Biol Invasions (2012) 14:1843–1850
DOI 10.1007/s10530-012-0194-0

ORIGINAL PAPER

Invasion risk posed by macroinvertebrates transported in ships' ballast tanks

Elizabeta Briski · Sara Ghabooli · Sarah A. Bailey · Hugh J. MacIsaac

Received: 23 September 2011 / Accepted: 11 February 2012 / Published online: 25 February 2012

Abstract Invasions by non-indigenous macroinvertebrates often cause ecological and economic problems, and commercial ships have been implicated as a principal mechanism for their dispersal. We explored the presence and species diversity of adult macroinvertebrates transported by transoceanic and coastal vessels arriving to ports on the Atlantic coast of Canada. We sampled 67 ballast tanks from 62 ships operating along discrete geographic pathways and tested whether mid-ocean exchange or voyage length affects the probability for translocation of macroinvertebrates. Additionally, we assessed the relationship between macroinvertebrate presence and the amount of sediment in ballast tanks. We document the presence of highly invasive European green crab (*Carcinus maenas*), mud crab (*Rhithropanopeus harrisii*), common periwinkle (*Littorina littorea*), soft shell clam (*Mya arenaria*) and blue mussel (*Mytilus galloprovincialis*) in ballast tanks of surveyed ships. Mid-ocean exchange did not affect macroinvertebrate occurrence, suggesting that current ballast water management regulations are ineffective for this taxonomic group. Viable individuals were recorded in vessels undertaking shorter voyages (average and maximum of 4.5 and 15 days, respectively) and presence was not related to the amount of sediment in tanks. While presence and densities of macroinvertebrates were low, invasion risk may nonetheless be significant during reproductive seasons owing to high fecundity of some taxa. The highest risk may be posed by decapods since gravid females may carry thousands to several million eggs per clutch, and after several weeks of brooding, two or more subsequent clutches may be fertilized by retained sperm from an earlier mating.

Keywords Ballast tanks · Crab · High fecundity · Mollusc · Non-indigenous species · Propagule pressure

E. Briski (✉) · S. A. Bailey
Great Lakes Laboratory for Fisheries and Aquatic Sciences, Fisheries and Oceans Canada, Burlington, ON L7R 4A6, Canada
e-mail: elizabeta.briski@dfo-mpo.gc.ca; elzabriski@yahoo.com

S. Ghabooli · H. J. MacIsaac
Great Lakes Institute for Environmental Research, University of Windsor, Windsor, ON N9B 3P4, Canada

Introduction

Human activities unintentionally translocate many aquatic species of which only a small percentage establish populations in new environments (e.g. Kolar and Lodge 2001; Colautti and MacIsaac 2004; Karatayev et al. 2009). In a recent review, Karatayev et al. (2009) showed that freshwater macroinvertebrate invaders are not a random selection of species, but rather are over-represented by molluscs and crustaceans.

Introduced molluscs can alter benthic community dynamics by out-competing native species (Branch and Steffani 2004; Ward and Ricciardi 2007), while crustaceans may adversely affect bivalves, molluscs and other crustaceans through predation, competition, and burrowing activities (e.g. Grosholz and Ruiz 1996). Furthermore, molluscs and crustaceans can cause major economic losses (Pimentel et al. 2005; Colautti et al. 2006a); for example, losses in Canada due to zebra (*Dreissena polymorpha*) and quagga mussels (*Dreissena bugensis rostriformis*) alone are estimated at more than $8 million CDN per year (Colautti et al. 2006a). Several molluscs and crustaceans have been listed in the Global Invasive Species Database of "100 of the World's Worst Invasive Alien Species" (ISSG 2011).

Macroinvertebrates may be transported by numerous vectors (Ricciardi 2006; Molnar et al. 2008), however ships have been implicated as a leading mechanism for aquatic non-indigenous species (NIS) introductions worldwide (Ricciardi 2006; Molnar et al. 2008; Hulme 2009). A diverse array of species are transported in ballast tanks or attached to external surfaces of ships' hulls (Humphrey 2008; Klein et al. 2010; Briski et al. 2010; Sylvester et al. 2011), and numerous studies report presence of unidentified bivalve and/or decapod larvae in ballast water. Thus, shipping may be a potent vector for dispersal of species including European green crab (*Carcinus maenas*), mud crab (*Rhithropanopeus harrisii*), and soft shell clam (*Mya arenaria*) (Carlton 1985; Duggan et al. 2006; Humphrey 2008; Darling et al. 2008; Simard et al. 2011). To our knowledge, no study has yet confirmed records of these species from ballast tanks, either as larvae or benthic adults.

Ballast water can contain significant amounts of suspended sediment that later settle to the tank bottom (Carlton 1985; Gollasch and Leppäkoski 1999) and provide suitable habitat for benthic organisms (Briski et al. 2010, 2011a). Invertebrate dormant stages are not influenced by voyage length, but survival of active invertebrates and diatoms is reduced by longer voyages (Reid et al. 2007; Humphrey 2008; Klein et al. 2010). Ballast water management regulations enacted by Canada, including requirements for mid-ocean exchange (MOE) and salt-water flushing are primarily aimed at reducing invasion risk of entrained microplankton and have not been evaluated heretofore for their efficacy against benthic macroinvertebrates (Government of Canada 2006).

Females of some aquatic macroinvertebrates—including molluscs and decapods—are capable of prolific egg production, with output ranging from thousands to millions of eggs per reproductive event (Stickney 1963; Hughes and Roberts 1980; Creaser and Clifford 1982; Morgan et al. 1983; Shields and Okazaki 1991; Grosholz and Ruiz 2002; Samuel and Soundarapandian 2010). Invasion theory identifies a close linkage between the number of propagules released in a new location and the success of an invader (Colautti et al. 2006b; Lockwood et al. 2009), thus the introduction of one pair of spawning molluscs or one gravid female decapod could pose a surprisingly high invasion risk. Further, decapod females can oviposit several masses (i.e. clutches) of fertilized eggs over several months after a single mating (Morgan et al. 1983; Shields and Okazaki 1991), complicating measures of propagule pressure and invasion risk.

Here, we report on the presence and species diversity of macroinvertebrates sampled from transoceanic and coastal ships arriving to the Atlantic coast of Canada. We sampled 67 ballast tanks of ships on particular geographic pathways to test three hypotheses: (1) invasion risk posed by macroinvertebrates from ships performing MOE and those exempt from MOE is equal; (2) the length of voyage does not affect macroinvertebrate survival inside tanks; and (3) the amount of sediment in tanks does not affect survival of macroinvertebrates in tanks.

Methods

As part of a larger study (see Briski et al. 2011a), we sampled ships following three distinct geographic pathways to Canadian ports: transoceanic ships performing MOE (TOE), coastal ships performing MOE (CE), and coastal ships without MOE (CNE) (Fig. 1). TOE ships were defined as ships arriving from any continent except North America. CE ships arrived from ports in the USA south of Cape Cod, Massachusetts. CNE ships arrived from American or Canadian ports north of Cape Cod. Ships were sampled opportunistically on arrival to Sept-Îles, Baie-Comeau and Port-Cartier in Quebec, Saint John in New Brunswick, and Halifax, Hantsport, Canso, Port Hawkesbury, Liverpool, Point Tupper and Sheet Harbour in Nova Scotia. In total, we collected samples

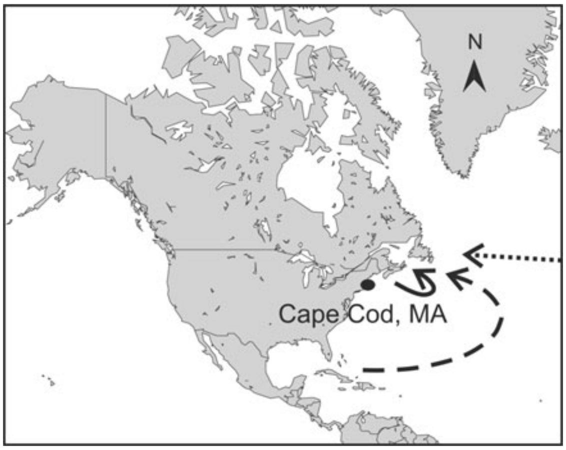

Fig. 1 Schematic representation of pathways utilized by ships arriving to the Atlantic region of Canada. *Dotted line* represents transoceanic ships with mid-ocean exchange (TOE), *dashed line* represents coastal ships with mid-ocean exchange (CE) and *solid line* represents coastal ships exempt from mid-ocean exchange (CNE). Vessels arriving from ports north of Cape Cod, Massachusetts (MA) are exempt from mid-ocean exchange requirements, if destined for specific Canadian ports on the Atlantic coast (Government of Canada 2006)

from 25 TOE, 21 CE and 21 CNE ships. Data contained in ballast water reporting forms, including previous dates and locations of ballast uptake and discharge, and last port-of-call, were used to determine ships' pathways and voyage length.

Tanks were entered immediately after deballasting occurred at Canadian ports, and all compartments of the tank were visually inspected for presence of larger macroinvertebrates such as cnidarians, decapods, gastropods, and bivalves, which were collected when found. Additionally, approximately 6 kg of sediment was collected from multiple, randomly-selected areas inside the ballast tank to check for smaller, buried taxa. As sediment tended to accumulate peripherally in tanks with only a thin layer spread across the tank bottom (mean and maximum depth of 3 mm and 25 cm, respectively), we were usually able to collect sediment from a range of depths. Sediment depth and percent cover inside ballast tanks were recorded and architectural diagrams of ships' tanks reviewed to estimate the amount of sediment carried per tank. Upon return to the laboratory, sediments were processed using a 500 μm sieve and sieve contents examined under a dissecting microscope. Macroinvertebrates were isolated and stored in 95% ethanol until later identification. Taxa smaller than 500 μm, mostly dormant stages of zooplankton, are reported in Briski et al. (2011a). Identification of animals was conducted using molecular methods as well as traditional morphological taxonomy; taxonomic experts were consulted when morphological identifications were uncertain. In addition, we measured carapace width (for crabs) or the length of the widest axis (other macroinvertebrates) for each individual.

One small piece of tissue was dissected from every individual and used for molecular identification. DNA was extracted following Elphinstone et al. (2003). Fragments of the mitochondrial genes COI and 16S were amplified using the universal COI primers LCO1490 and HCO2190 (Folmer et al. 1994), and 16S primers S1 and S2 (Palumbi 1996). PCR reactions and sequencing protocol followed Briski et al. (2011b). Recovered DNA sequences were blasted against those in the GenBank database (http://blast.ncbi.nlm.nih.gov/Blast.cgi) using the nucleotide blast (default parameters). In addition, COI sequences were compared to those in the Barcode of Life Database (http://www.barcodinglife.org), using the identification engine BOLD-IDS, with the option 'All Barcode Records on BOLD'.

Statistical analyses

We determined whether macroinvertebrate presence in ballast tanks was influenced by MOE using Pearson Chi-square analysis (SPSS version 11.5.0; SPSS). Separate logistic regression analyses were used to determine if the occurrence of macroinvertebrates was related to the length of the ships' voyage or the amount of sediment contained in ballast tanks (SPSS version 11.5.0; SPSS). All three ship pathways (i.e. TOE, CE and CNE) were pooled for the above mentioned logistic analyses, as Pearson Chi-square analysis showed no effect of MOE on the occurrence of macroinvertebrates (see "Results" section).

Results

Sixty-seven ballast tanks (62 ships) were sampled between May 2007 and August 2009. The average voyage length and amount of sediment in tanks of sampled ships were 8.94 days and 1.44 m^3, respectively (Table 1). Voyage length differed among ship pathways, being 14.08, 7.28, and 4.47 days, for TOE,

Table 1 Voyage length and amount of sediment per tank for all ships sampled in the study and for ships containing macroinvertebrates

	All ships						Ships containing macroinvertebrates					
	Number of samples	Mean	SE	Min	Max	Range	Number of samples	Mean	SE	Min	Max	Range
Voyage length (day)												
Total	67	8.94	0.83	1	44	43	7	4.57	1.79	2	15	13
TOE	25	14.08	1.38	6	44	38	1	15.00	N/A	N/A	N/A	N/A
CE	21	7.28	1.09	2	23	21	2	4.50	0.50	4	5	1
CNE	21	4.47	0.85	1	14	13	4	2.00	0.00	2	2	0
Sediment (m^3)												
Total	61	1.44	0.39	0.00	16.80	16.79	7	0.52	0.44	0.01	3.19	3.18
TOE	20	2.05	0.64	0.01	11.25	11.24	1	0.01	N/A	N/A	N/A	N/A
CE	20	2.25	0.95	0.00	16.80	16.80	2	1.59	1.58	0.01	3.19	3.18
CNE	21	0.08	0.01	0.00	0.26	0.26	4	0.12	0.03	0.04	0.18	0.14

Statistics are shown for all pathways (total), transoceanic ships which conducted mid-ocean exchange (TOE), coastal ships which conducted mid-ocean exchange (CE), and coastal ships exempt from mid-ocean exchange (CNE)

CE and CNE ships, respectively (Table 1). Macroinvertebrates were found in seven tanks (10.4%) with an average density of 2.8 individuals per tank when present, and no more than 3 individuals per species (Table 2). We were unable to detect any pattern with respect to where macroinvertebrates were found in tanks. The average voyage length and amount of sediment in tanks of ships containing macroinvertebrates were 4.57 days and 0.52 m^3, respectively (Table 1). Macroinvertebrates were found in ships from all three pathways (Tables 1, 2), but more than 50% of records were from tanks of CNE ships

Table 2 Macroinvertebrate taxa (with abundance and size) sampled from ballast tanks of ships arriving to the Atlantic coast of Canada, by vessel

Ship ID	Ship pathway	Voyage length (days)	Sediment per tank (m^3)	Taxon	Number per tank	Size (mm)
56	CE	5	0.0078	*Rhithropanopeus harrisii*	1	20
				Platynereis sp.	3	36, 42, 47
				Parahyotissa numisma*	2	21
				Mya arenaria	1	11
59	CNE	2	0.0405	***Carcinus maenas****	1	36
				Littorina littorea*	2	22
65	CNE	2	0.081	Actinaria, unidentified	1	21
				Neanthes virens	2	78
				Parahyotissa numisma*	1	19
				Mytilus galloprovincialis*	1	22
75	CE	4	3.186	*Platynereis* sp.	2	36, 41
77	TOE	15	0.0157	*Mya arenaria*	1	10
78	CNE	2	0.18	*Mya arenaria*	1	12
80	CNE	2	0.18	***Carcinus maenas****	1	39
				Mya arenaria	1	11

Ship particulars (pathway, voyage length and sediment amount) are included where *CE* coastal ships which conducted mid-ocean exchange *CNE* coastal ships exempt from mid-ocean exchange and *TOE* transoceanic ships which conducted mid-ocean exchange. Non-indigenous species are shown in bold; those able to tolerate ambient salinity in the recipient port are indicated (*)

(Table 2). Occurrence of macroinvertebrates in ballast tanks was not affected by MOE ($\chi^2 = 2.31$, $P = 0.12$), though logistic regression analysis revealed that voyage length was an important factor ($\chi^2 = 5.15$, $P = 0.02$). Even though the amount of sediment in tanks containing macroinvertebrates was lower than that where macroinvertebrates were not recorded (0.52 vs. 1.44 m^3, respectively; Table 1), the relationship between amount of sediment and presence in tanks was not significant ($\chi^2 = 0.72$, $P = 0.28$).

We identified nine taxa belonging to five taxonomic groups (Table 2). One crab (European green crab *C. maenas*), one gastropod (common periwinkle *Littorina littorea*) and two bivalves (*Parahyotissa numisma* and *Mytilus galloprovincialis*) recorded are considered non-native to the Atlantic coast of Canada (Table 2). While *P. numisma* is considered a tropical/subtropical species not likely to survive in Atlantic Canada, and *C. maenas* is already established in the region, *L. littorea* and *M. galloprovincialis* have not yet been reported from the region. A gravid female mud crab (*R. harrisii*) was found in a coastal ship that had conducted MOE (Fig. 2; Table 2). This species is native to Atlantic Canada (A Locke, Fisheries and Oceans Canada, personal communication), but is an established NIS in many locations in Europe and western North America. The native bivalve *M. arenaria* was the only species recorded from the TOE pathway, from a ship with voyage length of 15 days (Table 2).

Discussion

To our knowledge, this study provides the first evidence for transport of highly invasive macroinvertebrates including European green crab (*C. maenas*), mud crab (*R. harrisii*), common periwinkle (*L. littorea*), soft shell clam (*M. arenaria*) and blue mussel (*M. galloprovincialis*) in ships' ballast tanks (see also Briski et al. 2011a). Furthermore, ships' ballast appears to be an active vector for transport of adult macroinvertebrates to Canadian waters despite requirements for ballast water management by MOE. While *C. maenas* is already established in some areas of Atlantic Canada, its discovery in ships' ballast tanks poses concern for further inter-regional spread and possible introduction of individuals with novel

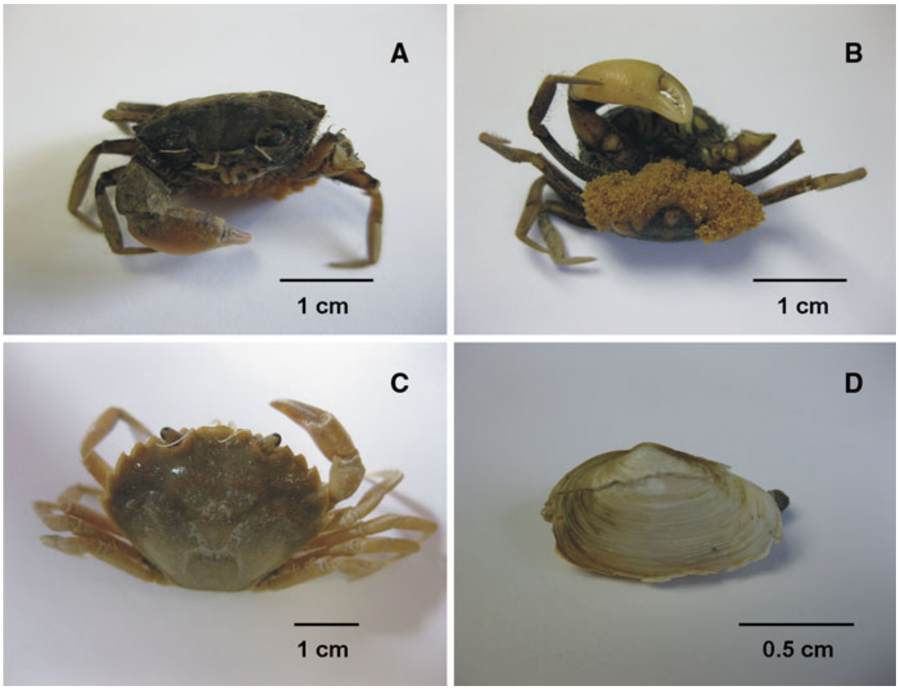

Fig. 2 Images of mud crab *Rhithropanopeus harrisii* (**a**), female mud crab carrying eggs (**b**), European green crab *Carcinus maenas* (**c**) and soft-shell clam *Mya arenaria* (**d**) found in ballast tanks of ships

genotypes that may be better adapted to the local environment, potentially enhancing further spread and/or negative impacts (Roman 2006; Simberloff 2009; Rossong et al. 2011). In addition to macroinvertebrates themselves, these species could potentially introduce 'fellow traveller' parasites (Torchin et al. 2001) or disease organisms (Payen and Bonami 1979); *R. harrisii*, for example, commonly carry strains of the white spot baculovirus (Payen and Bonami 1979).

The discovery of a live, gravid female *R. harrisii* in a ballast tank is particularly troubling discovery. This species may produce between 1,000 and 4,000 eggs per clutch, and females are able to release fertilized egg clutches up to four separate times following a single mating (Morgan et al. 1983). Thus, the propagule pressure exerted by such a female could approach that reported for juvenile stages previously ($\sim$30 individuals per m^3 of unidentified decapod larvae in ballast water of coastal ships arriving to the Atlantic coast of Canada; Humphrey (2008)). *R. harrisii* is native to Atlantic estuarine waters and larval development may take place under a broad range of environmental conditions, including salinities ranging from 2.5 to 40 ppt (Costlow et al. 1966); the collection of a viable individual after MOE and exposure to marine water above 30 ppt salinity highlights the tremendous salinity tolerance of this species. Reports of adults, zoeae and small juveniles of the mud crab in freshwater reservoirs in Texas (Howells 2001) indicate that the species could pose an invasion risk to the Laurentian Great Lakes if introduced. Several euryhaline species have already established in the Great Lakes, including the amphipod *Gammarus tigrinus*, which is also native to estuaries of the northwest Atlantic Ocean (Ricciardi and MacIsaac 2000; Kelly et al. 2006).

It is not known if adult macroinvertebrates can be entrained into, or discharged from, ballast tanks in a viable condition. It might be possible for smaller individuals to pass through the ballast pipes and pumping system intact, particularly if sea chest screens and strainers are damaged. At any rate, hatched larvae of *R. harrisii* remain free-swimming from 7 to 35 days depending on salinity and temperature (Goncalves et al. 1995), allowing easy ingress to or egress from ballast tanks. Although this study indicates that macroinvertebrates are transported in only 10% of ships and only at low density, invasion risk could be enhanced during reproductive seasons and/or when environmental conditions are conducive to survival inside ballast tanks. Many molluscs and decapods possess r-selected reproductive strategies, which are characterized by production of numerous offspring. These taxa may be best suited to overcoming environmental and demographic stochasticity associated with the invasion process, and rapidly achieving densities amenable to successful colonization in new habitiats (McMahon 2002). The highest risk may be posed by decapods not requiring simultaneous presence of both sexes inside tanks to produce free-swimming larvae (Morgan et al. 1983; Samuel and Soundarapandian 2010). In contrast, bivalves require both sexes for reproduction, and spawning in the often hostile conditions inside ballast tanks may be problematic.

Considering that macroinvertebrates were found only in ships completing shorter voyages (average of 4.5 days), environmental conditions in ballast tanks may be an important determinant of macroinvertebrate survival. Reid et al. (2007) and Klein et al. (2010) measured rapid declines in dissolved oxygen concentration inside ballast tanks to 2 mg/L within five to 7 days, with 90% of initial oxygen content lost within 10 days at temperatures above 20°C. As a result, environmental conditions in ballast tanks of ships on long, transoceanic voyages may inhibit survival. In contrast, environmental conditions may not be limiting in tanks of coastal vessels undertaking short voyages, during which ballasting activities (loading, discharging and/or conducting MOE) may occur every few days. Carver and Mallet (2004) showed that the infusion of nutrients and oxygen into tanks can enhance the total number of plankton species surviving at the end of voyage. While this may indicate macroinvertebrate transfers will be limited mainly to short distance dispersal, we caution that our study was conducted during warmer parts of the year when temperatures were always above 10°C and usually above 20°C. Reid et al. (2007) observed no decline in dissolved oxygen concentration in ballast water during a voyage in December at temperatures ranging between 3 and 5°C, thus the possibility exists for enhanced intercontinental survival during cooler seasons.

Acknowledgments We thank participating shipping companies and crews, the Shipping Federation of Canada, multiple port authorities and our sampling team: A.M. Weise, O. Casas-Monroy, N. Simard, J.-Y. Couture, M. Huot, O. Lacasse, D. Humphrey, C.Owens, S. Ross, C.D.A. van Overdijk and Dr. C. McKindsey. We are grateful to R. Tedla, S. Ross and H. Coker for laboratory support and to Dr. M. Cristescu for making available her laboratory resources. Great thanks to Dr. Paul Valentich-Scott for taxonomic assistance, and to Dr. Andrea Locke and an anonymous reviewer for helpful comments. This research was supported by NSERC's Canadian Aquatic Invasive Species Network, Transport Canada, Fisheries and Oceans Canada, by NSERC Discovery Grants to SAB and HJM, and by a DFO Invasive Species Research Chair to HJM.

References

Branch GM, Steffani CN (2004) Can we predict the effects of alien species? A case-history of the invasion of South Africa by *Mytilus galloprovincialis* (Lamarck). J Exp Mar Biol Ecol 300:189–215

Briski E, Bailey SA, Cristescu ME, MacIsaac HJ (2010) Efficacy of 'saltwater flushing' in protecting the Great Lakes from biological invasions by invertebrate eggs in ships' ballast sediment. Freshw Biol 55:2414–2424

Briski E, Bailey SA, MacIsaac HJ (2011a) Invertebrates and their dormant eggs transported in ballast sediments of ships arriving to the Canadian coasts and the Laurentian Great Lakes. Limnol Oceanogr 56:1929–1939

Briski E, Cristescu ME, Bailey SA, MacIsaac HJ (2011b) Use of DNA barcoding to detect invertebrate invasive species from diapausing eggs. Biol Invas 13:1325–1340

Carlton JT (1985) Transoceanic and interoceanic dispersal of coastal marine organisms: the biology of ballast water. Oceanogr Mar Biol 23:313–371

Carver CE, Mallet AL (2004) Investigating potential ballast water management strategies for ships travelling from Chesapeake Bay to ports in Nova Scotia. Transport Canada. Atlantic Ballast water sub-committee report. URL: http://www.ceaa.gc.ca/B4777C6B-docs/WP-1785-052.pdf

Colautti RI, MacIsaac HJ (2004) A neutral terminology for defining invasive species. Divers Distrib 10:135–141

Colautti RI, Bailey SA, van Overdijk CDA, Amundsen K, MacIsaac HJ (2006a) Characterised and projected costs of nonindigenous species in Canada. Biol Invas 8:45–59

Colautti RI, Grigorovich IA, MacIsaac HJ (2006b) Propagule pressure: a null model for biological invasions. Biol Invas 8:1023–1037

Costlow JD, Bookhout CG, Monroe R (1966) Studies on the larval development of the crab, *Rhithropanopeus harrisii* (Gould). I. The effect of salinity and temperature on larval development. Physiol Zool 39:81–100

Creaser EP, Clifford DA (1982) Life history studies of the sandworm, *Nereis virens* Sars, in the Sheepscot estuary, Maine. Fish B-NOAA 80:735–743

Darling JA, Bagley MJ, Roman J, Tepolt CK, Geller JB (2008) Genetic patterns across multiple introductions of the globally invasive crab genus *Carcinus*. Mol Ecol 17: 4992–5007

Duggan IC, Bailey SA, van Overdijk CDA, MacIsaac HJ (2006) Invasion risk of active and diapausing invertebrates from residual ballast in ships entering Chesapeake Bay. Mar Ecol Prog Ser 324:57–66

Elphinstone MS, Hinten GN, Anderson MJ, Nock CJ (2003) An inexpensive and high-throughput procedure to extract and purify total genomic DNA for population studies. Mol Ecol Notes 3:317–320

Folmer O, Black M, Hoeh W, Lutz R, Vrijenhoek R (1994) DNA primers for amplification of mitochondrial cytochrome *c* oxidase subunit I from diverse metazoan invertebrates. Mol Mar Biol Biotech 3:294–299

Gollasch S, Leppäkoski E (1999) Initial risk assessment of alien species in Nordic coastal waters. Nordic Council of Ministers, Copenhagen

Goncalves F, Ribeiro R, Soares AMVM (1995) Laboratory study of effects of temperature and salinity on survival and larval development of a population of *Rhithropanopeus harrisii* from the Mondego River estuary, Portugal. Mar Biol 121:639–645

Government of Canada (2006) Ballast water control and management regulations. Can Gazette 140(13). URL: http://gazette.gc.ca/archives/p2/2006/2006-06-28/html/sor-dors129-eng.html#a

Grosholz ED, Ruiz GM (1996) Predicting the impact of introduced marine species: Lessons from the multiple invasions of the European green crab *Carcinus maenas*. Biol Conserv 78:59–66

Grosholz ED, Ruiz GM (2002) Management plan for the European green crab. Aquatic nuisance species task force. URL: http://www.anstaskforce.gov/Species%20plans/GreenCrabManagementPlan.pdf. Accessed 7 July 2011

Howells R (2001) Introduced non-native fishes and shellfishes in Texas water: an updated list and discussion. Texas Parks and Wildlife Department, Management Data Series 188. URL: http://www.tpwd.state.tx.us/publications/pwdpubs/media/mds_inland/mds-188.pdf

Hughes RN, Roberts DJ (1980) Reproductive effort of winkles *(Littorina* spp.) with contrasted methods of reproduction. Oecologia 47:130–136

Hulme PE (2009) Trade, transport and trouble: managing invasive species pathways in an era of globalization. J Appl Ecol 46:10–18

Humphrey DB (2008) Characterizing ballast water as a vector for non-indigenous zooplankton transport. MSc Thesis, Univ. of British Columbia, Vancouver, B.C. Canada

Invasive Species Specialist Group (ISSG). (2011) Global invasive species database. IUCN Species Survival Commission. URL: http://www.issg.org. Consulted on 12 Aug 2011

Karatayev AY, Burlakova LE, Padilla DK, Mastitsky SE, Olenin S (2009) Invaders are not a random selection of species. Biol Invas 11:2009–2019

Kelly DW, Muirhead JR, Heath DD, MacIsaac HJ (2006) Contrasting patterns in genetic diversity following multiple invasions of fresh and brackish waters. Mol Ecol 15: 3641–3653

Klein G, MacIntosh K, Kaczmarska I, Ehrman JM (2010) Diatom survivorship in ballast water during trans-Pacific crossings. Biol Invas 12:1031–1044

Kolar CS, Lodge DM (2001) Progress in invasion biology: predicting invaders. Trends Ecol Evol 16:199–204

Lockwood JL, Cassey P, Blackburn T (2009) The more you introduce the more you get: the role of colonization pressure and propagule pressure in invasion ecology. Divers Distrib 15:904–910

McMahon RF (2002) Evolutionary and physiological adaptations of aquatic invasive animals: r selection versus resistance. Can J Fish Aquat Sci 59:1235–1244

Molnar JL, Gamboa RL, Revenga C, Spalding MD (2008) Assessing the global threat of invasive species to marine biodiversity. Front Ecol Environ 6:485–492

Morgan SG, Goy JW, Costlow JD (1983) Multiple ovipositions from single matings in the mud crab *Rhithropanopeus harrisii*. J Crustacean Biol 3:542–547

Palumbi S (1996) Nucleic acids II: the polymerase chain reaction. In: Hillis D, Mable B, Moritz C (eds) Molecular systematics. Sinauer, Sunderland, pp 205–247

Payen GG, Bonami J-R (1979) Mise en evidence de particules d'allure virale associees aux noyaux des cellules mesodermiques de la zone germinative testiculaire du crabe *Rhithropanopeus harrisii* (Gould) (Brachyoure, Xanthide). Rev Trav Inst Pech Marit 43:361–365

Pimentel D, Zuniga R, Morrison D (2005) Update on the environmental and economic costs associated with alien-invasive species in the United States. Ecol Econ 52:273–288

Reid DF, Johengen T, MacIsaac HJ, Dobbs FC, Doblin M, Drake L, Ruiz GM, Jenkins PT, Santagata S, van Overdijk CDA, Gray D, Ellis S, Hong Y, Tang Y, Thomson F, Heinemann S, Rondon S (2007) A final report for the project "Identifying, verifying, and establishing options for best management practices for NOBOB vessels". National Oceanic and Atmospheric Administration, Great Lakes Environmental Research Laboratory, and University of Michigan Cooperative Institute for Limnology and Ecosystems Research, Ann Arbor. URL: http://www.glerl.noaa.gov/res/Task_rpts/2004/aisreid04-1.html

Ricciardi A (2006) Patterns of invasion in the Laurentian Great Lakes in relation to changes in vector activity. Divers Distrib 12:425–433

Ricciardi A, MacIsaac HJ (2000) Recent mass invasions of the North American Great Lakes by Ponto-Caspian species. Trends Ecol Evol 15:62–65

Roman J (2006) Diluting the founder effect: cryptic invasions expand a marine invader's range. Proc R Soc B 273: 2453–2459

Rossong MA, Quijón PA, Snelgrove PVR, Barrett TJ, McKenzie CH, Locke A (2011) Regional differences in foraging behaviour of invasive green crab (*Carcinus maenas*) populations in Atlantic Canada. Biol Invas. doi: 10.1007/s10530-011-0107-7

Samuel NJ, Soundarapandian P (2010) Embryology of commercially important portunid crab *Scylla serrata* (Forskal). Curr Res J Biol Sci 2:38–41

Shields JD, Okazaki RK (1991) Fecundity and the reproductive potential of the yellow rock crab *Cancer Anthonyi*. Fish B-NOAA 89:299–305

Simard N, Plourde S, Gilbert M, Gollasch S (2011) Net efficacy of open ocean ballast water exchange on plankton communities. J Plankton Res 33:1378–1395

Simberloff D (2009) The role of propagule pressure in biological invasions. Annu Rev Ecol Evol Syst 40:81–102

Stickney AP (1963) Histology of the reproductive system of the soft-shell clam (*Mya arenaria*). Biol Bull 125:344–351

Sylvester F, Kalaci O, Leung B, Lacoursière-Roussel A, Murray CC, Choi FM, Bravo MA, Therriault TW, MacIsaac HJ (2011) Hull fouling as an invasion vector: can simple models explain a complex problem? J Appl Ecol 48: 415–423

Torchin ME, Lafferty KD, Kuris AM (2001) Release from parasites as natural enemies: increased performance of a globally introduced marine crab. Biol Invas 3:333–345

Ward JM, Ricciardi A (2007) Impacts of *Dreissena* invasions on benthic macroinvertebrate communities: a meta-analysis. Divers Distrib 13:155–165