Distribution and abundance of golden star tunicate (*Botryllus schlosseri*) and *Botrylloides sp.* on artificial and natural substrates at twelve sites in western Newfoundland, Canada.

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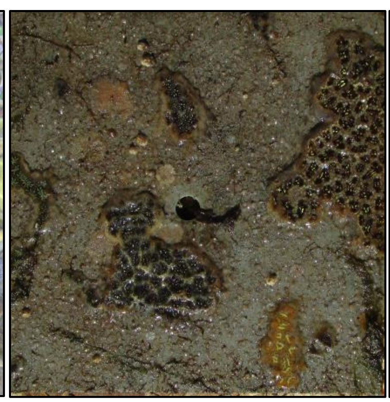
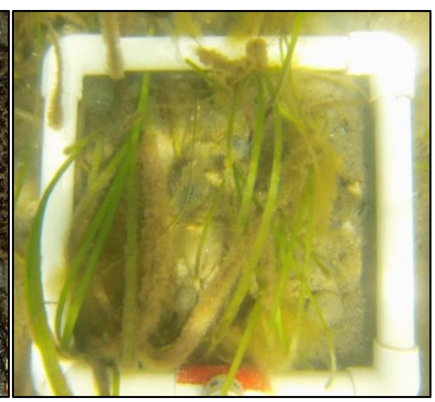

SUMMARY

The distribution and abundance of invasive colonial tunicates were documented at twelve sites along a ~270-km latitudinal gradient in western Newfoundland (NL), Canada. A combination of 10x10 and 15x15 cm polyvinyl chloride (PVC) panels were deployed on fixed and/or floating docks at each site between June and November 2014. The presence and abundance of invasive colonial tunicates were documented in September and November. Similar to patterns observed in 2013 (Caines personal observation), *Botryllus schlosseri* was found at Sites 1, 2 and 5, while *Botrylloides* sp. was observed at Site 2. Monthly sampling of 15x15 cm PVC panels, eelgrass (*Zostera marina*), rockweed (*Fucus* sp.), and kelp (*Saccharina latissima*) was conducted between September and November at Sites 1, 2 and 5 to determine spatial and temporal variation in the abundance of invasive colonial tunicates on artificial and natural substrates. Mean colony cover of *B. schlosseri* was 14.1 and 19.5% for panels sampled from floating docks in September for Sites 2 and 5, respectively, while mean cover of *Botrylloides* sp. was 3.7% on panels sampled from the fixed dock at Site 2. Interestingly, the frequency of occurrence for *B. schlosseri* colonies on rockweed increased from 20% in September to 100% in October at Site 2, while it decreased from 100% in September to 80% in October at Site 5. The frequency of *Botrylloides* sp. peaked at 50% and 70% on rockweed and kelp, respectively, in October at Site 2. The frequency of *B. schlosseri* on kelp specimens at Site 1 peaked at 40% in October, while kelp specimens from Site 2 had a peak colony frequency of 100% in September. Overall, the abundance of *B. schlosseri* on artificial and natural substrates was substantially lower at Site 1, which may be related to cooler sea temperature and increased wave exposure at this site.

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INTRODUCTION

The spread of non-indigenous species threatens marine bio-diversity and can lead to large scale changes in the structure and function of marine environments (Berman et al. 1992, Lambert et al. 1992, Harris & Tyrrell 2001, Levin et al. 2002, Bax et al. 2003, Molnar et al. 2008, McQuaid & Arenas 2009). The golden star tunicate (*Botryllus schlosseri*; herein golden star) was first observed in Arnolds Cove, Placentia Bay along Newfoundland's (NL) east coast in 1945, but remained undetected throughout much of insular NL for over 30 years (Fisheries and Oceans Canada 2013a). Since 2006, golden star has established widespread populations in Placentia Bay and more recently has been observed in isolated harbours in Hermitage, Fortune, and Conception Bays (Callahan et al 2010, Ma 2012, Fisheries and Oceans Canada 2013a). In 2007, a population of violet tunicate (*Botrylloides violaceus*) was found in Belleoram, Fortune Bay, but has not been confirmed elsewhere in NL (Fisheries and Oceans Canada 2013b). Intertidal and subtidal surveys in 2013 indicate that golden star and *Botrylloides* sp. (speculated to be violet tunicate) have expanded their range to the southwest coast of insular NL (Caines personal observations 2013). These marine invaders have been found to cause economic losses for the shellfish aquaculture industry and pose a high ecological risk to coastal ecosystems of Atlantic Canada (Carver et al. 2006).

Sessile marine invasive species have been shown to preferentially colonize artificial over natural substrates (Glasby et al. 2007, Tyrrell & Byers 2007, Dumont et al. 2011). Invasive colonial tunicates are commonly abundant on artificial structures or in anthropogenically disturbed areas. Artificial structures and disturbed areas provide a unique habitat where invasive organisms can establish source populations from which they inevitably invade natural substrates (Dumont et al 2011). The migration of these invaders from artificial to natural habitats can alter subtidal community structure and have significant ecological and economic impacts (Carver et al. 2006). Globally, eelgrass habitat is declining in response to pollution, physical disturbances, eutrophication, and introduced species (Hauxwell et al. 2001, Waycott et al 2009, Garbary et al. 2014). Recently, colonial and solitary invasive tunicates have been found to colonize eelgrass shoots reducing light transmittance, growth, survival and productivity (Wong & Vercaemer 2009). The fouling of eelgrass by invasive tunicates and the dislodgement of eelgrass shoots by the invasive green crab (*Carcinus maenas*) may result in large scale losses in this ecologically significant habitat in Atlantic Canada (Wong & Vercaemer 2009, Garbary et al. 2014).

Sea temperature strongly influences the population dynamics and biological interactions of invasive species in Atlantic Canada (Saunders & Metaxas 2007, 2008, Caines & Gagnon 2012, Matheson & Gagnon 2012). Furthermore, experiments and field observations have shown that climactic changes in ocean temperature will favour invasive species by increasing the magnitude of recruitment and likely competitive success (Stachowicz et al 2002, Rahel & Olden 2008, Saunders &Metaxas 2008, Sorte et al 2010). The recent introduction of botryllid tunicates to NL's southwest coast provides an opportunity to investigate the influence of sea temperature on the abundance, cover, and habitat utilization of these invaders. Newfoundland's west coast spans a ~450-km north-south axis. Sea temperature along this coast varies by up to 5^{o}C and has been shown to strongly influence the abundance and cover of invasive fouling organisms (Caines & Gagnon 2012). Understanding the relationship between sea temperature and abundance and cover of colonial tunicates on artificial and natural substrates in their newly invaded range will allow us to better elucidate the risk that these species pose to the relatively pristine environment along Newfoundland's west coast. This research aims to: 1) confirm the presence of golden star and violet tunicate in southwestern NL, 2) investigate spatial and temporal variability in the abundance and cover of colonies on polyvinyl chloride panels (PVC) at twelve sites along an ~270-km latitudinal gradient, and 3) determine the frequency of occurrence of colonies on eelgrass (*Zostera marina*), rockweed (*Fucus* spp.), and/or kelp (*Saccharina latissima*) at three sites along a ~120-km latitudinal gradient.

MATERIALS AND METHODS

Study sites

This study was conducted at twelve sites within western NL: 1) Port aux Basques, 2) Codroy, 3) St. David's, 4) St. George's, 5) Little Port Harmon, 6) Fox Island River, 7) Piccadilly, 8) Frenchman's Cove, 9) Lark Harbour, 10) Summerside, 11) Rocky Harbour, and 12) St. Paul's (Fig. 1) . Sites 1, 2, 3, 5, 6, 10, and 12 were located in protected harbours open to a bay or the Gulf of St. Lawrence from a small to moderately wide channel, while Sites 4, 7, 8, 9, and 11were

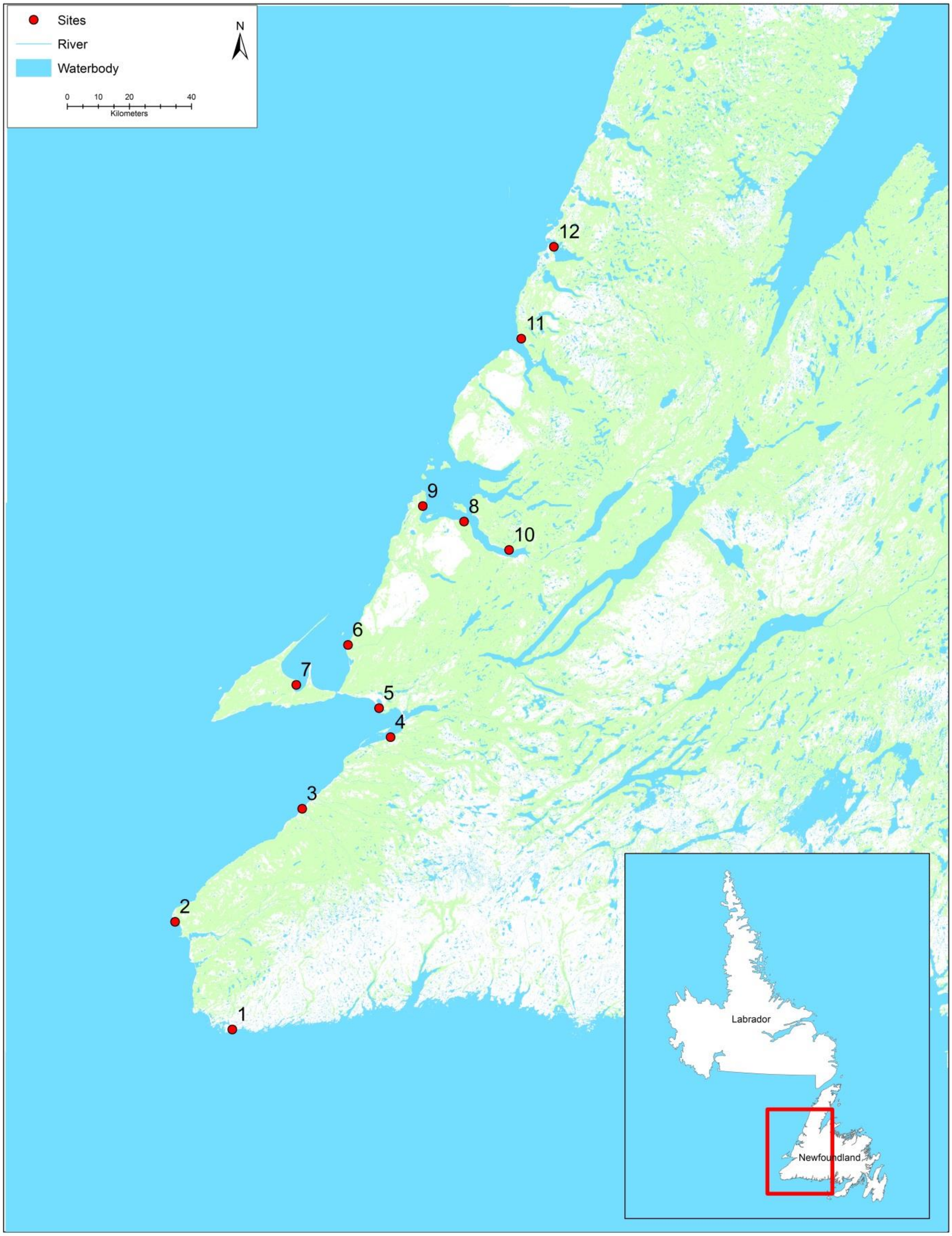

Figure 1: Location of the twelve study sites along the west coast of Newfoundland, Canada.

located in larger harbours that were open to a bay or the Gulf of St. Lawrence. All sites were selected based on the presence of either a fixed or floating dock.

Artificial substratum and physical data

To determine the distribution of invasive colonial tunicates in western NL, one to three 15x15 and/or 10x10 cm PVC towers were deployed from fixed docks at each of the twelve sites along the west coast of NL (Appendix A). Three towers were deployed at Sites 9 and 11, as there were two distinct docks within each community. A single tower was deployed at Sites 7 and 8, as these sites were added after the original project design. The towers consisted of a single 4 to 6 m strand of polypropylene rope that had one end attached to a 0.9 to 1.3 kg weight. The first PVC panel was placed 20 cm above the weight by passing the rope through a 1.0 cm hole drilled in the center of the panel. The remaining two panels were placed above the previous panel and spaced by 10 cm. Cable ties were passed through the braided rope below each panel to suspend at a specified height on the tower. The trailing end of the tower was tied to the fixed dock and was suspended 1.0 to 1.5 m below low tide. The towers at Sites 3, 9 and 10 were attached to floating docks, with a single tower at Site 10 being attached to a fixed dock. Towers were deployed at each site in late June or early July and were checked in September and November 2014. The tower at Site 7 was deployed in September and checked in November.

Invasive colonial tunicates were observed at Sites 1, 2, and 5 in the fall of 2013 (Caines personal observations). Additional PVC towers were deployed at these sites to better elucidate spatial and temporal variation in the population dynamics of these invaders in NL. Increasing the number of PVC panels facilities a more rigorous investigation of spatial variability between sites. In June, two towers with four 15x15 cm PVC panels were deployed from floating docks at each site, with an additional two towers with four 15x15 cm PVC panels deployed from floating docks in September. Two towers, one with three 15x15 cm panels and a second with three 10x10 cm PVC panels, were deployed from fixed docks at each site in June. Panels were sampled in September, October and November. These towers were constructed as previously described, with towers attached to floating docks deployed at 1.0 m below the water level and towers attached to fixed docks deployed 1.0 to 1.5 m below absolute low tide. Temperature data was collected every hour at Sites 1 to 6 and 8 to 12 with a Hobo Pendant data logger (Onset Computer Corporation ± 0.5 ^{o}C accuracy). Mean sea temperature was calculated between 3 July and 5 November for sites with available data. Salinity data was collected opportunistically with a

Hanna HI 9828 meter at 0.5, 1.0, and 1.5 m below the water level for each site during sampling periods in September, October, and November.

Images were taken of each panel using a Cannon Power Shot ELPH 150 IS and Nikon Coolpix P7100 during each sampling period. A 100 point grid overlay was added to each image using ImageJ with the Grid Overlay application. The presence or absence of tunicates at each point was used to determine the percent cover of tunicates on each panel.

Natural substratum

To determine spatial and temporal variation in the use of natural habitats by colonial invasive tunicates, samples of eelgrass (*Zostera marina*), rockweed (*Fucus* sp., mostly *F. vesiculosus*) and kelp (*Saccharina latissima*) were sampled from Sites 1, 2 and 5. Initial surveys were conducted at Sites 2 and 5 to locate eelgrass meadows in June. Additional surveys at Site 2 in September and October did not yield an eelgrass meadow. At Site 5, a moderate sized eelgrass meadow was detected near the dock structure and was sampled again in September and October. A GoPro Hero 3 Silver camera was attached to a 20x20 cm quadrat and was used to document the presence of invasive colonial tunicates on eelgrass shoots. Twenty haphazard quadrats were sampled within the eelgrass meadow from 1 to 2 m below absolute low tide. Still images were generated from each quadrat and used to determine the frequency of occurrence of colonial tunicates for each quadrat.

Ten rockweed specimens were sampled below the water level during spring low tide from each site in June, September, and October and ten kelp specimens were sampled between ~0.5 and 1.0 m below absolute low tide at Sites 1 and 2 in June, September, October, and November 2014. Rockweed samples were placed in individual bags and stored in a cooler on ice until they were analyzed the following day. Each rockweed specimen was visually inspected to determine the presence of each tunicate species. Kelp specimens were visually inspected in the field to determine the presence of each tunicate species.

RESULTS

Artificial substratum and physical data

A total of 273 PVC panels were sampled between June and November to determine the abundance and distribution of invasive colonial tunicates at twelve sites along western NL.

Similar to 2013, golden star was detected at Sites 1, 2 and 5 and *Botrylloides* sp. was limited to Site 2. Invasive colonial tunicates were not detected on PVC panels at any other site. The composition of the fouling community was similar between Sites 1, 2, 4, 5, 8, 9, and 11. The fouling community at these sites was dominated by bryozoans, hydrozoans, tunicates, jingle shells and spirorbid worms (Table 2, Appendix B).

Salinity was between 29 and 31 practical salinity units (PSU) for Sites 1, 2, 5, 7, 8, and 9 and was between 26 and 28 for Sites 3, 4, 11, and 12. Salinities between 14 and 16 were observed at Sites 6 and 10, which were near large freshwater sources (Table 1). Wave exposure at Sites 1, 2, and 5 was very low, while Sites 4, 6, 8, 9 and 11 have moderate to high wave exposure (Caines personal observations). A total of three towers were lost throughout the study, accounting for 8.3% of all towers deployed. Towers were lost between September and November and were likely due to increased wind speeds during the first week of November. Mean sea temperature between July and November peaked at 16.1 ^{o}C at Site 5 and decrease at sites further north and south (Fig. 2). Mean sea temperature between July and September explained 87.0% of the variation in golden star colony cover at sites 1, 2, and 5 (Appendix C).

The cover of golden star was 14.1 and 19.5% for panels sampled from floating docks at Sites 2 and 5, respectively, in September. Interestingly, the cover of golden star at Sites 2 and 5 was much lower for panels sampled from fixed docks, ranging from 0 to 2.4% in September (Figs. 3 and 4). Two small florets of golden star were observed on panels from Site 1 in September, but did not account towards cover as they did not fall within one of the intersection points in the grid (Appendix B). The cover of *Botrylloides* sp. peaked at 3.7% on fixed dock panels in September at Site 2 (Fig. 5). Interestingly, panels from Sites 2 and 5 in October and November had numerous recruits, but, due to limitations during picture analyses, these recruits were not accounted for with grid overlay sampling (Appendix B). Overall, the abundance of tunicate recruits (mostly the larger *Botrylloides* sp.) was estimated to be 2800.0 individuals m^{-2} PVC in October and 418.3 individuals m^{-2} PVC in November for fixed dock panels at Site 2 (Fig. 6).

Natural substratum

Initial site assessments discovered a small eelgrass meadow near the fixed and floating dock structures at Site 5. Unfortunately, the topography and benthos at Site 1 was not compatible

Table 1: Geographic coordinates, mean salinity, and mean temperature for twelve study sites along the west coast of Newfoundland, Canada.

Site name	Site number	Latitude	Longitude	Mean salinity (PSU)	Mean temperature (^{o}C)
Port aux Basques	1	47 34.456	59 08.327	30.63	13.04
Codroy	2	47 52.871	59 23.894	30.33	13.75
St. David’s	3	48 13.103	58 51.759	27.94*	na
St. George’s	4	48 25.860	58 29.132	28.00	15.27
Little Port Harmon	5	48 30.859	58 32.297	29.18	16.06
Fox Island River	6	48 41.716	58 40.769	15.20	15.53
Piccadilly	7	48 34.580	58 54.102	30.11	na
Frenchmen’s Cove	8	49 03.501	58 10.814	30.63	na
Lark Harbour	9	49 06.070	58 21.813	30.14	14.25
Summerside	10	48 58.684	57 58.808	14.51	15.33
Rocky Harbour	11	49 35.388	57 56.276	26.16	14.71
St. Paul’s	12	49 51.399	57 47.834	26.82	15.31

* Salinity data for Site 3 is based on a single sampling period. Measurements were taken during a rising tide, when a salt wedge likely forced freshwater further towards the mouth of Crabbe’s River, NL. Additional sampling would provide a more representative value.

Table 2: Dominant species observed from the fouling community on PVC panels sampled in September and November at twelve sites in western Newfoundland, Canada

Site	Dominant species
1	Golden star, spirorbid worms (*Spirorbis* sp.), *Membranipora membranacea*, caprellids, mussels (*Mytilus edulis*), *Cryptosula* sp., anemone (*Metridium* sp.), *Molgula* sp., jingle shells (*Anomia* sp.), *Tubularia* sp.
2	Golden star, *Botrylloides* sp., mussels (*Mytilus edulis*), *Bugula* sp., jingle shells (*Anomia* sp.), spirorbid worms (*Spirorbis* sp.), caprellids
3	Unidentified bryozoan
4	Jingle shells (*Anomia* sp.), *Membranipora membranacea*, *Electra pilosa*, spirorbid worms (*Spirorbis* sp.), *Tubularia* sp.
5	Golden star, barnacles (*Balanus* sp.), anemone (*Metridium* sp.), *Membranipora membranacea*, *Electra pilosa*, *Cribrilina annulata*, *Cryptosula* sp., jingle shells (*Anomia* sp.)
6	Barnacles (*Balanus* sp.), mussels (*Mytilus edulis*)
7	*Membranipora membranacea, Cribrilina annulata, Electra pilosa, Cryptosula* sp.
8	Barnacles (*Balanus* sp.), *Electra pilosa*, *Membranipora membranacea*, *Tubularia* sp., jingle shells (*Anomia* sp.), mussels (*Mytilus edulis*)
9	Barnacles (*Balanus* sp.), *Membranipora membranacea*, spirorbid worms (*Spirorbis* sp.), mussels (*Mytilus edulis*), anemones (*Metridium* sp.), *Electra pilosa*, jingle shells (*Anomia* sp.) *Cribrilina annulata*, *Cryptosula* sp. , *Bugula* sp.
10	Barnacles (*Balanus* sp.)
11	*Membranipora membranacea*, *Electra pilosa*, spirorbid worms (*Spirorbis* sp.), jingle shells (*Anomia* sp.), anemone (*Metridium* sp.) barnacles (*Balanus* sp.), mussels (*Mytilus edulis*)
12	*Electra pilosa*, mussel (*Mytilus edulis*), jingle shells (*Anomia* sp.), unidentified bryozoan

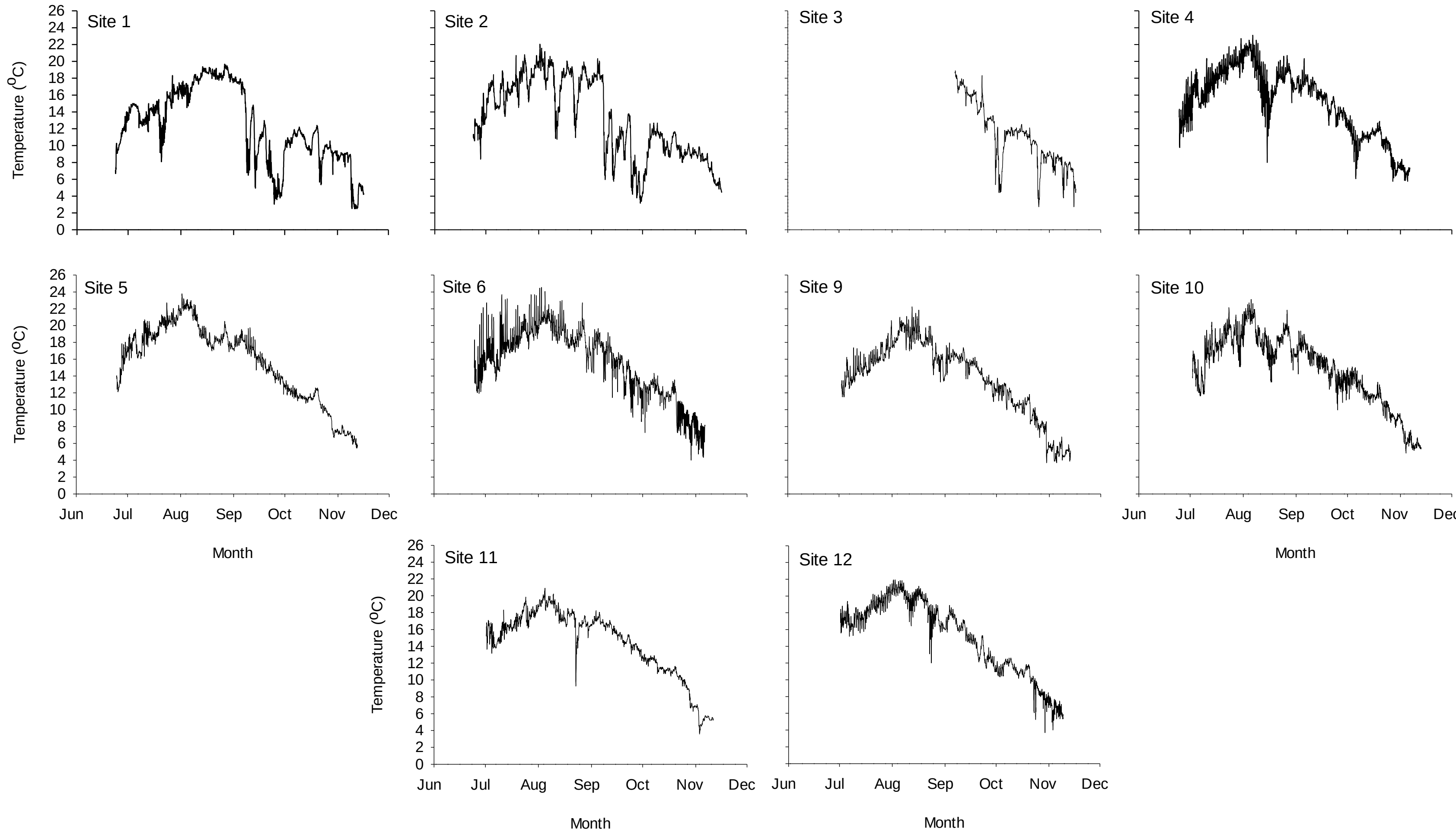

Figure 2: Hourly change in sea temperature at Sites 1 to 6 and 9 to 12 between June and November 2014. Note: Sites are arranged horizontally from south to north. Sites 1, 2, 5, 9 and 10 are deployed from floating docks and Sites 3, 4, 6, 11, and 12 are deployed from fixed docks.

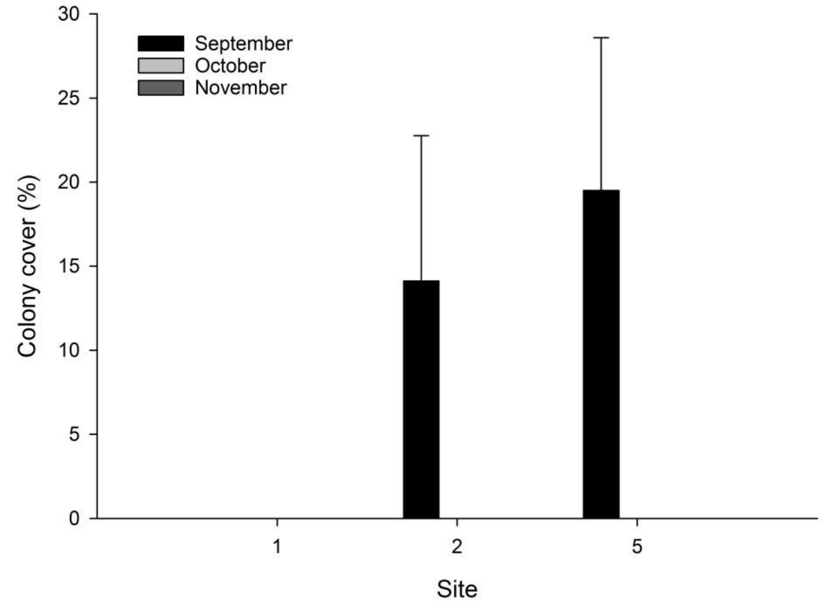

Figure 3: Change in the mean (± SE) colony cover of golden star on PVC panels sampled from floating docks at Sites 1, 2, and 5 in September, October, and November.

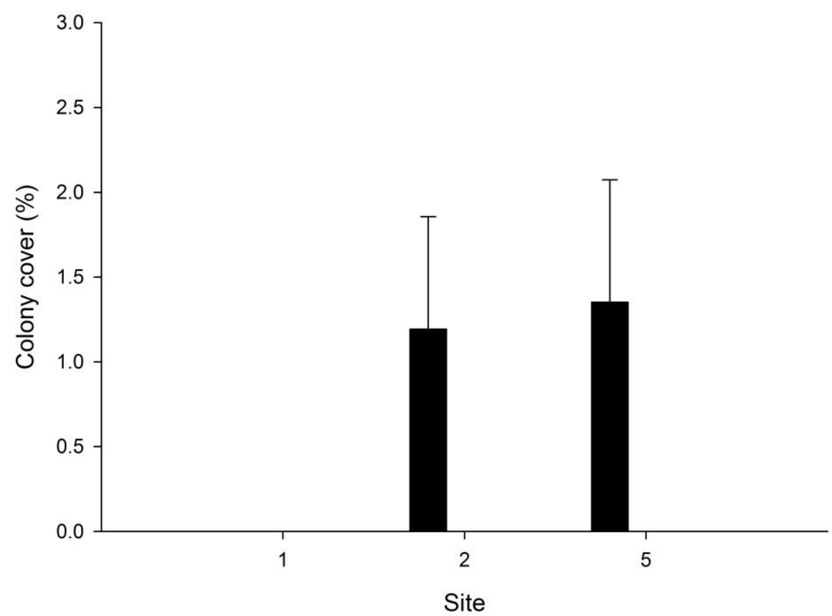

Figure 4: Change in the mean (± SE) colony cover of golden star on PVC panels sampled from fixed docks at Sites 1, 2, and 5 in September, October, and November.

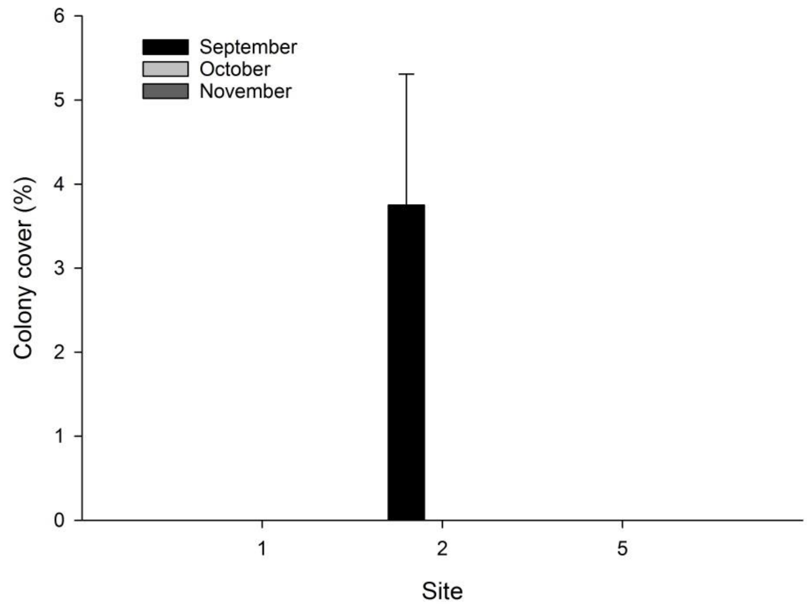

Figure 5: Change in the mean (± SE) colony cover of *Botrylloides* sp. on PVC panels sampled from fixed docks at Sites 1, 2, and 5 in September, October, and November.

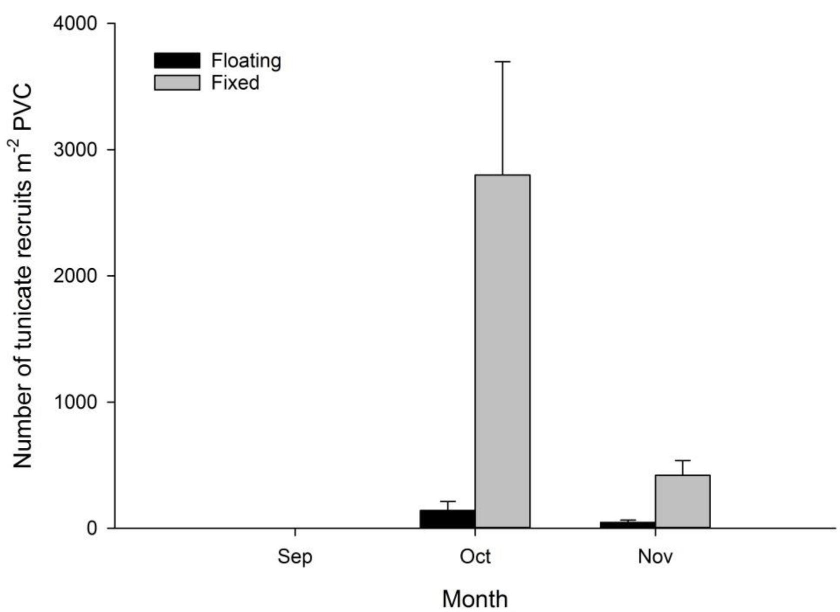

Figure 6: Change in the mean (± SE) abundance of tunicate recruits on PVC panels sampled from fixed docks at Site 2 in September, October, and November.

for eelgrass and site assessments in June, September, and October did not yield an eelgrass meadow in Codroy harbour or the Codroy estuary. Small patches of eelgrass were observed in Codroy harbour, but were beyond the sampling depth for this study. The frequency of golden star colonies on eelgrass peaked at 40.0% in September before decreasing to 15.0% in October at Site 5 (Fig. 7). Similarly, the frequency of occurrence of colonies on rockweed decreased from 100% in September to 80% in October (Fig. 8). The occurrence of golden star on rockweed increased from 20 to 100% and the occurrence of *Botrylloides* sp. increased from 10 to 50% at Site 2 between September and October (Fig. 8 and 9). No colonies were observed on rockweed specimens sampled at Site 1. The frequency of occurrence for golden star on kelp peaked at 40% in October for Site 1 and was 10 and 20% in September and November, respectively (Appendix B, Fig. 10). Colony frequency of golden star on kelp specimens from Site 2 was 80 to 100% in September and October before decreasing to 44% in November. Similarly, the frequency of *Botrylloides* sp. on kelp peaked at 70% in October before decreasing to 33% in November (Fig. 11).

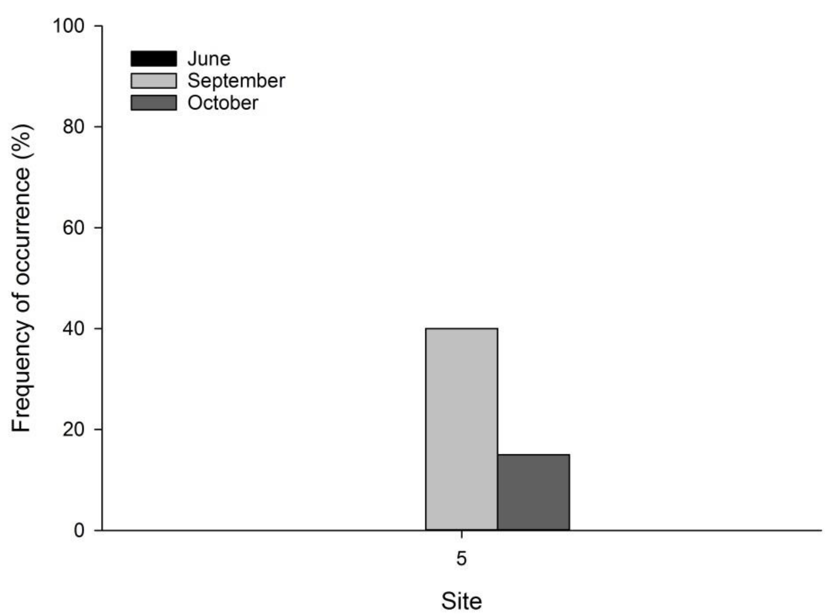

Figure 7: Change in the mean frequency of occurrence of golden star on eelgrass shoots sampled from Site 5 in June, September, and October.

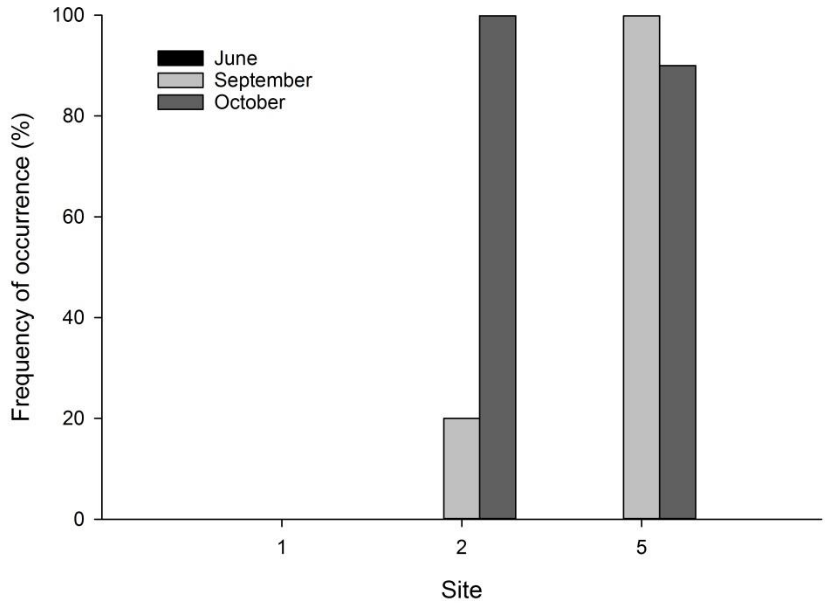

Figure 8: Change in the mean frequency of occurrence of golden star on rockweed fronds sampled from Sites 1, 2 and 5 in June, September, and October.

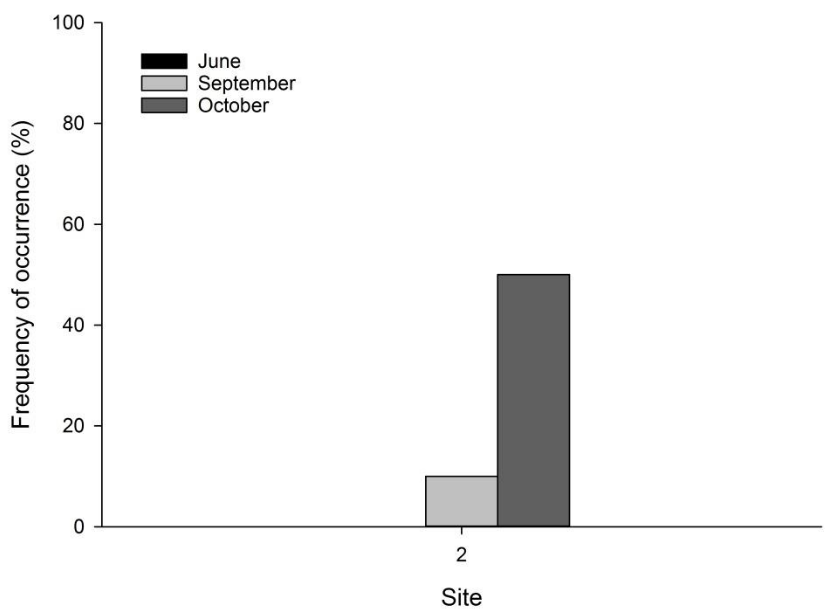

Figure 9: Change in the mean frequency of occurrence of *Botrylloides* sp. on rockweed fronds sampled from Site 2 in June, September, and October.

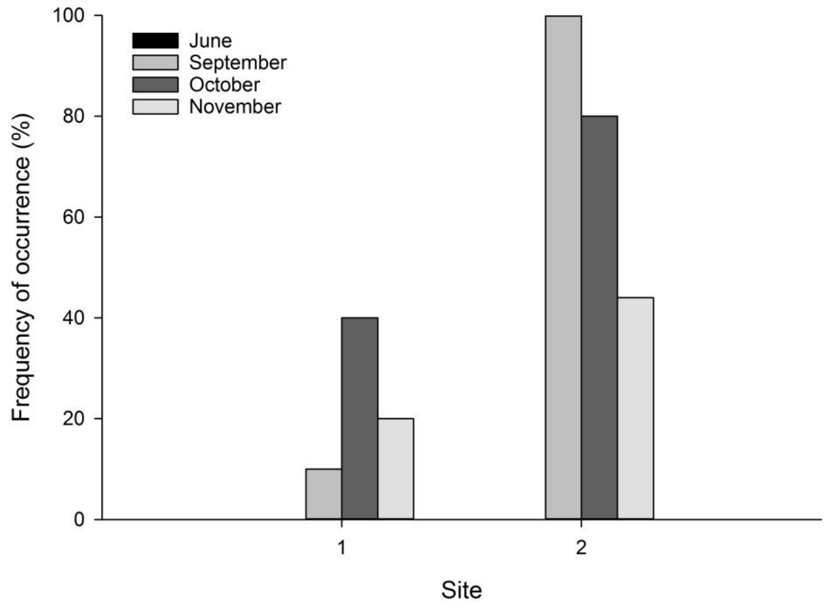

Figure 10: Change in the mean frequency of occurrence of golden star on kelp blades sampled from Sites 1 and 2 in June, September, October, and November.

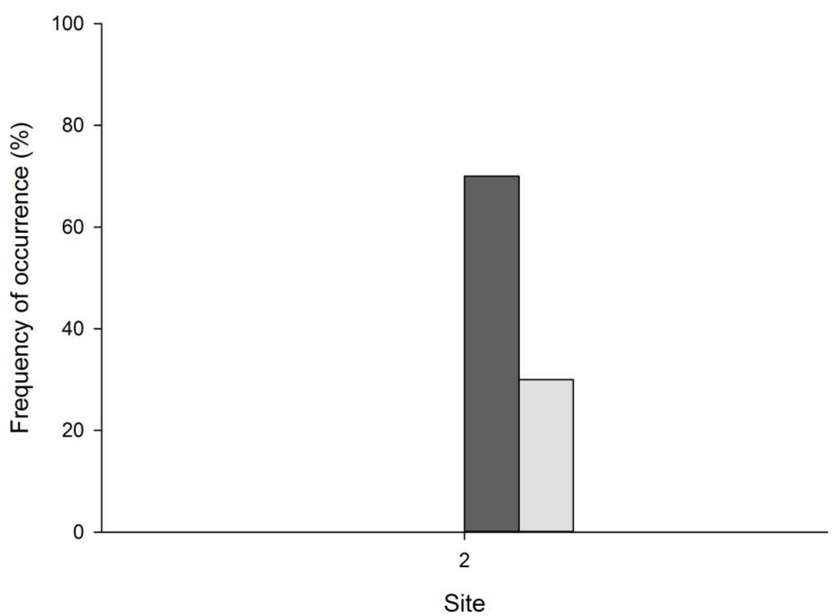

Figure 11: Change in the mean frequency of occurrence of *Botrylloides* sp. on kelp blades sampled from Site 2 in June, September, October, and November

DISCUSSION

Invasive colonial tunicates were observed at Sites 1, 2 and 5, with golden star found at all three sites and *Botrylloides* sp. observed at Site 2. Studies have utilized both sea temperature and salinity as predictive variables to determine suitable habitats and the potential range for both golden star and violet tunicates in the eastern Pacific (Epelbaum et al 2009). Low salinities, due to proximity to a large river and/or stream, will likely preclude the successful establishment of these invaders at Sites 3, 6, and 8. The fouling community, salinity, and sea temperature at invaded sites was similar to Sites 4, 9, 11, and 12, suggesting that future establishment of golden star or *Botrylloides* sp. is possible. These sites are further at risk, as pelagic herring and mackerel seiners move between northern and southern NL during peak recruitment in the fall (Caines personal observation). Although Sites 4, 9, 11, and 12 appear to match a number of the physiological requirements of both golden star and violet tunicate, these sites may be inhospitable due to increased wave exposure, difference in available food resources, and resource competition (food and space).

Latitudinal and annual variation in sea temperature has been shown to greatly influence the population dynamics and, subsequently, species interactions between invasive and native species (Stachowicz et al 2002, Rahel & Olden 2008, Saunders & Metaxas 2007, 2008, Sorte et al 2010, Caines & Gagnon 2012). Overall, the abundance and cover of golden star on PVC panels at Site 1 was substantially lower than Site 5, where sea temperature was on average 3.0 ^{o}C warmer. Overall, sea temperature was similar between Sites 1 and 2 during the entire study, but was on average 1.3 to 3.2 ^{o}C lower at Site 1 compared to Sites 2 and 5, respectively, between July and September. Temperature plays a key role in the growth and development of golden star and violet tunicate and likely was an important factor influencing the population dynamics of these species at Sites 1, 2 and 5 (Epelbaum et al 2009). Low recruit abundance and subsequent growth of these recruits during the summer and fall will likely be an important factor affecting the overwintering survival of colonies at Site 1. Interestingly, mean sea temperature during the summer and fall at Site 1 is similar to more northerly sites in NL, providing us a glimpse into the invasion dynamics of golden star at higher latitudes in Atlantic Canada (Caines & Gagnon 2012a,b).

The frequency of occurrence of golden star colonies on kelp increased from 10 to 40% between September and October at Site 1. Single small golden star florets were observed in September on PVC panels and kelp specimens, while colonies observed on kelp in October and November were larger and consisted of a number of zooid florets. A lack of recruits and small colonies on PVC in October and November suggests a single recruitment event was recorded at Site 1. Kelp specimens at Sites 2 and 5 were characterized by numerous colonies of varying sizes throughout the study, while PVC panels showed that tunicate recruitment peaked in October before decreasing in November. The abundance of tunicates on rockweed and kelp increased between September and October at Site 2, but decreased on eelgrass and rockweed at Site 5 during the same period. Warmer sea temperatures at Site 5 likely contributed to the earlier peak and overall percent cover of golden star compared to Site 2.

Habitat utilization will play an important role in the management and transport of colonial invasive tunicates in western Newfoundland. Artificial dock structures will provide a refuge from which these invaders can infiltrate natural substrates (Dumont et al 2011). Botryllid tunicates are characterized by lecithtrophic larvae that typically spend less than a few hours in the water column, which greatly reduces larval transport (Carver et al. 2006). Tunicates overcome this by colonizing boat hulls and rafting on dislodged algae and plants (Carver et al. 2006, Clarke-Murray et al 2011). The movement of fishing boats along the west coast of NL during the fall and rafting on dislodge kelp blades, rockweed fronds, and eelgrass shoots will inevitably result in the transport of these species to new habitats in western NL. Although these introductions may not have the same economic effect as seen in other parts of Atlantic Canada, they may have a detrimental effect on subtidal coastal environments (Carver et al. 2006). Globally, seagrasses are declining in response to multiple stressors, including biological interactions with aquatic invasive species (Hauxwell et al. 2001, Waycott et al 2009, Wong & Vercaemer 2009, Garbary et al. 2014). The recent introduction and future range expansion of colonial invasive tunicates in western NL will put additional pressure on ecologically significant eelgrass habitats, which may lead to large scale changes in the composition and structure of subtidal coastal communities.

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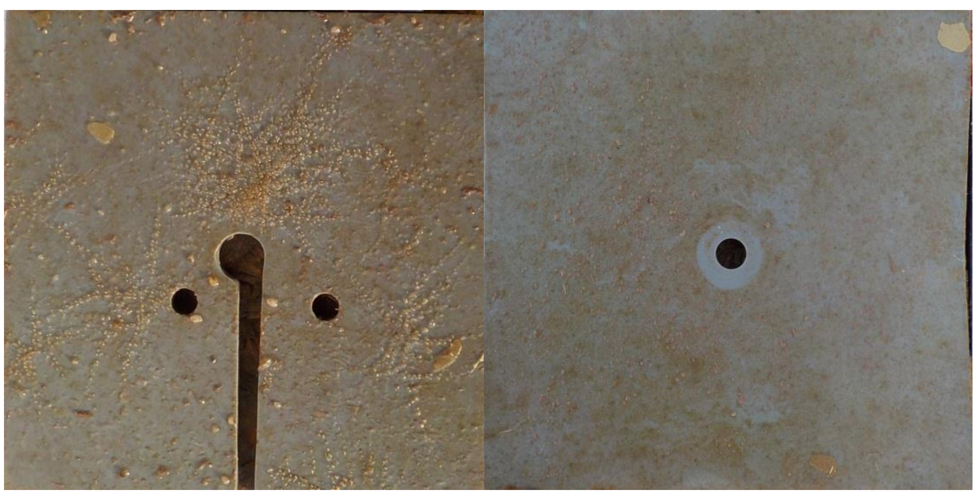
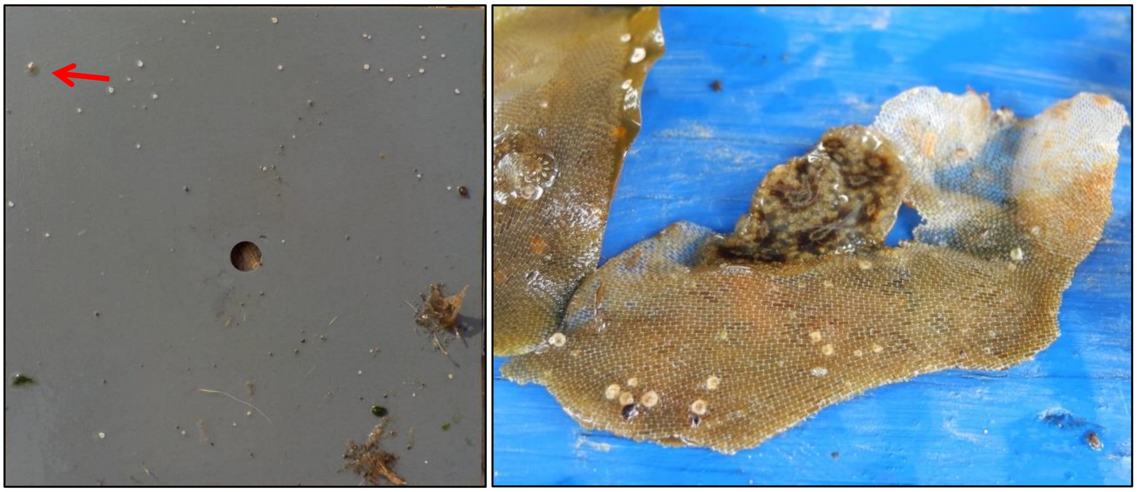
APPENDIX A: Tower design and deployment

Sampling tower: Image showing tower orientation and placement of plates during deployment from a floating dock at Site 2.

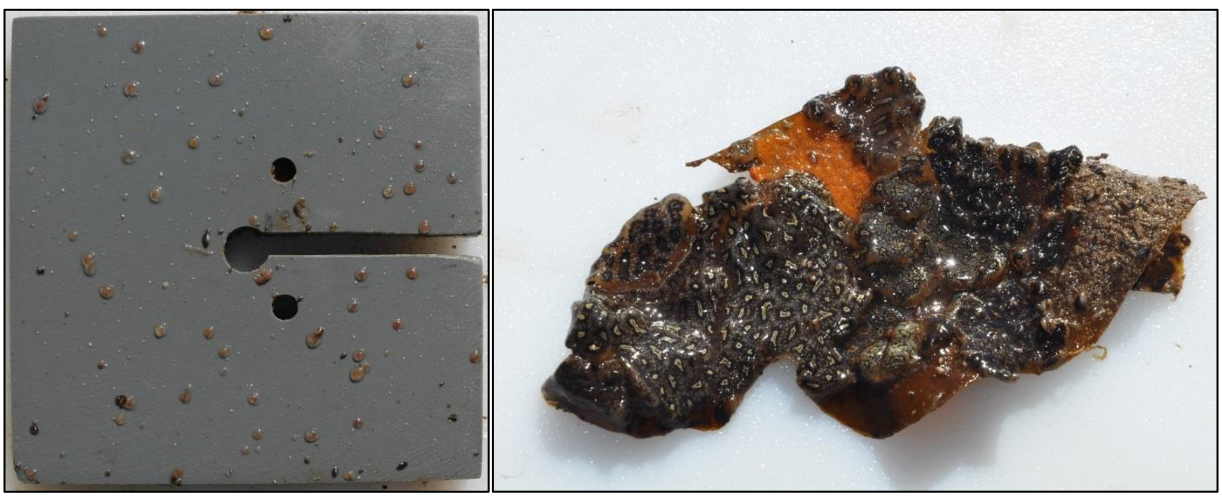


Sampling panels: Image showing 10x10 (left) and 15x15 cm (right) PVC panels used to sample fouling communities at twelve sites in western Newfoundland

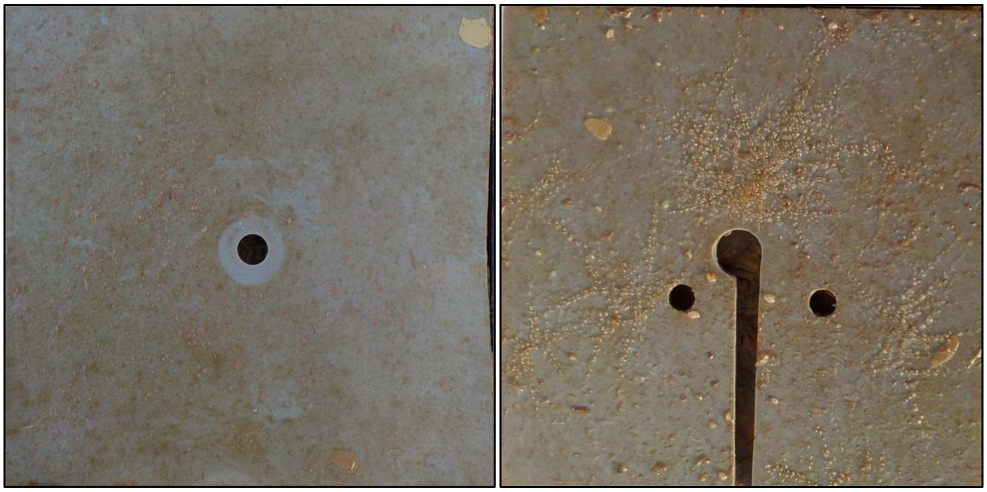
APPENDIX B: Representative images of fouling communities at twelve study sites



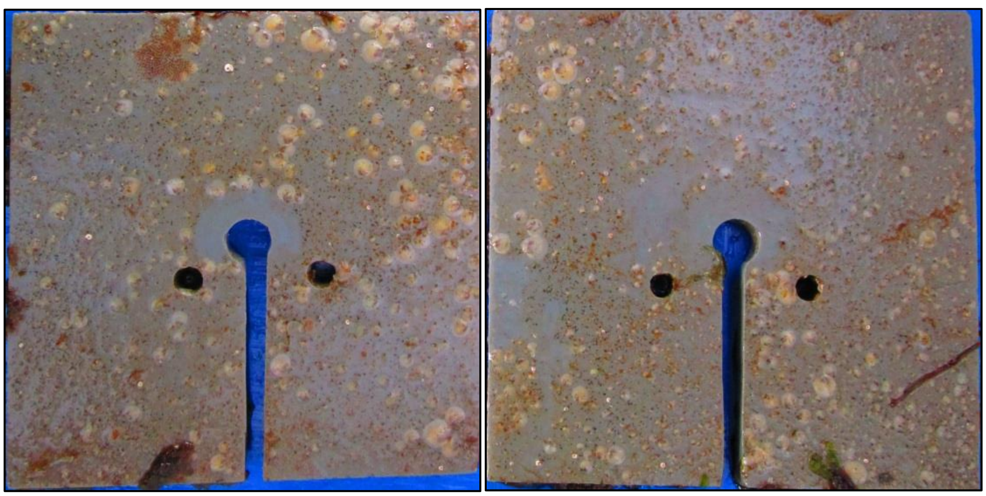
Site 1: Images showing a PVC panel with a single golden star floret (top left of image) sampled in September and a kelp specimen with a golden star colony sampled in November.



Site 2: Images showing a PVC panel with numerous tunicate recruits and a kelp specimen with golden star and *Botrylloides* sp. colonies sampled in October.

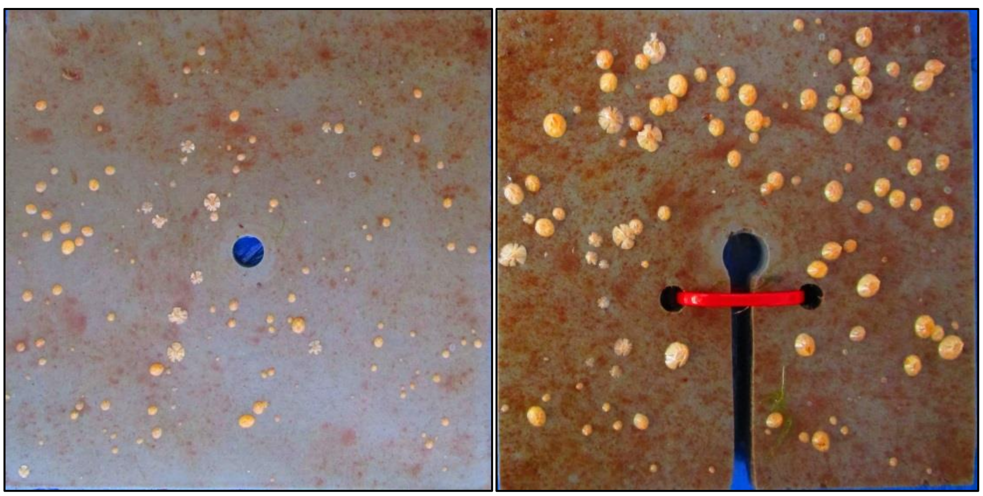


Site 3: Images showing two PVC panels with unidentified bryozoans and bivalves sampled in September.

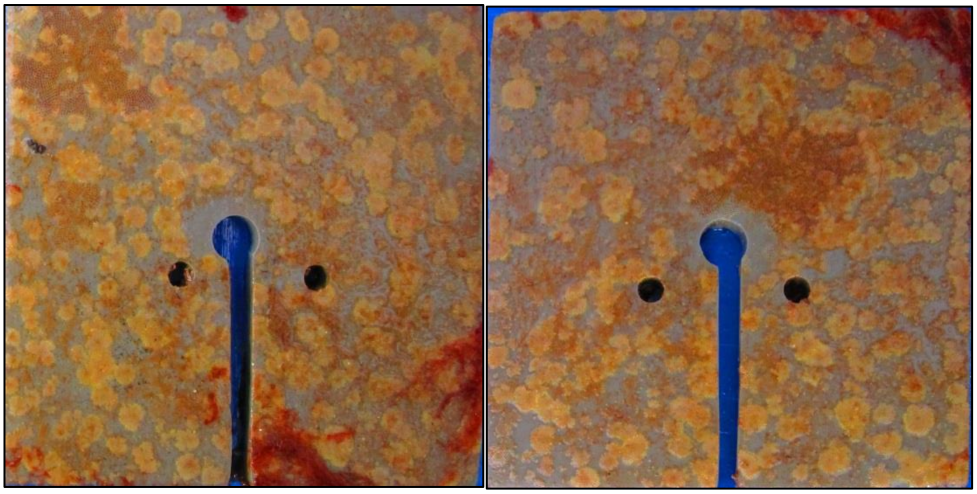


Site 4: Images showing two PVC panels with jingle shells (*Anomia* sp.), *Membranipora membranacea*, and spirorbid worms (*Spirorbis* sp.).

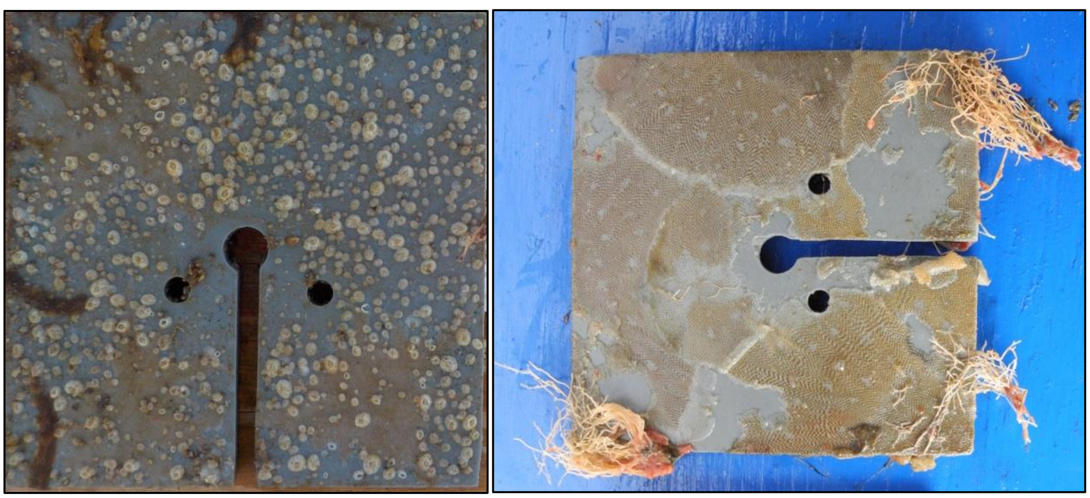
Site 5: Images showing golden star colonies growing on rockweed and a PVC panel in September.



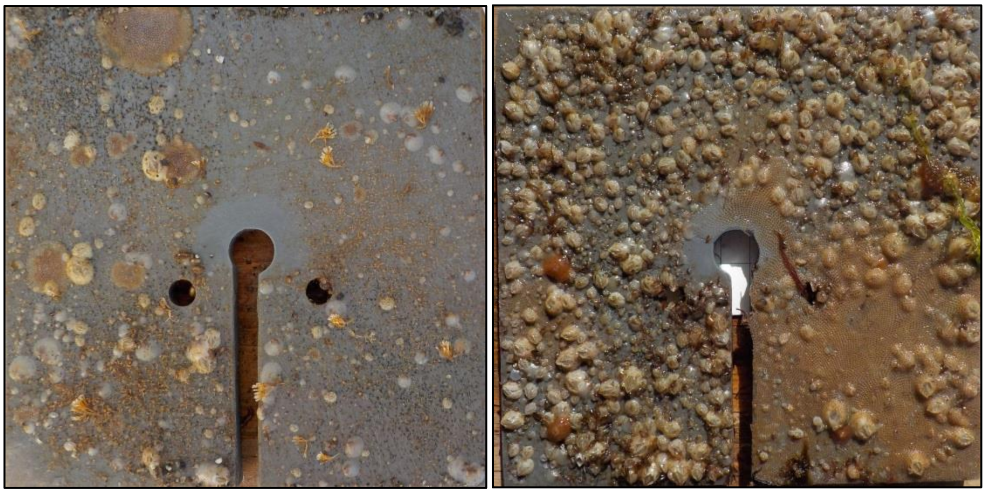
Site 6: Images showing barnacles (*Balanus* sp.) on PVC panels sampled in November.



Site 7: Image showing *Membranipora membranacea* and *Cribrilina annulata* growing on PVC panels sampled in November.



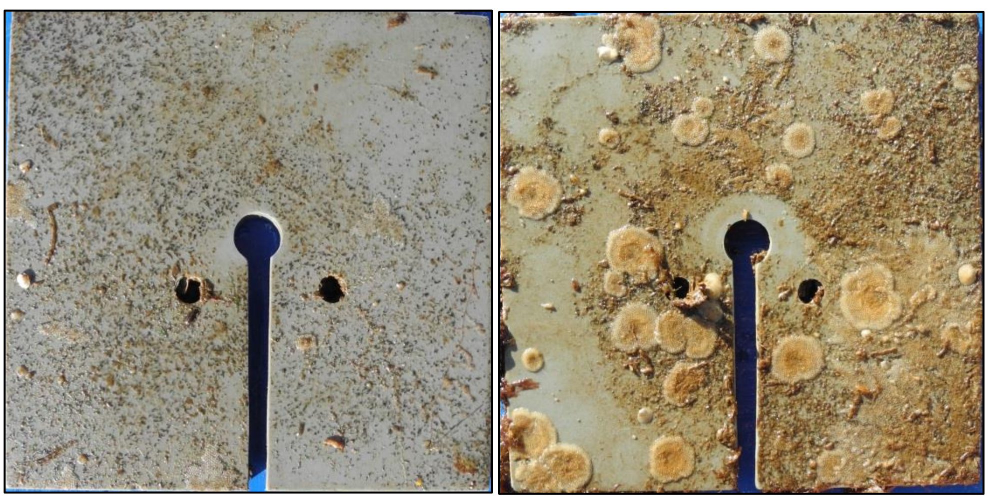
Site 8: Images showing *Electra pilosa*, jingle shells (*Anomia* sp.) and barnacles (*Balanus* sp.) sampled in September (left) and *Membranipora membranacea* and *Tubularia* sp. sampled in November (right).



Site 9: Images showing *Bugula* sp., *Cryptosula* sp., *Electra pilosa*, jingle shells (*Anomia* sp.), barnacles (*Balanus* sp.), *Membranipora membranacea*, and anemones (*Mytridium* sp.) on PVC panels sampled in September.

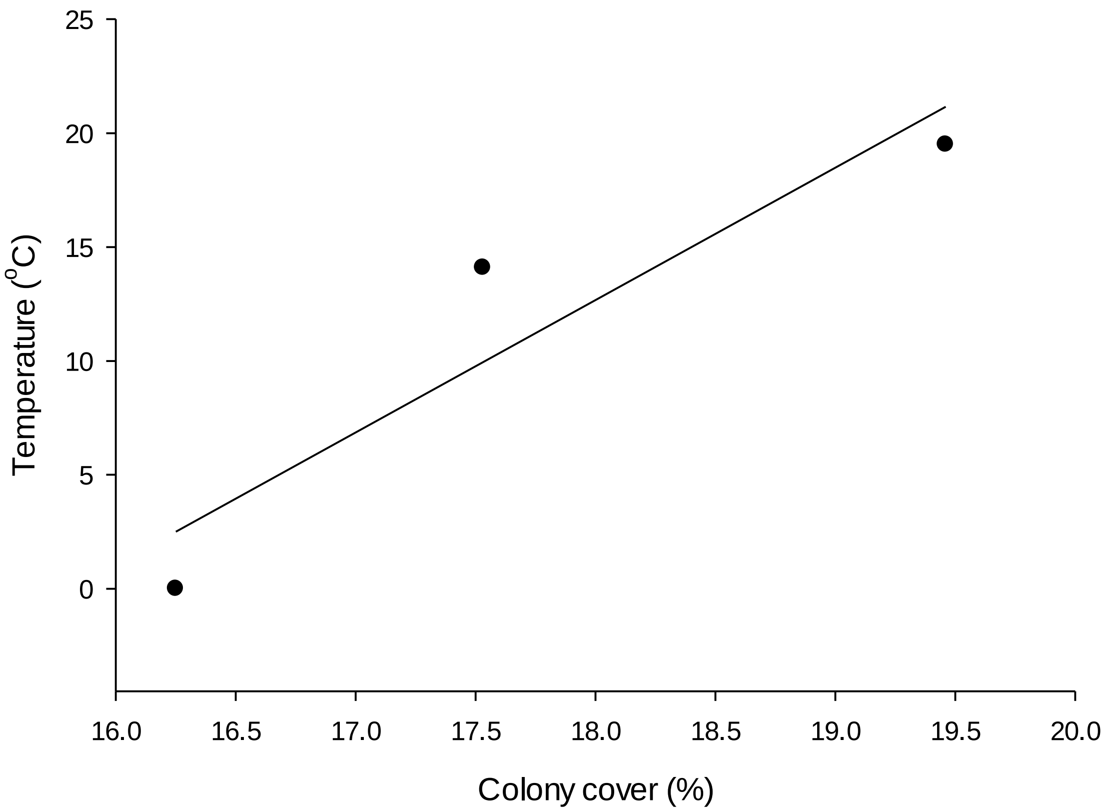
Site 10: Images showing barnacles (*Balanus* sp.) growing on PVC panels sampled in September.

Site 11: Images showing *Electra pilosa*, barnacles (*Balanus* sp.) spirorbid worms (*Spirorbis* sp.), and an unidentified bryozoan from PVC panels sampled in September and November.



Site twelve: Images showing *Electra pilosa*, jingle shells (*Anomia* sp.), and unidentified bryozoan on PVC panels sampled in November.

APPENDIX C: Regression of temperature and golden star colony cover in September



Regression analysis showing the relationship between mean sea temperature and golden star percent cover at Sites 1, 2, and 5. Line is the linear fit for the data ($r^2 = 0.870$).

Note: Mean temperature data used in regression model was based on values collected between 3 July and sampling date in September for Sites 1 and 2 (6 September) and Site 5 (2 September). Sample size was too small to run ANOVA analyses. Results should be interpreted cautiously. Further data collection is needed to accurately elucidate latitudinal differences in population dynamics.