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Research Article

First record of vase tunicate, *Ciona intestinalis* (Linnaeus, 1767), in coastal Newfoundland waters

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Abstract

Vase tunicate, *Ciona intestinalis* (Linnaeus, 1767) Type B (*sensu* Sato et al. 2012), was first recorded in Newfoundland (Canada) in Burin, Placentia Bay, by SCUBA divers conducting rapid visual surveys for invasive species on 19 September 2012. Follow-up surveys of Burin and adjacent communities were conducted in October and November 2012. *Ciona intestinalis* was present in relatively low abundance in Burin, but a well-established population was located in Little Bay, approximately 15 km north of Burin. Little Bay is located within Mortier Bay, an area of high vessel traffic and the possible introduction source of *C. intestinalis*. Further surveys are recommended to delineate the present distribution and monitor future dispersal of *C. intestinalis* in coastal Newfoundland waters. Due to the demonstrated high ecological and economic impacts of this species in other previously invaded regions, options for mitigating population growth and dispersal should be examined to minimize effects of this non-indigenous species.

Key words: non-native; invasive species; ascidian; sea squirt; survey

Introduction

Non-indigenous ascidians continue to be problematic globally as invasive species (Lambert 2002; 2007), as they often have wide environmental tolerances, rapid growth rates, early sexual maturity, and are prolific spawners (Lambert 2002). Invasive ascidians may have significant ecological and economic effects, by altering subtidal community structure and diversity (Lambert and Lambert 2003; Blum et al. 2007; Valentine et al. 2007) and by fouling man-made structures, especially bivalve aquaculture sites (LeBlanc et al. 2007; Ramsay et al. 2008; Rocha et al. 2009).

A variety of survey methods using qualitative and/or quantitative sampling techniques have been employed worldwide to detect introduced marine species (Campbell et al. 2007). Visual surveys by SCUBA divers have been demonstrated to be highly effective in detecting artificial *Ciona intestinalis* decoys (Kanary et al. 2010). Rapid visual surveys (RVS) were carried out opportunistically by SCUBA divers in September 2012 while conducting transect surveys for an ongoing Department of Fisheries and Oceans project: the Wharf and Breakwater Survey Project. These surveys supplemented the Aquatic Invasive Species (AIS) Harbour Survey Monitoring Program of the Department of Fisheries and Oceans and Memorial University. This paper reports the first detection of vase tunicate, *Ciona intestinalis* (Linnaeus, 1767), in coastal Newfoundland waters.

Methods

The Wharf and Breakwater Survey Project (WB Project) of Canada's Department of Fisheries and Oceans (DFO) investigates changes to habitat, fish, epibenthic macroinvertebrate, and macroalgae species associated with wharf or breakwater construction sites in Newfoundland. Since 2007, 17 sites (11 wharf/breakwater and 6 controls) have been surveyed by SCUBA divers annually in September along the coast of insular Newfoundland (Figure 1A). In 2012, while two divers conducted visual surveys and video recorded habitat along transects, a third diver conducted RVS at all project sites. RVS consisted of brief (15 – 20 min) dives to visually examine adjacent wharves, floating docks, breakwater structures, and boat hulls for the presence of aquatic invasive species.

Initial observation of *Ciona intestinalis* during one RVS prompted follow-up surveys in Burin and nearby communities. Follow-up surveys were conducted by DFO's Newfoundland and Labrador AIS Program, in collaboration with Memorial University of Newfoundland's Department of Ocean Sciences, hereafter referred to as the DFO/MUN AIS survey program. This program has been conducting AIS surveys in Newfoundland's coastal marine waters since 2006. During these AIS surveys a variety of techniques were employed including RVS, underwater video, photography, and quadrat scrapings of wharf structures by SCUBA divers, deployment of crab traps, and occasionally beach seining. Previous surveys by this program in 2011 (10 sites) and 2012 (25 sites) did not detect *C. intestinalis* (Figure 1A). In October 2012, divers conducted video transect surveys in Baine Harbour and Spanish Room, and RVS of public wharves and adjacent natural substrates in Burin, Little Bay, Little St. Lawrence, and St. Lawrence (Figure 1B). These RVS were similar to those conducted during the WB Project but were more extensive (30 – 45 min). They consisted of visual examination of public wharves, including wharf understructure and pilings, floating docks, adjacent boat hulls, and nearby natural substrates and observations were recorded post-dive. In November 2012, further surveys were completed by the DFO/MUN AIS survey team to delineate the range of *C. intestinalis* within Little Bay, and Mortier Bay.

Results and discussion

On 19 September 2012, four *Ciona intestinalis* specimens (Figure 2A) were identified during the WB Project RVS in Burin, Newfoundland (Figure 1). This was the first observation of *C. intestinalis* in Newfoundland waters (Ma et al. 2011; DFO 2012). This observation coincided with the first detection of *C. intestinalis* on collector plates in St. Pierre Harbour, St. Pierre and Miquelon, France (4 September 2012; F. Urtizberea, Department of Agriculture and Forestry, St. Pierre and Miquelon, France, pers. com.) approximately 80 km southwest of Burin (Figure 1B). St. Pierre and Miquelon has been a member of the AIS Zonal Monitoring Program since 2010 (B. Vercaemer, DFO, Dartmouth, Nova Scotia, Canada, pers. com.). Burin *C. intestinalis* specimens were attached to coffin box bryozoan, *Membranipora membranacea* (Linnaeus, 1767), another non-native species in Newfoundland waters present since the early 2000s (R. Hooper, MUN, St. John's, Newfoundland and Labrador, pers. com.). In turn, *M. membranacea* were attached to the native kelp, *Saccharina longicruris* (Bachelot de la Pylaie) Kuntze, 1891, growing on wharf and floating dock structures. Specimens were observed at 2 – 5 m depth at 15°C. Several small colonies (1 – 4 cm diameter) of the non-native golden star tunicate, *Botryllus schlosseri* (Pallas, 1766), were also observed on the floating dock, growing on *M. membranacea* attached to *S. longicruris* and on a car tire. *Botryllus schlosseri* has been reported in Placentia Bay since 2006 (Callahan et al. 2010; Ma et al. 2011). Green crab, *Carcinus maenas* (Linnaeus, 1758), an invasive species observed in Placentia Bay since 2007 (Klassen and Locke 2007), was not observed at the Burin site, but local people reported seeing this species in the area. The Burin site was previously surveyed on 20 September 2007, during DFO/MUN AIS surveys. At that time only indigenous ascidian species were observed, including *Aplidium glabrum* (Verrill, 1871), *Ascidia* sp., *Boltenia echinata* (Linnaeus, 1767), and great numbers of large *Halocynthia pyriformis* (Rathke, 1806). During the 2012 WB Project RVS, many large *H. pyriformis* appeared dead or dying, although many small individuals were observed on the outer wharf pilings. One small *Molgula* sp. was also observed attached to a piece of *S. longicruris*. Through the remainder of the RVS, the only invasive species observed was *M. membranacea,* which was detected at all sites, except Renews, and both Terra Nova sites (see Figure 1A), although it was observed at other locations in Terra Nova in 2012 (P. Sargent, per. obs.).

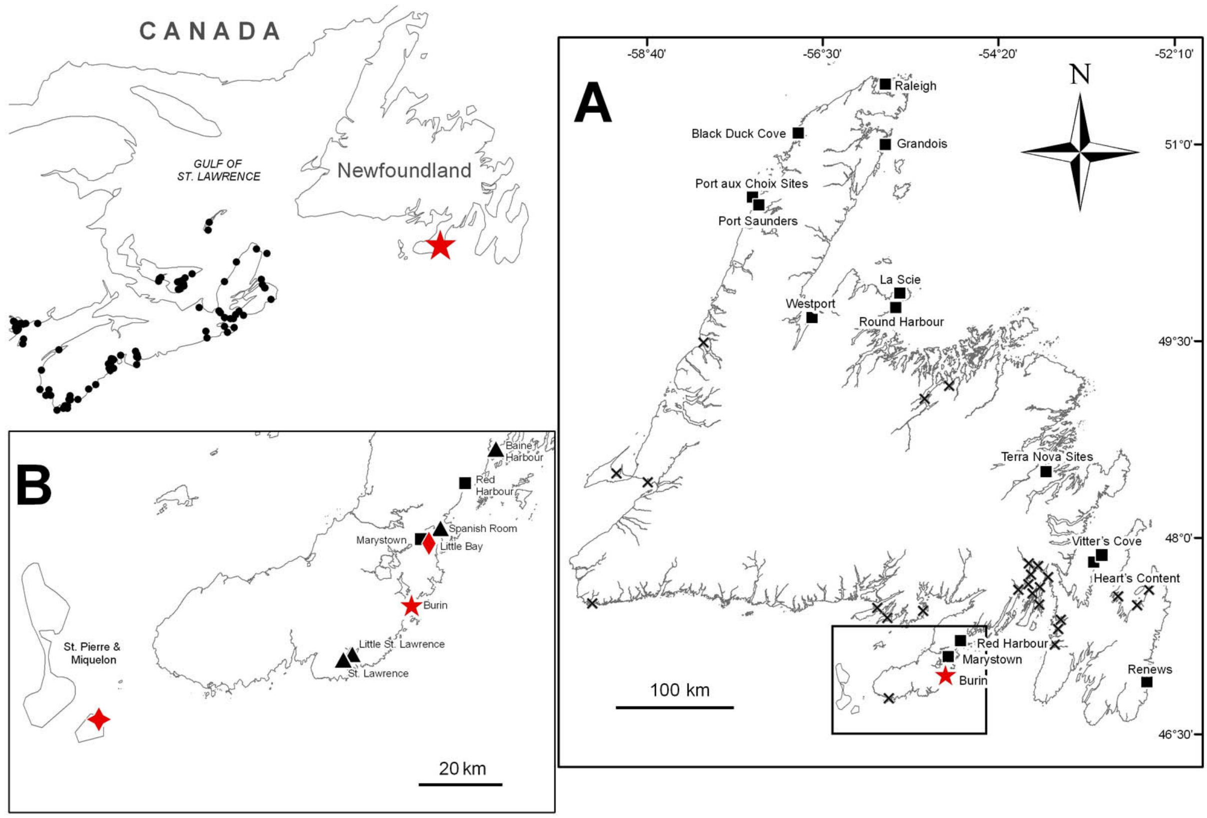

Figure 1. *Ciona intestinalis* distribution in eastern Canada from the National Aquatic Invasive Species Database (DFO 2012) (●) and collection site of specimens in Burin, Newfoundland on 19 September 2012 (★): A – Inset of insular Newfoundland showing Wharf and Breakwater Survey Project sites (■), and Department of Fisheries and Oceans and Memorial University of Newfoundland Aquatic Invasive Species Survey sites 2011 – 2012 (×); B – Inset of southern Burin Peninsula, Newfoundland showing follow-up surveys (▲), collection site in St. Pierre Harbour, St. Pierre and Miquelon, France on 4 September 2012 (F. Urtizberea, pers. com.; ✦) and site of *C. intestinalis* observations in Little Bay, Newfoundland on 18 October 2012 (♦).

Three *C. intestinalis* specimens collected from Burin were preserved in ethyl alcohol (anhydrous). Two specimens were deposited in The Rooms, Provincial Museum of Natural History Annex (St. John's, Newfoundland and Labrador, Canada) under Catalogue #NFM CH-1. The remaining specimen was dissected to examine gonads and estimate sexual maturity. Presence of red ocelli, 6 on the atrial siphon and 8 on the buccal siphon (Van Name 1945; Figure 2A), lack of white pigment flecks in the body wall (Lambert and Lambert 1998; Figure 2B), and presence of single subterminal refringent bodies in the follicle cells of the eggs (Byrd and Lambert 2000; Figure 2C) confirmed species identification as *C. intestinalis*. Genomic studies have revealed the presence of two cryptic species of *C. intestinalis*, presently referred to as Types A and B (Sato et al. 2012). Yellow pigmentation on the distal end of the siphons (Figure 2A) and absence of tubercles on the sides of the siphons (Figure 2B) further identified the Burin specimen as *C. intestinalis* Type B (Sato et al. 2012). This individual was approximately 70 mm in length. Its vas deferens were white with sperm and its oviducts and ovary were full of eggs (170 µm diameter; Figure 2D). It was sexually mature, and capable of reproduction (G. Lambert, University of Washington, Seattle, Washington, USA, pers. com.).

Follow-up DFO/MUN AIS video transect surveys on 17 October 2012 did not detect *C. intestinalis* at Baine Harbour or Spanish Room (Figure 1B). The Burin public wharf site was

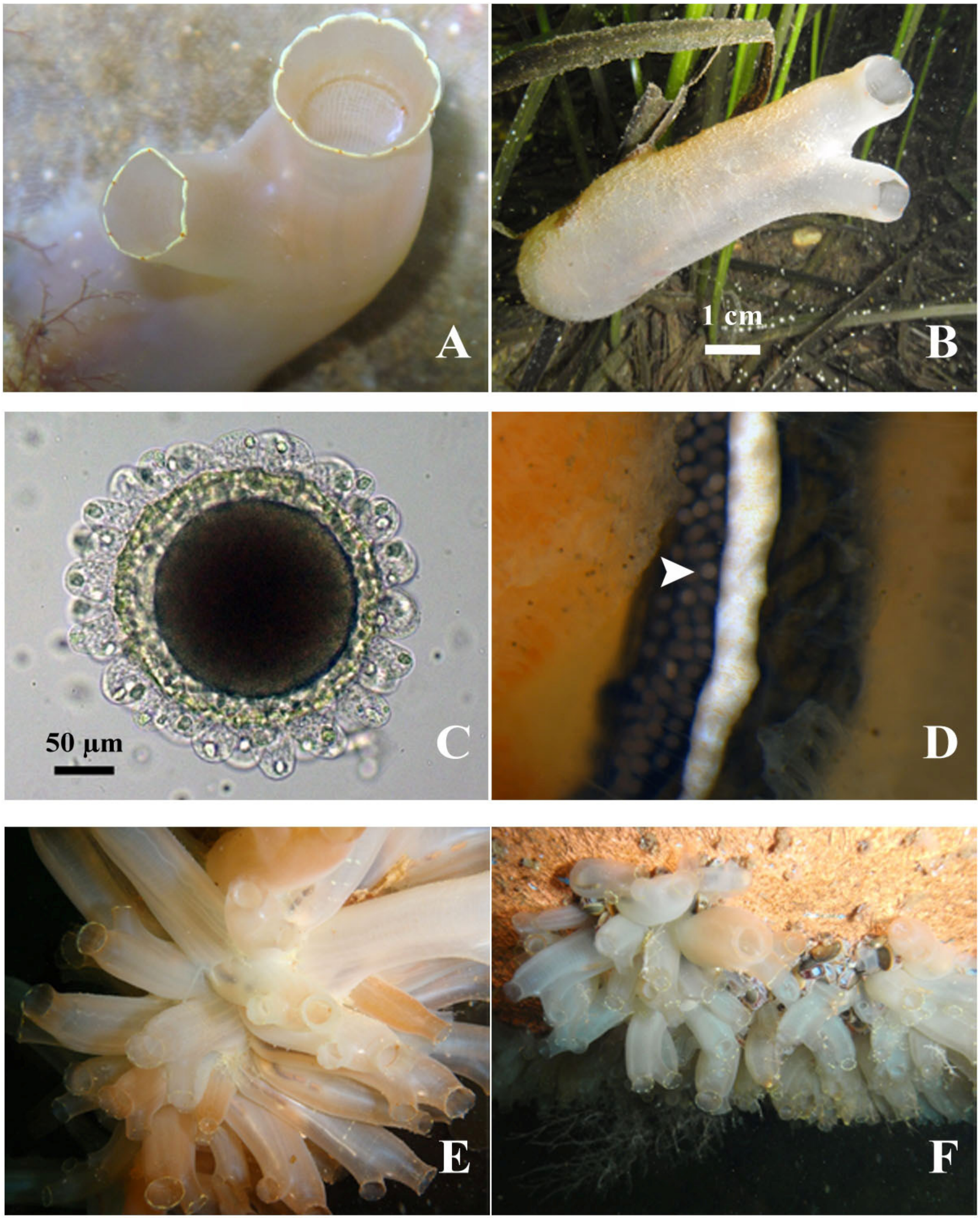

Figure 2. Photographs of *Ciona intestinalis* discovered in Newfoundland: **A** – *C. intestinalis* specimen from the public wharf in Burin showing red ocelli and yellow pigmentation on distal end of siphons; **B** – *C. intestinalis* attached to *Zostera marina* showing absence of tubercles on sides of siphons; **C** – *C. intestinalis* egg showing follicle cells with single subterminal refringent bodies; **D** – *C. intestinalis* showing oviduct with eggs (arrowhead) and white sperm duct (right); **E** – *C. intestinalis* specimens attached to the wharf in Little Bay; **F** – *C. intestinalis* specimens attached to a boat hull in Little Bay. **A** – Photograph by P. Sargent; **B**, **E**, **F** – Photographs by R. O'Donnell; **C**, **D** – Photographs by T. Wells.

re-examined on 18 October 2012 and divers observed that *C. intestinalis* was widespread at this site, but in low densities, attached to kelps, *S. longicruris* and *Agarum cribosum* (Bory de Saint-Vincent, 1826), mussels, *Mytilus* sp., wharf structure, and sea cucumbers, *Cucumaria frondosa* (Gunnerus, 1770). *Ciona intestinalis* was not detected at Little St. Lawrence or St. Lawrence. Only indigenous *Ascidia callosa* (Stimpson, 1852) and *Molgula complanata* (Alder and Hancock, 1870) were observed on kelp attached to the public wharf in St. Lawrence. In Little Bay, large and small individuals of *C. intestinalis* were detected in very high densities with up to 100% coverage on the undersurface wharf structure and pilings, kelp, and mussels. Most individuals were attached to permanent wharf structures (Figure 2E), but they were also attached to the two adjacent floating docks and hull of one vessel at the wharf (Figure 2F). *Ciona intestinalis* were also observed attached to blades of eelgrass, *Zostera marina* (Linnaeus, 1753), near the wharf (< 1 km away) individually (Figure 2B) and in clumps of 5 – 10 animals and to the stipe of an individual *S. longicruris*. Given the abundance and variation of size classes of *C. intestinalis* observed, it is likely to have been established in Newfoundland waters for more than one year. Further surveys conducted 26 – 27 November 2012 found *C. intestinalis* throughout Little Bay, except mid-harbour where depth increased and substrate changed to sand with little to no vegetation. The public and shipyard wharves, immediately adjacent the WP Project site in Marystown were also surveyed. Small *C. intestinalis* were observed near the base of pilings on the public wharf and on mussels and ladders of the adjacent private wharf. General abundance in Marystown was much lower than in Little Bay.

The present observations of *C. intestinalis* in Newfoundland waters are approximately 375 km east of previous reports of this species in Cape Breton, Nova Scotia (DFO 2012; Figure 1). Swimming larvae of *C. intestinalis* are planktonic for only 1 – 6 days before settlement (Svane and Havenhand 1993) which suggests planktonic dispersal is limited. Kanary et al. (2011) predicted planktonic dispersal of *C. intestinalis* may be up to 6 km per generation. Limited planktonic dispersal and extensive distance between the Burin Peninsula population and other Atlantic Canadian populations of this species suggests vessel traffic was the likely transmission vector.

Little Bay is a small, sheltered embayment of Mortier Bay, 3 km east of Marystown, the largest community on the Burin Peninsula. Mortier Bay is a busy area for vessel traffic and may be the point of introduction for *C. intestinalis*. Invasive ascidians survive particularly well in sheltered harbours where they may rapidly reproduce and further colonize (Lambert and Lambert 1998). In Little Bay, environmental conditions may have been exceptionally favourable to allow the establishment and colonization of *C. intestinalis*, whereas nearby Marystown and Spanish Room are more exposed and may be less favourable (Figure 1B). However, as other parts of Mortier Bay remain unsurveyed, undetected populations of *C. intestinalis* may exist elsewhere.

Atlantic Canadian waters are considered at high risk for establishment of *C. intestinalis* (Therriault and Herborg 2008), and this species poses a serious threat to the shellfish aquaculture industry (Ramsay et al. 2008). Suspended shellfish culture sites inadvertently provide abundant available substrate and refuge from predators with little interspecific competition (Vercaemer et al. 2011), thus increasing survival of non-native tunicates (Carver et al. 2003). In mussel aquaculture, tunicates, such as *C. intestinalis*, may compete with mussels for food and result in decreased mussel size and condition and increased mortality rates (Daigle and Herbinger 2009). In addition, the associated costs of tunicate removal results in lost revenue for mussel growers (Daigle and Herbinger 2009). In Atlantic Canada, *C. intestinalis* was first reported as a significant biofouling problem for the mussel aquaculture industry in Nova Scotia in 1997 (Cayer et al. 1999). It was later detected in Prince Edward Island in 2004 and by 2007 had colonized its east coast in epidemic proportions, and replaced the clubbed tunicate, *Styela clava* Herdman, 1881, as the most problematic invasive species to the local mussel aquaculture industry (Ramsay et al. 2008).

Unlike their neighbours in other parts of Atlantic Canada, the Newfoundland mussel aquaculture industry has not had to deal with the problem of invasive ascidians to date. Also, unlike their Atlantic Canadian counterparts, the Newfoundland mussel aquaculture industry must contend with higher transportation costs associated with getting their product from the island to mainland markets. Thus any extra costs associated with the removal of invasive ascidians during harvesting and processing would be especially problematic to the industry (D. Green, Newfoundland Aquaculture Industry Association, St. John's, Newfoundland and Labrador, pers. com.). There were 53 mussel aquaculture sites licensed in Newfoundland in 2012, including 5 sites located in Placentia Bay (DFA 2012) within 90 – 100 km of Little Bay. Given the limited dispersal range of *C. intestinalis* (Kanary et al. 2011), it would take more than 7 years to reach these sites through natural dispersal alone. However, due to the high volume of commercial and recreational vessel traffic moving within Placentia Bay, this time may be significantly shortened unless utmost vigilance is taken on behalf of users of this bay.

A variety of environmental factors influence growth, reproduction and survival in ascidians. Temperature and salinity are factors that influence distribution, persistence, and spread of *C. intestinalis* (Vercaemer et al. 2011). In general, *C. intestinalis* has wide environmental tolerances for temperature (-1 to 30ºC) and salinity (12 to 40) (Carver et al. 2006). However, these wide ranges likely overestimate tolerances of specific populations (Vercaemer et al. 2011). For example, *C. intestinalis* from southern Nova Scotia do not tolerate long term exposure to salinity ≤ 20 and temperature ≥ 24ºC (Vercaemer et al. 2011). *Ciona intestinalis* is essentially an annual species, living 12 – 18 months (Millar 1952). Individuals that settle in May and June may spawn by August of the same year, and mature individuals may spawn continually at water temperatures > 8ºC (Carver et al. 2003). In sheltered waters, two generations per year may be produced (Dybern 1965; Vercaemer et al. 2011). As with spawning, recruitment occurs when mean water temperature exceeded 8ºC (Howes et al. 2007; Ramsay et al. 2009). In Prince Edward Island, *C. intestinalis* recruitment occurs mid-June to late November (Ramsay et al. 2009) while in Nova Scotia it occurs late June to mid-November (Howes et al. 2007). Howes et al. (2007) also found that *C. intestinalis* recruitment in southern Nova Scotia is highest in sheltered locations at depths of 4.5 – 8.5 m. Environmental conditions in southern Nova Scotia and eastern PEI are especially suitable for invasive ascidians (Carver et al. 2003; Locke et al. 2007; Sephton et al. 2011), but it has yet to be determined whether environmental conditions within coastal Newfoundland waters will allow *C. intestinalis* to attain invasive status. Temperature and salinity data collected from 2009 – 2012 from three nearshore locations at the northern end of Placentia Bay (Arnold's Cove: 47°45'30.65"N, 53°59'16.97"W – Ma 2012, Lowen et al. in prep.; North Harbour: 47°50'46.24"N, 54°05'49.12"W – McKenzie, unpublished data; Come by Chance Point: 47°47'22.80"N, 54°02'56.40"W – SmartBay 2013) and one exposed location near the mouth of the bay (46°58'52.80"N, 54°41'16.80"W – SmartBay 2013) indicated suitable conditions for *C. intestinalis*. Salinity in Placentia Bay ranged from 30.0 – 32.0 from 2009 – 2011, but peaked at 35.8 in 2012 at the outermost exposed site. Water temperature, collected at 0.5 m depth, annually ranged from -1 to 21.3ºC throughout the bay. In all years, average monthly water temperatures suitable for *C. intestinalis* spawning and recruitment (> 8ºC) occurred from July to October. However, suitable water temperatures extended from June to October in 2009 and June to November in 2012. Warmer summers with little freshwater input, as in 2012, may extend the spawning and recruitment periods of *C. intestinalis* and further its spread within Placentia Bay. Sheltered areas within Placentia Bay, especially those adjacent to Mortier Bay, or such sites along vessel traffic routes from Mortier Bay, may be at risk of colonization by *C. intestinalis.*

Future effects of global warming and increases in vessel transportation will only increase the likelihood of spread of invasive species (Myerson and Mooney 2007; Stachowicz et al. 2002). Until current regulations regarding domestic discharge of ballast water and movement of hull-fouled vessels between ports change, the spread of non-indigenous species will continue to escalate (Lambert and Lambert 1998). It is therefore crucial to have a system in place focused on early detection, prevention, and public awareness to recognize when and where introductions occur.

Based on potential impacts posed by *C. intestinalis* in Newfoundland waters, the following measures are recommended. Immediate and annual AIS surveys are recommended throughout Mortier Bay and adjacent harbours to detect other colonization sites and monitor changes in current distribution and abundance of this and other non-native species. *Ciona intestinalis* and other non-native species including a green macroalgae, *Codium fragile* (Suringar) Hariot, 1889, are present in nearby St. Pierre and Miquelon (F. Urtizberea, Department of Agriculture and Forestry, St. Pierre and Miquelon, France, pers. com.; Figure 1B). As there is regular ferry service between St. Pierre

Harbour and Fortune, Newfoundland, AIS surveys are recommended to determine if these species have been introduced to Fortune Bay. Early detection is crucial to mitigation measures since invasive ascidians, once established, provide a local source of larvae from which they may perpetuate the invasion regionally (Lambert 2002, 2007). The public should be made aware of potential economic and ecological impacts in order to minimize further anthropogenic transmission of *C. intestinalis*. Mitigation measures should be prepared immediately to alleviate potential problems caused by this species. The aquaculture industry, in consultation with government departments, should also prepare a contingency plan to prepare for further spread of *C. intestinalis*. Continued use of RVS with collaborative partners is also recommended. Due to economic and logistic constraints, researchers studying aquatic invasive species cannot possibly survey all locations regularly. Qualitative survey methods such as RVS provide rapid evaluations of the presence of non-native species in an area and baseline information that may be improved upon by follow-up AIS surveys (Campbell et al. 2007). Therefore, RVS performed by collaborative partners while conducting their own fieldwork may increase chances of early detection. This in turn may enhance implementation of mitigation tools against population growth and further spread of these species.

Projects like the WB Project represent a useful means to conduct RVS for invasive species to supplement the DFO/MUN AIS survey program. Invasive ascidians thrive on wharf structures, floats and boat hulls in sheltered harbours (Lambert 2007). Increased vessel transportation in recent years has led to increases in supporting infrastructure providing more available space upon which opportunistic ascidian larvae may settle (Lambert and Lambert 1998) with little competition from native species (Lambert 2007). The WB Project focuses on such sites with broad scale coverage of insular Newfoundland. Its continuation for another four years, and the opportunity for further RVS, increases the chances of early detection and effectiveness of monitoring aquatic invasive species in Newfoundland.

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Supplementary material

The following supplementary material is available for this article.

Appendix 1. *Ciona intestinalis* distribution records in eastern Canada.

Appendix 1. *Ciona intestinalis* distribution records in eastern Canada from the National Aquatic Invasive Species Database (DFO 2012), St. Pierre and Miquelon, France (F. Urtizberea pers. com.), and this study.

Location Name	Coordinates		Database ID Code	Country	Province	Data Manager
	Latitude	Longitude				
Back Bay	45.056	-66.864	18761	Canada	NB	Legresley
Beaver Harbour	45.069	-66.740	18762	Canada	NB	Legresley
Brandy Cove	45.082	-67.085	18763	Canada	NB	Legresley
Dipper Harbour	45.094	-66.417	18778	Canada	NB	Legresley
Fairhaven	44.964	-67.008	18765	Canada	NB	Legresley
Friars Rd	44.904	-66.969	18774	Canada	NB	Legresley
Head Harbour	44.945	-66.920	18767	Canada	NB	Legresley
Indian Island	44.935	-66.969	18768	Canada	NB	Legresley
Ingalls Head	44.661	-66.757	18769	Canada	NB	Legresley
Leonardville	44.972	-66.953	18770	Canada	NB	Legresley
Letete	45.051	-66.896	18771	Canada	NB	Legresley
Lord's Cove	45.006	-66.947	18764	Canada	NB	Legresley
Man of War wharf	44.916	-66.942	18772	Canada	NB	Legresley
North Head	44.763	-66.749	18773	Canada	NB	Legresley
St Andrews Harbour	45.068	-67.053	18775	Canada	NB	Legresley
Wallace Cove	45.047	-66.805	18776	Canada	NB	Legresley
Burin	47.031	-55.172	19357	Canada	NL	Wells
Little Bay	47.163	-55.112	19358	Canada	NL	Wells
Marystown	47.167	-55.015	19356	Canada	NL	Wells
Alder Point	46.309	-60.284	1594	Canada	NS	Sephton
Arichat	45.511	-61.018	1270	Canada	NS	Sephton
Armdale	44.636	-63.613	19025	Canada	NS	Sephton
Auld's Cove	45.646	-61.434	8222	Canada	NS	Bernier
Bedford Basin	44.745	-63.664	1255	Canada	NS	Sephton
Blanche	43.548	-65.432	18134	Canada	NS	Sephton
Camp Cove	43.724	-65.841	19000	Canada	NS	Sephton
Cape Canso	45.335	-60.986	19001	Canada	NS	Sephton
Chester	44.538	-64.238	19002	Canada	NS	Sephton
Chester Basin	44.559	-64.306	18084	Canada	NS	Sephton
Cheticamp	46.627	-61.016	1595	Canada	NS	Sephton
Clark's Harbour	43.445	-65.635	19003	Canada	NS	Sephton
Corkum's Island	44.361	-64.334	1259	Canada	NS	Sephton
Country Harbour	45.232	-61.759	19022	Canada	NS	Sephton
Daniel's Island	44.494	-64.307	4282	Canada	NS	Sephton
Dartmouth	44.682	-63.612	19007	Canada	NS	Sephton
D'Escousse	45.589	-60.962	19004	Canada	NS	Sephton
Digby	44.630	-65.752	19005	Canada	NS	Sephton
Dingwall	46.903	-60.460	19006	Canada	NS	Sephton
Eddy Point	45.521	-61.264	1762	Canada	NS	Sephton
Eel Lake	43.827	-65.908	18855	Canada	NS	Sephton
Gabarus	46.820	-60.155	1593	Canada	NS	Sephton
Gunning Cove	43.681	-65.340	1753	Canada	NS	Sephton
Havre Boucher	45.689	-61.985	18137	Canada	NS	Sephton
Hebb Point	44.391	-64.273	4296	Canada	NS	Sephton
Hubbards	44.638	-64.059	1742	Canada	NS	Sephton
Indian Point	44.456	-64.307	19008	Canada	NS	Sephton
Ingomar	43.563	-65.362	17967	Canada	NS	Sephton
Inverness	46.230	-61.317	1596	Canada	NS	Sephton
Leg Point	44.473	-64.320	4284	Canada	NS	Sephton
Little Harbour, St. Peter's Bay	45.583	-60.741	19021	Canada	NS	Sephton
Lockeport	43.700	-65.107	19009	Canada	NS	Sephton
Louisbourg	45.918	-59.989	19010	Canada	NS	Sephton
Lower Sandy Point	43.680	-65.301	1751	Canada	NS	Sephton
Lower South Cove	44.342	-64.319	18100	Canada	NS	Sephton
Lunenburg	44.375	-64.307	19011	Canada	NS	Sephton
Lunenburg	44.411	-64.322	1773	Canada	NS	Sephton

Appendix 1 (continued).

Location Name	Coordinates		Database ID Code	Country	Province	Data Manager
	Latitude	Longitude				
Lunenburg, Lower South Cove	44.323	-64.306	18099	Canada	NS	Sephton
Mader's Cove	44.436	-64.352	4300	Canada	NS	Sephton
Mahone Bay	44.448	-64.374	1744	Canada	NS	Sephton
Marriott's Cove	44.549	-64.265	4279	Canada	NS	Sephton
Martin Cove	44.399	-64.311	4289	Canada	NS	Sephton
Martin's River	44.483	-64.327	18087	Canada	NS	Sephton
Meteghan	44.194	-66.167	19012	Canada	NS	Sephton
Moose Harbour	44.019	-64.664	1747	Canada	NS	Sephton
North Sydney	46.140	-60.178	18136	Canada	NS	Sephton
Oak Island	44.521	-64.305	4281	Canada	NS	Sephton
Port Bickerton East	45.105	-61.723	1269	Canada	NS	Sephton
Port La Tour	43.498	-65.470	1752	Canada	NS	Sephton
Port Mouton	43.919	-64.843	1748	Canada	NS	Sephton
Prince's Inlet	44.412	-64.320	18096	Canada	NS	Sephton
Purcell's Cove	44.621	-63.580	17978	Canada	NS	Sephton
Queensport	45.345	-61.256	1271	Canada	NS	Sephton
Robin's Cove	45.506	-61.100	19024	Canada	NS	Sephton
Sambro	44.479	-63.600	19015	Canada	NS	Sephton
Shelburne	43.758	-65.322	19016	Canada	NS	Sephton
St. Peter's	45.661	-60.874	19017	Canada	NS	Sephton
Stonehurst W	44.373	-64.223	4248	Canada	NS	Sephton
Sydney	46.140	-60.168	19018	Canada	NS	Sephton
Upper Port la Tour	43.506	-65.362	1261	Canada	NS	Sephton
Venus Cove	45.615	-61.390	19019	Canada	NS	Sephton
Wedgeport	43.714	-65.969	17970	Canada	NS	Sephton
Whitehead	45.233	-61.159	19023	Canada	NS	Sephton
Yarmouth Bar	43.816	-66.148	19020	Canada	NS	Sephton
Boughton River	46.256	-62.443	15548	Canada	PEI	Bernier
Brudenell	46.173	-62.548	17905	Canada	PEI	Watson
Brudenell Bay	46.181	-62.551	1024	Canada	PEI	McKindsey
Brudenell River	46.186	-62.557	1133	Canada	PEI	Smith
Brudenell/Montague	46.177	-62.593	15456	Canada	PEI	Bernier
Cardigan Bay	46.135	-62.465	15398	Canada	PEI	Bernier
Cardigan River	46.201	-62.512	15452	Canada	PEI	Bernier
Charlottetown Harbour	46.230	-63.122	15546	Canada	PEI	Bernier
Gaspereaux	46.096	-62.455	15444	Canada	PEI	Bernier
Georgetown	46.167	-62.535	15373	Canada	PEI	Bernier
Montague	46.167	-62.641	15419	Canada	PEI	Bernier
Montague River	46.167	-62.527	15552	Canada	PEI	Bernier
Montague Bay	46.172	-62.542	1035	Canada	PEI	McKindsey
Murray	46.020	-62.600	15562	Canada	PEI	Bernier
Murray Harbour	46.028	-62.558	15438	Canada	PEI	Bernier
Murray River	46.021	-62.587	15557	Canada	PEI	Bernier
Nine Mile Creek	46.150	-63.180	15544	Canada	PEI	Bernier
Souris	46.347	-62.249	15561	Canada	PEI	Bernier
St. Mary's Bay	46.125	-62.505	15554	Canada	PEI	Bernier
West River	46.205	-63.160	15543	Canada	PEI	Bernier
BHA	47.226	-61.873	18680	Canada	QC	Simard
Cap aux Meules	47.377	-61.852	2315	Canada	QC	Simard
St. Pierre	46.776	-56.175	N/A	France	SPM	Urtizberea