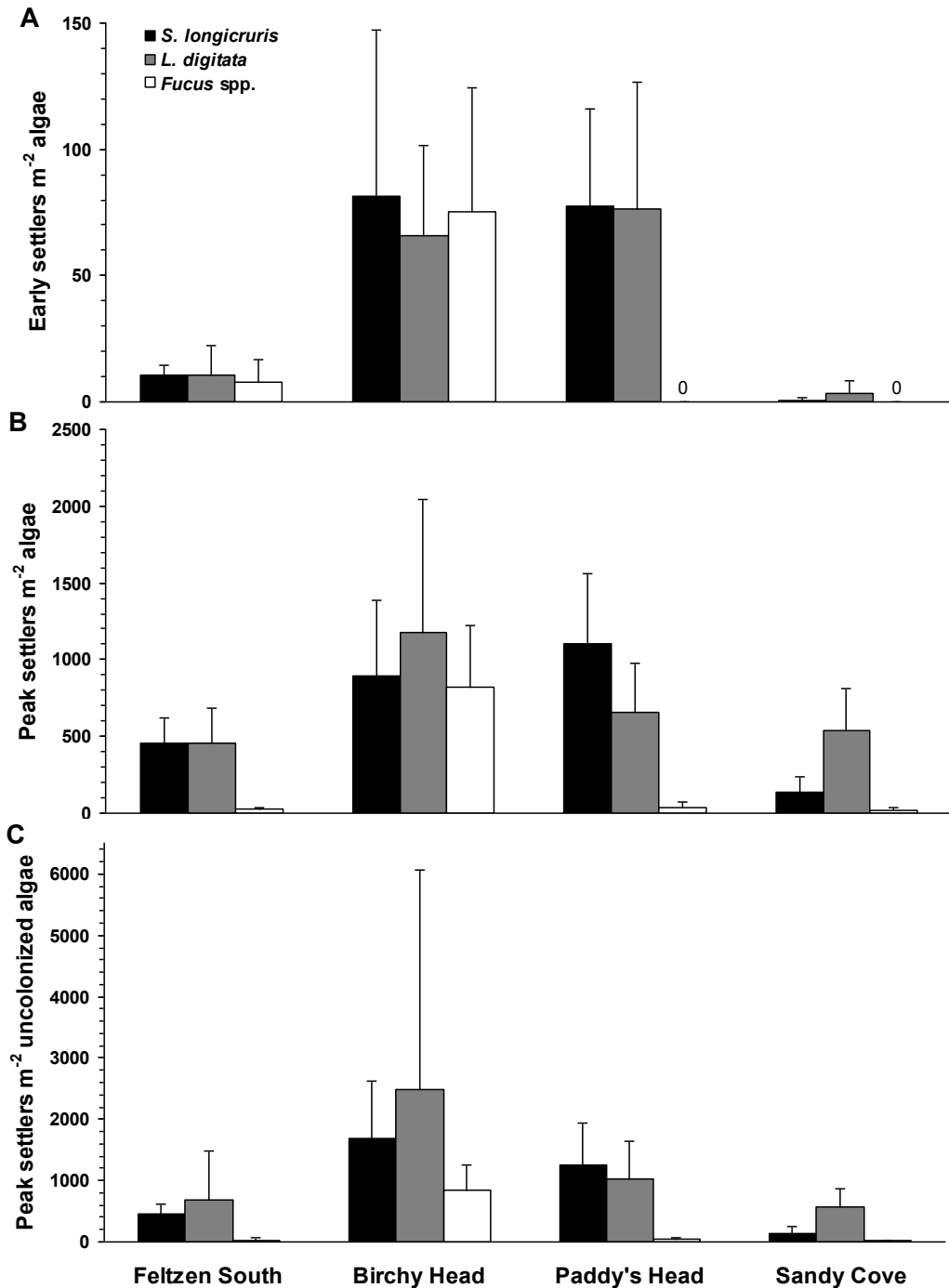
Fig. 2.3 Abundance of settlers of *Membranipora membranacea* per unit area of algae on three algal taxa (*Saccharina longicruris*, *Laminaria digitata*, *Fucus evanescens* at Birchy Head, Paddy's Head, and Sandy Cove, and *F. serratus* at Feltzen South) at each of four sites on the southern shore of Nova Scotia, showing: (A) early settlers m^{-2} algae (August 2008; mean + SD, N = 8 - 11), (B) peak settlers m^{-2} algae (October 2008; mean + SD, N = 9 - 11), and (C) peak settlers m^{-2} uncolonized algae (October 2008; mean + SD, N = 9 - 11), where "uncolonized algae" is area of thallus not occupied by mature colonies of *M. membranacea*.

Table 2.3 Results of 2-way ANOVA examining the effects of algal taxon (fixed factor, 3 levels: *Saccharina longicruris*, *Laminaria digitata*, *Fucus* spp.) and site (fixed factor, 4 levels: Feltzen South, Birchy Head, Paddy's Head, Sandy Cove) on abundance of early settlers m^{-2} algae (August 2008), peak settlers m^{-2} algae (October 2008), and peak settlers m^{-2} uncolonized algae (October 2008) of *Membranipora membranacea*. *Sl = S. longicruris*; *Ld = L. digitata*; *Fs = Fucus* spp.; FS = Feltzen South; BH = Birchy Head; PH = Paddy's Head; SC = Sandy Cove. Bold font indicates significant *p* values, $\alpha = 0.01$ (see Methods). Only significant differences in post hoc tests are shown (at $p < 0.05$).

Effect	MS	$F_{(df)}$	*p*	Tukey's HSD	
Early settlers m^{-2} algae					
Algal taxon	21.79	$35.02_{(2, 92)}$	**< 0.001**	FS:	*Sl > Fs*
				PH:	*Sl, Ld > Fs*
				SC:	*Ld > Sl, Fs*
Site	46.96	$75.50_{(3, 92)}$	**< 0.001**	*Sl*:	FS, BH, PH > SC
				Ld:	BH, PH > FS > SC
				Fs:	BH > FS > PH, SC
Algal taxon x Site	9.135	$14.69_{(6, 92)}$	**< 0.001**		
Error	0.622				
Peak settlers m^{-2} algae					
Algal taxon	14.38	$135.1_{(2, 108)}$	**< 0.001**	FS, PH:	*Sl, Ld > Fs*
				SC:	*Ld > Sl > Fs*
Site	4.798	$45.05_{(3, 108)}$	**< 0.001**	*Sl*:	FS, BH, PH > SC
				Fs:	BH > FS, PH, SC
Algal taxon x Site	1.828	$17.16_{(6, 108)}$	**< 0.001**		
Error	0.107				
Peak settlers m^{-2} uncolonized algae					
Algal taxon	17.07	$142.2_{(2, 108)}$	**< 0.001**	as for Peak settlers m^{-2} algae for all sites	
Site	6.306	$52.52_{(3, 108)}$	**< 0.001**	*Sl*:	BH, PH > FS > SC
				Ld:	BH > FS, SC
				Fs:	BH > FS, PH, SC
Algal taxon x Site	1.495	$12.45_{(6, 108)}$	**< 0.001**		
Error	0.120				
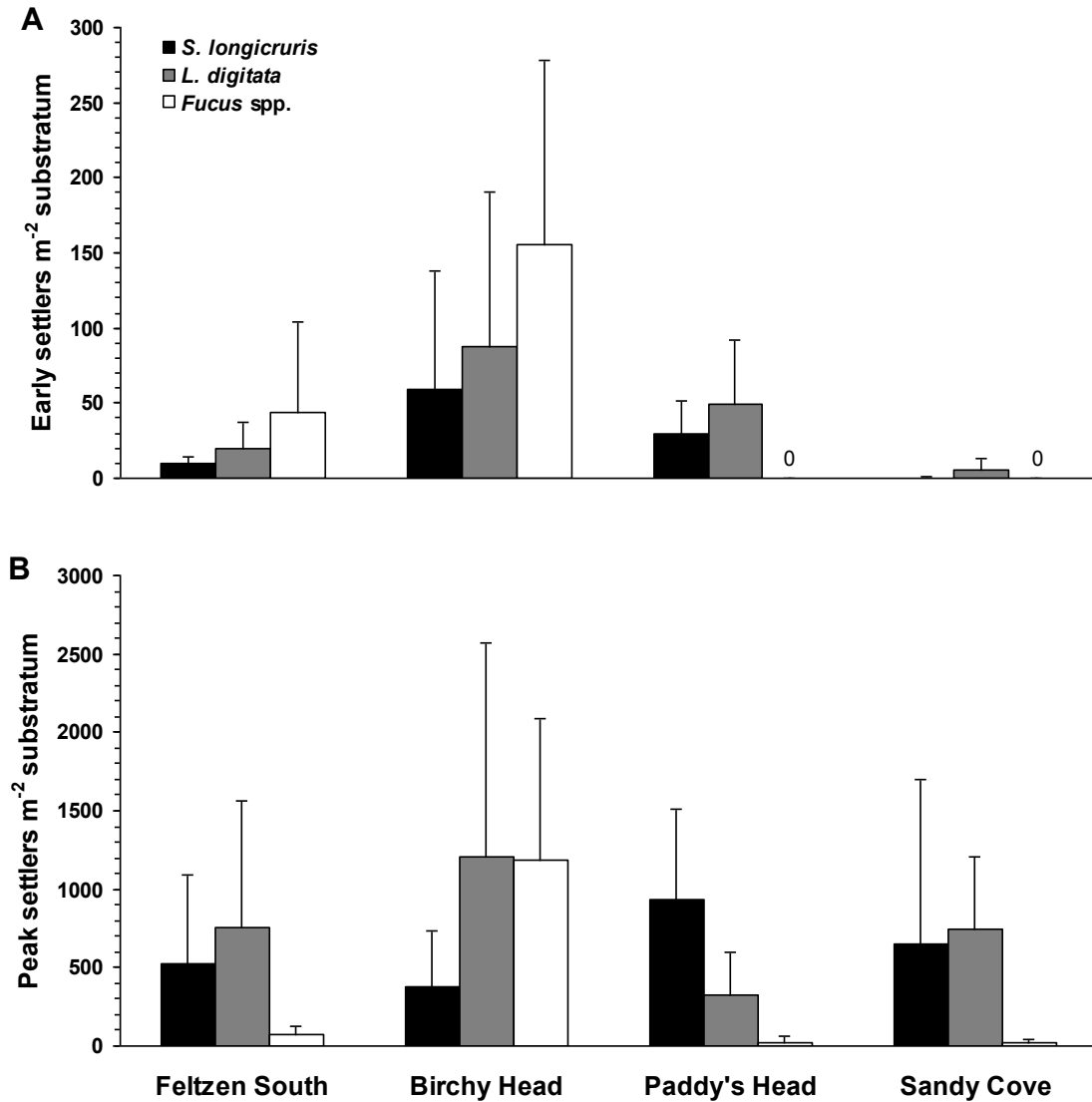
Fig. 2.4 Abundance of settlers of *Membranipora membranacea* per unit area substratum on each of three algal taxa (*Saccharina longicruris*, *Laminaria digitata*, and *Fucus* spp.) at each of four sites on the southern shore of Nova Scotia, showing: (A) early settlers m^{-2} substratum (Aug 2008; mean + SD, N = 8 - 11), and (B) peak settlers m^{-2} substratum (Oct 2008; mean + SD, N = 9 - 11).

**Table 2.4** Results of 2-way ANOVA examining the effects of algal taxon (fixed factor, 3 levels: *Saccharina longicruris*, *Laminaria digitata*, *Fucus* spp.) and site (fixed factor, 4 levels: Feltzen South, Birchy Head, Paddy's Head, Sandy Cove) on abundance of early settlers m^{-2} substratum (August 2008) and peak settlers m^{-2} substratum (October 2008) of *Membranipora membranacea*. Abbreviations as in Table 2.3; bold font indicates significant *p* values, $\alpha = 0.01$ (see Methods). Only significant differences in post hoc tests are shown (at $p < 0.05$).

Effect	MS	$F_{(df)}$	*p*	Tukey's HSD
Early settlers m^{-2} substratum				
Algal taxon	13.39	15.74 $_{(2, 92)}$	**< 0.001**	PH: *Sl, Ld > Fs*
				SC: *Ld > Sl, Fs*
Site	44.70	52.55 $_{(3, 92)}$	**< 0.001**	*Sl*: FS, BH, PH > SC
				Ld: BH, PH > SC
				Fs: BH > FS > PH, SC
Algal taxon x Site	9.214	10.83 $_{(6, 92)}$	**< 0.001**	
Error	0.851			
Peak settlers m^{-2} substratum				
Algal taxon	12.34	46.55 $_{(2, 108)}$	**< 0.001**	FS, SC: *Sl, Ld > Fs*
				BH: *Ld, Fs > Sl*
				PH: *Sl > Ld > Fs*
Site	2.560	9.653 $_{(3, 108)}$	**< 0.001**	*Sl*: PH > BH
				Fs: BH > FS > PH
Algal taxon x Site	3.610	13.61 $_{(6, 108)}$	**< 0.001**	
Error	0.265			
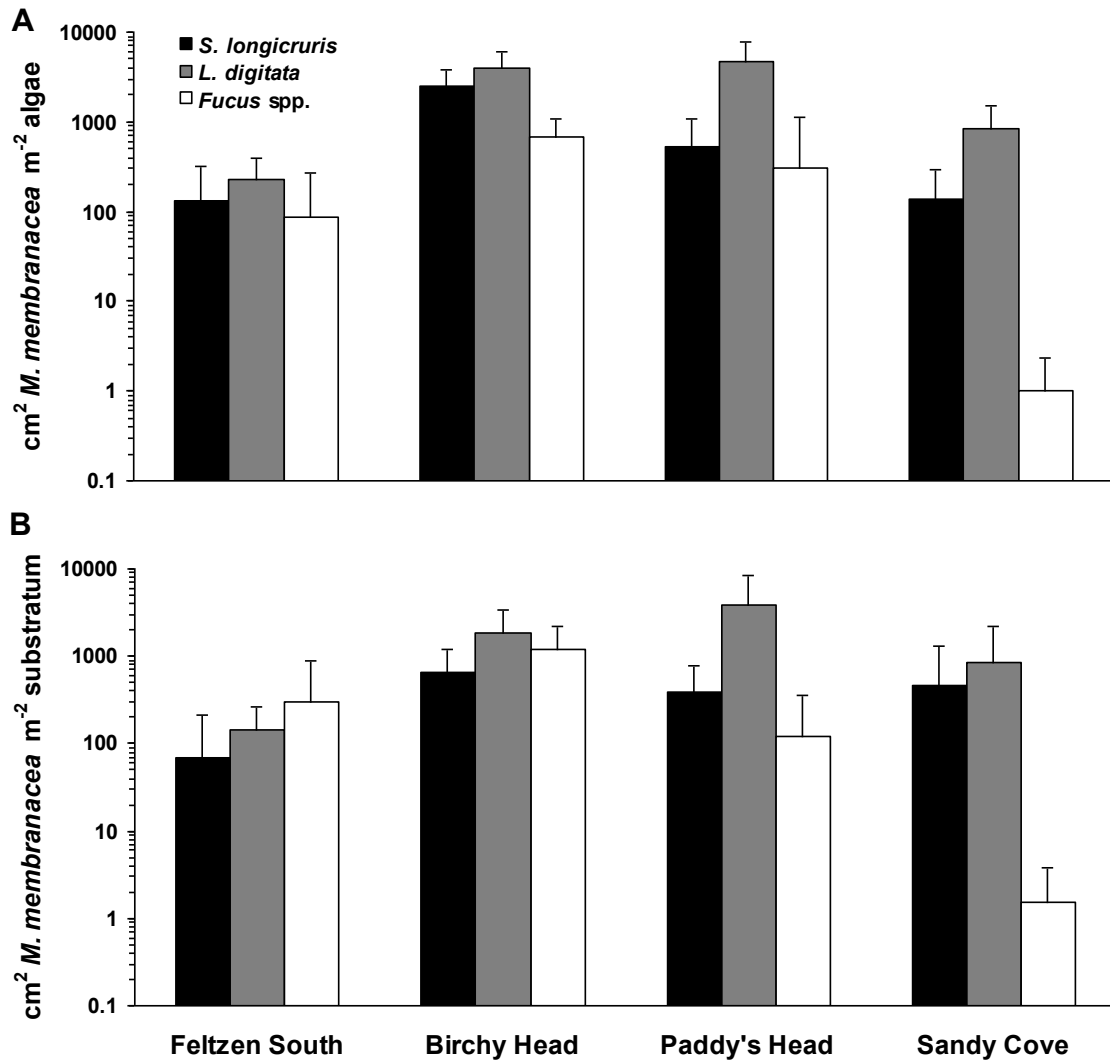
Fig. 2.5 Mean area of pre-winter cover of *Membranipora membranacea* on three algal taxa (*Saccharina longicruris*, *Laminaria digitata*, and *Fucus* spp.) at each of four sites on the southern shore of Nova Scotia (Nov-Dec 2008; + SD, N = 9 - 10), (A) per unit area of algae, and (B) per unit area substratum.

Table 2.5 Results of 2-way ANOVA examining the effects of algal taxon (fixed factor, 3 levels: *Saccharina longicruris*, *Laminaria digitata*, *Fucus* spp.) and site (fixed factor, 4 levels: Feltzen South, Birchy Head, Paddy's Head, Sandy Cove) on pre-winter cover cm^2 m^{-2} algae and pre-winter cover cm^2 m^{-2} substratum (November, December 2008) of *Membranipora membranacea*. Abbreviations as in Table 2.3; bold font indicates significant *p* values, $\alpha = 0.01$ (see Methods). Only significant differences in post hoc tests are shown (at $p < 0.05$).

| Effect	MS	$F_{(df)}$	p	Tukey's HSD
Pre-winter cover cm^2 m^{-2} algae				
Algal taxon	59.61	33.43 $_{(2, 105)}$	**< 0.001**	*Ld > Sl > Fs*
Site	24.30	13.63 $_{(3, 105)}$	**< 0.001**	BH > PH, SC, FS
Algal taxon x Site	3.217	1.804 $_{(6, 105)}$	0.105	
Error	1.783			
Pre-winter cover cm^2 m^{-2} substratum				
Algal taxon	39.78	20.49 $_{(2, 105)}$	**< 0.001**	*Ld > Sl > Fs*
Site	17.31	8.918 $_{(3, 105)}$	**< 0.001**	BH > PH, SC, FS
Algal taxon x Site	4.478	2.307 $_{(6, 105)}$	0.039	
Error	1.941			
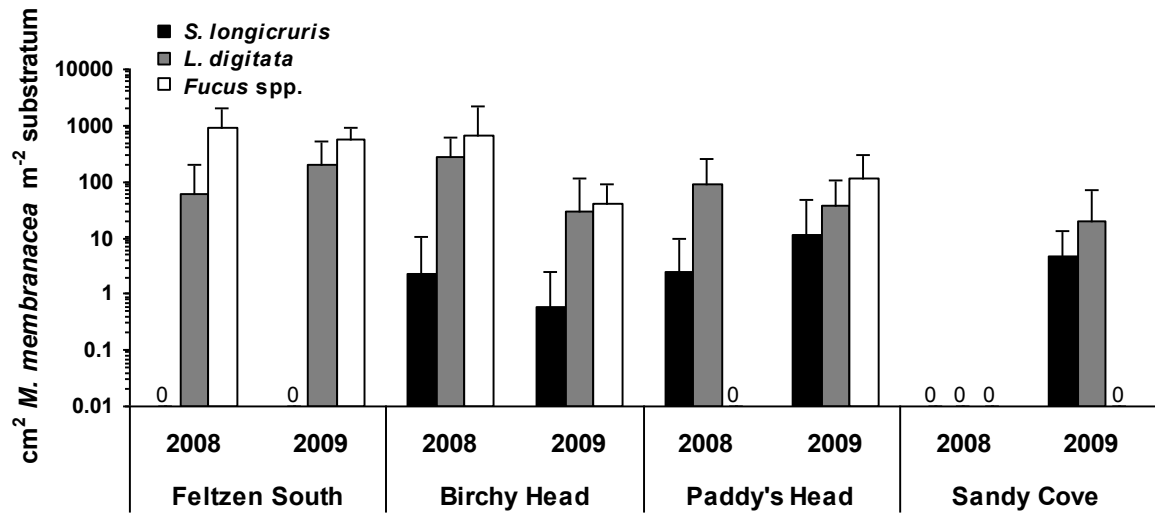
Fig. 2.6 Mean area of post-winter cover of colonies of *Membranipora membranacea* per unit area substratum on three algal taxa (*Saccharina longicruris*, *Laminaria digitata*, and *Fucus* spp.) at each of four sites on the southern shore of Nova Scotia in Jul 2008 and 2009 (+ SD, N = 7 - 11).

Table 2.6 Results of 3-way ANOVA examining the effects of algal taxon (fixed factor, 3 levels: *Saccharina longicruris*, *Laminaria digitata*, *Fucus* spp.), site (fixed factor, 4 levels: Feltzen South, Birchy Head, Paddy's Head, Sandy Cove), and year (random factor, 2 levels: 2008, 2009) on post-winter cover cm^2 m^{-2} substratum (July 2008, 2009) of *Membranipora membranacea*. Abbreviations as in Table 2.3; bold font indicates significant *p* values, $\alpha = 0.01$ (see Methods). Only significant differences in post hoc tests are shown (at $p < 0.05$).

Effect	MS	$F_{(df)}$	*p*	Tukey's HSD
Post-winter cover cm^2 m^{-2} substratum				
Algal taxon	68.358	$235.4_{(2, 2)}$	**0.004**	FS 2008: *Fs > Ld, Sl*
				2009: *Fs > Ld > Sl*
				BH 2008: *Ld, Fs > Sl*
				2009: *Fs > Ld, Sl*
				PH 2009: *Ld, Fs > Sl*
Site	34.497	$2.192_{(3, 3)}$	0.268	*Ld* 2008: BH > PH, FS, SC
				2009: FS > SC, BH; PH > BH
				Fs 2008: FS > BH > PH, SC
				2009: FS > BH, PH > SC
Year	6.657	$0.862_{(1, 0.64)}$	0.585	*Ld* FS: 2008 < 2009
				BH: 2008 > 2009
				SC: 2008 < 2009
				Fs PH: 2008 < 2009
Algal taxon x Site	23.994	$2.885_{(6, 6)}$	0.111	
Algal taxon x Year	0.290	$0.035_{(2, 6.00)}$	0.966	
Site x Year	15.736	$1.895_{(3, 6.01)}$	0.231	
Algal taxon x Site x Year	8.318	$4.496_{(6, 207)}$	**< 0.001**	
Error	1.850			

2.5 Discussion

We found that the invasive epiphytic bryozoan *Membranipora membranacea* settles, grows, and overwinters on *Fucus* spp. in its introduced range, as it does in native habitats of the Northeast Atlantic, and that it occurs on these substrates at magnitudes significant to populations in Nova Scotia. There was substantial spatial and temporal variation in the abundance of *M. membranacea*, suggesting that the relative importance of kelp and *Fucus* substrates at different stages of the population cycle of this bryozoan depends on various factors, such as larval supply and algal abundance and distribution, which can, in turn, vary temporally and spatially.

At sites where kelps were plentiful and distributed near *Fucus*, settlers were more abundant on kelps than on *F. evanescens*. This pattern did not hold, however, for sites with more open canopies resulting from low abundance of kelp or spatial separation of kelps and *Fucus*. Settlement of benthic invertebrates can be affected by numerous physical and biological factors, and larval preferences for specific substrate characteristics can strongly affect distribution patterns (McKinney and McKinney 1993). Larvae of *M. membranacea* are reported to settle preferentially on kelps, and this may be due to a combination of chemical and physical characteristics of these substrates. The presence of kelp tissue can induce settlement of *M. membranacea* in the laboratory (Stricker 1989). However, we found that the magnitude of settlement on *Fucus in situ* can equal or exceed that measured on kelps. Larvae of *M. membranacea* can be physically filtered by high kelps, which will then decrease larval supply beneath the canopy in kelp beds (Duggins et al. 1990). Consequently, settlers are likely to be abundant on *Fucus* spp. where kelps are sparse or distributed separately, as a result of the absence of this "kelp filtration" effect. Where kelps are particularly sparse, as under severe defoliation, it is possible that levels of settlement on taxa other than kelp may be further increased by limitation of space for settlement on kelps.

When adjusted for algal abundance at a particular site, settler abundance generally increased more on *Fucus* spp. than on kelps at both Feltzen South and Birchy Head. At Feltzen South, high abundance of settlers per unit area of substratum can be attributed to the dense and extensive distribution of *F. serratus* there, supplying higher surface area per unit area of benthos than kelps. *Fucus* is much less abundant at Birchy Head than at

Feltzen South; however, as kelps are particularly sparse at that site, they host fewer settlers per unit area of benthos than *F. evanescens*. Kelps were the principal substrates for settlement, both per unit area of algae and per unit area of substratum, at sites with closed kelp canopies, where settlers occur in particularly low abundance on *Fucus*.

While the algal taxa included in this study occurred over different depths at different sites, *Fucus* spp. were generally found at shallower depths than kelps. Consequently, spatial interspersion of substrates was not possible to the same extent at all sites. Several factors that vary with depth in subtidal habitats, other than algal substrate, may affect patterns in the abundance of *M. membranacea*. For example, temperature varies with depth and affects growth of *M. membranacea* (Saunders and Metaxas 2009a), as does settlement (Saunders and Metaxas 2007). We do not believe our results are compromised by the algal distributions for several reasons. At Feltzen South, and to some extent at Sandy Cove, distributions of all algal taxa overlapped. At Birchy Head, both *F. evanescens* and *L. digitata* occurred primarily at <1.5 m, ensuring interspersion. To minimize the effect of depth, *Fucus* and kelps were sampled at overlapping depths at all sites, and kelps were generally sampled at shallow and moderate depths (<8 m), except at Paddy's Head on one occasion during early settlement (Table 2.1). While Saunders and Metaxas (2007) found settlement on kelp to be significantly greater at 8 and 12 m than at 4 m depth at Paddy's Head throughout the settlement period, such differences were not consistent spatially and temporally across sites in St. Margarets Bay. If differential settlement occurred across depths, confounding differences detected here between *Fucus* and kelps, this would most likely only have occurred for Paddy's Head during early settlement when sampling of kelps extended to 14 m (sampling during other stages at this site occurred at <8 m).

Settlers were generally more abundant at sites in St. Margarets Bay than at other sites outside the bay, particularly during early settlement. While micro-hydrodynamic and larval behavioural processes, and substrate availability are important determinants of patterns at settlement at local scales, physical processes and larval transport have greater effects on the magnitude of settlement at larger scales (Pineda 2000). Larval supply was not quantified in this study; however, differences in timing and abundance of larvae among sites, and inside and outside St. Margarets Bay, are likely, as physical

mechanisms such as diffusion and advective transport, and upwelling and downwelling may vary across the study area. Saunders and Metaxas (2007) found that temperature was an important factor in determining patterns of settlement of *M. membranacea* in Nova Scotia, and suggested that high settlement within St. Margarets Bay at increased temperatures may be associated with larval retention. As *M. membranacea* is particularly abundant in St. Margarets Bay, larval retention would provide high supply to sites within the bay, and explain locally enhanced settlement. We detected fewer settlers at Sandy Cove than at most other sites on all algal taxa early in the settlement period, suggesting lower larval supply. Differences between Paddy's Head and Sandy Cove in timing and abundance of newly settled colonies have been previously observed on kelps (Saunders and Metaxas 2009b). As there were no significant differences in abundance of peak settlers among sites (e.g. for *L. digitata*), larval supply likely was high throughout the study area later in the settlement season.

Remarkably, despite the variation in patterns of settlement among sites, by late autumn, pre-winter cover was distributed similarly among algal taxa and sites. The pattern of decreasing cover from *L. digitata* to *S. longicruris* to *Fucus* spp. suggests differences among these taxa either in the growth or loss of colonies. Autumn cover of *M. membranacea* on *L. digitata* is greater than on other kelp species (including *S. longicruris*) and has been attributed to lower loss of colonies via erosion on *L. digitata* due to the relative stability of this kelp species (Saunders and Metaxas 2009b). Differential growth of *M. membranacea* colonies has not been observed on *S. longicruris* and *L. digitata*, and is unlikely because of the similarity of these kelps (Saunders and Metaxas 2009b). However, several differences between kelps and *Fucus* make differential growth between these two substrate types likely. Whereas space is abundant on kelps, *Fucus* thalli are relatively small and divided by branching. Additionally, space is more likely to be occupied by other epiphytic species on *Fucus* due to slower growth and the stability of this alga. Growth of *M. membranacea* is likely limited on *Fucus* spp. due to reduced space availability, and restriction of colony size and growth rate on this substrate (Saunders and Metaxas 2009a; Yorke and Metaxas submitted).

While post-winter cover on kelps in 2009 was an order of magnitude lower than pre-winter cover in 2008, colony cover on *Fucus* remained stable over that period,

suggesting high overwintering survival of *M. membranacea* on substrates other than kelp. Patterns of post-winter cover on kelps and *Fucus* spp. can largely be explained by retention or erosion of algal tissue, as it relates to the growth strategies of each algal taxon. Kelps grow basally and erode distally, and growth of kelps in this region is particularly rapid in late winter and early spring (Mann 1972; Krumhansl and Scheibling in press). Thus, frond tissue of *S. longicruris* and *L. digitata* is replaced during winter, allowing these species to shed epiphytes from fronds. Conversely, growth of fucoids is apical with minimal erosion and breakage, and growth occurs exclusively during summer when active photosynthesis is possible (Mann 1973). Differential retention may occur among kelps, and colonies of *M. membranipora* can overwinter in greater abundance on *L. digitata* than on *S. longicruris* (Saunders and Metaxas 2009b), likely because of reduced rates of breakage of the former which is more resilient to wave action. The incidence of post-winter cover on kelps reported here is principally due to retention of colonies on stipes and the bases of fronds. However, deeper refuges may exist on kelps where growth is slower and erosion is reduced due to less intense wave action.

Differences among sites and between years in patterns of post-winter cover per unit area substratum may be explained by factors affecting abundance of both *M. membranacea* and algae. For example, as post-winter cover was low on all algal taxa at Sandy Cove in 2008, low larval supply at that site in the previous season may explain our observations. However, this cannot be confirmed as settlement and pre-winter cover were not measured in 2007. Erosion rates of *S. longicruris* and *L. digitata* are highly variable in this region, and are likely sources of variation in post-winter cover on kelps, as erosion is related to both site exposure and cover of *M. membranacea* (Krumhansl and Scheibling in press). Overwintered *M. membranacea* on *Fucus* was greatest at Feltzen South, and this is associated with high abundance of *F. serratus* at that site. Despite site-specific and annual variation, where settlement and growth of *M. membranacea* occur on *Fucus* spp., and where this alga occurs in abundance, *Fucus* represents an important refuge for the overwintering of colonies in this region.

Successful overwintering on *Fucus* is important to the population dynamics of *M. membranacea* in Nova Scotia in multiple ways. These refuges can preserve local populations of this invasive bryozoan through winter in years when severe defoliation

removes colonies on kelps. In turn, where defoliation has been severe enough to prevent regrowth of kelps in the following season, *Fucus* can also provide a non-kelp substrate for settlement and growth. Furthermore, physical characteristics of *Fucus* substrates likely accelerate reproduction in *M. membranacea*. Whereas colonies of *M. membranacea* normally do not become reproductive until they are large and mature (approximately 2500 mm^2, aged 40 - 60 days in the East Pacific) (Harvell 1992), colonies that are crowded by conspecifics reproduce sexually, regardless of colony size and age (Harvell and Grosberg 1988). Colonies of *M. membranacea* on *Fucus* spp. in Nova Scotia may therefore reproduce earlier in the season than those on kelps, supplying larvae that can settle on kelps in spring and early summer. The timing of settlement can greatly affect *M. membranacea* populations, and models have revealed that early onset of settlement in Nova Scotia can cause exponential increases in colony abundance, size, and cover later in the season (Saunders et al. 2010).

Settlement, growth, and overwintering of *M. membranacea* on *Fucus* spp. also has implications for local spread of this invasive bryozoan. Although kelps are relatively continuously distributed along the Atlantic coast of Nova Scotia, there are areas in the region where kelps are sparse or absent and algal assemblages are dominated by *Fucus* spp. (Watanabe et al. 2009). The presence of *M. membranacea* on both *F. evanescens* and *F. serratus* in Nova Scotia suggests that these algal species may act as vectors for range expansion.

We have demonstrated that an understanding of the population dynamics of *M. membranacea* in Nova Scotia should extend beyond kelps to include the occurrence of this invasive bryozoan on *Fucus* spp. and possibly other algal taxa. We have also demonstrated that the relative importance of kelp and fucoid substrates varies both intra- and inter-annually, as well as spatially. In summary, we observed abundant settlement on both kelps and *Fucus* spp., and showed that *Fucus* can be an important substrate for settlement, specifically at locations with open kelp canopies. We propose that *Fucus* provides an important refuge for overwintering colonies, particularly where defoliation of kelps has been extensive, and characteristics of this substrate likely facilitate early reproduction and local spread. Quantification of *M. membranacea* on these algal taxa has expanded our understanding of the population dynamics of this bryozoan in Nova Scotia

and has revealed mechanisms that contribute to the success and spread of this invasive species.

2.6 Acknowledgements

Chapter 3

Interactions Between an Invasive and a Native Bryozoan (*Membranipora membranacea* and *Electra pilosa*) Species on Kelp and *Fucus* Substrates in Nova Scotia[1]

3.1 Abstract

The invasive bryozoan *Membranipora membranacea* interacts with native *Electra pilosa* on two substrates in northwest Atlantic subtidal habitats: highly dynamic and fast-growing kelps; and smaller, more stable, and slow-growing fucoid algae. We quantified the relative abundance and evaluated encounter outcomes of the two bryozoans on kelp and *Fucus* at four sites in Nova Scotia. We also examined the effects of substrate (kelp, *Fucus*), temperature (7, 10, 13ºC), and food (limited, unlimited) on growth rates of *E. pilosa* in the laboratory and the field. *Membranipora membranacea* was relatively more abundant on kelps than on *Fucus*, and competitive standoffs were more frequent than expected, with no differences between substrates. Growth of *E. pilosa* was faster on *Fucus* than kelp and increased with temperature, and was slower than that of *M. membranacea*. Large colony size, faster growth, and strong overgrowth abilities likely interact on kelps to ensure success of the invasive bryozoan; however, growth on non-kelp substrates is limited by space availability, and restriction of colony size and growth rate.

3.2 Introduction

Invasive species have been described as a major threat to biodiversity and have been associated with significant ecological and economic impacts. Our understanding of species introductions is expanding, and recent research has revealed both complexity and paradox in the nature of species invasions (Fridley et al. 2007; Stachowicz and Byrnes 2006). Much of the research on biological invasions has focused on cases where the introduced species are very successful and disruptive, and, in most instances, this has led

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[1] Chapter 3 has been submitted as a research article to *Marine Biology*:
Yorke, A.F. and Metaxas, A. Interactions between an invasive and a native bryozoan (*Membranipora membranacea* and *Electra pilosa*) species on kelp and *Fucus* substrates in Nova Scotia.
Dr. Metaxas (thesis supervisor, Dalhousie University) assisted with experimental design, provided advice on statistical analyses, and assisted with manuscript editing.

to a more thorough understanding of the biology of the invader than of certain native species. However, research on species introductions should also focus on native species, and include contexts where the presence of introduced species may not be noticeable due to their limited abundance and / or low impacts on native communities. Knowledge of the biology of both native and introduced species is essential for understanding their interactions, and these interactions may be important to the population dynamics of one or both groups. Additionally, research performed in less obvious contexts may reveal factors that limit the success and spread of invasive species.

Membranipora membranacea is a colonial encrusting bryozoan and an invasive species in subtidal habitats of the northwest Atlantic. Colonies encrust the blades of native kelps, making them brittle and causing defoliation by storms (Dixon et al. 1981). This defoliation can result in dramatic changes to native kelp beds, particularly in seasons following warmer winters when population outbreaks of *M. membranacea* occur due to increased survival of overwintering colonies, as well as earlier and higher settlement and recruitment of new colonies (Berman et al. 1992; Lambert et al. 1992; Saunders and Metaxas 2007, 2008; Scheibling and Gagnon 2009). An invasive green alga, *Codium fragile* ssp. *fragile*, colonizes spaces opened by defoliation and, once established, can prevent recolonization by kelps (Levin et al. 2002; Scheibling and Gagnon 2006). Previous research in the northwest Atlantic (mainly the Gulf of Maine, USA, and Nova Scotia, Canada) has focused on the impacts of *M. membranacea* on the native community, and almost all studies have been on the occurrence of the bryozoan on the kelps *Saccharina longicruris* and *Laminaria digitata* (Lambert et al. 1992; Levin et al. 2002; Saunders and Metaxas 2007, 2008, 2009a, 2009b; Scheibling and Gagnon 2009; Scheibling and Gagnon 2006).

In addition to kelps, *M. membranacea* occurs on two *Fucus* species in the northwest Atlantic, the native *F. evanescens* and the introduced *F. serratus*. These non-kelp substrates provide contrasting conditions from kelps for colonial epiphytes in this region. Whereas kelps can sustain great losses of biomass during autumn storms and re-grow quickly in winter and early spring, providing a large, seasonally variable amount of primary space, fucoid algae have much lower intrannual variation in biomass, slower growth rates, and smaller surface area (Johnson and Mann 1988; Mann 1972; McCook

and Chapman 1991). Further, the characteristics of the surface available for settlement varies between the two types of substrates. *Fucus* thalli are composed of narrow, branching segments with uneven surface topography, while kelp blades are broad and continuous with smooth surfaces.

In addition to its interaction with different algal substrates, *M. membranacea* encounters other epiphytic species in its introduced range (Berman et al. 1992; Chavanich and Harris 2000), including the native cheilostome bryozoan, *Electra pilosa*. Colonies of *E. pilosa* are generally smaller than those of *M. membranacea*, and *E. pilosa* occurs on kelps in this region, such as *S. longicruris*, *L. digitata*, and *Agarum cribrosum*, as well as non-kelp substrates, such as *F. evanescens*, *F. serratus*, and *Chondrus crispus* (personal observations). The percentage cover of *E. pilosa* and *M. membranacea* on kelps, as well as the proportion of kelps and *C. crispus* harboring these epiphytes, have been previously measured in the Gulf of Maine (Berman et al. 1992; Lambert et al. 1992). However, the relative abundance of *M. membranacea* and *E. pilosa* has not been compared between kelp and *Fucus* substrates.

Electra pilosa has not been associated with any significant effect on the population dynamics of *M. membranacea* in the northwest Atlantic, and is considered to be competitively subordinate to the invasive bryozoan in this region (Berman et al. 1992). Encounters between *M. membranacea* and *E. pilosa* have been previously examined on kelps in the Gulf of Maine (Berman et al. 1992), where *M. membranacea* overgrew *E. pilosa* in most competitive encounters. Conversely, in the northeast Atlantic, where *M. membranacea, E. pilosa,* and *F. serratus* are native, *M. membranacea* does not commonly overgrow *E. pilosa*, and is virtually excluded by the bryozoan epiphyte assemblage on *F. serratus* in the lower intertidal zone (O'Connor et al. 1980; Okamura 1988). It is unknown, however, how these introduced and native bryozoans interact on *Fucus* substrates in subtidal habitats of the northwest Atlantic. Measures on *F. serratus* in the northeast Atlantic were collected in intertidal habitats and may underestimate the competitive ability of *M. membranacea*, as the bryozoan is mainly a subtidal species (Best and Thorpe 1986). Interactions between *M. membranacea* and *E. pilosa* may also differ between regions due to differences in bryozoan species richness; epiphyte assemblages in the northeast Atlantic, where *M. membranacea* and *E. pilosa* are both

native, include several species of cheilostome and ctenostome bryozoans (Best and Thorpe 1986; O'Connor et al. 1980; Okamura 1988), whereas *E. pilosa* and *M. membranacea* are the only bryozoan epiphytes commonly observed in subtidal habitats in Nova Scotia (Watanabe et al. 2009). Substrate may also affect competitive encounters, and because studies set in the northeast and northwest Atlantic have been on different substrates, comparisons between outcomes on kelp and *Fucus* substrates have not been possible.

Although *M. membranacea* is considered competitively dominant in its introduced range, several outcomes of encounters with *E. pilosa* may occur. Outcomes such as stand-offs and competitive reversals can prevent competitive dominance and monopolization of resources, thereby preserving biodiversity (Walters and Wethey 1986). Therefore, whereas encounters between native and introduced bryozoans may not be important for the population dynamics of *M. membranacea* on kelps, the outcomes may be very significant for the dynamics of *E. pilosa* both on kelp and *Fucus* substrates.

A critical factor affecting outcomes of competitive encounters, and regulating space occupation by colonial encrusting animals, is growth. Indeterminate outcomes have been observed more frequently among colonial bryozoans than in other taxa, such as sponges and tunicates, likely due to the highly directional nature of bryozoan growth (Quinn 1982). The success of *M. membranacea* in subtidal habitats in Nova Scotia has been attributed to high growth rate of colonies (e.g. up to 12 mm $\cdot$ day^{-1}; Saunders and Metaxas, 2009a) and their ability to reach large sizes (Saunders and Metaxas 2009a), a combination that is possible on kelps but not on *Fucus* substrates.

As *E. pilosa* is a cosmopolitan species (Nikulina et al. 2007), several studies have examined factors that regulate its growth. The effects of such factors as temperature, food availability, flow, and the presence of conspecific and heterospecific neighbors on the growth of *E. pilosa* have been examined in both laboratory and field settings, but mostly in the northeast Atlantic (Bayer et al. 1994; Hermansen et al. 2001; Menon 1972; Menon 1974; Okamura 1988; Okamura 1992). In these studies, higher temperatures have resulted in greater feeding and growth rates in *E. pilosa* (Menon 1972; Menon 1974), but over wide temperature ranges that extend above temperatures observed in Nova Scotia (Saunders and Metaxas 2007). Food concentration can also have an effect on the growth

rate of *E. pilosa* at algal concentrations that overlap with the range quantified in coastal
Nova Scotia (Bayer et al. 1994; Metaxas and Scheibling 1996). The effect of algal
substrate (kelp and *Fucus* species) on growth of *E. pilosa* has not been examined,
however, and the interactive effects of temperature and food within ranges observed in
the northwest Atlantic have not been tested.

In the present study, we examine: (1) the relative abundance of the bryozoans *M.
membranacea* (introduced in Nova Scotia) and *E. pilosa* (native in Nova Scotia) on kelp
and *Fucus* substrates; (2) outcomes of encounters between these two bryozoans on
various substrates in different seasons; (3) the effects of substrate, temperature, and food
on growth of *E. pilosa* in the laboratory; and (4) the effect of temperature on growth of *E.
pilosa* in the field. We compare our findings on factors affecting the growth of *E. pilosa*
to those on *M. membranacea* obtained in similar (and thus directly comparable)
laboratory and field experiments (Saunders and Metaxas 2009a). Such a comparison
allows us to reveal possible interactions between factors that may affect the relative
abundance of these two species within the epiphytic community, and that may regulate
the population dynamics of the native bryozoan. Our study provides insight into
mechanisms of coexistence of native and introduced species.

3.3 Materials and Methods

3.3.1 Study Sites

We sampled 5 sites in and near St. Margarets Bay, Nova Scotia, Canada, where
the population dynamics of *M. membranacea* have been studied extensively before
(Saunders and Metaxas 2007, 2008, 2009a, 2009b; Scheibling and Gagnon 2009) (Fig.
3.1). Birchy Head (44º34.5'N, 64º02.5'W) and The Lodge (44º33.3'N, 64º01.9'W),
located on the western shore of St. Margarets Bay, have moderately sloping granite
substrata with boulders and cobbles; however, the topography of The Lodge is more
varied than at Birchy Head with granite outcroppings and large boulders. The kelps
Saccharina longicruris and *Laminaria digitata* occur at ~4 - 14 m depth, and shallower
areas (<4 m) are dominated by turf algae interspersed with patches of *Fucus evanescens*
(unpublished data). Paddy's Head (44º31.6'N, 63º57.2'W), on the eastern shore of St.
Margarets Bay, is more steeply sloping than the other two sites and is dominated by

abundant mixed kelps at depths >4 m, including *S. longicruris*, *L. digitata*, and *Agarum clathratum*. Shoreward, *F. evanescens* occurs sparsely interspersed with turf algae at depths <3 m. Sandy Cove (44º27.8'N, 63º42.5'W) is located 20 km east of St. Margarets Bay at the mouth of Terence Bay in Pennant Bay. The site includes an outer slope with bedrock ridges dominated by abundant mixed kelps (*S. longicruris*, *L. digitata*, and *A. clathratum*) at depths >4 m, as well as a gently sloping sheltered inner cove with *F. evanescens* occurring at ~4 m on a cobble bottom. Feltzen South (44º19.9'N, 64º16.9'W) is located on the southern shore of Nova Scotia near the mouth of Lunenburg Bay, and 30 km southwest of St. Margarets Bay. The site consists of a gentle slope of bedrock (4 - 6 m depth, ~150 m offshore) covered by an extensive, dense meadow of *Fucus serratus*. Offshore, a steep bedrock ridge runs parallel to the shore (6 - 10 m depth) with *S. longicruris* and *L. digitata* occurring down to a sand and gravel bottom at ~11 m depth.

3.3.2 Relative Abundance of Epiphytic Bryozoans

The relative abundances of *M. membranacea* and *E. pilosa* on algal substrates were measured with SCUBA at all sites, except The Lodge, in Nov/Dec 2008 during the annual period of high colony cover. At each site, all thalli of each of *S. longicruris*, *L. digitata*, and *F. evanescens* (or *F. serratus* at Feltzen South) were collected from 7 – 10 independent, randomly positioned 0.5-m^2 quadrats (0.25-m^2 for *F. serratus*) along transects, transferred to dry tubs at the surface, and transported to the laboratory where they were placed in tanks with running ambient seawater. For each quadrat, all colonies of *M. membranacea* and *E. pilosa* were traced on a single randomly selected side of kelp thalli, or on approximately 0.50 m^2 surface area for *Fucus* spp. All kelp thalli were sampled for quadrats containing ≤ 5 kelps; otherwise 5 thalli were randomly selected, except for one quadrat where 9 thalli were sampled. The tracings were photographed with a Nikon Coolpix 995 digital camera, and measured using SigmaScan Pro Image Analysis 5.0 (SPSS) software. Mean percent cover of colonies on algae was calculated for each quadrat for both *M. membranacea* and *E. pilosa*. Relative bryozoan abundance was calculated as the ratio of *M. membranacea* cover to total bryozoan cover for quadrats with non-zero values for *M. membranacea* cover.

3.3.3 Encounter Outcomes Between M. membranacea *and* E. pilosa

Approximately 40 thalli of each of *S. longicruris*, *L. digitata*, *F. evanescens*, and *F. serratus* were collected with SCUBA between Nov 2008 and Oct 2009 from Feltzen South, The Lodge, and Birchy Head (where *M. membranacea* and *E. pilosa* co-occur on the algal substrates) and frozen for storage. In the laboratory, encounters between pairs of heterospecific colonies were examined on algal samples thawed in ambient seawater, and outcomes were scored as: (1) *wins* or *losses*, when any part of the leading edge of a colony covered the apertures of another colony; or (2) *standoffs / ties,* when colonies were contacting but no overgrowth was apparent, or when both colonies overgrew one another. As single colonies of *M. membranacea* often encounter multiple colonies of *E. pilosa*, a single colony was randomly selected from all encountering *E. pilosa* colonies to be paired with each colony of *M. membranacea*. Additionally, the outcomes of all encounters along the leading edges of colonies were scored for 28 and 93 colonies of *M. membranacea* and *E. pilosa*, respectively.

3.3.4 Growth of E. pilosa*: Laboratory Experiments*

Colonies of *E. pilosa* growing on kelps (*S. longicruris* and *L. digitata*) and *F. evanescens* were collected with SCUBA from The Lodge and Paddy's Head, respectively, in Nov 2009, transported in 4ºC seawater, and placed in running ambient seawater tables within 3 h of collection. Algal thalli were cut into segments: kelp fronds were trimmed with scissors to sections approximately 20 x 30 cm; and *Fucus* thalli were separated at the bases of stipes or at holdfasts. Five colonies were selected on each segment and labelled with small cable ties.

Six kelp segments and 6 *Fucus* segments, each with 5 colonies, were placed in each of 6 flow-through seawater tanks, randomly assigned to a single treatment combination of three temperatures (7, 10, and 13ºC) and two food treatments (limited and unlimited food) (Table 3.1). Colonies in the limited food treatment were maintained in filtered seawater, whereas colonies in the unlimited food treatment were fed *Tetraselmis chuii* ($\sim 1 \times 10^6$ cells ml^{-1} culture) supplied constantly by a drip system. The "unlimited food" algal concentration was identical to the one used by Saunders and Metaxas (2009a)

for *M. membranacea*, and is above the range of phytoplankton concentrations measured in coastal Nova Scotia from March to November ($10^0 - 10^4$ cells ml^{-1}) (Metaxas and Scheibling 1996). Algal segments were gently stirred by hand once daily, altering their position within the tank and providing additional flow over colonies. Edges of segments were trimmed regularly to discard deteriorated material. Temperature was measured in each tank throughout the experiment with data loggers (Onset Co. Pendant; accuracy ±0.47ºC). Labelled colonies were photographed at the initiation of the experiment (day 0), and then every 5 days for 30 days (day 30), using an Olympus SZ-61 dissecting microscope with a mounted Nikon Coolpix 995 digital camera.

To determine growth of each colony, the perimeter of the colony including only completed zooids (those with a visibly formed base of the frontal wall where the distal portion of the loop is continuous) was traced on images taken at day 0. New completed zooids beyond that perimeter that were present on images at day 30 (to maximize growth and minimize error in growth measurements) were counted and used to calculate growth rate (zooids·day^{-1}). Some colonies were discarded before completion of the experiment due to degradation of algal substrates, and of the 30 colonies initially placed in each treatment, we sampled 20 colonies to ensure balance across groups. For treatments where fewer than 20 colonies were present on day 30, we used photographs recorded at as early as 15 days to measure growth for a total of 41 colonies. Initial colony surface area was measured from images on day 0, using SigmaScan Pro Image Analysis 5.0 (SPSS) software. Additionally, size of zooids grown during the experimental period was measured for 20 colonies both on kelp and *Fucus* species, at each of 7 and 13ºC, and in both limited and unlimited food.

3.3.5 Growth of E. pilosa*: Field Experiments*

We measured growth rate of *E. pilosa in situ* at The Lodge, using the technique described in Saunders and Metaxas (2009a) for *M membranacea*. Briefly, large plastic bags, were used to enclose kelps (*S. longicruris* and *L. digitata*) with ~1 x 10^3 − 1 x 10^4 colonies of *E. pilosa* per frond, cinched around the kelp stipes and injected with the calcium-binding dye Alizarin Red S (Sigma Aldrich). The dye was delivered at a concentration of approximately 200 g · ml^{-1}. Following dye injection, bags were closed

tightly around the stipes and left for approximately 20 h, after which they were removed. Colonies were allowed to grow at each of 8 and 12 m depth from 16 Sep to 3 Oct 2009 (17 days) and from 22 Oct to 9 Nov 2009 (18 days) (Table 3.1). Temperature was measured at 10-min intervals by data loggers, affixed to the substratum 1 - 5 m from dyed colonies at each depth, and averaged at daily intervals. At the end of each growth period, kelps with colonies were removed, transported to the laboratory in dry tubs, and frozen for storage within 12 h of collection. Kelps were later thawed in ambient seawater and colonies were photographed with a digital camera mounted on a dissecting microscope. Growth was measured on images by counting zooids completed during the growth period (as for the laboratory experiment), and growth rate was calculated as newly completed zooids·day^{-1}. New growth was discernible in dyed colonies because skeletal material present during the application of Alizarin Red S (previous growth) was stained pink (see Saunders and Metaxas 2009a).

3.3.6 Statistical Analyses

The effects of substrate (fixed factor: *S. longicruris*, *L. digitata*, and *Fucus* spp.) and site were examined by two-way ANOVA on the following: (1) percent cover of *M. membranacea* on algae, (2) percent cover of *E. pilosa* on algae, and (3) relative abundance of *M. membranacea* and *E. pilosa*. Site was also treated as a fixed factor because sites were selected for specific characteristics, such as algal composition, algal abundance, and proximity to St. Margarets Bay [and in some instances to be identical to the ones in Saunders and Metaxas (2007, 2008)].

Differences in the frequency of outcomes of encounters between *M. membranacea* and *E. pilosa* among combinations of substrates, sites, and seasons, were tested using a chi-square test of homogeneity. Additionally, if *M. membranacea* is competitively dominant, we would expect colonies to win over *E. pilosa* in all, or nearly all, encounters. To test this hypothesis, we used a chi-square goodness of fit test on the combined data from all substrates, sites and seasons. We used a conservative minimum expected frequency of 5 for both "stand-off / tie" and "*E. pilosa* wins" categories to fulfill the assumptions of the analysis (Zar 1999).

The effects of substrate (2 levels: kelp and *Fucus*), temperature (3 levels: 7, 10, and 13ºC), and food (2 levels: limited and unlimited) on growth rate were examined using 3-way ANCOVA, with initial colony size as a covariate, and the relationship between initial colony size and growth rate was determined using a simple linear regression. As there were no significant interactions (4-, 3- or 2-way) between the factors and the covariate, the ANCOVA model was revised to only retain the main factors, colony size as a covariate, and interactions among factors. Our measures of growth rate by quantification of new zooids did not incorporate zooid area; however, previous research has shown that this may be affected by various factors. We therefore used 3-way ANOVA to examine the effects of substrate (kelp and *Fucus*), temperature (7 and 13ºC), and food (limited, unlimited) on zooid size. Growth rates measured in the field were compared using 2-way ANOVA with growth period and depth as factors. Growth rates measured in both the laboratory and the field on the same substrate (kelp) and at similar temperatures were compared using 1-way ANOVA (see Table 3.5 for groups).

For the analyses, percent cover of colonies on algae and relative abundance of *M. membranacea* (proportion of *M. membranacea* cover to total bryozoan cover) were arcsine-square root transformed, and growth rates of *E. pilosa* and initial colony sizes in the laboratory were $\log(x + 0.001)$ -transformed to reduce heterogeneity of variances as detected by Levene's test ($p < 0.001$). However, since homogeneity was not achieved through transformation, a more conservative $\alpha_{crit} = 0.01$ was used for these analyses. Relative abundances of *M. membranacea* and growth rates of *E. pilosa* in the laboratory were also not distributed normally according to the Shapiro-Wilk test, but ANOVA is robust to deviations from normality (Zar 1999). Growth rates of *E. pilosa* measured in the field, and growth rates measured in the laboratory and field at similar temperatures (~11ºC and ~13º), were square-root transformed to attain homogeneity of variance, as well as normality for field growth rates only. Statistical analyses were conducted using SPSS 15.0. Where appropriate, homogeneous subsets were identified using Tukey's HSD test.
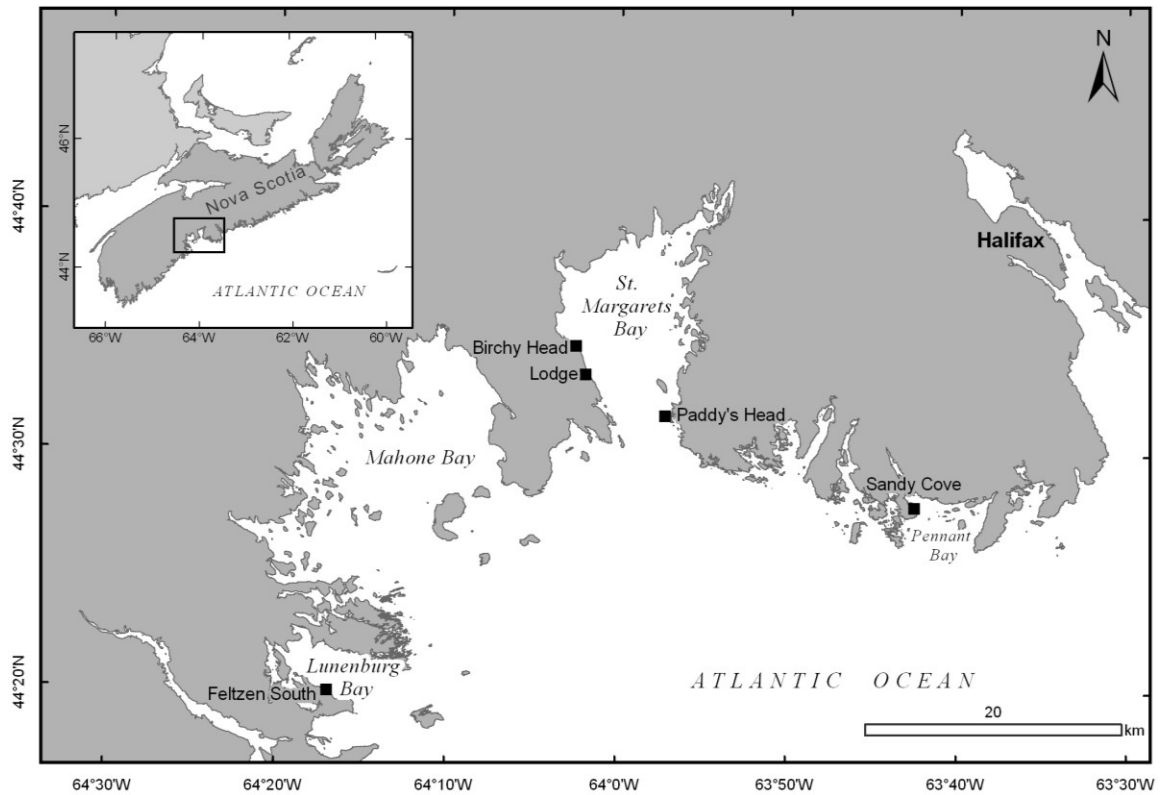
Fig. 3.1 Study area on the southern shore of Nova Scotia, Canada showing sampling sites in St. Margarets Bay (Lodge, Birchy Head, and Paddy's Head), Pennant Bay (Sandy Cove), and Lunenburg Bay (Feltzen South). (© Department of Natural Resources Canada. All rights reserved.)

Table 3.1 Details of laboratory and field experiments conducted to measure growth rate of *Electra pilosa*. (Kelp: *Saccharina longicruris*, *Laminaria digitata*; SMB: St. Margarets Bay; NS: Nova Scotia; n/a: not applicable.) Laboratory: N values are listed in the same order as temperature treatments; Field: Temperatures and N values are listed in the same order as depths.

Location	Time period	Substrate	Food	Depth (m)	Temperature (°C, mean ± SD)	N	Technique
Laboratory							
Dalhousie University	16 Nov-16 Dec 09	Kelp	Unlimited	n/a	7.4 ± 0.1, 10.4 ± 0.4, 13.4 ± 0.2	20, 20, 20	Imaging of tagged colonies
	16 Nov-16Dec 09	Kelp	Limited	n/a	7.3 ± 0.1, 10.2 ± 0.5, 13.3 ± 0.0	20, 20, 20	Imaging of tagged colonies
	17 Nov-17 Dec 09	*Fucus evanescens*	Unlimited	n/a	7.4 ± 0.1, 10.4 ± 0.4, 13.4 ± 0.2	20, 20, 20	Imaging of tagged colonies
	17 Nov-17 Dec 09	*Fucus evanescens*	Limited	n/a	7.3 ± 0.1, 10.2 ± 0.5, 13.3 ± 0.0	20, 20, 20	Imaging of tagged colonies
Field							
Lodge, SMB, NS	16 Sep-3 Oct 09	Kelp	n/a	8, 12	13.4 ± 1.3, 12.4 ± 1.6	20, 20	Dye
	22 Oct-9 Nov 09	Kelp	n/a	8, 12	11.0 ± 0.5, 11.1 ± 0.4	20, 20	Dye

3.4 Results

3.4.1 Relative Abundance of Epiphytic Bryozoans

Cover of bryozoan colonies on algal substrates ranged from $0.01 \pm 0.00\%$ (mean $\pm$ SE, N = 10) on *F. evanescens* at Sandy Cove to $44.6 \pm 10.6\%$ (N = 9) on *L. digitata* at Paddy's Head for *M. membranacea*, and from $0.00 \pm 0.00\%$ (N = 9) on *S. longicruris* and *L. digitata* at Paddy's Head to $7.3 \pm 6.9\%$ (N = 10) on *S. longicruris* at Feltzen South for *E. pilosa* (Fig. 3.2). The proportion of *M. membranacea* cover to total bryozoan cover ranged from $4.1 \pm 2.7\%$ (mean $\pm$ SE, N = 9) on *F. evanescens* at Sandy Cove to $100 \pm 0\%$ (N = 7, 9) on *S. longicruris* and *L. digitata* at Paddy's Head.

There was a significant interaction between sites and substrates on percent cover of *M. membranacea* on algae ($F_{6, 105} = 8.578$, $p < 0.001$). At Birchy Head, *M. membranacea* cover was significantly higher on *L. digitata* than *S. longicruris*, and higher on *S. longicruris* than *F. evanescens* (Tukey's HSD test, $p < 0.05$). At Paddy's Head and Sandy Cove, *M. membranacea* cover was higher on *L. digitata* than on both *S. longicruris* and *F. evanescens*. There were no significant differences among substrates at Feltzen South (Tukey's HSD test, $p > 0.05$). Cover of *M. membranacea* was significantly higher at Birchy Head than at all other sites for *S. longicruris*, higher at Paddy's Head and Birchy Head than at Sandy Cove and Feltzen South on *L. digitata*, and higher at Birchy Head than at Sandy Cove for *Fucus* spp.

There were significant effects of both site and substrate on percent cover of *E. pilosa* on algae (Site: $F_{3, 105} = 12.93$, $p < 0.001$; Substrate: $F_{2, 105} = 9.30$, $p < 0.001$) with no significant interaction between sites and substrates ($F_{6, 105} = 1.21$, $p = 0.308$). Cover of *E. pilosa* was significantly greater on *Fucus* spp. than on both kelp species, and significantly higher at Feltzen South than at all other sites (Tukey's HSD test, $p < 0.05$).

There was also a significant interaction between sites and substrates on the relative abundance of *M. membranacea* ($F_{6, 99} = 4.75$, $p < 0.001$). At all sites, the proportion of *M. membranacea* was significantly lower on *Fucus* substrates than on both kelp species (Tukey's HSD test, $p < 0.05$), but there were no significant differences between kelp species for any site (Tukey's HSD test, $p > 0.05$). Relative abundance of *M. membranacea* on *Fucus* substrates was greater at Birchy Head than at all other sites,

whereas, for kelp substrates, relative abundance was lower at Feltzen South than at all other sites.

3.4.2 Encounter Outcomes Between M. membranacea and E. pilosa

Most encounters were wins by *M. membranacea*, and the least frequent encounters were losses (Fig. 3.3). The frequency distributions of encounter outcomes did not vary among different combinations of substrate, site and sampling season ($\chi^2_8 =$ 10.559, $p > 0.05$). However, analysis of frequencies of encounters pooled across all combinations showed that the distribution of outcomes differed from those expected if *M. membranacea* nearly always won (Table 3.2). In particular, the observed frequency of standoffs / ties was greater than expected. Examination of encounters between individual *M. membranacea* colonies and multiple *E. pilosa* colonies revealed that outcomes can differ along the growing edge of a single *M. membranacea* colony (2 - 6 encounters per *M. membranacea* colony; *M. membranacea* wins: 69.6 ± 6.3%, mean ± SE, N=93; standoffs / ties: 29.7 ± 6.3%; *E. pilosa* wins = 0.7 ± 0.7%).

3.4.3 Growth of E. pilosa: Laboratory Experiments

Growth rates ranged from 0 to 2.83 zooids·day^{-1}, with the slowest growth occurring on kelp at 7ºC with unlimited food, and the fastest on *Fucus* at 7ºC with unlimited food (Fig. 3.4). The effect of initial colony size (*CS*) on growth rate (*GR*) was positive ($GR = 0.005CS + 0.150$; R$^2 = 0.032$; $p = 0.006$; N = 240), although this effect was marginally non-significant at $\alpha = 0.01$ (see Methods; Table 3.3; Fig. 3.4). There was a significant interaction between substrate and temperature on growth rate. Growth was slower on kelp than *Fucus* at 7 and 13ºC (Tukey's HSD tests, $p < 0.05$). On kelps, there was no difference in growth rate among temperatures, but on *Fucus* growth rate was higher at 7ºC than 10ºC.

The size of *E. pilosa* zooids grown in the laboratory ranged from 0.095 to 0.251 mm^2 (Fig. 3.5). Substrate and temperature both had significant effects on zooid size (Table 3.4); zooid surface area was larger on kelps than on *Fucus*, and at 7ºC than at 13ºC (Tukey's HSD tests, $p < 0.05$). There was no significant effect of food treatment, and no significant interactions among factors.

3.4.4 Growth of E. pilosa*: Field Experiments*

Field growth rates of *E. pilosa* colonies ranged from 0.11 to 3.05 zooids·day^{-1} (Fig. 3.6), overlapping those in the laboratory (0 to 2.83 zooids·day^{-1}). There was a marginally significant interaction between growth period and depth on growth rate ($F_{1, 76}$ = 4.306, p = 0.041); however, there were no significant differences between depths for each growth period (Tukey's HSD tests, $p > 0.05$). Growth was significantly faster during the earlier period than the later period (Tukey's HSD tests, $p < 0.05$), and the earlier period was associated with warmer mean temperatures (Fig. 3.6).

When growth of *E. pilosa* on kelp was compared at similar temperatures, field growth rates were generally higher than laboratory growth rates. Within each temperature group (~11°C and ~13°C; Table 3.5), significant differences in growth rate were found (1-way ANOVA: ~11°C: $F_{3, 76}$ = 5.85, p = 0.001; ~13°C: $F_{2, 57}$ = 37.66, $p <$ 0.001). Specifically, at ~11°C, growth was faster in the field than in the laboratory when food was limited (Tukey's HSD test, $\alpha < 0.05$), but did not vary from growth in the laboratory in unlimited food (Tukey's HSD test, $\alpha > 0.05$). At ~13°C, growth was faster in the field than in the laboratory for both limited and unlimited food.
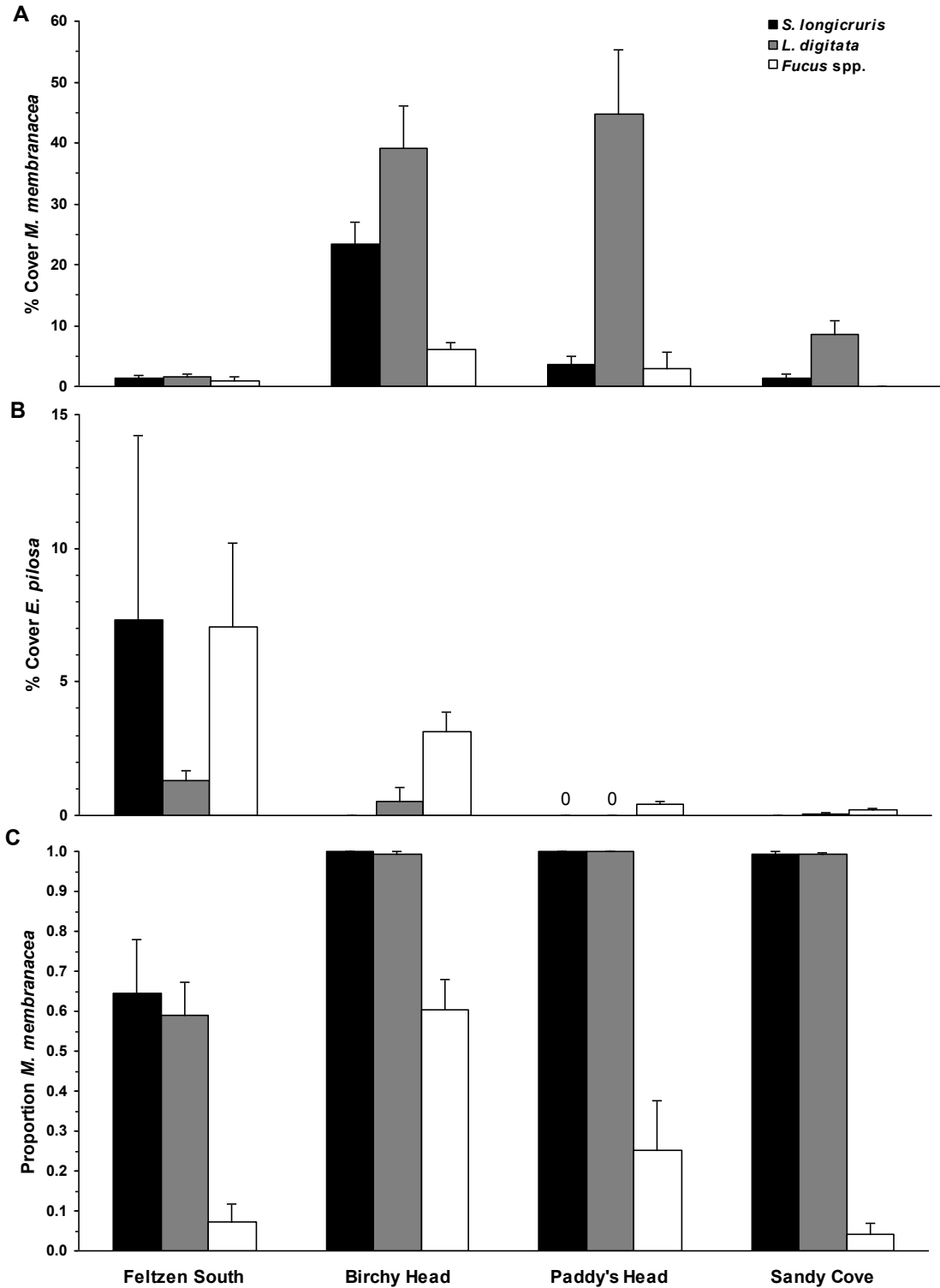
Fig. 3.2 Mean percent cover of (A) *Membranipora membranacea*, and (B) *Electra pilosa*, on algal substrates (+ SE, N = 9 - 10), and (C) mean ratio of *M. membranacea* cover to total bryozoan (*M. membranacea* and *E. pilosa*) cover (+ SE, N = 7 - 10), on two species of kelp and two species of *Fucus,* at each of four sites in Nova Scotia, Canada, sampled in Dec 2008.
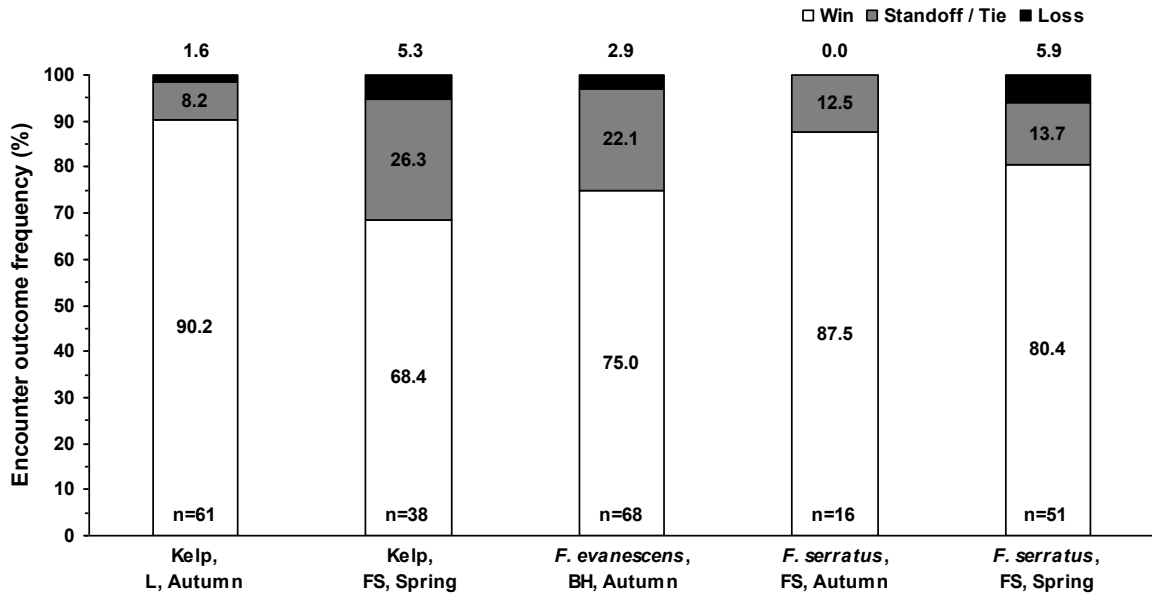
Fig. 3.3 Outcomes of encounters between *Membranipora membranacea* and *Electra pilosa* on different algal substrates (Kelp: *Saccharina longicruris*, *Laminaria digitata*), sampled at 3 different sites in autumn 2008 and spring 2009. L: The Lodge; FS: Feltzen South; BH: Birchy Head. "Win": *M. membranacea* overgrows *E. pilosa*; "Standoff / tie": no overgrowth or both *M. membranacea* and *E. pilosa* overgrow one another; "Loss": *E. pilosa* overgrows *M. membranacea*.

Table 3.2 Chi-square goodness of fit test of the frequency of encounter outcomes across sampling groups showing observed outcomes and outcomes expected if *Membranipora membranacea* nearly always wins (a nominal minimum value of 5 instead of 0 was used for the expected frequencies for "Standoff / tie" and "*Electra pilosa* wins" to meet the assumptions of the test).

	Observed	Expected	$[(\text{Observed-Expected})^2] / \text{Expected}$
M. membranacea wins	187	224	6.112
Standoff / tie	39	5	231.2
E. pilosa wins	8	5	1.800
N	234	234	$\chi^2_0 = 239.1$
			$\chi^2_{0.05} = 5.991$
			$df = 2$
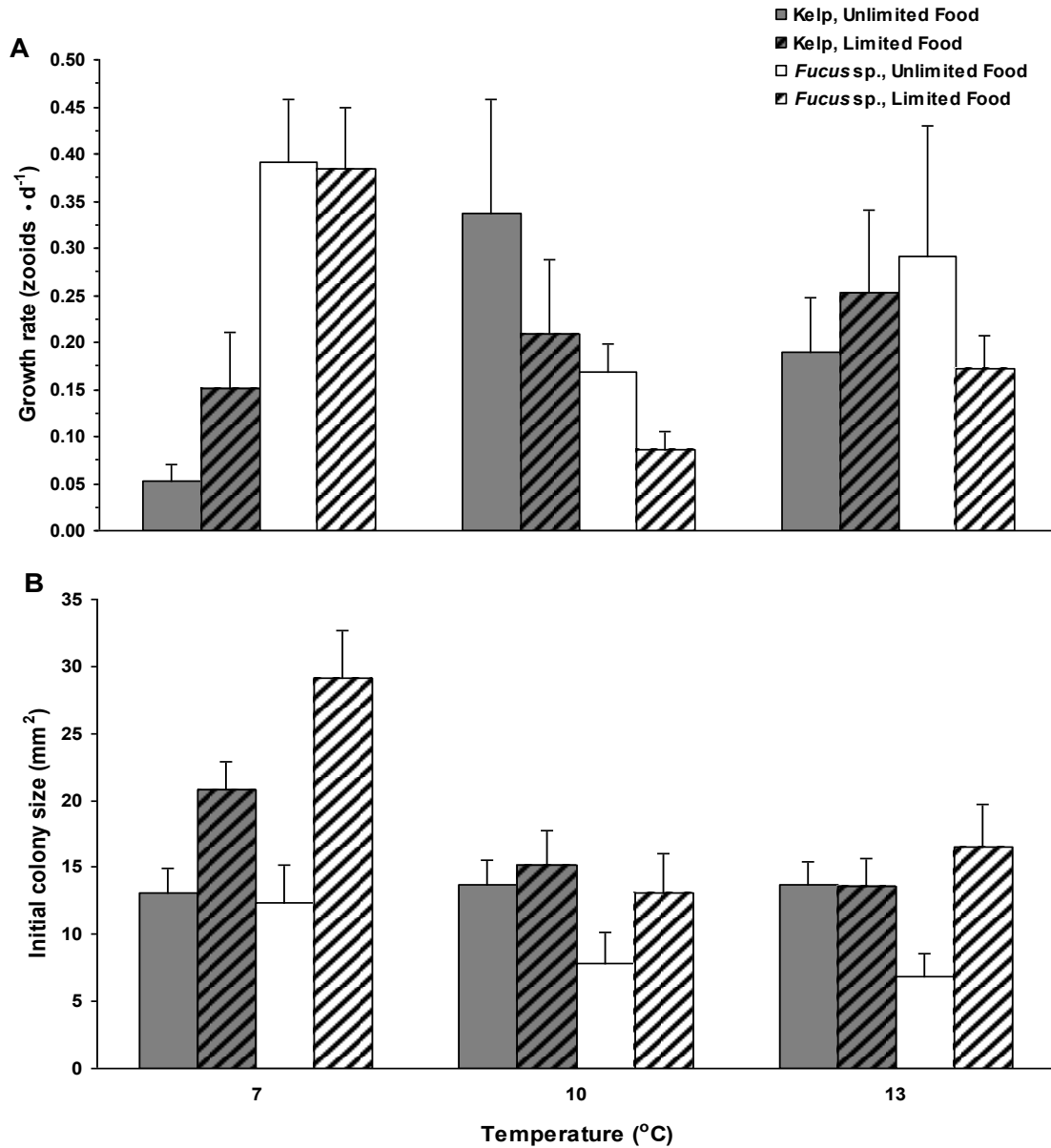
Fig. 3.4 (A) Mean growth rate (+ SE, N = 20) and (B) mean initial size (+ SE, N = 20) of colonies of *Electra pilosa* on kelp (*Saccharina longicruris*, *Laminaria digitata*) and *Fucus* measured in the laboratory at each of 3 temperatures and each of 2 food treatments.

Table 3.3 Results of 3-way ANCOVA examining the effects of substrate, temperature, and food on growth rates of *Electra pilosa* in the laboratory with initial colony size as a covariate. Bold font indicates significant p values. Because of variance heterogeneity, $\alpha = 0.01$ (see Methods).

Factor	MS	df	F	p
Colony size	4.662	1	5.986	0.015
Substrate	26.55	1	34.09	**<0.001**
Temperature	0.328	2	0.421	0.657
Food	1.339	1	1.719	0.191
Substrate x Temperature	3.960	2	5.084	**0.007**
Substrate x Food	1.638	1	2.103	0.148
Temperature x Food	1.855	2	2.382	0.095
Substrate x Temperature x Food	0.561	2	0.720	0.488
Error	0.779	227		
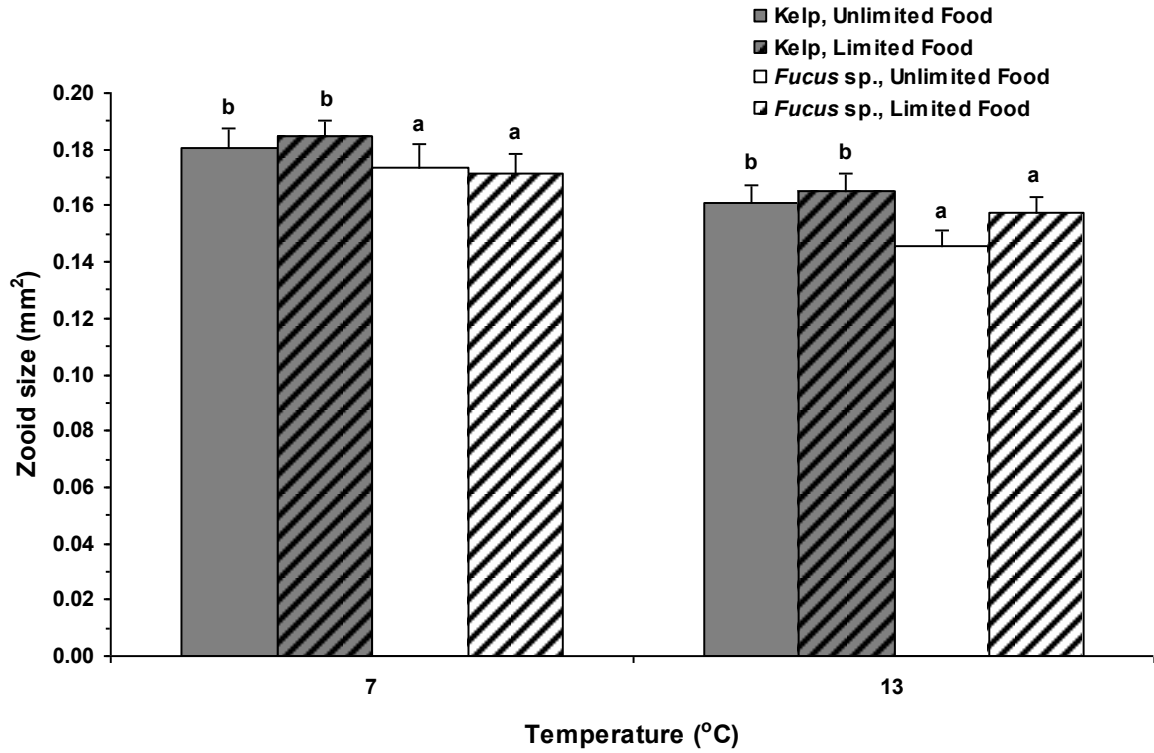
Fig. 3.5 Mean zooid size (+ SE, N = 20; except for Kelp, 7℃ - Unlimited Food: N = 17) of colonies of *Electra pilosa* grown on kelp (*Saccharina longicruris*, *Laminaria digitata*) and *Fucus* in the laboratory at each of 2 temperatures and each of 2 food treatments. Letters above bars indicate homogeneous subsets among treatments within each temperature, identified using Tukey's HSD test, $\alpha = 0.05$.

Table 3.4 Results of 3-way ANOVA examining the effects of substrate, temperature, and food on size of *Electra pilosa* zooids grown in the laboratory. Bold font indicates significant *p* values.

Factor	MS	df	*F*	*p*
Substrate	0.005	1	6.009	**0.015**
Temperature	0.016	1	20.75	**<0.001**
Food	0.001	1	1.000	0.319
Substrate x Temperature	0.000	1	0.027	0.870
Substrate x Food	0.000	1	0.009	0.925
Temperature x Food	0.001	1	0.647	0.423
Substrate x Temperature x Food	0.001	1	0.671	0.414
Error	0.001	149		
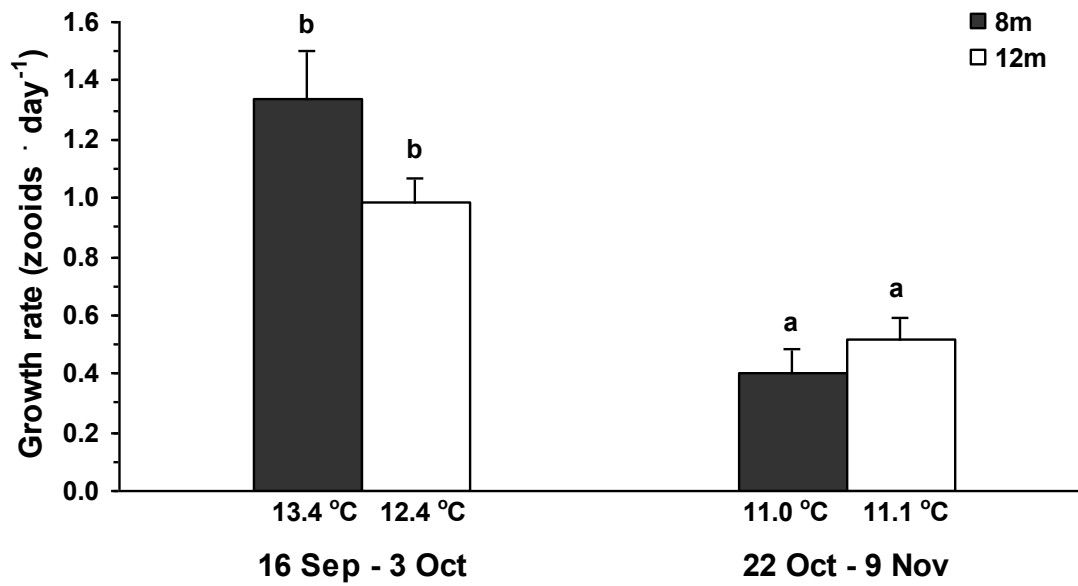
Fig. 3.6 Mean growth rate (+ SE, N = 20) of colonies of *Electra pilosa* measured in the field at The Lodge in St. Margarets Bay, Nova Scotia, Canada, in autumn 2009. Temperatures shown are means for the growth period. Letters above bars indicate homogeneous subsets, identified using Tukey's HSD test, α = 0.05.

Table 3.5 Growth rates of *Electra pilosa* colonies measured on kelp (*Saccharina longicruris* and *Laminaria digitata*) in the laboratory and the field at similar temperatures.

Group	Setting	Mean Temperature (ºC)	Growth Rate (zooids · day^{-1}; mean ± SE)	*N*
~ 11ºC	Laboratory, Unlimited Food	10.4	0.337 ± 0.122	20
	Laboratory, Limited Food	10.2	0.209 ± 0.078	20
	Field, 22 Oct-9 Nov 09 (18 d), 8 m	11.0	0.403 ± 0.079	20
	Field, 22 Oct-9 Nov 09 (18 d), 12 m	11.1	0.519 ± 0.075	20
~ 13ºC	Laboratory, Unlimited Food	13.4	0.190 ± 0.058	20
	Laboratory, Limited Food	13.3	0.252 ± 0.088	20
	Field, 16 Sep-3 Oct 09 (17 d), 8 m	13.4	1.338 ± 0.167	20

3.5 Discussion

3.5.1 Patterns in the Relative Abundance of the Two Bryozoans

We found that *M. membranacea* was generally more abundant than *E. pilosa* on kelps, whereas *E. pilosa* was relatively more abundant on *Fucus* species in most cases. During the sampling period, *M. membranacea* made up nearly the total bryozoan cover on kelps (*S. longicruris* and *L. digitata*) at three of four sites, consistent with previous studies that indicate peaks in abundance of *M. membranacea* from October to December (Saunders and Metaxas 2009b; Scheibling and Gagnon 2009). The population dynamics of *E. pilosa* in this region, and its seasonal patterns of abundance on kelp and *Fucus* substrates, are unknown.

The observed patterns of relative abundance observed are likely affected by several factors, including differential effects of kelp and *Fucus* substrates on settlement, growth and competitive interactions of *M. membranacea* and *E. pilosa*. Settlement patterns may differ between kelp and *Fucus* for either bryozoan because of chemical or physical differences. For example, larvae of *M. membranacea* are reported to exhibit preference for kelp species, such as *S. longicruris* (Seed 1976; Stricker 1989), although the mechanisms behind the observed preference have not been identified. Settlers of *M. membranacea* on *F. serratus* are usually found toward the bases of thalli, and growth is directed proximally, in the opposite direction of frond extension (Ryland and Stebbing 1971), eventually limiting colony size as the bases of thalli are reached. Conversely, most settlers of *E. pilosa* are found toward frond apices, and early growth occurs linearly, parallel to the longitudinal axes of fronds and generally in an apical direction, thus not limiting colony size (Ryland and Stebbing 1971). In contrast, settlement of *M. membranacea* on kelps occurs near the bases of thalli, allowing for colony expansion near the location of basal frond extension and during blade growth (Brumbaugh et al. 1994). These differences suggest that *E. pilosa* may be better adapted for colony expansion on *Fucus* and *M. membranacea* on kelps.

Several characteristics of *Fucus* substrates may affect colony expansion of *M. membranacea* negatively. Saunders and Metaxas (2009a) identified a positive, exponential relationship between growth rate and colony size for *M. membranacea*, and

reported colony sizes up to two orders of magnitude larger on kelps than those typically observed for *E. pilosa* in these habitats. The dynamics of kelp beds in our region provide rapidly growing, large substrates with high seasonal availability of space. However, space is much more limited on *Fucus* species, and divided due to algal branching. Colonies of *M. membranacea*, therefore, cannot grow as large as on kelp and cannot reach maximum growth rates. Additionally, due to the relative stability of *Fucus* substrates, epiphytes can occupy space on thalli over many years, potentially fostering higher native species richness and allowing slow-growing organisms, such as colonies of *E. pilosa*, to reach larger sizes, thereby reducing free space. Both native species richness and resource (space) availability can affect invasibility of subtidal habitats (Stachowicz et al. 1999; Chavanich and Harris 2000), and colonization of *Fucus* thalli by *E. pilosa* and other epiphytes is likely to reduce growth rates of *M. membranacea* colonies, even in cases where overgrowth occurs.

Differences in relative abundance of the two bryozoans were also recorded among sites for each substrate type. Factors such as larval supply, food availability, turbidity, and flow were not quantified across sites, but can affect the abundance of both *M. membranacea* and *E. pilosa* (O'Connor et al. 1979; Saunders and Metaxas 2009a). At Feltzen South, where lower relative abundance of *M. membranacea* occurred on kelps than at other sites, kelp blades are longer, broader, and thinner than at other sites (unpublished data), indicative of kelps growing in low energy environments (Gerard and Mann 1979). Kelp morphology, in combination with heavy silting repeatedly observed on algae, suggest that Feltzen South is considerably more sheltered than the other study sites. This, in turn, may affect the abundance of *M. membranacea* more than that of *E. pilosa*, for which high abundance has been reported for low flow environments (O'Connor et al. 1979). The higher relative abundance of *M. membranacea* on *Fucus* at Birchy Head than at other sites, may be associated with characteristics of the kelp canopy at that site. Kelps were less abundant at Birchy Head than at other sites due to a defoliation event in autumn 2007 (Saunders and Metaxas 2009b; Metaxas, personal observations; unpublished data). Settlement of *M. membranacea* on *Fucus* may have been greater there relative to other sites, due to the openness of the kelp canopy and/or a shortage of space on the sparse kelps, increasing larval supply to *Fucus* substrates.

3.5.2 Competitive Interactions Between the Two Bryozoans

Membranipora membranacea is competitively superior to *E. pilosa*, and competition from *E. pilosa* is not a determining factor in the population dynamics of the introduced bryozoan (Berman et al. 1992; Saunders and Metaxas 2007, 2009a). Because space can be limiting for encrusting species in sessile marine invertebrate communities, interference competition commonly involves overgrowth (Dayton 1971; Buss and Jackson 1979; Konar and Iken 2005). However, we have shown that it is not the case that *M. membranacea* nearly always wins in encounters with *E. pilosa*. Standoffs, in particular, were more frequent than expected. In previous studies, the frequency of standoffs was within the range found here, but overgrowth of *M. membranacea* by *E. pilosa* was much higher (> 20% on kelps) (Berman et al. 1992) than in our study (< 6 % on kelps and *Fucus*).

Where competitive reversals and standoffs are frequent, they facilitate coexistence and contribute significantly to the preservation of diversity (Walters and Wethey 1986). Therefore, whereas the occurrence of standoffs and overgrowth of *M. membranacea* by *E. pilosa* may not be important to the population dynamics of the invasive species in this region, these outcomes are significant for the preservation of populations of the native *E. pilosa*. It is important to note that in cases where *E. pilosa* overgrows *M. membranacea*, it is unlikely that an entire *M. membranacea* colony will be overgrown, whereas overgrowth of *E. pilosa* by *M. membranacea* frequently results in mortality of the native bryozoan colony because of its small size.

Competitive outcomes in various sessile marine invertebrates have been determined by factors such as differential growth (barnacles) (Connell 1961a; Connell 1961b) and size (bryozoans, ascidians, sponges, corals) (Connolly and Muko 2008; Jackson 1979; Russ 1982), encounter angle (bryozoans) (Jackson 1979), thickness (sponges) (Bell and Barnes 2003), age (bryozoans) (Buss 1980), and substrate topography (bryozoans) (Walters and Wethey 1986). It is probable that faster growth rates and larger colony sizes are the main factors contributing to the competitive dominance of *M. membranacea* over *E. pilosa*, and these characteristics likely explain the predominant overgrowth observed in this and other studies (Berman et al. 1992). The

frequent occurrence of standoffs reported here may be related to encounter angle; for example, where directions of colony growth are parallel, overgrowth may occur less frequently. Colony thickness has been shown to contribute to competitive ability in the tropical cheilostome bryozoan *Antropora tincta* (Buss 1980). Thickness is associated with colony age, and may differ between substrates for native *E. pilosa*. We observed large, thick *E. pilosa* colonies on *Fucus* thalli that appeared to have grown over multiple seasons in high density with conspecifics, in contrast to the small colonies of the current season typically observed on the more dynamic kelp substrates. Physical differences between substrates may also contribute to competitive outcomes. Walters and Wethey (1986) showed that competitive reversals between bryozoans (*E. pilosa* and *Alcyonidium* species) can occur due to elevation differences caused by substrate topography. Whereas kelp surfaces are flat, blades of *F. serratus* possess a prominent midrib, and this feature may affect outcomes.

3.5.3 Patterns in Growth Rate

For the native bryozoan *E. pilosa*, growth rates were slow and ranged from 0.00 to 3.05 zooids·day^{-1}. In other studies, growth rates were equal to or much greater than those reported here [approximately 0.75 to 14.85 zooids·day^{-1}, calculated from data reported by Hermansen et al. (2001)]; however, these experiments were performed at higher temperatures (approximately 14 to 15ºC), in some cases on artificial substrates, and generally with larger colonies than those used here, all conditions associated with increased growth rates of cheilostome bryozoans (Hermansen et al. 2001; Menon 1972; Saunders and Metaxas 2009a). Most notably, we found that *E. pilosa* grows much more slowly than *M. membranacea*. Conversions of growth rates reported here, based on mean zooid length of 0.55 ± 0.01 mm (mean ± SE, N = 24), indicate that *E. pilosa* colonies grew from approximately 0.00 to 1.68 mm·day^{-1}, whereas growth of *M. membranacea* has been recorded from 0.01 to 12 mm·day^{-1} (Saunders and Metaxas 2009a).

We detected a positive association between growth rate and temperature for *E. pilosa* colonies growing in the field, but a positive relationship between these factors in the laboratory was not pronounced, possibly because of the range of temperatures tested. *Electra pilosa* has a wide distribution, occurring in the Arctic, North Atlantic and Indo-

Pacific oceans and the Mediterranean Sea (Nikulina et al. 2007). Because it occurs over a wide range of temperatures, and has a high capacity for thermal acclimation (Menon 1972), the range of 7 to 13 ºC may have been too narrow to detect strong effects of temperature in the laboratory, particularly since laboratory growth rates were slow compared with those measured in the field. A distinct positive effect of temperature on growth rate has been found for *E. pilosa* over a wider range of temperatures than those tested here (6 to 22 ºC; Menon 1972), and a similar relationship has been reported for other bryozoan species (Menon 1972; O'Dea and Okamura 1999; Saunders and Metaxas 2009a). Saunders and Metaxas (2009a) also found a significant effect of temperature on growth of newly settled and mature *M. membranacea* colonies both in the laboratory and the field, suggesting a stronger effect on *M. membranacea* than for *E. pilosa* within this temperature range. Colony size explained more variation in growth rate of *M. membranacea* colonies than temperature (Saunders and Metaxas 2009a), whereas, in our study, initial colony size had a marginally non-significant positive effect on growth rate of *E. pilosa*. The colonies of *M. membranacea* tested, however, covered a wider range of colony sizes than those used here, including newly settled colonies to established colonies up to 192 mm in diameter. Colony size of *E. pilosa* may have a stronger effect on growth rate at sizes beyond those included in this study that, however, only occur in habitats outside our study area.

Growth rate of *E. pilosa* was not affected significantly by food concentration. Studies of other cheilostome bryozoans, including *M. membranacea*, have shown that at low temperatures, food availability does not affect growth rate, and this is likely due to low metabolic rate (O'Dea and Okamura 1999; Saunders and Metaxas 2009a).

The faster growth of *E. pilosa* on *Fucus* than on kelps has not been recorded before and may be the result of physical differences between the surfaces of these substrates. Macroalgae produce extracellular organic matter, and as rates of exudation are greater for kelps than for *Fucus* species, the quantity of mucus on the surfaces of these substrates may differ (Sieburth 1969). Although the effect of mucus on bryozoan growth is not known, growth rate has been found to be higher on artificial substrates (that are free of mucus) than on kelp blades (Hermansen et al. 2001; Manriquez and Cancino 1996).

We found that growth rates of *E. pilosa* measured in the field were significantly higher than laboratory growth rates in similar conditions (i.e. equivalent temperatures), and this difference has been recorded for other bryozoans including *M. membranacea* (Hermansen et al. 2001; Okamura 1992; Saunders and Metaxas 2009a). Differences were not large, however, and the range of field growth rates in this study overlapped those measured in the laboratory. Laboratory growth may be negatively affected by factors such as disturbance to colonies during transport, inferior algal substrate quality, and reduced flow. The methods of growth measurement used here differed between laboratory and field conditions and may have further contributed to the variation observed.

The inverse relationship we observed between temperature and zooid size in *E. pilosa* has been previously observed in this species (Menon 1972), as well as for *Conopeum seurati* (O'Dea and Okamura 1999) and *C. reticulum* (Menon 1972). The development of larger zooids at lower temperatures may be related to reduced metabolic rate and growth (Kinne 1963; Menon 1972). Weak effects of salinity on zooid size have also been recorded, but no effects of food, mean growth rate, reproductive state, neighboring colonies, or flow (O'Dea and Okamura 1999; Okamura 1992). The effect of substrate on zooid size had not been previously tested for bryozoans, and we found that zooids of *E. pilosa* colonies are larger on kelp than on *Fucus* substrates across temperature and food treatments. The association between reduced metabolic and growth rates and larger zooid size, proposed for temperature-related patterns, may also apply to substrate-related patterns, since growth was slower on kelps than on *Fucus*. It is important to note, however, that our measurements of growth rates were based on production of new zooids. These measures likely underestimate expansion of colony area at lower temperatures and on kelp substrates because zooid area was greater under such conditions. Therefore, differences detected here between substrates at lower temperatures, for example, are partially reflective of differences in colony morphology rather than differences in rates of colony expansion.

In summary, we have shown that where space is not limited (i.e. on kelps), large colony size, relatively fast growth, and strong overgrowth abilities likely interact to ensure success of the invasive bryozoan, *M. membranacea*, and competitive dominance

over colonies of the native bryozoan, *E. pilosa*. Factors that can limit the success of *M. membranacea* on alternative non-kelp substrates include reduced space availability, and restriction of colony size and growth rate. Mechanisms of coexistence between *M. membranacea* and *E. pilosa* include competitive standoffs that can preserve native diversity. This study underscores the importance of incorporation of alternative contexts into invasion research, which can reveal factors involved in the resilience of native communities.

3.6 Acknowledgements

Chapter 4

Discussion

This thesis describes research on interactions between the invasive epiphytic bryozoan *Membranipora membranacea* and species not previously considered important to its dynamics in its introduced range. It has revealed mechanisms that regulate success and expansion of this species, as well as its persistence, and has identified factors that may limit its dominance in rocky subtidal habitats. An understanding of factors affecting the population dynamics of this invasive bryozoan is particularly important as this species has a dramatic effect on the dynamics of native kelp beds in Nova Scotia.

Several factors can contribute to the success of *M. membranacea* in native kelp beds. Growth rates of *M. membranacea* exceed those measured for the native bryozoan, *Electra pilosa*, by nearly an order of magnitude. The invasive bryozoan also shows evidence of competitive superiority as it overgrows *E. pilosa* in approximately 70 - 90 % of encounters. *Membranipora membranacea* exhibits generalist attributes in its use of kelp and non-kelp substrates, and this enables its expansion into habitats where kelps are absent. I have recorded the extent of its occurrence on *Fucus evanescens* and *F. serratus*; it may also occur on other non-kelp substrates, further enabling the alongshore spread of *M. membranacea* to kelp-dominated regions, through non-kelp habitat.

Membranipora membranacea has persisted as a dominant epiphyte in rocky subtidal habitats of the northwest Atlantic despite almost complete removal at times of its preferred substrate, kelp, through defoliation. Larvae occur in abundance in late summer and autumn in Nova Scotia, despite removal of colonies from kelp blades during winter. The occurrence of *M. membranacea* on *Fucus* substrates identifies an important refuge with reduced susceptibility to erosion and defoliation in fall and winter storms. Settlement and growth of *M. membranacea* on non-kelp substrates also ensures retention of the invasive bryozoan in years when primary substrates (kelps) are absent or sparse due to severe defoliation. Removal of kelps does not constrain usable substrates for *M. membranacea* in the following season; rather, open kelp canopies enable settlement on *Fucus* as well as kelp substrates.

In addition to identifying mechanisms that regulate the dominance of this invasive bryozoan, this research has also revealed limitations to the success of *M. membranacea* in

subtidal communities in our region. I suggest that reduced space availability, and restriction of colony size and growth rate, likely limit the success of *M. membranacea* on non-kelp substrates. While *M. membranacea* is the dominant space holder on kelps, its relative abundance to *E. pilosa* is lower on *Fucus* spp. The native bryozoan can also limit resource use by *M. membranacea* through standoffs which occur in approximately 8 - 26 % of encounters, ensuring coexistence of the invasive and native epiphytes.

 This research has identified the potentially significant role of a substrate used by *Membranipora membranacea* not studied to date. It has clarified the role of non-kelp in the population dynamics of this invasive bryozoan in Nova Scotia. This study has also increased our understanding of the ecology of a native bryozoan that interacts with *M. membranacea*, identifying factors that influence their relative abundance. Introduced species can occupy various locations within non-native communities. *Membranipora membranacea* provides a case study on the importance of investigating invasive species in various contexts, including those where their impacts are not obvious, to ensure a more complete understanding of patterns and processes involved in species invasions.

References

Audet D, Davis DS, Miron G, Moriyasu M, Benhalima K, Campbell R (2003) Geographical expansion of a nonindigenous crab, *Carcinus maenas* (L.), along the Nova Scotian shore into the Southeastern Gulf of St. Lawrence, Canada. J Shellfish Res 22: 255-262

Bayer MM, Cormack RM, Todd CD (1994) Influence of food concentration on polypide regression in the marine bryozoan *Electra pilosa* (L.) (Bryozoa: Cheilostomata). J Exp Mar Biol Ecol 178: 35-50

Bell JJ, Barnes DKA (2003) The importance of competitor identity, morphology and ranking methodology to outcomes in interference competition between sponges. Mar Biol 143: 415-426

Berman J, Harris L, Lambert W, Buttrick M, Dufresne M (1992) Recent invasions of the Gulf of Maine: three contrasting ecological histories. Conserv Biol 6: 435-441

Best MA, Thorpe JP (1986) Effects of food particle concentration on feeding current velocity in six species of marine bryozoa. Mar Biol 93: 255-262

Bird CJ, Dadswell MJ, Grund DW (1993) First record of the potential nuisance alga *Codium fragile* ssp. *tomentosoides* (Chlorophyta, Caulerpales) in Atlantic Canada. Proceedings of the Nova Scotia Institute of Science 40: 11-17

Brumbaugh DR, West JM, Hintz JL, Anderson FE (1994) Determinants of recruitment by an epiphytic marine bryozoan: field manipulations of flow and host quality. In: WH Wilson Jr, SA Sticker, GL Shinn (eds) Reproduction and development of marine invertebrates. John Hopkins University Press, Baltimore, pp 287-301

Buss LW (1980) Competitive intransitivity and size-frequency distributions of interacting populations. Proc Natl Acad Sci USA 77: 5355-5359

Buss LW, Jackson JBC (1979) Competitive networks: nontransitive competitive relationships in cryptic coral reef environments. Am Nat 113: 223-234

Chavanich S, Harris LG (2000) Potential impact of the introduced bryozoan, *Membranipora membranacea*, on the subtidal snail, *Lacuna vincta*, in the Gulf of Maine. In: J Pederson (ed) Marine bioinvasions: Proceedings of the first national conference. MIT Sea Grant College Program, Cambridge, pp 157-163

Colautti RI, MacIsaac HJ (2004) A neutral terminology to define 'invasive' species. Divers Distrib 10: 135-141

Connell JH (1961a) Effects of competition, predation by *Thais lapillus*, and other factors on natural populations of the barnacle *Balanus balanoides*. Ecol Monogr 31: 61-104

Connell JH (1961b) The influence of interspecific competition and other factors on the distribution of the barnacle *Chthamalus stellatus*. Ecology 42: 710-723

Connolly SR, Muko S (2008) Space preemption, size-dependent competition, and the coexistence of clonal growth forms. Eco Soc America 84: 2979-2988

Coyer JA, Peters AF, Stam WT, Olsen JL (2003) Post-ice age recolonization and differentiation of *Fucus serratus* L. (Phaeophyceae; Fucaceae) populations in Northern Europe. Mol Ecol 12: 1817-1829

Dayton PK (1971) Competition, disturbance, and community organization: the provision and subsequent utilization of space in a rocky intertidal community. Ecol Monogr 41: 351-389

Dixon J, Schroeter SC, Kastendiek J (1981) Effects of the encrusting bryozoan, *Membranipora membranacea*, on the loss of blades and fronds by the giant kelp, *Macrocystis pyrifera* (Laminariales). J Phycol 17: 341-345

Duggins DO, Eckman JE, Sewell AT (1990) Ecology of understory kelp environments. II. Effects of kelps on recruitment of benthic invertebrates. J Exp Mar Biol Ecol 143: 27-45

Edelstein T, Greenwell M, Bird CJ, McLachlan J (1971) Investigations of the marine algae of Nova Scotia. X. Distribution of *Fucus serratus* L. & some other species of *Fucus* in the Maritime Provinces. Proc NS Inst Sci 27: 33-42

Elton, CS (1958) The ecology of invasions by plants and animals. Methuen, London

Fridley JD, Stachowicz JJ, Naeem S, Sax DF, Seabloom EW, Smith MD, Stohlgren TJ, Tilman D, Von Holle B (2007) The invasion paradox: reconciling pattern and process in species invasions. Ecology 88: 3-17

Gerard VA, Mann KH (1979) Growth and production of *Laminaria longicruris* (Phaeophyta) populations exposed to different intensities of water movement. J Phycol 15: 33-41

Harding, S (1992) After the neutrality ideal: science, politics, and "strong objectivity". Soc Res 59: 567-587

Harding, S (1998) Women, science, and society. Science 281: 1599-1600

Harris LG, Tyrrell MC (2001) Changing community states in the Gulf of Maine: synergism between invaders, overfishing and climate change. Biol Invasions 3: 9-21

Harvell CD (1992) Inducible defenses and allocation shifts in a marine bryozoan. Ecology 73: 1567-1576

Harvell CD, Grosberg RK (1988) The timing of sexual maturity in clonal animals. Ecology 69: 1855-1864

Hermansen P, Larsen PS, Riisgård HU (2001) Colony growth rate of encrusting marine bryozoans (*Electra pilosa* and *Celleporella hyalina*). J Exp Mar Biol Ecol 263: 1-23

Hierro JL, Maron JL, Callaway RM (2005) A biogeographical approach to plant invasions: the importance of studying exotics in their introduced and native range. J Ecol 93: 5-15

Jackson JBC (1979) Overgrowth competition between encrusting cheilostome ectoprocts in a Jamaican cryptic reef environment. J Anim Ecol 48: 805-823

Johnson CR, Mann KH (1988) Diversity, patterns of adaptation, and stability of Nova Scotian kelp beds. Ecol Monogr 58: 129-154

Kinne O (1963) The effects of temperature and salinity on marine and brackish water animals. I. Temperature. Oceanogr Mar Biol A Rev 1: 301-340

Konar B, Iken K (2005) Competitive dominance among sessile marine organisms in a high arctic boulder community. Polar Biol 29: 61-64

Krumhansl KA, Scheibling RE (in press) Detrital production in Nova Scotian kelp beds: patterns and processes. Mar Ecol Prog Ser

Lambert W, Levin PS, Berman J (1992) Changes in the structure of a New England (USA) kelp bed: the effects of an introduced species? Mar Ecol Prog Ser 88: 303-307

Larson BMH (2008) The war of the roses: demilitarizing invasion biology. Eco Soc America 3: 495-500

Larson BMH (2007) Who's invading what? Systems thinking about invasive species. Can J Plant Sci 87: 993-999

Levin PS, Coyer JA, Petrik R, Good TP (2002) Community-wide effects of nonindigenous species on temperate rocky reefs. Ecology 83: 3182-3193

Mack RN, Simberloff D, Lonsdale WM, Evans H, Clout M, Bazzaz FA (2008) Biotic invasions: causes, epidemiology, global consequences, and control. Ecol Appl 10: 689-710

Mann KH (1972) Ecological energetics of the seaweed zone in a marine bay on the Atlantic coast of Canada. II. Productivity of the seaweeds. Mar Biol 14: 199-209

Mann KH (1973) Seaweeds: their productivity and strategy for growth. Science 182: 975-981

Manriquez PH, Cancino JM (1996) Bryozoan-macroalgal interactions: do epibionts benefit? Mar Ecol Prog Ser 138: 189-197

McCook LJ, Chapman ARO (1991) Community succession following massive ice-scour on an exposed rocky shore: effects of *Fucus* canopy algae and of mussels during late succession. J Exp Mar Biol Ecol 154: 137-169

McKinney FK, McKinney MJ (1993) Larval behaviour and choice of settlement site: correlation with environmental distribution pattern in an erect bryozoan. Facies 29: 119-131

Menon NR (1972) Heat tolerance, growth and regeneration in three North Sea bryozoans exposed to different constant temperatures. Mar Biol 15: 1-11

Menon NR (1974) Clearance rates of food suspension and food passage rates as a function of temperature in two North Sea bryozoans. Mar Biol 24: 65-67

Metaxas A, Scheibling RE (1996) Spatial heterogeneity of phytoplankton assemblages in tidepools: effects of abiotic and biotic factors. Mar Ecol Prog Ser 130: 179-199

Nikulina EA, Hanel R, Schäfer P (2007) Cryptic speciation and paraphyly in the cosmopolitan bryozoan *Electra pilosa* - impact of the tethys closing on species evolution. Mol Phylogenet Evol 45: 765-776

O'Connor RJ, Seed R, Boaden PJS (1979) Effects of environment and plant characteristics on the distribution of bryozoa in a *Fucus serratus* L. community. J Exp Mar Biol Ecol 38: 151-178

O'Connor RJ, Seed R, Boaden PJS (1980) Resource space partitioning by the bryozoa of a *Fucus serratus* L. community. J Exp Mar Biol Ecol 45: 117-137

O'Dea A, Okamura B (1999) Influence of seasonal variation in temperature, salinity and food availability on module size and colony growth of the estuarine bryozoan *Conopeum seurati*. Mar Biol 135: 581-588

Okamura B (1988) The influence of neighbors on the feeding of an epifaunal bryozoan. J Exp Mar Biol Ecol 120: 105-123

Okamura B (1992) Microhabitat variation and patterns of colony growth and feeding in a marine bryozoan. Ecology 73: 1502-1513

Pineda J (2000) Linking larval settlement to larval transport: assumptions, potentials, and pitfalls. Oceanogr E Pac 1: 61-81

Quinn JF (1982) Competitive hierarchies in marine benthic communities. Oecologia 54: 129-135

Ricciardi A, Rasmussen JB (1998) Predicting the identity and impact of future biological invaders: a priority for aquatic resource management. Can J Fish Aquat Sci 55: 1759-1765

Russ GR (1982) Overgrowth in a marine epifaunal community: competitive hierarchies and competitive networks. Oecologia 53: 12-19

Ryland JS (1959) Experiments on the selection of algal subsrates by polyzoan larvae. J Exp Biol 36: 613-631

Ryland JS (1962) The association between polyzoa and algal substrata. J Anim Ecol 31: 331-338

Ryland JS, Stebbing ARD (1971) Settlement and orientated growth in epiphytic and epizoic bryozoans. In: DJ Crisp (ed) Fourth European marine biology symposium. Cambridge University Press, Cambridge, pp 105-123

Saier B, Chapman AS (2004) Crusts of the alien bryozoan *Membranipora membranacea* can negatively impact spore output from native kelps (*Laminaria longicruris*). Bot Mar 47: 265-271

Sakai AK, Allendorf FW, Holt JS, Lodge DM, Molofsky J, With KA, Baughman S, Cabin RJ, Cohen JE, Ellstrand NC, et al. (2001) The population biology of invasive species. Annu Rev Ecol Syst 32: 305-332

Saunders M, Metaxas A (2007) Temperature explains settlement patterns of the introduced bryozoan *Membranipora membranacea* in Nova Scotia, Canada. Mar Ecol Prog Ser 344: 95-106

Saunders M, Metaxas A (2008) High recruitment of the introduced bryozoan *Membranipora membranacea* is associated with kelp bed defoliation in Nova Scotia, Canada. Mar Ecol Prog Ser 369: 139-151

Saunders MI, Metaxas A (2009a) Effects of temperature, size, and food on the growth of *Membranipora membranacea* in laboratory and field studies. Mar Biol 156: 2267-2276

Saunders MI, Metaxas A (2009b) Population dynamics of a nonindigenous epiphytic bryozoan *Membranipora membranacea* in the western North Atlantic: effects of kelp substrate. Aquat Biol 8: 83-94

Saunders MI, Metaxas A, Filgueira R (2010) Implications of warming temperatures for population outbreaks of a nonindigenous species (*Membranipora membranacea*, Bryozoa) in rocky subtidal ecosystems. Limnol Oceanogr 55: 1627-1642

Scheibling RE (1984) Echinoids, epizootics and ecological stability in the rocky subtidal off Nova Scotia, Canada. Helgol Mar Res 37: 233-242

Scheibling RE, Gagnon P (2006) Competitive interactions between the invasive green alga *Codium fragile* ssp. *tomentosoides* and native canopy-forming seaweeds in Nova Scotia (Canada). Mar Ecol Prog Ser 325: 1-14

Scheibling RE, Gagnon P (2009) Temperature-mediated outbreak dynamics of the invasive bryozoan *Membranipora membranacea* in Nova Scotian kelp beds. Mar Ecol Prog Ser 390: 1-13

Scheibling RE, Hennigar AW, Balch T (1999) Destructive grazing, epiphytism, and disease: the dynamics of sea urchin- kelp interactions in Nova Scotia. Can J Fish Aquat Sci 56: 2300-2314

Schiebinger L (1997) Creating sustainable science. Osiris 12: 201-216

Schwaninger HR (1999) Population structure of the widely dispersing marine bryozoan *Membranipora membranacea* (Cheilostomata): implications for population history, biogeography, and taxonomy. Mar Biol 135: 411-423

Seed R (1976) Observations on the ecology of *Membranipora* (Bryozoa) and a major predator *Doridella steinbergae* (Nudibranchiata) along the fronds of *Laminaria saccharina* at Friday Harbor, Washington. J Exp Mar Biol Ecol 24: 1–17

Sieburth JM (1969) Studies on algal substances in the sea. III. The production of extracellular organic matter by littoral marine algae. J Exp Mar Biol Ecol 3: 290-309

Simons AM (2003) Invasive aliens and sampling bias. Ecol Lett 6: 278-280

Stachowicz JJ, Byrnes JE (2006) Species diversity, invasion success, and ecosystem functioning: disentangling the influence of resource competition, facilitation, and extrinsic factors. Mar Ecol Prog Ser 311: 251-262

Stachowicz JJ, Whitlatch RB, Osman RW (1999) Species diversity and invasion resistance in a marine ecosystem. Science 286: 1577-1579

Stebbing ARD (1973) Competition for space between the epiphytes of *Fucus serratus* L. J Mar Biol Assoc UK 53: 247-261

Stricker SA (1989) Settlement and metamorphosis of the marine bryozoan *Membranipora membranacea*. Bull Mar Sci 45: 387-405

Walters LJ, Wethey DS (1986) Surface topography influences competitive hierarchies on marine hard substrata: a field experiment. Biol Bull 170: 441-449

Watanabe S, Scheibling RE, Metaxas A (2009) Contrasting patterns of spread in interacting invasive species: *Membranipora membranacea* and *Codium fragile* off Nova Scotia. Biol Invasions 12: 2329-2342

Wright JT, Davis AR (2006) Demographic feedback between clonal growth and fragmentation in an invasive seaweed. Ecology 87: 1744-1754

Yorke AF, Metaxas A (submitted) Interactions between an invasive and a native bryozoan (*Membranipora membranacea* and *Electra pilosa*) species on kelp and *Fucus* substrates in Nova Scotia. Mar Biol

Zar JH. (1999) Biostatistical analysis. Prentice-Hall, NJ