



Université du Québec
à Rimouski

**ÉVALUATION DU RISQUE POTENTIEL D'INTRODUCTION D'ESPÈCES NON-
INDIGÈNES DE MÉSOZOOPLANCTON SUITE AU DÉVERSEMENT DES EAUX
DE BALLAST D'UN NAVIRE DOMESTIQUE DANS L'ARCTIQUE CANADIEN**

Mémoire présenté
dans le cadre du programme de maîtrise en océanographie
en vue de l'obtention du grade de maître ès sciences

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« Ohne Kampf, kein Sieg ».

« Sans bataille il n'y a pas de victoire ».

« Без муки нет науки ».

« Sans tourment il n'y a pas de science ».

« Век живй — век учись ».

« On n'a jamais fini d'apprendre ».

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AVANT-PROPOS

Ce projet de recherche sur l'introduction de zooplancton non-indigène s'insère dans le cadre du projet *Understanding ballast water as a pathway for introduction of aquatic invasive species (AIS) in the Arctic*, sous l'égide du programme SPERA (*Strategic Program for Ecosystem-based Research and Advice*), portant sur les impacts écosystémiques des activités anthropiques. Les résultats de ce projet de recherche ont été présentés sous forme de présentations orales et d'affiches au cours de différents congrès de recherche nationaux et internationaux.

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Tremblay, P.; Rochon, A.; Winkler, G.; Howland, K.; Simard, N.; Bailey, S. 2015. « Ships' ballast water as a potential vector of invasive zooplankton introduction in the Canadian Arctic ». *14^{ième} Assemblée Générale annuelle de Québec-Océan*. 10 au 11 octobre 2015. Québec (Canada).

Tremblay, P.; Rochon, A.; Winkler, G.; Howland, K.; Simard, N.; Bailey, S. 2016. « Ballast water of domestic ships as a vector of invasive zooplankton introduction in Nunavik ». *Congrès des étudiants du GEOTOP 2016*. 19 au 20 mars 2016. Montréal (Canada).

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Tremblay, P.; Rochon, A.; Winkler, G.; Howland, K.; Simard, N.; Bailey, S. 2016. « Assessment of nonindigenous species introduction risk through ballast water discharge of a domestic ship in the Canadian Arctic ». *ICES Annual Science Conference 2016*. 19 au 23 septembre 2016. Riga (Lettonie).

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Tremblay, P.; Rochon, A.; Winkler, G. 2016. « Les eaux de ballast de navires domestiques comme vecteur d'introduction d'espèces mésozooplanctoniques non-indigènes dans l'est de l'Arctique canadien ». *Présentation des résultats de maîtrise, Institut des Sciences de la Mer de Rimouski*. 6 septembre 2016. Rimouski (Canada).

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RÉSUMÉ

Le déversement d'eau de ballast est la source d'un grand nombre d'introductions d'espèces non-indigènes (ENI) au Canada et dans le monde. Ces eaux sont transportées dans les réservoirs de ballast de navires peu chargés, où les organismes présents peuvent être relâchés en-dehors de leur aire de répartition naturelle lors du déballastage. Le développement de l'Arctique et l'augmentation conséquente du trafic maritime, combinés à la réduction du couvert de glace, sont susceptibles d'augmenter à long terme les risques d'introductions d'ENI. Le règlement sur la gestion de l'eau de ballast, via la *Loi sur la marine marchande du Canada*, a été conçu pour limiter l'introduction d'ENI par les navires internationaux. Ceux-ci sont tenus d'échanger leur eau de ballast d'origine côtière en milieu océanique ouvert, limitant ainsi les risques d'introduction d'ENI côtières. Or, les navires domestiques, naviguant exclusivement dans la zone économique exclusive canadienne, en sont présentement exemptés. Nous évaluons les risques d'introduction d'ENI de mésozooplancton (MZP) dans les eaux côtières de l'est de l'Arctique canadien en mesurant la densité, la richesse et la diversité du MZP dans l'eau de ballast transportée par un navire domestique canadien, le *M/V Arctic*. Ce navire transporte des eaux prélevées au port de Québec (QC) et les déverse régulièrement au port de Baie Déception (QC). L'évaluation de l'efficacité de deux zones d'échange volontaire d'eau de ballast, les détroits de Jacques-Cartier et de Belle-Isle, a démontré que les réservoirs échangés contenaient généralement une plus grande densité totale en organismes, comparativement aux réservoirs inchangés (contrôles). Dans les réservoirs inchangés, la communauté mésozooplanctonique a été soumise à une forte mortalité en raison de l'effet du vieillissement des eaux de ballast. Toutefois, la composition des espèces dans les réservoirs échangés était similaire à la communauté du port de destination (Baie Déception), alors que les réservoirs non-échangeés contenaient une plus grande densité d'ENI. L'information relative au projet contribuera à l'amélioration des connaissances sur le transport d'ENI en Arctique et à la révision des lois sur la gestion des eaux de ballast.

Mots clés : Déversement d'eau de ballast; Espèces non-indigènes; Zooplancton; Arctique canadien; Nunavik; Port de Baie Déception; Port de Québec; Navires domestiques; Échange volontaire d'eau de ballast; Détroit de Belle-Isle; Détroit de Jacques-Cartier

ABSTRACT

Ballast water discharge is a major source of nonindigenous species (NIS) introductions globally and in Canadian waters. This water is carried in ballast tanks of ships loaded with little or no cargo, where surviving organisms may be released in new environments upon deballasting. Arctic economic development and consequential rise in marine traffic, combined with sea ice reduction, may increase the risks of introductions over the long term. Ballast water exchange (BWE) regulations, under the *Canada Shipping Act*, were designed to limit NIS introductions in Canadian waters by international vessels, which are required to conduct mid-ocean ballast water exchange. However, domestic ships, navigating exclusively within Canadian waters, are currently exempt from these regulations. Here, we assessed the potential risks of NIS introduction to the Eastern Arctic by quantifying species density and diversity of mesozooplankton (MZP) in ballast water carried by a domestic ship, the *M/V Arctic*, which regularly transits between Quebec City and Deception Bay (QC). Evaluation of two voluntary BWE zones in Jacques Cartier Strait and Strait of Belle Isle revealed lower efficacy in reducing MZP density compared to unexchanged (control) water ballasted in Quebec City. In the unexchanged ballast water tanks, the MZP community experienced high mortality due to increasing ballast water age. However, species composition in exchanged tanks was similar to the community of the port of destination (Deception Bay), whereas unexchanged tanks transported higher densities of NIS. Data obtained through this study will provide basic information on NIS transport to the Arctic and contribute to the revision of regulations.

Keywords: Ballast water discharge; Non-indigenous species; Zooplankton; Eastern Canadian Arctic; Nunavik; Deception Bay harbor; Quebec City harbor; Domestic vessels; Voluntary ballast water exchange; Strait of Belle Isle; Jacques Cartier Strait

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LISTE DES ACRONYMES

Français	Anglais
ÉEB	Échange d'eau de ballast
ENI	Espèce non-indigène
MZP	Mészooplancton
<i>M/V</i>	Navire à moteur
	BWE Ballast water exchange
	NIS Non-indigenous species
	MZP Mesozooplankton
	M/V Motor vessel
	df Degrees of freedom
	SS Sum of squares
	MC Monte Carlo test
	PERM PERMANOVA test
	ANOVA Analysis of variance
	Mo Months
	Si Sites
	Tr Treatments
	BACI Before-After-Control-Impact
	MDS Multidimensional scaling

INTRODUCTION GÉNÉRALE

LES EAUX DE BALLAST ET LEURS IMPACTS DANS LE MONDE

Depuis des siècles, le ballastage des navires était assuré par le chargement de ballast solide, comme par exemple la pierre ou le sable. Au début du 20^{ième} siècle, avec l'avènement des navires à coque d'acier à fort tonnage et des technologies de pompage, l'eau a remplacé complètement le ballast solide pour des raisons de sécurité et de logistique maritime (Transport Canada, 2010a ; Jing *et al.*, 2012). Toutefois, les rejets de ces eaux de ballast, dans le cadre d'un transport commercial maritime mondial, ont été globalement la source d'un grand nombre d'introductions d'espèces non-indigènes (ENI) et le principal vecteur d'introductions en eaux côtières canadiennes (Locke *et al.*, 1993 ; Ruiz *et al.*, 2000 ; Molnar *et al.*, 2008 ; Chan *et al.*, 2012 ; Katsanevakis *et al.*, 2013 ; Pêches et Océans, 2014). L'eau de ballast est pompée en grands volumes par les cargos contenant peu ou pas de marchandises et transportée temporairement dans les réservoirs de ballast afin d'obtenir une profondeur optimale de fonctionnement des hélices et de contrôler gîte, tirant d'eau et stabilité du navire. Le poids supplémentaire ainsi ajouté permet d'abaisser la ligne de flottaison du navire, procurant stabilité pendant les manœuvres ou pour permettre de passer sous certaines structures, comme par exemple les ponts. Le transport par voie maritime assurant plus de 80% du transit mondial de marchandises, de grandes quantités d'eau de ballast, estimées à 4 milliards de tonnes, sont déversées chaque année dans les ports du globe (Tsolaki et Diamadopoulos, 2010 ; Jing *et al.*, 2012 ; Great Lakes Information Center, 2015). On estime que plus de 7000 espèces sont quotidiennement en transit via les eaux de ballast (David et Gollasch, 2011). Le Canada, à lui seul, recevrait un volume estimé à 46 millions de tonnes métriques par année d'eau de ballast provenant d'outre-mer (McCollin *et al.*, 2007). Les organismes côtiers présents dans l'eau de port sont alors transportés de la région donatrice (port d'origine) à la région réceptrice (port de

destination), peuvent survivre au trajet et être relâchés au déballastage, lors du chargement du navire (**Figure 1**). Il peut ainsi y avoir reproduction et établissement de ces communautés, en-dehors de leur aire de répartition et de dispersion naturelle. Les eaux et les sédiments de ballast peuvent contenir des organismes de tailles variables, allant des plantes et animaux microscopiques jusqu'aux crabes, moules et poissons (MIT Sea Grant, 2006). L'introduction d'ENI peut occasionner d'importantes conséquences écologiques et économiques irréversibles, comme la perturbation des réseaux trophiques, la perte de biodiversité, l'extinction d'espèces indigènes et le déclin de pêches ou d'exploitations aquacoles (Chan *et al.*, 2012 ; Pêches et Océans Canada, 2014 ; Ruiz *et al.*, 1997 ; Organisation Maritime Internationale, 2004 ; Casas-Monroy, 2014).

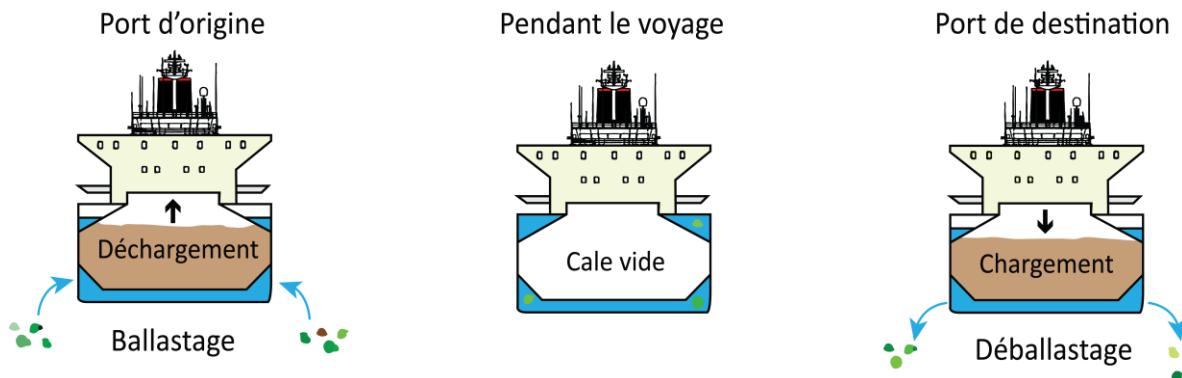


Figure 1. Fonctionnement des eaux de ballast (d'après Laget, 2016)

Les ENI peuvent perturber les réseaux trophiques et les relations de compétition autour desquelles la communauté indigène s'était originellement structurée (Centre Saint-Laurent, 1996). Plus de 145 espèces non-indigènes auraient été introduites dans les Grands Lacs, dont plus de 80% via les eaux de ballast (Fofonoff *et al.*, 2003). On estime qu'environ 10% des introductions aquatiques ont eu d'importants impacts sur le plan environnemental (Ricciardi et Kipp, 2008). Parmi les exemples frappants d'introductions d'ENI ayant eu d'importantes conséquences écologiques et économiques dans les Grands Lacs figure le cas de la moule zébrée (*Dreissena polymorpha*), qui a été aperçue pour la première fois en Amérique du Nord en 1988 dans le lac Sainte-Claire (Ontario). Cette espèce envahissante redoutable, originaire du bassin ponto-caspien, aurait été introduite par

déballastage et dispersée dans tout le bassin des Grands Lacs et dans l'Est des États-Unis via la dispersion naturelle et la navigation de plaisance. Chaque femelle de cette espèce très prolifique a la capacité de produire entre 30 000 et 100 000 oeufs par année, ce qui explique son succès de colonisation. Le stade larvaire (véligère) de cette espèce est planctonique ($\geq 70 \mu\text{m}$) et est porté par les courants (Lucy, 2006). La moule zébrée serait à la source de la disparition de 13 espèces indigènes dans le lac Sainte-Claire, ainsi que de la quasi-extinction de 10 espèces indigènes dans le lac Érié. Cette espèce se fixe en colonies très denses sur une grande variété de substrats. Ainsi, elle peut boucher les prises d'eau municipales, industrielles et hydroélectriques. Des densités supérieures à 10 000 individus/ m^2 ont été observées (Ruiz *et al.*, 2007). L'activité de filtration des colonies de moules zébrées (de 5 à 400 ml d'eau par heure par moule) diminue fortement la quantité de phytoplancton disponible pour les autres organismes, augmente la transparence de l'eau, diminue la quantité d'oxygène dissous et augmente les concentrations en nutriments dissous (Levinton et Waldman, 2006 ; Elliot *et al.*, 2008). Les frais associés à la gestion de cette espèce envahissante (râclage des colonies, utilisation de chlore) sont de plusieurs millions de dollars par année au Canada seulement (Bureau du vérificateur général du Canada, 2007 ; Ministère des Forêts, de la Faune et des Parcs, 2013 ; Centre Saint-Laurent, 1996). Dans le bassin des Grands Lacs, le contrôle des colonies autour des prises d'eau sur une période de dix ans aurait coûté 3,1 G \$US (Office of Technology Assessment, 1993). Au Canada, les pertes économiques associées à 16 ENI importantes ont été estimées entre 13,3 et 34,5 G \$CAD an⁻¹ (Colautti *et al.*, 2006a).

LES EAUX DE BALLAST DANS UNE RÉGION EN MUTATION

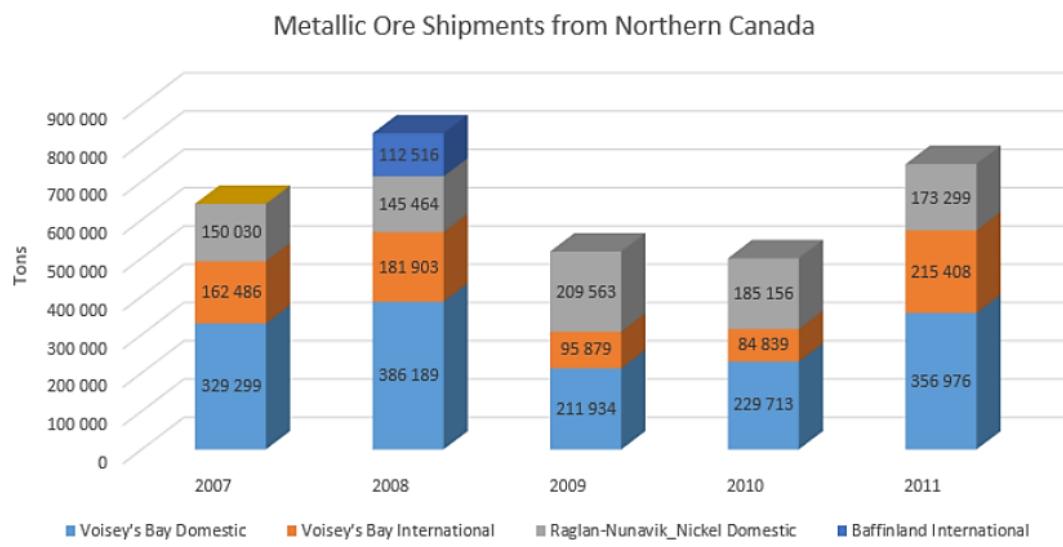
Alors qu'aucun établissement d'ENI introduite par déballastage n'a encore été observé dans les eaux arctiques canadiennes, l'introduction de 10 espèces non-indigènes a déjà été rapportée ailleurs dans le monde pour les zones arctique et subarctique (Chan *et al.*, 2012). 207 introductions ont été recensées pour les zones antarctique et subantarctique (Frenot *et al.*, 2005). Il a également été démontré que certaines espèces tempérées pouvaient survivre

aux conditions extrêmes de température et d’englacement des eaux antarctiques (Lewis *et al.*, 2006 ; Lee et Chown, 2009). Ainsi, la rudesse de l’environnement arctique n’empêcherait pas que certaines espèces généralistes et eurythermes puissent s’y établir. De plus, plusieurs auteurs estiment que le développement des ressources de l’Arctique et l’augmentation conséquente du trafic maritime seraient susceptibles d’augmenter les risques futurs d’introduction, à travers l’accroissement de la quantité d’eau de ballast déversée (Chan *et al.*, 2012 ; Chan *et al.*, 2014 ; Ware *et al.*, 2015). Or, les activités minières canadiennes au-delà du 55^{ième} parallèle sont actuellement faibles, avec moins d’un million de tonnes de minerai extraites de l’Arctique annuellement (**Figure 2**). Seulement trois des dix mines actuellement en exploitation dans la région utilisent la voie maritime comme méthode d’acheminement du minerai (**Tableau 1**). Toutefois, sept projets de développement minier devraient utiliser les voies maritimes arctiques, une fois les nouvelles mines mises en service (**Tableau 1**). Or, l’incertitude liée à l’exploitation minière dans l’Arctique est très grande, en raison de la valeur du minerai, des coûts élevés de production en région éloignée, des conditions climatiques difficiles, des investissements, des permis d’exploration et d’exploitation et des négociations avec les communautés inuites locales. Tous ces facteurs viennent compromettre la réalisation des projets miniers. Ainsi, il n’est présentement pas possible de démontrer avec certitude que l’industrie minière sera à l’origine d’une augmentation du trafic maritime dans l’Arctique canadien. Une fois les chantiers projetés mis en service, le trafic maritime devrait néanmoins augmenter de manière significative (Tétu *et al.*, 2015). Bien que peu de projets voient présentement le jour, la prospection minière dans la région demeure toutefois importante (**Figure 3**).

Tableau 1. Les projets miniers dans l'Arctique canadien (Têtu *et al.*, 2015)

Mining activities	Exploration and feasibility study	Development and construction of infrastructure	Active mines	Actives mines for which exports of raw materials are based on a shipping logistics through the Canadian Arctic waters	Inactive and unexploited mines for which exports of raw materials will be based on a shipping logistics through the Canadian Arctic waters
Iron ore	1	3	0	—	3
Base/industrial metals (copper, nickel, zinc)	2	3	5	3	4
Uranium ore	2	1	0	—	0
Precious metals (gold and silver)	3	2	5 ^a	0	0
Total	8	9	10	3	7

^aJericho Diamond mine and Ulu and Lupin's gold mine are both closed; Elgin Mining Inc. closed the mine which is 'under care and maintenance' since February 2005 and Shear Diamonds Ltd. reassess the viability of reopening the Jericho Diamonds mine.

Figure 2. Quantités annuelles de minerai (tonnes) extraites de l'Arctique canadien (Têtu *et al.*, 2015)

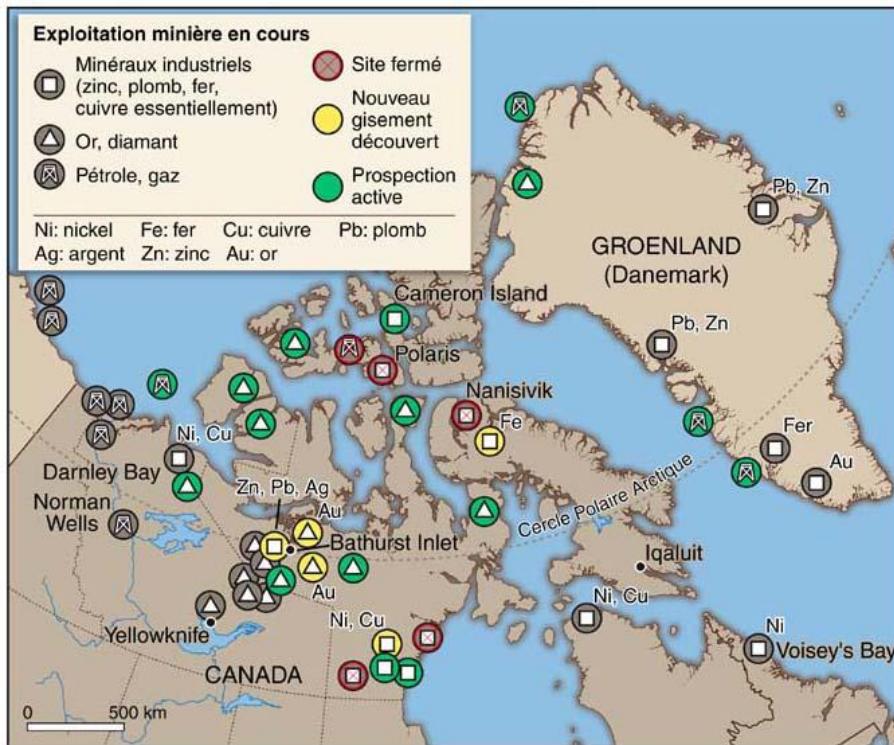


Figure 3. Mines en exploitation et prospection minière dans l'Arctique canadien (Lasserre et Rivard, 2007)

De plus, plusieurs auteurs estiment que l'effet d'un potentiel développement économique serait exacerbé par l'allongement de la saison de navigation, dû au déclin du couvert de glace saisonnier, en lien avec les changements climatiques dans l'Arctique (Niimi, A.J., 2004 ; Smith *et al.*, 2013; Pêches et Océans Canada, 2014 ; Chan *et al.*, 2014 ; Ware *et al.*, 2015). Certains modèles de simulation du climat suggèrent que le réchauffement dans l'Arctique rendrait cette région naturellement plus propice aux introductions biologiques et que les risques d'introduction seraient particulièrement élevés là où le trafic maritime transfère des organismes d'une zone biogéographique à une autre (Arctic Council, 2009 ; Smith et Stephenson, 2013). Le réchauffement des eaux de surface et les variations de salinité projetés pour cette région favoriseraient l'introduction d'ENI. Ces changements pourraient réduire les barrières environnementales qui jusqu'ici empêchaient la colonisation naturelle par les espèces tempérées. Ces effets pourraient aussi s'appliquer aux espèces qui sont transportées artificiellement par le biais des activités humaines (Ware *et al.*, 2015). La moyenne annuelle de l'étendue de glace dans l'Arctique

aurait diminué de 3,7%/décennie entre 1979 et 2007. Entre 2003 et 2007, le volume de la banquise aurait diminué de 42% pour l'automne et de 21% pour l'hiver (Spreen *et al.*, 2011). L'étendue estivale de la banquise, au minimum de glace, est également en diminution (**Figure 4**), se qui s'est traduit par une réduction de l'âge et de l'épaisseur de la glace de mer. Les prévisions actuelles tendent vers une domination progressive de la glace de première année, qui viendrait remplacer la glace pluriannuelle, plus épaisse (**Figure 5**). Certaines prédictions font même état d'un océan Arctique complètement libre de glace pendant l'été, en lien avec la disparition de la glace pluriannuelle (Arctic Council, 2009). Actuellement, plusieurs détroits de l'Archipel arctique canadien sont parfois libres de glace pendant la saison estivale (Bourbonnais et Lasserre, 2015). Toutefois, l'augmentation de la mobilité et de la fragilité de la banquise, due à son amincissement, présente l'inconvénient d'augmenter la fréquence des événements de déformation de la couverture de glace (+ 51%/décennie entre 1979 et 2007), occasionnant ainsi la formation de crêtes de pression, qui sont d'importants obstacles à la navigation en raison de leur épaisseur importante (10 à 12 m) (Rampal *et al.*, 2009 ; Bourbonnais et Lasserre, 2015). De plus, la fréquence des débris d'icebergs, difficilement détectables et dangereux pour la navigation, serait pour sa part également en hausse. La navigation dans les zones libres de glace en hiver expose les navires aux risques liés aux embruns verglaçants, qui menacent leur stabilité. Bref, même si les changements observés dans le couvert de glace de l'Arctique offrent désormais un accès amélioré et une saison de navigation allongée, il a été démontré que les conditions de navigation dans l'Arctique restent toujours périlleuses et difficiles à rentabiliser, surtout en période hivernale (Arctic Council, 2009 ; Bourbonnais et Lasserre, 2015). D'ici 2020, il n'est pas prévu que les grands cargos internationaux utilisent le passage du Nord-Ouest pour le transport de marchandises (Bureau du vérificateur général du Canada, 2014). L'ouverture de nouvelles routes maritimes en Arctique n'est prévue que pour 2040 à 2059 (Smith et Stephenson, 2013). Ainsi, une augmentation du trafic maritime dans l'Arctique ne concerne que la courte saison estivale et dépendra plutôt des marchés et de l'investissement dans le secteur minier que du changement climatique.

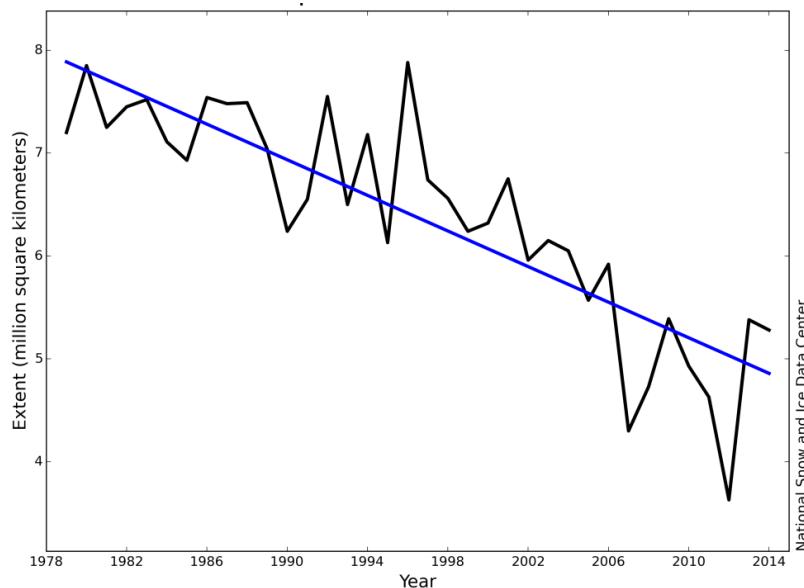


Figure 4. Diminution observée du minimum de glace en septembre dans l'Arctique entre 1979 et 2014 (NSIDC, 2016a)

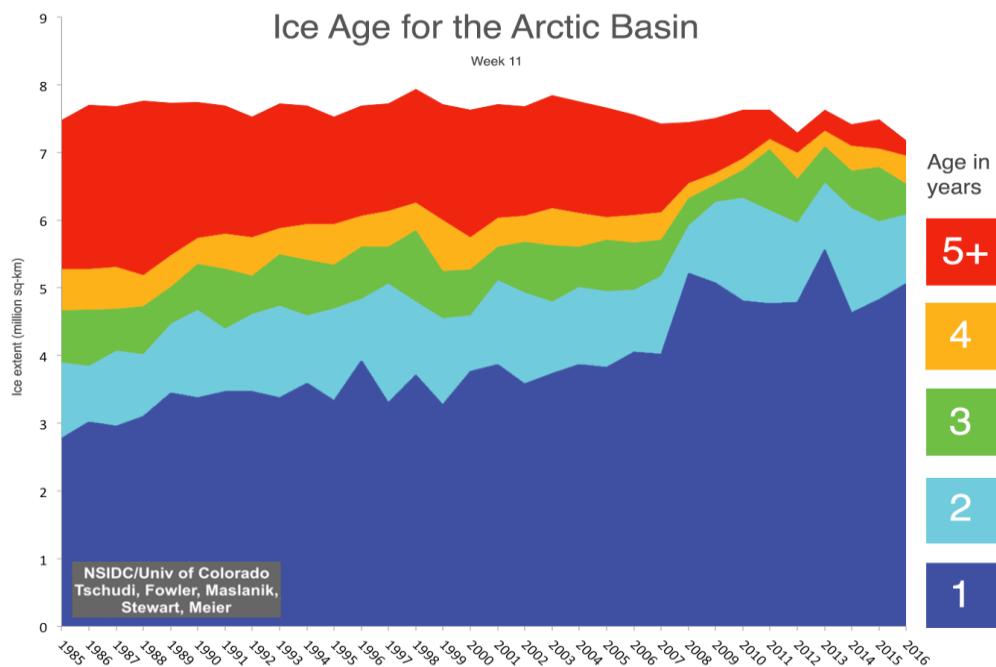
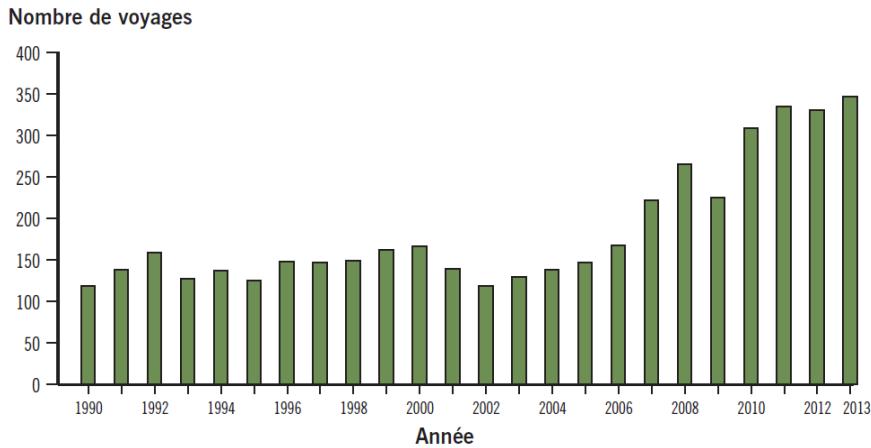


Figure 5. Composition en mars de la banquise arctique en fonction de l'âge de la glace entre 1985 et 2016 (NSIDC, 2016a)

D'un autre côté, l'augmentation du trafic observée entre 1990 et 2013 (**Figure 6**) est plutôt due aux navires n'étant pas équipés de réservoirs de ballast (Bureau du vérificateur général du Canada, 2014). Stewart *et al.* (2015) suggèrent que l'augmentation du traffic et donc des volumes d'eau déversés dans l'Arctique canadien sera d'origine internationale et non domestique; ainsi les navires impliqués auront déjà échangé leur eau de ballast en pleine mer, à l'extérieur de la zone économique exclusive canadienne. Les volumes d'eau de ballast déversés dans l'est de l'Arctique canadien devraient tripler si le projet d'ouverture de mine de fer à Mary River (île de Baffin) est réalisé. Un total de 662 000 m³ an⁻¹ devrait être déversé durant la saison de navigation libre de glace dans le port de Milne Inlet. Or, ce minerai sera acheminé via des navires transocéaniques en provenance de Rotterdam, qui auront préalablement échangé leurs eaux de ballast dans l'Atlantique en milieu océanique ouvert. Ces eaux seront ensuite traitées, même si les traitements envisagés n'ont pas encore été spécifiés. De plus, l'efficacité des traitements en question n'a pas encore été testée en milieu arctique. Le projet en question engendrera le déversement de 20 400 000 m³ an⁻¹ d'eau de ballast, correspondant à 74 fois le volume total moyen déversé dans tout l'Arctique canadien entre 2005 et 2008 (Stewart *et al.* 2015). Le *M/V Nunavik*, mis en service en 2014 pour acheminer vers la Chine le nickel de la mine de Canadian Royalties à partir de Baie Déception via le passage du Nord-Ouest, est pour sa part équipé d'un système de traitement d'eau de ballast. Il s'agissait en 2014 d'une première pour un navire domestique canadien (Nunatsiaq News, 2014).



Source : Estimations de la Garde côtière au 31 décembre 2013

Figure 6. Nombre de voyages effectués par voie maritime dans l'Arctique canadien entre 1990 et 2013 (Bureau du vérificateur général du Canada, 2014)

LÉGISLATION CANADIENNE SUR LES EAUX DE BALLAST ET LES NAVIRES DOMESTIQUES

Selon Ware *et al.* (2015), l'actuelle gestion des eaux de ballast au niveau global ne permet pas de prévenir l'introduction d'ENI dans l'Arctique. Le Règlement sur le contrôle et la gestion de l'eau de ballast, à travers la Loi sur la marine marchande du Canada, a été conçu pour limiter l'introduction d'ENI par les navires internationaux (venant de l'étranger) en eaux côtières canadiennes. Il prévoit l'échange d'eau de ballast (ÉEB) d'origine étrangère en milieu océanique ouvert, causant ainsi la mort de 80 à 100% des organismes côtiers par choc osmotique dû au changement brusque de salinité (Chan *et al.*, 2012 ; Chan *et al.*, 2014 ; DiBacco *et al.*, 2012). Inversement, les espèces océaniques ne survivent généralement pas en zone côtière pour les mêmes raisons (Chan *et al.*, 2014 ; Niimi, 2004). Cette pratique permet en théorie de réduire la densité et la diversité des organismes présents dans les réservoirs (Ware *et al.*, 2015). En pratique, l'ÉEB en milieu marin réduit les risques d'introduction de manière efficace entre les écosystèmes d'eau douce. Or, l'efficacité est moindre lorsque le trajet relie deux ports situés en eau salée (Roy *et al.*, 2012 ; Ware *et al.*, 2015). Cependant, les navires domestiques, naviguant entre les ports de la Zone Économique Exclusive canadienne (ZEEC), sont exemptés de toute gestion de leurs eaux

de ballast (Ministère de la Justice du Canada, 2014). Ceux-ci présentent pourtant un potentiel élevé de survie des organismes, en raison de la durée généralement plus courte de leurs trajets, et peuvent contribuer à l'introduction secondaire d'ENI déjà établies et d'espèces indigènes des ports d'origine vers les hautes latitudes (Chan *et al.*, 2012 ; Pêches et Océans Canada, 2014 ; DiBacco *et al.*, 2012). Ainsi, certains navires réalisent actuellement l'échange côtier sur une base volontaire pour réduire les risques d'introduction (Chan *et al.*, 2012). Or, l'efficacité de ces échanges est mise en doute, vu la similarité environnementale entre les zones d'échange et les ports de destination, ainsi que la variabilité saisonnière dans les risques d'introduction (Casas-Monroy, 2014). On estime présentement que les risques d'introduction par les navires domestiques seraient faibles. Toutefois, un doute subsiste, dû à l'insuffisance d'échantillons biologiques d'eau de ballast pour cette région. L'avis scientifique sur l'introduction d'ENI de Pêches et Océans Canada Sciences (2014) recommande ainsi l'échantillonnage biologique pour les voies d'entrée (navires) arctiques, d'où l'intérêt du présent projet. En comparaison, les risques d'introduction en Arctique par les navires transocéaniques seraient faibles, principalement en raison de l'âge élevé des eaux de ballast qu'ils transportent (15 jours et plus) et de leur obligation de réaliser l'échange d'eau de ballast en milieu océanique ouvert (Chan *et al.*, 2012 ; Chan *et al.*, 2014). Or, pour les navires domestiques, cette période est généralement plus courte (environ sept jours pour le *M/V Arctic*), augmentant ainsi les risques de survie et les stocks d'individus (Casas-Monroy, 2014 ; Chan *et al.*, 2014). L'âge des eaux de ballast est le principal facteur influençant la diversité et la densité du zooplancton (Chan *et al.*, 2014). De surcroît, les ports domestiques canadiens fourniraient les plus grands stocks de propagules dans l'Arctique, en raison notamment de l'exemption d'ÉEB et d'une plus grande similarité entre les conditions environnementales des ports d'origine et de destination. Finalement, plusieurs espèces d'ENI à haut potentiel de dommages écologiques ont été recensées dans les ports qui sont directement reliés à l'Arctique par les navires domestiques (ex: moule zébrée, *Dreissena polymorpha*, à Québec) (Chan *et al.*, 2012 ; Environnement Canada, 2000).

GESTION DES EAUX DE BALLAST

Tous les navires naviguant en eaux canadiennes se doivent d'enregistrer toute opération impliquant leurs eaux de ballast (OMI, 2004). Les informations concernant les volumes transportés, la méthode d'échange, la date, l'heure, ainsi que les coordonnées des échanges doivent être archivées dans le *Ballast Water Logbook*. Il existe deux méthodes principales d'échange d'eau de ballast :

- **L'échange séquentiel** consiste à pomper, dans un premier temps, la totalité des eaux du réservoir en-dehors du réservoir de ballast (déballastage complet). Une fois cette étape terminée, le réservoir est rempli à nouveau avec l'eau de la zone d'échange. Cette méthode ne permet toutefois pas de se débarrasser des eaux résiduelles et des sédiments de ballast, qui sont difficilement délogeables du fond des cales. De plus, le fait de vider temporairement complètement un réservoir de ballast peut compromettre la sécurité et la manoeuvrabilité du navire. Lors du déballastage, le ballottement des eaux à l'intérieur du réservoir à moitié vide peut causer d'importants chocs sur la coque et peut compromettre la stabilité du navire. C'est la méthode employée par l'équipage du *M/V Arctic* pour réaliser l'échange de ses eaux de ballast sur une base volontaire.
- **L'échange par circulation** requiert une prise et une sortie d'eau fonctionnant simultanément pour assurer la circulation de l'eau. Le triple du volume initial doit être pompé pour assurer un renouvellement suffisant des eaux. L'eau de ballast initiale est donc déballastée et remplacée simultanément par l'eau d'échange qui est pompée. Comme cette méthode n'implique pas la vidange complète du réservoir, elle ne compromet pas la sécurité du navire (Transport Canada, 2011).

MOTIVATIONS DE L'ETUDE

Bien que l'augmentation du trafic maritime arctique dans un avenir proche ne soit encore pour l'instant qu'une prévision remplie d'incertitudes, il s'agit toutefois d'une éventualité à long terme. Quoique le déversement d'eau de ballast dans cette région soit encore très faible (environ 275 130 m³/an), cela représente toutefois une réalité récente (Chan *et al.*, 2012). Il est entendu qu'il s'agit d'un vecteur d'introduction d'ENI dominant et que celui-ci peut avoir d'importants impacts irréversibles sur les écosystèmes côtiers. Dans une région où les collectivités sont étroitement dépendantes des ressources marines pour leur sécurité alimentaire (chasse et pêche de subsistance), le dossier des eaux de ballast suscite un intérêt grandissant (Tétu *et al.*, 2015). Car bien que l'industrie, les collectivités et le secteur public s'entendent sur la nécessité de développer l'Arctique canadien, il y a pression pour que ce développement se fasse de manière à en minimiser les impacts. Ainsi, cette étude s'insère dans un projet plus vaste de Pêches et Océans Canada, visant à fournir les connaissances de base sur une région où l'échantillonnage n'en est qu'à ses débuts. Si l'augmentation dans un avenir proche des volumes d'eau de ballast déversés en Arctique n'est pour l'instant qu'une possibilité, il est toutefois important d'apporter une étude préliminaire des ENI y étant actuellement déversées. Étant donné que cette région est encore pratiquement libre d'envahisseurs biologiques, il importe donc de démontrer le potentiel d'invasion que représente le trafic maritime actuel (Goldsmith *et al.*, 2014). De plus, le fait qu'aucune ENI n'ait encore été observée dans l'Arctique canadien pourrait être dû au manque de suivi rigoureux (Molnar *et al.*, 2008 ; Chan *et al.*, 2012 ; Chan *et al.*, 2014 ; Ware *et al.*, 2015). De plus, les écosystèmes de l'océan Arctique seraient plus vulnérables aux perturbations en raison de réseaux trophiques plus courts qu'aux latitudes tempérées et tropicales (Cangelosi *et al.*, 2007). L'impact écologique lié à des introductions d'ENI pourrait donc y être plus marqué. Finalement, peu d'études portent sur l'importance du transport d'ENI par ballast aux hautes latitudes (Jing *et al.*, 2012 ; Chan *et al.*, 2014 ; Ware *et al.*, 2015). Un seul échantillonnage de la composition biologique des eaux de ballasts a été réalisé pour les eaux côtières canadiennes arctiques (Chan *et al.*, 2014).

Pour comprendre les risques potentiels d'introduction d'ENI en eaux côtières arctiques, nous nous intéressons au contenu en mésozooplankton (MZP) des eaux de ballast déversées dans un important port de la région, Baie Déception (QC). Celui-ci présente les plus hauts risques d'introduction car il se retrouve au premier rang des ports de l'Arctique canadien en termes de volumes d'eau de ballast domestique déversés, avec 80% du volume total domestique ($60\ 144\ m^3/\text{an}$). Ce port recevait également entre 2005 et 2008 environ 22% des eaux totales (domestiques et internationales) déversées dans l'Arctique canadien. De plus, 12 espèces non-indigènes à fort potentiel de dommages écologiques seraient reliées à Baie Déception par le trafic domestique (Chan *et al.*, 2012). Le projet concerne un seul navire domestique, le *M/V Arctic*, visitant Baie Déception à l'année longue (Fednav, 2016a) (**Figure 7**). Normalement, les mines de l'Arctique stockent le minerai tout au long de l'hiver et l'exportent une fois la saison de navigation entamée. Or, étant donné que le concentré de nickel produit par la mine Raglan et stocké au dépôt du port de Baie Déception se dégrade s'il n'est pas traité rapidement, il doit être acheminé sur une base régulière durant toute l'année. En 2004, ce transport effectué à longueur d'année était le seul enregistré pour l'Arctique, avec la mine de nickel de Norilsk (Fédération de Russie) (Arctic Council, 2009).

OBJECTIFS ET HYPOTHESES

Objectif général : L'objectif général de cette étude est de caractériser la densité et la composition des espèces du mésozooplancton présent dans les eaux de ballast du *M/V Arctic* pour évaluer le risque potentiel d'introduction d'espèces non-indigènes dans le port de Baie Déception (QC). Ce navire est utilisé comme exemple de navire domestique canadien transitant dans l'Arctique canadien durant toute l'année. La notion de risque potentiel concerne les espèces non-indigènes déversées au port de Baie Déception, selon leur gradient de tolérance thermique et haline. L'information relative au projet contribuera à l'amélioration des connaissances sur le transport d'ENI aux hautes latitudes et à la révision des lois actuelles sur la gestion des eaux de ballast de navires domestiques.

- **Objectif spécifique #1 :** Évaluer la variabilité mensuelle estivale des communautés mésozooplanctoniques présentes dans les réservoirs du *M/V Arctic* au port d'origine (Québec). Les paramètres évalués seront la densité, la diversité et la richesse en espèces du mésozooplancton. Une attention particulière sera portée à la présence d'espèces non-indigènes notoires déjà décrites dans la littérature.
- **Hypothèse #1 :** Nous allons tester deux hypothèses déjà émises par Basu *et al.* (1999) et Irigoien *et al.* (2004) dans d'autres contextes. La densité totale du mésozooplancton sera maximale au milieu de la saison estivale. Pendant ce pic de densité, la diversité et la richesse en espèces seront minimales. Par ailleurs, la densité des ENI sera maximale pendant cette période.

- **Objectif spécifique #2 :** Évaluer l'effet et l'efficacité de l'échange volontaire d'eau de ballast réalisé par le *M/V Arctic* dans le golfe du Saint-Laurent. Le Site 1 (détroit de Jacques-Cartier) et le Site 2 (détroit de Belle-Isle) seront testés selon leur efficacité à réduire la densité totale, la diversité et le nombre d'espèces non-indigènes de mésozooplancton. De plus, évaluer l'effet du vieillissement des eaux de ballast sur la densité totale et la diversité du mésozooplancton dans les réservoirs inchangés (contrôles).
- **Hypothèse #2 :** L'efficacité de l'échange d'eau de ballast à réduire le nombre d'espèces non-indigènes sera élevée en raison de la forte dissimilarité environnementale entre le port d'origine (eau douce) et les zones d'échange (eau salée). L'échange d'eau de ballast dans l'environnement marin du golfe du Saint-Laurent devrait éliminer les espèces non-indigènes dulcicoles par choc osmotique. Toutefois, l'efficacité de l'échange d'eau de ballast à réduire la densité totale de mésozooplancton sera faible, car l'échange en milieu côtier, généralement productif, fournira une eau riche en propagules. De plus, l'eau de ballast échangée dans le golfe aura un âge d'eau de ballast moindre et donc une plus grande densité totale en organismes (Pêches et Océans Canada, 2014 ; Casas-Monroy, 2014). L'échange d'eau de ballast devrait également augmenter la diversité du mésozooplancton car les eaux marines sont généralement plus diversifiées que les eaux douces (Lehman, 1988 ; Fernando *et al.*, 1990). Finalement, le vieillissement de l'eau de ballast dans les réservoirs contrôles devrait diminuer la densité totale et la diversité du mésozooplancton (Wonham *et al.*, 2001 ; Humphrey, 2008 ; Chan *et al.*, 2014).

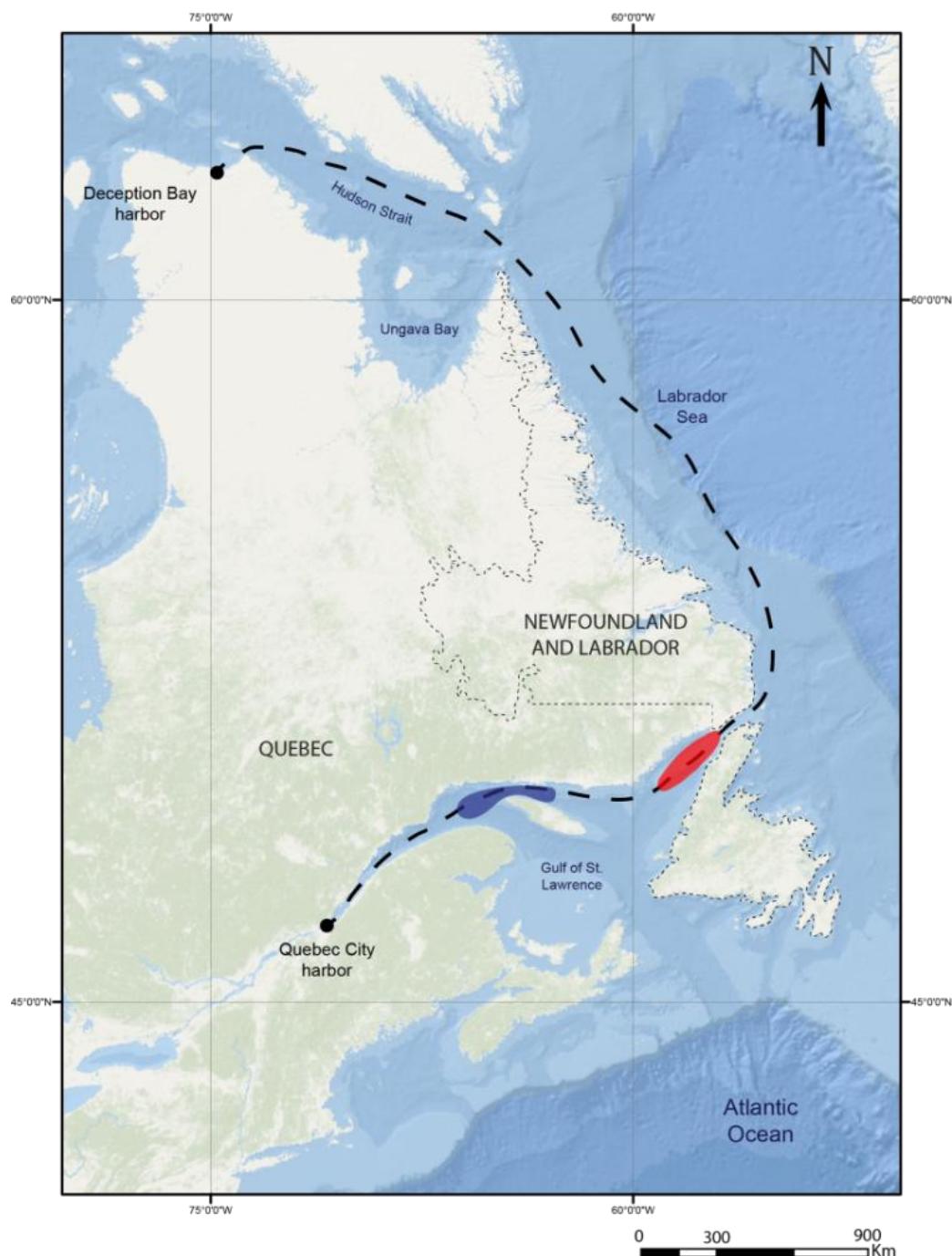


Figure 7. Le trajet du *M/V Arctic*, ainsi que les deux zones d'échange d'eau de ballast testées. En bleu : Détroit de Jacques-Cartier (Site #1). En rouge : Détroit de Belle-Isle (Site #2) (Laget, 2016)

CHAPITRE 1

CANADIAN DOMESTIC VESSELS AS A VECTOR OF INTRODUCTION OF MESOZOOPLANKTONIC AQUATIC INVASIVES: AN ASSESSMENT OF VOLUNTARY BALLAST WATER EXCHANGE EFFICACY

1.1. CONTEXT OF THE STUDY AND RESEARCH OBJECTIVES

Ballast water discharge is a major vector of nonindigenous species (NIS) introductions globally and in Canadian waters (Locke *et al.*, 1993; Ruiz *et al.*, 2000; Molnar *et al.*, 2008; Chan *et al.*, 2012; Katsanevakis *et al.*, 2013; Fisheries and Oceans Canada, 2014). Large volumes of waters are pumped by cargo ships carrying little or no cargo and transported temporarily inside the ballast tanks to control ship stability, draft, trim and list. Entrained coastal organisms from the port of origin may survive the ship's voyage and be released in new environments after deballasting in the port of destination. Since sea shipping corresponds to 80% of global cargo transit, large volumes of ballast water, estimated at 4 billion tons, are discharged every year in numerous ports around the world (Tsolaki and Diamadopoulos, 2010; Jing *et al.*, 2012; Great Lakes Information Center, 2015). Introductions of NIS can have serious irreversible economic (between 13.3 and 34.5 G \$CAD/year in Canada due to 16 NIS), and most importantly ecological impacts, such as the disruption of food webs, decreases in biodiversity, extinction of indigenous species and the decline of fisheries and aquaculture operations (Ruiz *et al.*, 1997; International Maritime Organization, 2004; Chan *et al.*, 2012; Fisheries and Oceans Canada, 2014; Casas-Monroy *et al.*, 2014).

Although no establishment of ship-mediated NIS has been recorded in the Canadian Arctic, economic development and consequential rise in sea traffic, combined with sea ice reduction, could increase the rate of introductions. Mining activities beyond the 55th parallel north are still low, with less than a million tons of ore extracted yearly. However, mining exploration in the Arctic is active, with several projects underway. Nevertheless, there is great uncertainty regarding mining development because of regional constraints. Prices of ore, high production costs in remote regions, harsh weather conditions, investment, exploration and operating licenses and negotiations with local communities are all factors that have limited the development of mines in this region. However, mining exploration is currently a fact, and if projected mines are commissioned, there will be a significant increase in shipping traffic (Tétu *et al.*, 2015). Moreover, the effects of potential economic development are predicted to be exacerbated by the lengthening of the shipping season, due to the reduction of sea ice cover with climate change in the Arctic (Fisheries and Oceans Canada, 2014; Chan *et al.*, 2014; Niimi, A.J., 2004; Smith *et al.*, 2013). Mean annual ice extent has decreased by 3.7% between 1979 and 2007. Between 2003 and 2007, the volume of the pack ice decreased by 42% for fall and 21% for winter (Spreen *et al.*, 2011). The summer sea ice extent is decreasing, resulting in a reduction of ice age and thickness, with many straits of the Canadian Arctic Archipelago now being ice-free during summer (Bourbonnais and Lasserre, 2015).

An increase in maritime traffic in the near future is a prediction filled with uncertainty, but it is likely to increase in the long term. Although ballast water discharge in the region remains very low ($\approx 275\text{--}130\text{ m}^3\text{ year}^{-1}$, Chan *et al.*, 2012), this discharge is a new and recent phenomenon for much of the Arctic Basin. It is now understood that ballast water is a dominant vector of NIS introductions, causing potentially irreversible impacts on coastal ecosystems. In a region of the world where communities rely closely on resources of the sea for food security through subsistence fishing and hunting, ballast water issues are of increasing interest because of potential damage on local ecosystems (Tétu *et al.*, 2015). There is growing enthusiasm for the development of the Arctic, which is accompanied by local pressure for sustainable development. Thus, the aim of this study is to provide basic

knowledge on potential mesozooplankton (MZP) introductions, in an area where baseline coastal sampling for NIS is at an early stage, if not non-existent. If an increase in ballast water discharge in the Canadian Arctic over the short term remains only a possibility, knowledge of actual NIS transport is crucial. Given that this region does not contain any known marine invaders, it is relevant to estimate the invasion potential related to the actual shipping traffic. Moreover, the fact that no marine NIS have been recorded for the Canadian Arctic may be due to the lack of rigorous monitoring (Molnar *et al.*, 2008; Chan *et al.*, 2012; Chan *et al.*, 2014; Goldsmith *et al.*, 2014). Very few studies have been conducted on ballast-borne NIS at high latitudes. Only a single sampling effort of biological composition of ballast water has been conducted for Canadian coastal arctic waters (Chan *et al.*, 2014; Chan *et al.*, 2015).

Ballast water exchange (BWE) regulations for international vessels, under the "Canada Shipping Act", were designed to limit NIS introductions in Canadian waters. However, domestic ships, navigating within Canadian waters, are currently exempt. To assess potential risks of NIS introduction in arctic coastal waters, we evaluated MZP communities of discharged ballast water in an important port of the eastern Arctic, Deception Bay (QC). This port was targeted in the study due to its high risks for introduction, receiving the highest volume of domestic ballast water in the Canadian Arctic ($60\ 144\ m^3\ year^{-1}$, 80% of total). Furthermore, 12 high-impact NIS species are connected to Deception Bay via domestic vessels (Chan *et al.*, 2012). Here, we quantified species density, richness and diversity of MZP in ballast water carried by the *M/V Arctic*, a domestic vessel visiting Deception Bay year-round.

Main objective: The main objective of this study was to characterize density and species composition of mesozooplankton present in the ballast water tanks of the *M/V Arctic* to assess potential risks of NIS introductions in Deception Bay (QC). An evaluation of the influence of its ballast water exchange was also conducted. This ship served as an example of a Canadian domestic vessel regularly transiting in Canadian Arctic waters. Potential risks of introduction relate to nonindigenous species brought in Deception Bay in relation with their salinity and temperature tolerance thresholds. Data obtained through this study will improve our knowledge on NIS introductions in the Canadian Arctic and contribute to the revision of regulations on ballast water management for domestic vessels.

Specific objective #1: Evaluate summer monthly variability in mesozooplankton communities present in the ballast water tanks of the *M/V Arctic* at the port of origin (Quebec City harbor). MZP communities will be characterized according to total density, diversity, species richness and species composition based on densities. Special attention will be given to taxa already described in the literature as notorious invaders.

Hypothesis #1: We will test two hypotheses already issued by Basu *et al.* (1999) and Irigoien *et al.* (2004) in other contexts. Total MZP density in the port of origin will peak during midsummer. During this peak in density, diversity and species richness will be lowest. Quebec City harbor will also have maximal NIS density during this period.

Specific objective #2: Evaluate the effect and efficacy of voluntary ballast water exchange performed by the *M/V Arctic* in the Gulf of St. Lawrence. Exchange sites 1 (Jacques Cartier Strait) and 2 (Strait of Belle Isle) will be tested to assess their efficacy in reducing MZP density, diversity, species richness and nonindigenous species occurrence. The effect of ballast water aging on MZP density and diversity in the unexchanged (control) ballast tanks will also be evaluated.

Hypothesis #2: Efficacy of ballast water exchange in reducing the occurrence of nonindigenous species is expected to be high due to high environmental mismatch between the port of origin (freshwater) and the exchange sites (marine). Exchanging in marine waters from the Gulf of St. Lawrence should eliminate freshwater nonindigenous species. However, efficacy of ballast water exchange in reducing total MZP density will be low, because exchanging in rich coastal waters should provide large amounts of propagules, compared to oceanic offshore exchange. Also, exchanged ballast water will have lower ballast water age and thus lower mortality, with probably higher total MZP density (Pêches et Océans Canada, 2014; Casas-Monroy, 2014). Moreover, since marine environments are generally more diverse than freshwater, ballast water exchange is expected to increase mesozooplankton diversity (Lehman, 1988; Fernando *et al.*, 1990). Finally, ballast water aging should reduce total MZP density and diversity (Wonham *et al.*, 2001; Humphrey, 2008; Chan *et al.*, 2014).

1.2. MATERIAL AND METHODS

1.2.1. VESSEL AND EXPERIMENTAL DESIGN

Sampling effort was focused on the *M/V Arctic* (IMO # 7517507), a Canadian domestic ship built in 1978 and owned by Fednav Shipping Enterprise. This PC-4 type icebreaker was designed to sail year-round through first-year sea ice dotted with multi-year ice. This ship has been transporting nickel concentrate between Deception Bay harbor (QC) and the Port of Quebec City since 1998 (Glencore, 2013). It also carries supplies destined for Glencore-owned Raglan Mine Complex on the way back to Deception Bay. This cargo vessel is 221 m (725 ft) long and 23 m (75 ft) wide, with a draught of 15.24 m (50 ft). It has 20 ballast tanks that can hold a total volume of 28161.2 m³ of ballast water (**Figure 8**) (Fednav, 2016b). Sampling was carried out monthly from summer to fall 2015. During the three summer trips (June, July and August), our sampling team was aboard the *M/V Arctic* for the entire voyage between Quebec City harbor and Deception Bay. Sampling of the ballast water tanks for these three trips was conducted before and after exchange in two voluntary BWE zones. For September and October, sampling was conducted by our team only in Quebec City harbor and by Raglan Mine technicians upon arrival of the ship in Deception Bay. Thus, for these two months, only samples corresponding to the beginning and end of the trips were collected in the control, Site 1 and Site 2 tanks, as well as in Quebec City and Deception Bay harbors. For each of the five trips, three ballast tanks and the two ports were sampled. However, there was an issue in September and the Raglan Mine technicians did not sample the ship upon arrival in Deception Bay. The three tanks were selected in accordance to their role in our experimental design (**Table 2**). One ballast tank had its waters exchanged in Site 1 (Jacques Cartier Strait), another in Site 2 (Strait of Belle Isle), and the third remained unexchanged for the whole trip, serving as control

treatment. The control tank was filled with freshwater uploaded in Quebec City. At the end of each trip, information regarding BWE practices, compiled in the ship's *Ballast Water Logbook*, was retrieved by our sampling team upon request to the first officer. This information included: date, hour and coordinates of BWE (beginning and end), as well as method of deballasting and capacity of each exchanged ballast tank. Precise BWE locations can be viewed in **Appendix 1**. Also, see **Appendix 2** and **Figure 8** for a visual representation of the experimental design and of the tanks used.

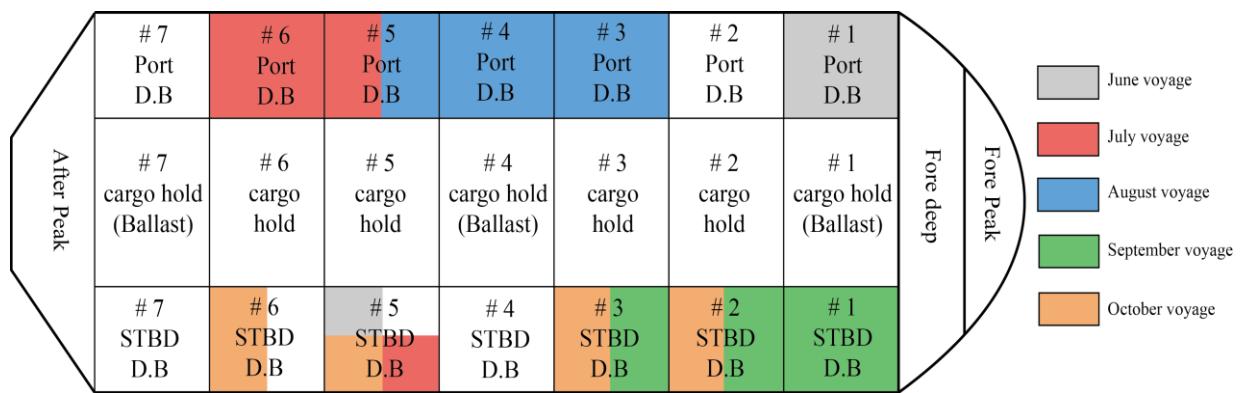


Figure 8. The *M/V Arctic*'s ballast tanks and their use during the trips (Port = Port side; STBD = Starboard side). D.B. = Double bottom ballast water tank.

Table 2. Information on experimental design and the *M/V Arctic*'s trips (S = Starboard; P = Port side; Site #1 = Jacques Cartier Strait; Site #2 = Strait of Belle Isle)

Trips	Dates	Exchanged tank (Site 1)	Exchanged Tank (Site 2)	Control tank
June	June 2 nd - 15 th 2015	3S	1P	5S
July	July 9 th - 20 th 2015	5P	6P	5S
August	August 13 th - 21 st 2015	3P	5P	4P
September	Start only (September 3 rd and 4 th 2015)	1S	3S	2S
October	September 30 th - October 11 th 2015	5S	6S	2S

1.2.2. DEPTH AND PHYSICAL PARAMETERS OF PORT AND BALLAST TANK WATERS

Depths of the water column in the harbors and ballast tanks were measured using a graduated rope with an attached lead weight. A vertical continuous profile of the water column characteristics from the surface to 2m above the bottom was obtained by lowering a CTD probe ([©] Seabird Electronics SBE_19 Plus V1 #4927) (speed: 1 m s⁻¹) and measuring temperature, conductivity, depth, fluorescence, and dissolved oxygen. The probe was lowered approximately 2 m from the bottom to avoid sediment resuspension. For comparison and to serve as backup, temperature and salinity of water collected from three [©]Niskin bottles (surface, middle and bottom of water column) were also measured using a thermosalinometer probe [©]WTW Cond 3210 Set 2 and water samples were also later analysed with an [©]Autosal device at Maurice Lamontagne Institute, Mont-Joli (Canada). All manipulations were carried out on the *M/V Arctic*'s deck. All information regarding sampling was written down on sampling forms (*Ship Sampling Field Log*, or *Log sheets*) and transformed into digital format ([©]Excel).

1.2.3. SAMPLING OF MESOZOOPLANKTON (MZP)

MZP samples were collected using two *Nitex* plankton nets of 80 µm mesh size. A net with a diameter of 0.3 m was used for sampling of the ballast tanks. A larger net (0.5 m diameter), was used to collect port samples. A minimal volume of 1000 L of ballast and port water was filtered for each sample, estimated according to net diameter and sampled depth (often requiring multiple vertical net tows). The net was thoroughly rinsed top-down with a wash bottle filled with prefiltered (20 µm) ballast water taken from the sampled tank, to avoid contamination between sampled tanks. The codend was then rinsed and its contents poured on an 80 µm sieve. Organisms were preserved in 95% ethanol, transferred to pre-identified 500 ml plastic bottles and sealed with electrical tape. After 24 hours, the ethanol was replaced by sieving the organisms and rinsing them back into the bottle with a

fresh supply of ethanol, to ensure better preservation (95% final concentration). Samples were stored at room temperature and kept away from light exposure.

1.2.4. IDENTIFICATION OF MESOZOOPLANKTONIC SPECIES

For traditional taxonomy work, each sample was split into subsamples before analysis using a *Motoda* splitter, due to the high density of organisms (Cusson, 2011). One subsample was then placed under a [°]*Leica MZ12₅* binocular microscope for specimen identification. Photographs of key specimens were taken with a [°]*Wild Heer-Brugg* digital camera to confirm identification and for personal reference. Zooplanktonic organisms were identified to the lowest taxonomic level possible, at a magnification power of 1000x. For more precise analyses on single specimens, a [°]*Zeiss Scope A1 (AXIO)* microscope was used occasionally. The analysis of some marine samples was conducted at DFO's Maurice-Lamontagne Institute in Mont-Joli (Canada), with expert taxonomists available for advice on problematic specimens. A minimum of 500 individuals were counted and identified in each subsample, to obtain representative densities. Several online identification keys and compilations of photographs and descriptions from numerous unidentified sources were used as a reference for taxonomy (Conway, 2006; Guittard and Joly, 1999; Haney *et al.*, 2013; Hudson *et al.*, 2003; Lesko *et al.*, 2003a; Lesko *et al.*, 2003b). Calanoid and cyclopoid copepods were also identified and classified according to copepodid development stages using the charts provided in Conway (2006). MZP density estimates were expressed by the number of individuals per cubic meter of water sampled (Ware *et al.*, 2015). Zooplankton taxa were classified as indigenous or non-indigenous to Deception Bay (in which they were to be discharged) using a database of previously identified MZP from the Canadian Arctic based on taxonomic and biogeographic literature (Grainger, 1962; Grainger., 1965; Roddie *et al.*, 1984; Smith and Schnak-Schiel, 1990; United States Department of the Interior, 1990; Hudon *et al.*, 1993; Basu *et al.*, 1999; Harvey *et al.*, 2001; Lesko *et al.*, 2003ab; Winkler *et al.*, 2003; Dufour and Ouellet, 2007; Cusson, 2011; Harvey *et al.*, 2011; Pinel-Alloul *et al.*, 2011; Estrada *et al.*, 2012; Ware *et al.*, 2015;

WoRMS, 2015; Marine Species Identification Portal, 2016a; Marine Species Identification Portal, 2016b; Global Biodiversity Information Facility, 2016; Kipp *et al.*, 2016; Ocean Biogeographic Information System, 2016). See **Appendices 3-4** for a complete list of taxa by ballast water uptake site and harbor.

1.2.5. STATISTICAL ANALYSES

Analyses performed

We used the Permutational Multivariate Analysis of Variance (PERMANOVA; PRIMER 6.1.11 software) statistical approach to test for differences among MZP communities (date and location) because of its insensitivity to the presence of many zeros and its distribution-free assumptions. The complement of the Bray-Curtis dissimilarity coefficient was used to produce similarity matrices because it is well suited for species density data and ignores double zeros, which were common in our MZP database. The “Reduced model” permutation method was used because it has the best statistical power and has the most accurate Type 1 error for complex designs. For each analysis, a maximum of 9999 permutations were set. Monte Carlo simulations were done when the number of possible permutations was low. For each PERMANOVA analysis, multidimensional scaling (MDS) was used for complementary visual analyses (Kruskal and Whish, 1978). *Post-hoc* pair-wise tests were done when a significant difference was observed to see which samples were different from each other. Similarity Percentages (SIMPER) analysis was subsequently used to identify species responsible for the differences among samples (Clarke, 1993). Also, the Analysis of Similarity (ANOSIM) test was conducted to test for differences in the taxonomic composition among groups of samples (Clarke and Green, 1988). This test can be applied to one-way and two-way crossed and nested experimental designs. The R-value generated by this analysis ranges from -1 to 1, with 0 meaning no difference, 1 indicating large between-group differences and -1 indicating large intra-group differences (Clarke and Warwick, 2001). Additionally, the analysis of variance (ANOVA; JMP Pro 12.0.1 software) method was used on univariate data, such as MZP diversity,

species richness and total density. When significant differences were found, *post-hoc* Tukey tests were conducted to see which groups differed from each other. For some objectives, linear regressions were used to test for the effect of an independent variable (ballast water age) on MZP community parameters. All linear regression results are given using the adjusted R^2 , which takes into account the amount of predictor variables in the model (Ayanso, 2014). All univariate analyses were conducted using JMP® Pro 12.0.1 software. The level of significance used for all tests was $P = 0.05$. Basic assumptions for parametric tests were tested using the Shapiro-Wilk test for the normality of residuals and visual evaluation for the homogeneity of variance.

Data transformation and dependant variables

Depending on the analyses, MZP density data were transformed in various ways to answer different questions. Raw data were left untransformed for information concerning the whole MZP community's species composition based on densities. Fourth root transformations were done to give rare taxa a greater impact. Presence/absence transformation was required to observe differences based on species occurrence only. Furthermore, all transformations were made on data including and excluding copepod nauplii. As copepod larval stages are difficult to identify to the species level with traditional taxonomy, they were counted as individuals in the "copepod nauplii" category, which included freshwater as well as marine copepod nauplii. In the case when these larvae form an important part of the MZP community, they can sometimes mask the signal of difference between freshwater and marine samples (exchanged and unexchanged tanks, in this case). For this reason, when results did not show any significant results, data without copepod nauplii was alternatively used to see if results would differ. This modification can however take away an important part of the total MZP density. Some analyses were also conducted on another dependant variable, MZP diversity, which was expressed using the absolute values of the Shannon-Wiener Diversity Index (H') (Estrada *et al.*, 2012). This index is defined as a measure of species composition, both in terms of the number of species and

their relative densities (Harvey *et al.*, 2000). It incorporates both species richness and evenness. The value of H' (with natural logarithm) ranges from 1.5 for communities with low species richness and evenness to 3.5 for communities with high species richness and evenness. Diversity data were left untransformed, as suggested in the literature, because the interpretation of a transformed index would be unpractical (McDonald, 2003). Finally, some analyses were performed using MZP species richness (S), which relates to the total number of species present (Estrada *et al.*, 2012). This index of diversity is widely used in ecology, because it is more sensitive to the presence of rare taxa than other indices. We used this index to detect MZP community changes (Harvey *et al.*, 2000).

Obj. #1: Evaluate summer monthly variability in MZP communities present in the ballast water tanks of the M/V Arctic in the port of origin (Quebec City harbor)

A one-way replicated permutational multivariate analysis of variance (PERMANOVA) with the fixed factor “Month” with five levels (June, July, August, September, October) was conducted to compare MZP communities (species composition based on densities) between months (Anderson, 2001; Clarke and Gorley, 2006). The input variable was the Bray-Curtis similarity matrix of community density data. Since the three tanks (“Control”, “Site 1” and “Site 2”) for the experiment were filled up in Quebec City harbor at the beginning of each trip, they were treated as replicates for each of the five sampled months. Homogeneity of variance was tested by using the permutation analysis of multivariate dispersion (PERMDISP) routine before performing PERMANOVA (Anderson, 2001). The “Unrestricted permutation of raw data” permutation method was used for this analysis, as recommended by the software itself for one-way designs.

A one-way ANOVA analysis on total MZP density (ind. m^{-3}) was also performed. The fixed factor “Month” had five levels (June, July, August, September and October). Densities were fourth root transformed to meet basic normality of residuals and homogeneity of variance ANOVA assumptions (Quinn and Keough, 2002). A *post-hoc* Tukey test was later applied to determine which months differed. The same one-way

ANOVA analysis was also performed on MZP diversity, to observe a potential effect of seasonality on plankton diversity, which was expressed using the Shannon-Wiener diversity index. Finally, the same analysis was done with MZP species richness (number of species present). Data was left untransformed, as ANOVA assumptions were met. For all analyses, Quebec City harbor values were excluded, as they sometimes differed greatly from ballast water tank values and because samples were collected using a different method (0.5 m plankton net). Divergence between harbor and tank samples was probably due to the tanks being filled progressively, several days before sampling. These ballast waters had already several days of age when we initially sampled them and for this reason they should not be expected to be comparable with harbor values. Analyses were focused on ballast tank values because they represent the true risks of introduction, since their contents are deballasted in Deception Bay.

Analyses concerning seasonal changes in risks of MZP introduction were focused only on MZP communities of ballast tanks in the port of origin (Quebec City harbor), since this was the only replicated site. Sampling at all other sites (Exchange sites 1 & 2, and Deception Bay harbor) was unreplicated (due to operational constraints of the vessel only one tank was available for each treatment), so we could not test for the effect of seasonality. Testing for seasonal variability requires replicates for each month, so no effect of seasonality on BWE efficacy or MZP survival was tested (Alain Caron, professor/statistician at UQAR, personal communication). Furthermore, Quebec City harbor values were excluded from all analyses because strong currents made the calculated filtered volumes less reliable due to wide net angle, possibly influencing total organism density. The bottom of the water column was difficult to detect during sampling, so considerable time was needed to lower the net to the bottom. Time during which strong water inflow went through the net and was filtered without any measurement (no flowmeter was used), obviously resulting in inaccurate filtered volume and thus overestimated total MZP density.

Obj. #2A: Evaluate the effect of ballast water age on MZP diversity and total density

To test the effect of ballast water age on MZP mortality, total density from the control tanks during June, July and August were used, as these were the only trips where data were available at all stations along the ship's route (five stations per trip). Densities were converted to percentages (ranks) to eliminate heterogeneity between the three trips. Ballast water age was estimated with the initial sampling in Quebec City corresponding to 0 days of age. A linear regression between ballast water age (days) and total MZP density (%), as well as diversity (Shannon-Wiener index) was made to observe a possible effect of aging on these two variables. These three trips could not be treated as replicates because they were done on the same ship (repeated measures). For this reason, the "Month" fixed factor with three levels ("June", "July" and "August") was added in the model. However, the interaction "Month x Ballast water age" was not included, due to the lack of replication. The basic assumption concerning the normality of residuals was not met for the analysis on total MZP density. Data transformations were not possible, since densities were already expressed in percentages (ranks). The residuals were not normally distributed because of an important outlier in the month of July. The control tank at Site 1 before exchange had an increase in MZP density corresponding to 511% of initial density, probably due to the hatching of copepod nauplii. In fact, copepod nauplii went from 3055 to 19 774 ind. m⁻³. *Eurytemora carolleae* stage I copepodids, closely related to the nauplius stage, also increased from 204 to 3195 ind. m⁻³. This shows strong evidence that the drastic increase of total MZP density between Quebec City and Site 1 before exchange was due to a massive hatching of *Eurytemora carolleae* nauplii. For this reason, a decision was made to remove this outlier from July data. Normality of residuals was then met and the ANOVA was performed.

Obj. #2B: MZP communities of the control tanks in Quebec City and Deception Bay

A multivariate two-way PERMANOVA analysis was used to see if control tanks differed in MZP density/species composition between the beginning and the end of each trip. The fixed factor “Station” had two levels, “Quebec City” and “Deception Bay”. The fixed factor “Month” had three levels, “June”, “July” and “August”. Raw density data including copepod nauplii and presence/absence data including copepod nauplii were used for this analysis.

Obj. #2C: Comparison of MZP communities in exchanged tanks immediately after exchange and upon arrival in Deception Bay

We conducted a three-way PERMANOVA analysis on Site 1 and Site 2 tanks after exchange and before arrival in Deception Bay. Our goal was to see if the communities ballasted in the exchange zones would stay the same after several days, upon deballasting in Deception Bay. The fixed factor “Station” analysis included two levels, “After exchange” and “Deception Bay”. The fixed factor “Tank” had two levels, “Site 1” and “Site 2”. Since we used repeated measures (monthly trips) and no replicates for this analysis, the “Month” factor was also added in the analysis, with the three levels “June”, “July” and “August”. Raw density data including copepod nauplii and presence/absence data including copepod nauplii were used.

Obj. #2D: Effect of ballast water exchange on MZP communities (BACI design)

We used an unreplicated randomized block design for these multivariate analyses. Sampling was conducted during three summer trips (June, July, August), acting as our blocks. Ballast water studies in partnership with the industry often require opportunistic and limited sampling time, thus a replicated design was not possible. The ship being the vector of NIS introductions, multiple ships would be required for a replicated approach. A pseudoreplicated approach would have required several tanks for each treatment, which was impossible due to operational constraints. The three trips could not be treated as

replicates since they were made with the same vessel. For this reason, each statistical analysis included the “Month” block, which corresponded to each monthly trip. This analysis was designed as a Before-After-Control-Impact (BACI), consisting of three crossed factors (Keough and Mapstone, 1995; Smith *et al.*, 1993). The “Month” fixed factor had three levels, “June”, “July”, “August”, corresponding to each trip where sampling was continuous. The “Treatment” fixed factor had three levels, “Before Exchange”, “After Exchange” and “Control”. The “Site” fixed factor had two levels, “Site 1” and “Site 2”; corresponding respectively to the Straits of Jacques Cartier and Belle Isle. All the interactions between these three factors were also added in the analysis. The goal of this analysis was to see the effect of BWE in our two sites, relative to the control tanks. Fourth root transformed density data including copepod nauplii and presence/absence data including copepod nauplii were used for this analysis.

Obj. #2E: Comparison of MZP communities in the two ballast water exchange zones (Sites 1 & 2)

A two-way PERMANOVA analysis was conducted on data from the exchanged tanks immediately after exchange. Our goal was to see if MZP communities would differ between the two tested ballast water exchange zones. The “Site” fixed factor had two levels, “Site 1” and “Site 2”, corresponding respectively to the Straits of Jacques Cartier and Belle Isle. The “Month” fixed factor had three levels, “June”, “July” and “August”.

Obj. #2F: MZP diversity and total density of exchanged and control tanks upon arrival in Deception Bay

We performed a two-way ANOVA to see if total MZP density and diversity would differ between the exchanged and the control tanks upon arrival in Deception Bay. The “Treatment” fixed factor had two levels, “Exchanged” and “Control”. The “Exchanged” level included both Site 1 and Site 2 tanks; they were undifferentiated for this analysis since our goal was to compare control and exchanged tanks. In other words, the two sites were treated as a single one, being the Gulf of St. Lawrence. Since the trips were unreplicated, we added the “Month” block, with three levels, “June”, “July” and “August”. A third factor, “Month*Treatment”, would have been the interaction between the two first factors. However, we were not able to construct the model because of issues with degrees of freedom. This interaction had to be eliminated due to the lack of replication, which is required to evaluate its error. Total MZP density was fourth root transformed to give rare taxa a greater impact. Our goal with this analysis was to prove statistically that exchanging ballast water increased MZP density.

Obj. #2G: MZP communities of the three tanks upon arrival in Deception Bay

A two-way PERMANOVA analysis was performed to see if the MZP community would vary between the three ballast tanks before deballasting in Deception Bay. The fixed factor “Tank” had three levels, “Site 1”, “Site 2” and “Control”. To evaluate the effect of the repeated measures, we also added the fixed factor “Month”, with three levels, “June”, “July” and “August”. Copepod nauplii were excluded from the MZP density/species composition analysis, because they masked the differences between freshwater and marine ballast water tanks. Raw density data without copepod nauplii and presence/absence data with copepod nauplii were used for this analysis.

1.3. RESULTS

1.3.1. OBJ. #1: EVALUATE SUMMER MONTHLY VARIABILITY IN MZP COMMUNITIES PRESENT IN THE BALLAST WATER TANKS OF THE M/V ARCTIC IN THE PORT OF ORIGIN (QUEBEC CITY HARBOR).

Summer monthly variability of MZP communities and total density in the three tanks filled in Quebec City harbor

Total MZP density differed significantly among months (ANOVA; $P < 0.001$; **Table 3**). The month of August differed from all the other months, with much higher total MZP density (**Table 4**, **Figure 9**), and June had significantly lower total MZP density than September (*post-hoc* Tukey test) (**Table 4**, **Figure 9**). Species composition of MZP based on densities showed relatively clear groupings of replicates by month (**Figure 10**), whereas no particular pattern was observed based on MZP species presence/absence data (**Figure 11**). This pattern was supported by the statistical analyses, which also showed monthly differences in MZP species composition based on densities (raw density data including copepod nauplii; PERMANOVA, $P = 0.001$; ANOSIM, R-value = 0.778) and occurrence (presence/absence data including copepod nauplii; PERMANOVA, $P = 0.001$; ANOSIM, R-value = 0.629) (**Table 5**). Copepod nauplii, *Bosmina longirostris*, mussel veligers and *Eurytemora carolleae* (in order of importance) were responsible for almost 99% of the dissimilarity between months of strongest dissimilarity (June and August), with lower abundance in June and higher abundance in August (raw density data including copepod nauplii; SIMPER, 98.48 %) (**Table 6**). June and August always showed high dissimilarity with all the other months (**Table 7**). This is mainly explained by lower density in June and higher density in August of *Eurytemora carolleae*, of copepod nauplii, of the cladoceran *Bosmina longirostris* and of mussel veligers, compared to other months (raw density data including copepod nauplii; SIMPER) (**Table 6**).

Table 3. ANOVA table of results for Objective #1

Obj. #1 - Summer monthly variability of total MZP density of the three tanks filled in Quebec City harbor (June-October 2015)

ANOVA – Fourth root transformed total MZP density

Source	df	SS	Pseudo-F	Prob. > F
Month	4	232.06187	30.7825	< 0.0001*
Residuals	10	18.84690		
Total	14	250.90877		

Obj. #1 - Summer monthly variability of MZP diversity of the three tanks filled in Quebec City harbor (June-October 2015)

ANOVA – Shannon-Wiener (H')

Source	df	SS	Pseudo-F	Prob. > F
Month	4	0.5202000	2.0159	0.1681
Residuals	10	0.6451333		
Total	14	1.1653333		

Obj. #1 - Summer monthly variability of MZP species richness of the three tanks filled in Quebec City harbor (June-October 2015)

ANOVA – Shannon-Wiener (H')

Source	df	SS	Pseudo-F	Prob. > F
Month	4	248.26667	5.7117	0.0117*
Residuals	10	108.66667		
Total	14	356.93333		

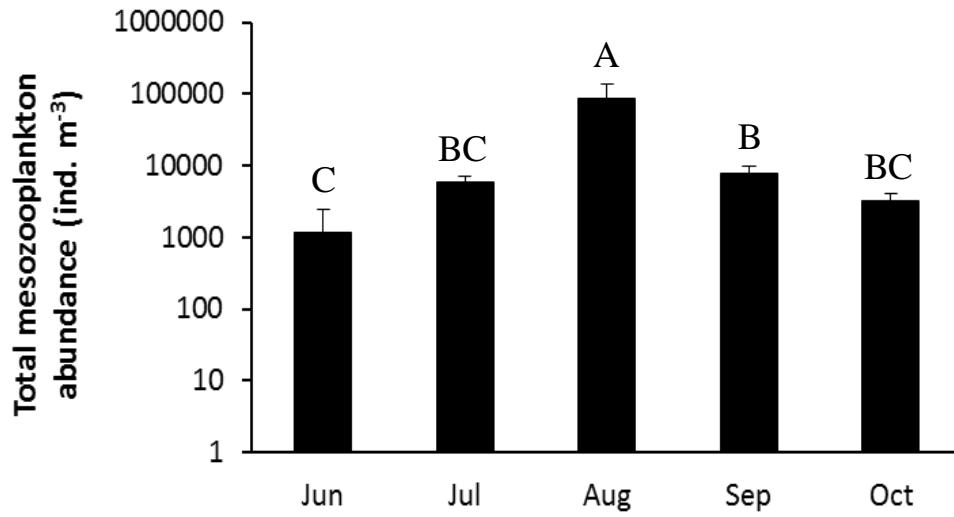


Figure 9. Mean MZP density of the three ballast water tanks filled in Quebec City harbor. Levels not connected by the same letters are significantly different ($P < 0.05$)

Table 4. Results of *post-hoc* Tukey test for total MZP density for the three tanks filled in Quebec City harbor. Levels not connected by the same letters are significantly different ($P < 0.05$)

Level		Least mean square
Aug	A	16.895557
Sep	B	9.418720
Jul	B C	8.703069
Oct	B C	7.469093
Jun	C	5.268406

Table 5. PERMANOVA table of results and pairwise tests for Objective #1

Obj. #1 - Comparison of MZP communities observed in the three tanks filled in Quebec City harbor (June-October 2015)

PERMANOVA – Raw density data including copepod nauplii (untransformed)

Source	df	SS	Pseudo-F	P(perm)	perms	P(MC)
Months	4	29708	8.3833	0.0001*	9881	0.0001
Residuals	10	8859.2				
Total	14	38567				

PERMANOVA – Presence/Absence data including copepod nauplii

Source	df	SS	Pseudo-F	P(perm)	perms	P(MC)
Months	4	9849.7	2.7585	0.0001*	9843	0.0005
Residuals	10	8926.8				
Total	14	18777				

Pair-wise tests – Raw density data including copepod nauplii (untransformed)

Groups	t	P(perm)	Unique perms	P (MC)
June - July	2.3879	0.1036	10	0.0238*
June - August	2.6387	0.0990	10	0.0148*
June - September	2.6019	0.0940	10	0.0175*
June - October	1.9532	0.0966	10	0.0484*
July - August	4.7059	0.0951	10	0.0019*
July - September	2.8896	0.1020	10	0.0095*
July - October	2.1212	0.1001	10	0.0551
August - September	4.5084	0.1014	10	0.0018*
August - October	3.9586	0.1011	10	0.0027*
September - October	3.1577	0.0967	10	0.0085*

Pair-wise tests – Presence/Absence data including copepod nauplii

Groups	t	P(perm)	Unique perms	P (MC)
June - July	1.2517	0.1997	10	0.2237
June - August	2.4679	0.0976	10	0.0160*
June - September	1.7724	0.0989	10	0.0601
June - October	1.6948	0.0989	10	0.0735
July - August	1.7588	0.0991	10	0.0745
July - September	1.2602	0.1017	10	0.2218
July - October	1.4358	0.1020	10	0.1335
August - September	1.9996	0.0954	10	0.0383*
August - October	1.8435	0.0982	10	0.0523
September - October	1.3632	0.2048	10	0.1687

Table 6. Complete SIMPER table of results for Objective #1

	Group June	Group July				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Veligers	42,93	2854,91	42,55	3,73	50,69	50,69
Copepod nauplii	459,82	1785,62	20,23	1,56	24,1	74,79
<i>Eurytemora carolleeae</i>	14,55	598,68	8,86	3,25	10,55	85,34
<i>Diacyclops thomasi</i>	432,02	15,45	5,11	0,77	6,09	91,44
	Group June	Group August				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Copepod nauplii	459,82	46704,4	50,27	7,11	51,05	51,05
<i>Bosmina longirostris</i>	45,57	19761,01	22,13	3,02	22,47	73,51
Veligers	42,93	10993,71	12,83	7,94	13,03	86,54
<i>Eurytemora carolleeae</i>	14,55	10452,83	12,26	4,49	12,45	98,99
	Group July	Group August				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Copepod nauplii	1785,62	46704,4	45,74	5,94	53,14	53,14
<i>Bosmina longirostris</i>	292,4	19761,01	20,52	3,01	23,84	76,98
<i>Eurytemora carolleeae</i>	598,68	10452,83	10,79	4,62	12,53	89,51
Veligers	2854,91	10993,71	8,52	8,44	9,9	99,41
	Group June	Group September				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Veligers	42,93	5524,97	62,34	4,63	69,63	69,63
<i>Bosmina longirostris</i>	45,57	1101,4	10,72	1,21	11,97	81,61
Copepod nauplii	459,82	769,34	6,14	1,45	6,86	88,46
<i>Diacyclops thomasi</i>	432,02	0	4,23	0,79	4,72	93,19
	Group July	Group September				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Veligers	2854,91	5524,97	19,54	4,33	50,84	50,84
Copepod nauplii	1785,62	769,34	8,35	1,27	21,73	72,57
<i>Bosmina longirostris</i>	292,4	1101,4	5,62	0,9	14,62	87,19
<i>Eurytemora carolleeae</i>	598,68	258,47	2,55	2,51	6,63	93,81
	Group August	Group September				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Copepod nauplii	46704,4	769,34	45,81	6,22	56,31	56,31
<i>Bosmina longirostris</i>	19761,01	1101,4	19,02	2,79	23,38	79,69
<i>Eurytemora carolleeae</i>	10452,83	258,47	10,91	4,77	13,41	93,1
	Group June	Group October				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Copepod nauplii	459,82	1393,97	26,19	1,57	33,18	33,18
Veligers	42,93	858,38	21,86	1,66	27,69	60,87
<i>Bosmina longirostris</i>	45,57	518,56	10,72	1,77	13,58	74,45
<i>Diacyclops thomasi</i>	432,02	0	7,91	0,84	10,02	84,47
<i>Eurytemora carolleeae</i>	14,55	247,56	6,15	1,84	7,79	92,26
	Group July	Group October				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Veligers	2854,91	858,38	22,58	4,81	49,79	49,79
Copepod nauplii	1785,62	1393,97	11,23	1,37	24,75	74,55
<i>Eurytemora carolleeae</i>	598,68	247,56	3,98	2,65	8,79	83,33
<i>Bosmina longirostris</i>	292,4	518,56	3,27	1,25	7,22	90,55
	Group August	Group October				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Copepod nauplii	46704,4	1393,97	47,75	6,48	51,82	51,82
<i>Bosmina longirostris</i>	19761,01	518,56	20,93	2,94	22,71	74,54
<i>Eurytemora carolleeae</i>	10452,83	247,56	11,62	4,58	12,61	87,15
Veligers	10993,71	858,38	11,42	9,38	12,39	99,54
	Group September	Group October				
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Veligers	5524,97	858,38	42,46	6,34	69,11	69,11
Copepod nauplii	769,34	1393,97	7,96	1,51	12,95	82,06
<i>Bosmina longirostris</i>	1101,4	518,56	6,78	1,04	11,04	93,09

Table 7. Average dissimilarity in MZP species composition based on densities of the three tanks filled in Quebec City harbor (raw density data including copepod nauplii, SIMPER)

	June	July	August	September	October
June					
July	83.95				
August	98.48	86.08			
September	89.52	38.43	81.35		
October	78.95	45.35	92.13	61.44	

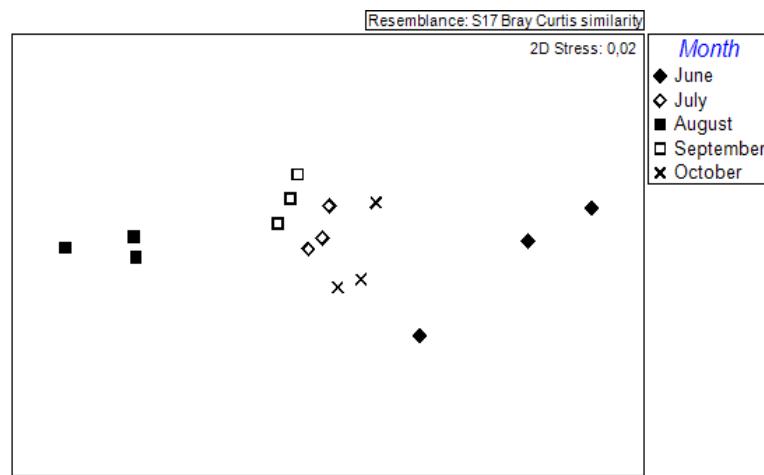


Figure 10. Non-metric multi-dimensional scaling (MDS) ordination of the three ballast water tanks filled in Quebec City harbor (Raw density data including copepod nauplii)

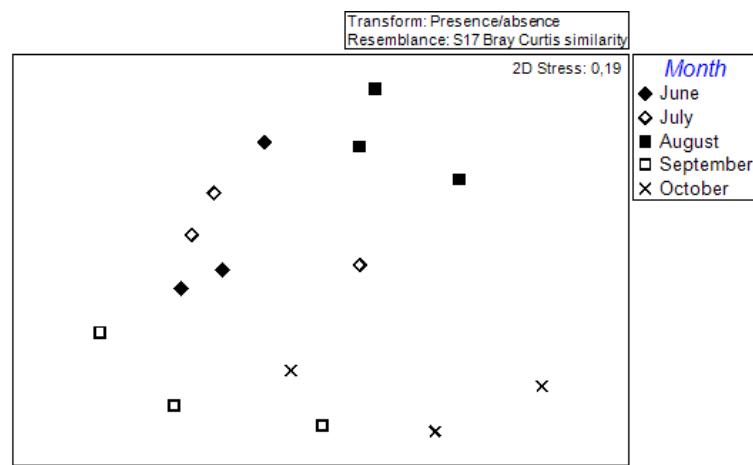


Figure 11. Non-metric multi-dimensional scaling (MDS) ordination of the three ballast water tanks filled in Quebec City harbor (Presence/absence data including copepod nauplii)

Summer monthly variability of MZP diversity in the three tanks filled in Quebec City harbor

There was high variability in diversity among replicates and no clear pattern between months (**Figure 12**). No monthly differences in MZP diversity were observed (ANOVA, P = 0.1681; **Table 3**).

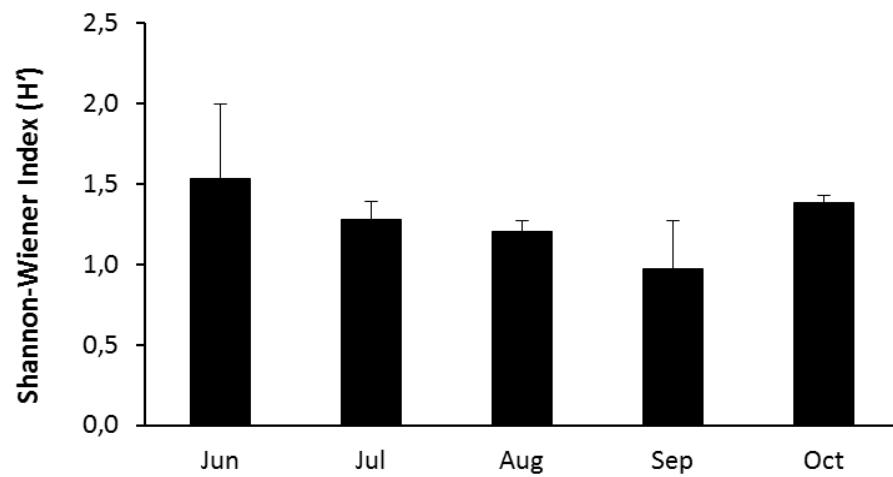


Figure 12. Mean MZP diversity of the three ballast water tanks filled in Quebec City harbor

Summer monthly variability of MZP species richness in the three tanks filled in Quebec City harbor

The month of August, also associated with the peak in total MZP density, had the lowest species richness of all months (**Figure 13**). Monthly differences in MZP species richness were observed (ANOVA, $P = 0.0117$; **Table 3**). The month of August differed in terms of species richness from all the other months, except October (*post-hoc* Tukey test, $P < 0.05$) (**Table 8**).

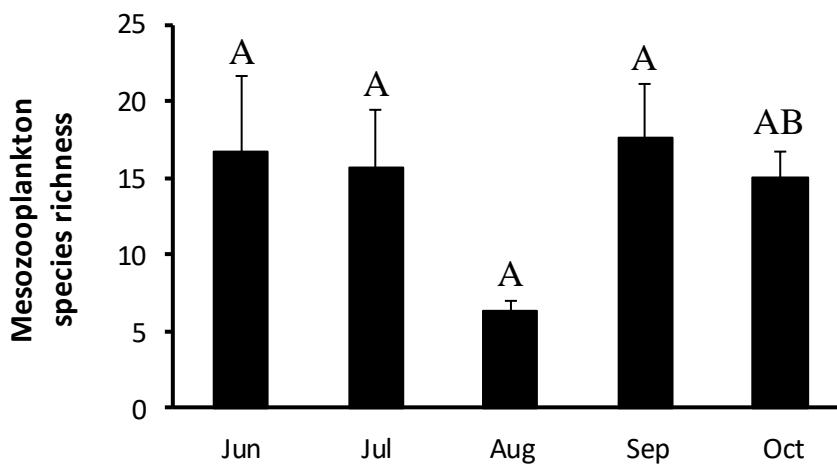


Figure 13. Mean MZP species richness of the three ballast ballast water tanks filled in Quebec City harbor

Table 8. Results of *post-hoc* Tukey test for MZP species richness for the three tanks filled in Quebec City harbor. Levels not connected by the same letters are significantly different ($P < 0.05$)

Level		Least Mean Square
Sep	A	17.666667
Jun	A	16.666667
Jul	A	15.666667
Oct	A B	15.000000
Aug	B	6.333333

1.3.2. OBJ. #2: EVALUATE THE EFFECT AND EFFICACY OF VOLUNTARY BALLAST WATER EXCHANGE (BWE) PERFORMED BY THE *M/V ARCTIC* IN THE GULF OF ST. LAWRENCE

A. Effect of ballast water age on MZP diversity and total density

Punctual increases in total MZP density in control tanks were observed en route during all three trips (**Figure 14**). Despite this, ballast water age was negatively correlated with total MZP density (Linear regression, $R^2 = -0.472$, $P = 0.033$; **Table 9**). However, there was an additional significant effect of the trip in our model of density (ANOVA, $P = 0.0391$; **Table 9**) with July and August differing (Tukey *post-hoc* test; $P < 0.05$, **Table 10**). A weak, albeit non-significant negative correlation was also observed between ballast water age and MZP diversity (Linear regression, $R^2 = -0.3314$; $P = 0.0734$; **Table 9**) (**Figure 15**).

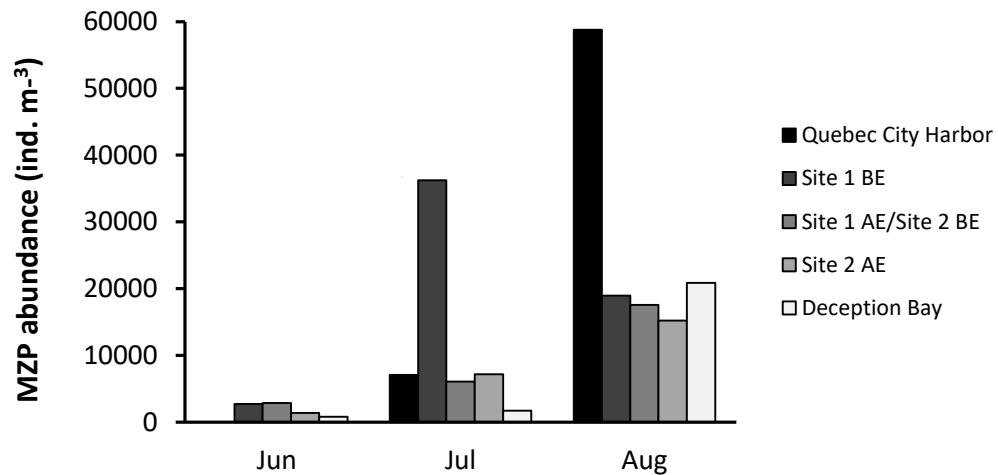


Figure 14. Total MZP density in the control tanks at all stations during June, July and August trips (Site 1 = Jacques Cartier Strait; Site 2 = Strait of Belle Isle; BE = Before Exchange; AE = After Exchange; Jun = June; Jul = July; Aug = August)

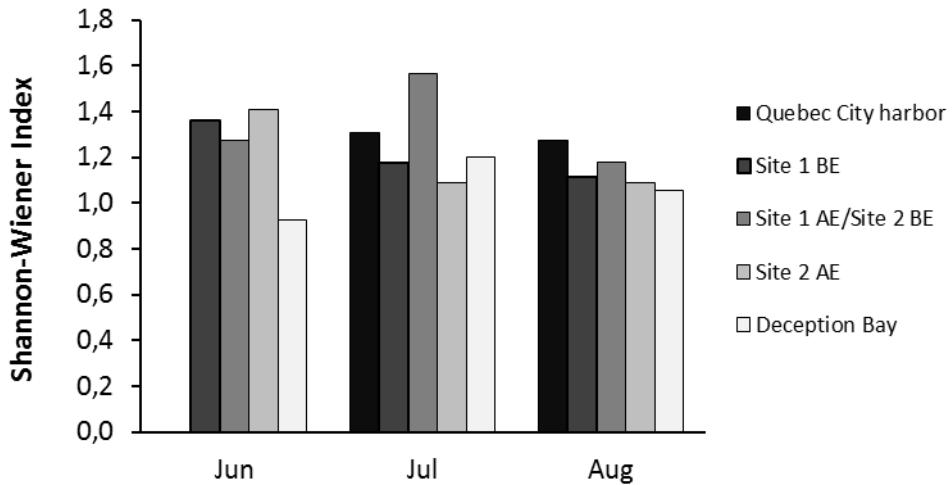


Figure 15. Continuous MZP diversity in the control tanks during June, July and August trips (Site 1 = Jacques Cartier Strait; Site 2 = Strait of Belle Isle; BE = Before Exchange; AE = After Exchange; Jun = June; Jul = July; Aug = August)

Table 9. Linear regression table of results for Obj. #2A

Obj. #2A – Effect of ballast water age on total MZP density

Linear regression – Ballast water age and ranked total MZP density (%)

Source	df	SS	Pseudo-F	Prob. > F
Model	3	8958.803	4.5712	0.0330*
Residuals	9	5879.560		
Total	12	14838.363		

Test of effects

Source	df	SS	Pseudo-F	Prob. > F
Ballast water age	1	6139.8	9.3984	0.0134*
Months	2	6200.6	4.7457	0.0391*

Obj. #2A – Effect of ballast water age on MZP diversity

Linear regression – Ballast water age and MZP diversity (H')

Source	df	SS	Pseudo-F	Prob. > F
Model	3	0.1689	3.1488	0.0734
Residuals	10	0.1788		
Total	13	0.3477		

Test of effects

Source	df	SS	Pseudo-F	Prob. > F
Ballast water age	1	0.1252	7.0023	0.0245*
Month	2	0.1090	3.0472	0.0926

Table 10. Results of *post-hoc* Tukey test for the effect of the months in our ballast water age on total MZP density model. Levels not connected by the same letters are significantly different ($P < 0.05$)

Level		Least Mean Square
July	A	91.160659
June	A B	69.971667
August	B	34.954139

B. MZP communities of the control tanks in Quebec City and Deception Bay harbors

The control tanks at the two stations had similar MZP species composition based on densities at the beginning and end of June and August trips. July controls differed more between stations because of higher mortality (76%, compared to 71% for June and 64% for August) experienced by copepod nauplii, *Bosmina longirostris* and mussel veligers, which contributed 95% of the dissimilarity between stations (raw density data including copepod nauplii; SIMPER) (**Figure 16**). Both June and July trips showed greater dissimilarity in terms of species occurrence between stations than the August trip (**Figure 17**). However, no significant differences between stations were detected based on statistical comparisons for density (raw density data including copepod nauplii; PERMANOVA, $P = 0.4209$; ANOSIM, R-value = -0.185) and species occurrence (presence/absence data including copepod nauplii; PERMANOVA, $P = 0.418$; ANOSIM, R-value = -0.148) (**Table 11**). Intra-group variability was greater than inter-group variability (ANOSIM; -0.185), meaning that control tanks showed greater difference between trips than between the two stations (groups). Copepod nauplii, *Bosmina longirostris* and mussel veligers (in order of importance) were responsible for 78% of the dissimilarity between stations (raw density data including copepod nauplii; SIMPER, 71.33). Since these three taxa were dominant, they were highly affected by mortality throughout the trips. June and August were the two most dissimilar months, with copepod nauplii, *Bosmina longirostris*, mussel veligers and *Eurytemora carolleae* (in order of importance) being responsible for 97% of dissimilarity (raw density data including copepod nauplii; SIMPER, 94.18).

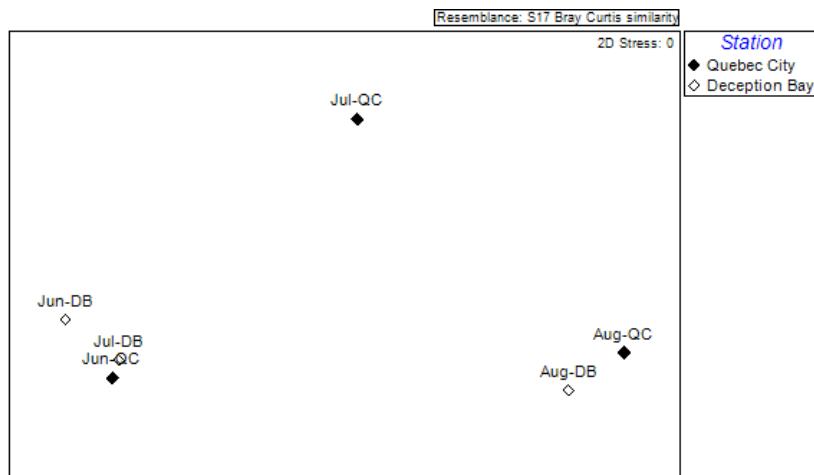


Figure 16. Non-metric multi-dimensional scaling (MDS) ordination of the control tanks in Quebec City harbor and before deballasting in Deception Bay for the June, July and August trips (raw data including copepod nauplii). QC = Quebec City harbor; DB = Upon arrival in Deception Bay; Jun = June; Jul = July; Aug = August

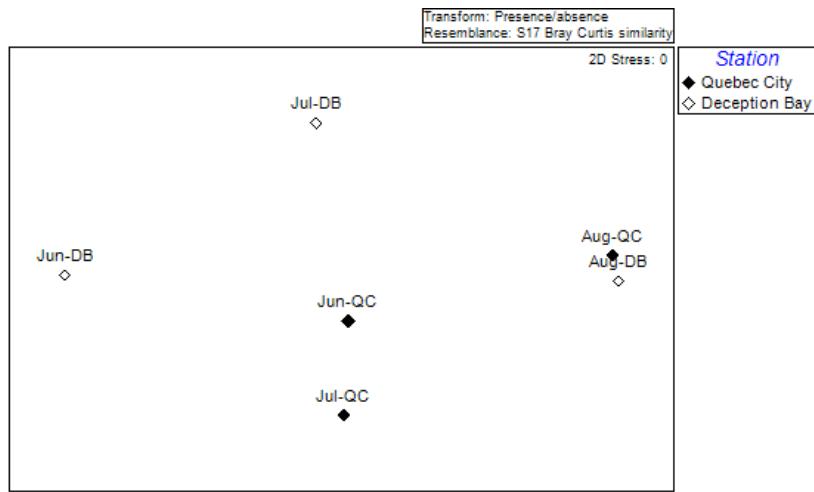


Figure 17. Non-metric multi-dimensional scaling (MDS) ordination of the control tanks in Quebec City harbor and upon arrival in Deception Bay for the June, July and August trips (Presence/absence data including copepod nauplii). QC = Quebec City harbor; DB = Upon arrival in Deception Bay; Jun = June; Jul = July; Aug = August

Table 11. PERMANOVA table of results for Obj. #2B

Obj. #2B - Comparison of MZP communities of control tanks in Quebec City harbor and Deception Bay (June-August 2015)

PERMANOVA – Raw density data including copepod nauplii (untransformed)						
Source	df	SS	Pseudo-F	P(perm)	perms	P(MC)
Station	1	1706.1	1.0908	0.3617	38	0.4209
Month	2	9850.8	3.1492	0.1372	60	0.1159
Residuals	2	3128.1				
Total	5	14685				

PERMANOVA – Presence/Absence data including copepod nauplii						
Source	df	SS	Pseudo-F	P(perm)	perms	P(MC)
Station	1	1200.5	1.0945	0.3680	37	0.4180
Month	2	4374.1	1.995	0.1346	60	0.2061
Residuals	2	2193.7				
Total	5	7768.2				

C. MZP communities of exchanged tanks immediately after exchange and upon arrival in Deception Bay

No differences in MZP species composition based on densities (raw data including nauplii; PERMANOVA, $P = 0.5689$; ANOSIM, R -value = -0.137; **Table 12**) or species occurrence (presence/absence data including copepod nauplii; PERMANOVA, $P = 0.1232$; ANOSIM, R -value = 0.222; **Table 12**) were observed between exchange and arrival in Deception Bay. There were also no significant differences between trips (raw data including nauplii; PERMANOVA, $P = 0.5689$). Tanks exchanged in Site #2 always show greater similarity between stations than tanks exchanged in Site #1 (**Figure 18**), even though there was no significant difference due to the site of exchange (**Table 12**).

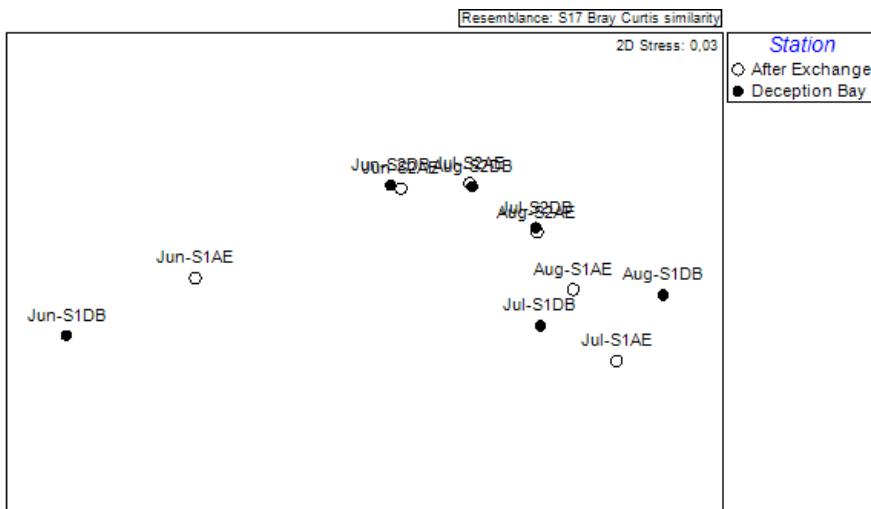


Figure 18. Non-metric multi-dimensional scaling (MDS) ordination of the two exchanged ballast water tanks after exchange and upon arrival in Deception Bay (raw data including copepod nauplii). Jun = June; Jul = July; Aug = August; S1 = Site 1; S2 = Site 2; AE = After exchange; DB = Upon arrival in Deception Bay

Table 12. PERMANOVA table of results for Obj. #2C

Obj. #2C - Comparison of MZP communities in exchanged tanks after exchange and upon arrival in Deception Bay (June-August 2015)

PERMANOVA – Raw density data including copepod nauplii (untransformed)

Source	df	SS	Pseudo-F	P(perm)	perms	P(MC)
Station (St)	1	280.27	0.7124	0.5689	9431	0.5483
Tank (Ta)	1	3401.1	8.6444	0.0565	9432	0.0548
Month (Mo)	2	6061.3	7.7029	0.0788	6941	0.0565
St x Ta	1	49.892	0.1268	0.8015	2070	0.8346
St x Mo	2	467.37	0.5940	0.7117	9955	0.7047
Ta x Mo	2	2226.5	2.8295	0.1625	9954	0.1597
Residuals	2	786.88				
Total	11	13273				

PERMANOVA – Presence/Absence data including copepod nauplii

Source	df	SS	Pseudo-F	P(perm)	perms	P(MC)
Station (St)	1	1692.0	3.3711	0.1232	9424	0.1227
Tank (Ta)	1	1468.5	2.9258	0.1508	9416	0.1490
Month (Mo)	2	2229.8	2.2212	0.2042	6898	0.2031
St x Ta	1	283.47	0.5648	0.6653	2094	0.6510
St x Mo	2	836.65	0.8335	0.5899	9950	0.5877
Ta x Mo	2	1654.2	1.6478	0.2803	9941	0.2787
Residuals	2	1003.8				
Total	11	9168.4				

D. Effect of ballast water exchange on MZP communities (BACI design)

MZP species composition based on densities (fourth root transformed) showed groupings of unexchanged and control tanks, as well as a grouping of exchanged tanks (**Figure 19**). Only the PERMANOVA analysis performed on presence/absence transformed data had no significant effect of interactions between factors (**Table 13**). Thus, only these results will be interpreted here, since interpretation with significant interactions between factors is rather complex (meaning that the response variable is also influenced by the interaction between the predictor variables in the model). Differences in MZP species occurrence (presence/absence data including copepod nauplii; PERMANOVA, $P = 0.0026$; ANOSIM, Global R-value = 0.546; **Table 13**) between Control and After exchange treatments were observed. However, these differences were also observed between trips (presence/absence data including copepod nauplii; PERMANOVA, $P = 0.0006$; ANOSIM, Global R-value = 0.303; **Table 13**), due to seasonal differences in communities and density. *Oithona similis*, *Bosmina longirostris*, mussel veligers, *Microsetella norvegica* and *Eurytemora carolleeae* (in order of importance) were responsible for 74% of the dissimilarity between control and exchanged tanks (raw density data excluding copepod nauplii; SIMPER, 90.47). *Bosmina longirostris*, *Eurytemora carolleeae*, mussel veligers, *Oithona similis* and *Microsetella norvegica* were responsible for 82% of dissimilarity between the two most dissimilar months, June and August (raw density data excluding copepod nauplii; SIMPER, 92.61).

Table 13. PERMANOVA table of results and pair-wise tests for Obj. #2D (BACI design)

Obj. #2D – Effect of ballast water exchange (June-August 2015)					
PERMANOVA – Presence/Absence data including copepod nauplii					
Source	df	SS	Pseudo-F	P(perm)	perms
Treatment (Tr)	2	16640	14.542	0.0026*	9927
Site (Si)	1	1040.6	1.8188	0.1557	9951
Month (Mo)	2	7492.6	6.5482	0.0006*	9955
Tr x Si	2	1490.3	1.3024	0.2868	9924
Tr x Mo	4	3185.6	1.3920	0.1834	9911
Si x Mo	2	1388.4	1.2134	0.3269	9935
Tr x Si x Mo	4	2063.5	0.9017	0.5992	9930
Residuals	6	3432.7			
Total	23	38944			

Pair-wise tests – Presence/Absence data including copepod nauplii					
Groups (Mo)	t	P(perm)	Unique perms		
June - July	1.7581	0.0504	9900		
June -August	3.9794	0.0043*	9903		
July - August	2.1476	0.0246*	9905		

Pair-wise tests – Presence/Absence data including copepod nauplii					
Groups (Tr)	t	P(perm)	Unique perms		
Before exc. - Control	1.325	0.1592	9751		
Before exc. - After exc.	No test				
Control – After exc.	4.8297	0.0009*	9747		

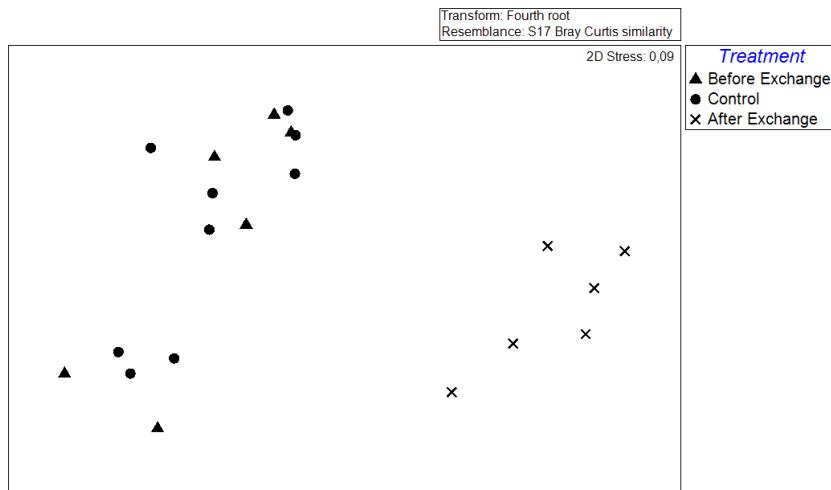


Figure 19. Non-metric multi-dimensional scaling (MDS) ordination of the three tanks before and after exchange at Site 1 & 2 during June, July and August trips (fourth root transformed data including copepod nauplii)

E. MZP communities in the two ballast water exchange zones (Sites 1 & 2)

Species composition based on MZP densities and species occurrence showed no clear grouping patterns (**Figures 20-21**) and no statistical differences were observed between sites based on densities (raw data including copepod nauplii; PERMANOVA, $P = 0.2307$; ANOSIM, Global R-value = 0.259; **Table 14**) and species occurrence (presence/absence data including copepod nauplii; PERMANOVA, $P = 0.3455$; ANOSIM, Global R-value = 0.185; **Table 14**).

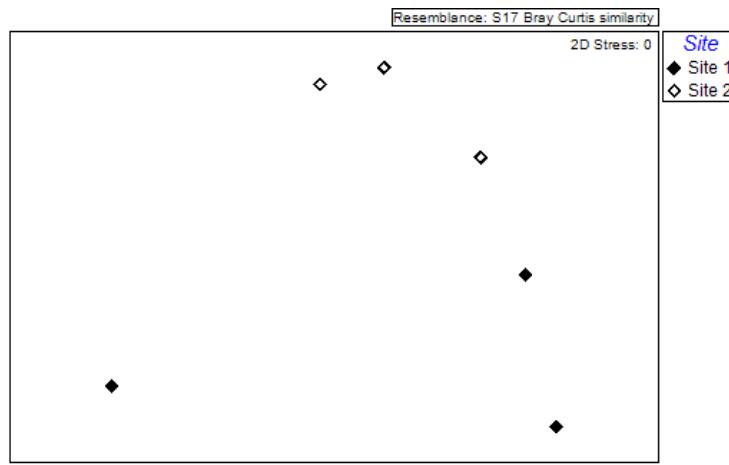


Figure 20. Non-metric multi-dimensional scaling (MDS) ordination of the two exchanged tanks immediately after exchange at Sites 1 & 2 during June, July and August trips (raw data including copepod nauplii)

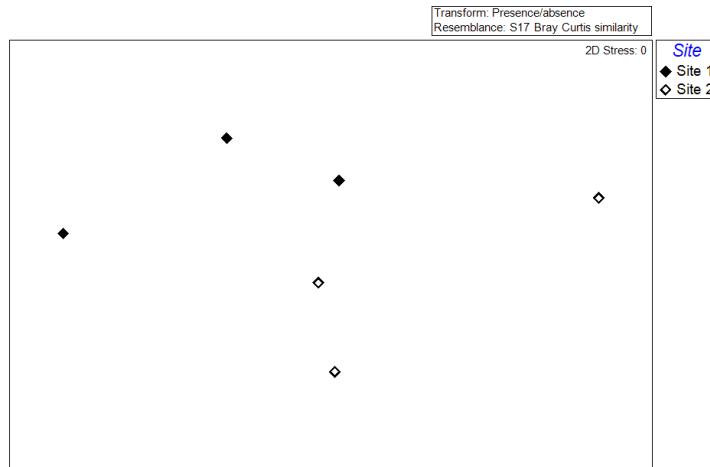


Figure 211. Non-metric multi-dimensional scaling (MDS) ordination of the two exchanged tanks immediately after exchange at Sites 1 & 2 during June, July and August trips (Presence/Absence data including copepod nauplii)

Table 14. PERMANOVA table of results for Obj. #2E

Obj. #2E - Comparison of MZP communities in the exchanged tanks after exchange (June-August 2015)

PERMANOVA – Raw density data including copepod nauplii (untransformed)

Source	df	SS	Pseudo-F	P(perm)	perms	P(MC)
Site	1	1486.0	1.9853	0.2307	38	0.2382
Month	2	2644.2	1.7663	0.3483	60	0.3214
Residuals	2	1497.0				
Total	5	5627.2				

PERMANOVA – Presence/Absence data including copepod nauplii

Source	df	SS	Pseudo-F	P(perm)	perms	P(MC)
Site	1	1129.3	1.3959	0.327	38	0.3455
Month	2	1580.0	0.9765	0.540	60	0.5107
Residuals	2	1618.0				
Total	5	4327.3				

F. MZP total density and diversity of exchanged and control tanks upon arrival in Deception Bay

Total density of remaining MZP propagules at Deception Bay was higher in exchanged tanks compared to controls (**Figure 22**). However, there was no clear tendency for MZP diversity (**Figure 23**). There was an effect of the treatment (ballast water exchange) on total MZP density (ANOVA, $P = 0.0358$; **Table 15**), but no effect of the month (ANOVA, $P = 0.0702$; **Table 15**), indicating that ballast water exchange increased total MZP density, regardless of the trip. There was no effect of the treatment on MZP diversity (ANOVA, $P = 0.9977$; $P < 0.05$, **Table 15**).

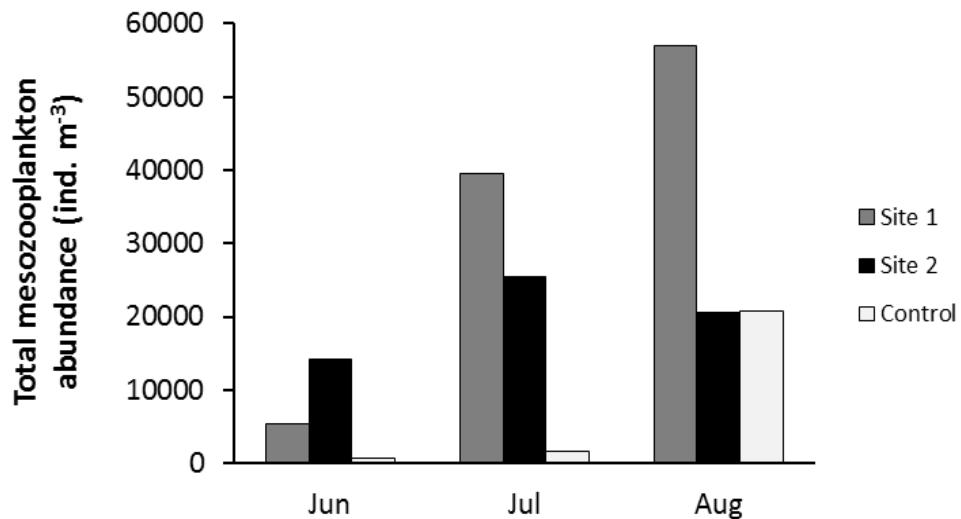


Figure 22. Total mesozooplankton density of the three tanks upon arrival in Deception Bay

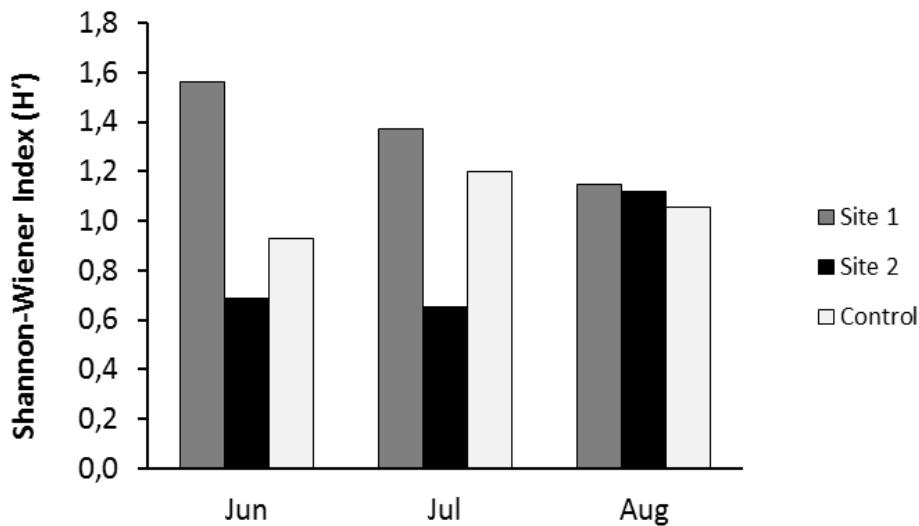


Figure 23. Mesozooplankton diversity of exchanged and control tanks upon arrival in Deception Bay

Table 15. ANOVA table of results for Obj. #2F

Obj. #2F - Total MZP density in exchanged and control tanks upon arrival in Deception Bay (June-August 2015)

ANOVA – Fourth root transformed total MZP density

Source	df	SS	Pseudo-F	Prob. > F
Model	3	74.046	6.4657	0.0358*
Residuals	5	19.087		
Total	8	93.133		

Test of effects

Source	df	SS	Pseudo-F	Prob. > F
Month	2	36.131	4.7325	0.0702
Treatment	1	37.915	9.9322	0.0253*

Obj. #2F - MZP diversity in exchanged and control tanks upon arrival in Deception Bay (June-August 2015)

ANOVA – Shannon-Wiener (H')

Source	df	SS	Pseudo-F	Prob. > F
Model	3	0.00549	0.0131	0.9977
Residuals	5	0.69819		
Total	8	0.70369		

Test of effects

Source	df	SS	Pseudo-F	Prob. > F
Treatment	1	0.00161	0.0115	0.9188
Month	2	0.00389	0.0139	0.9862

G. MZP communities of the three tanks upon arrival in Deception Bay

MZP species composition based on densities (excluding copepod nauplii) and species occurrence showed clear groupings of exchanged tanks and dissimilarity of control tanks between months (**Figures 24-25**). Differences in MZP species composition based on densities (raw density data without copepod nauplii; PERMANOVA, $P = 0.0475$; ANOSIM, Global R-value = 0.416; **Table 16**) and species occurrence (presence/absence data including copepod nauplii; PERMANOVA, $P = 0.0329$; ANOSIM, Global R-value = 0.49; **Table 16**) were observed between tanks. However, *post-hoc* comparisons did not show significant differences between pairs of tanks.

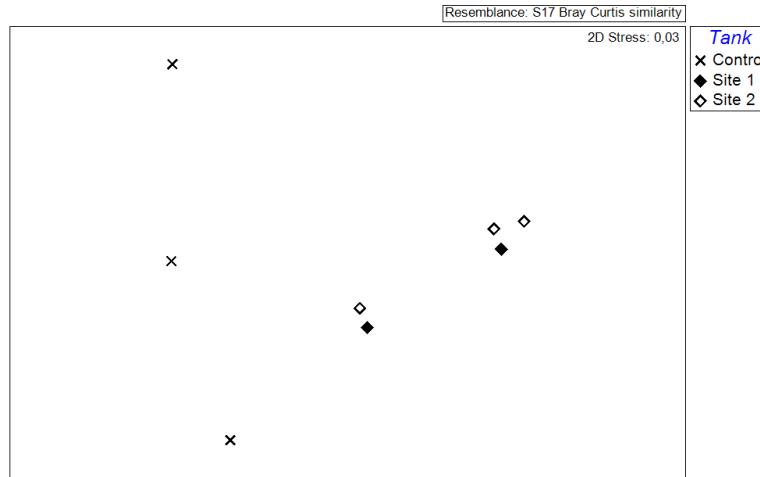


Figure 24. Non-metric multi-dimensional scaling (MDS) ordination of the three tanks upon arrival in Deception Bay for June, July and August trips (raw data excluding copepod nauplii)

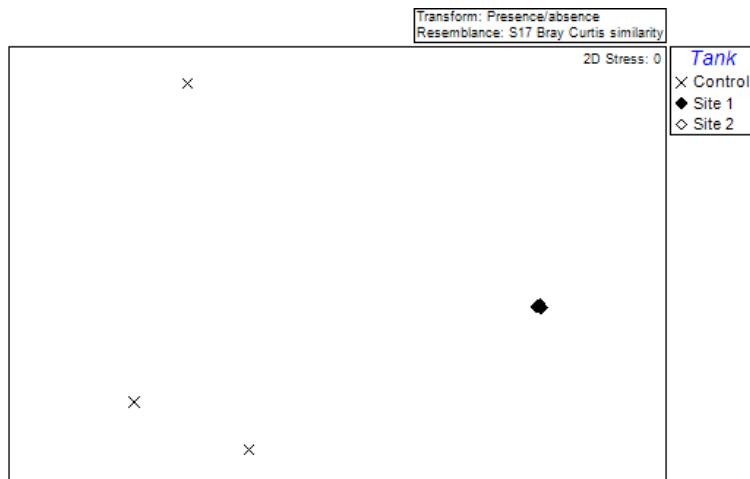


Figure 25. Non-metric multi-dimensional scaling (MDS) ordination of the three tanks upon arrival in Deception Bay for June, July and August trips (Presence/Absence data including copepod nauplii)

Table 16. PERMANOVA table of results and pair-wise tests for Obj. #2G

Obj. #2G - MZP communities in the three ballast water tanks upon arrival in Deception Bay (June-August 2015)

PERMANOVA – Raw density data excluding copepod nauplii (untransformed)

Source	df	SS	Pseudo-F	P(perm)	perms
Tank	2	11448	2.181	0.0475*	6171
Month	2	7680.1	1.463	0.1726	6108
Residuals	4	10498			
Total	8	29626			

PERMANOVA – Presence/Absence data including copepod nauplii

Source	df	SS	Pseudo-F	P(perm)	perms
Tank	2	8687.7	4.4489	0.0329*	5987
Month	2	2700.2	1.3827	0.2058	6116
Residuals	4	3905.6			
Total	8	15294			

Pair-wise tests – Raw density data excluding copepod nauplii (untransformed)

Groups (Tanks)	t	P(perm)	Unique perms	P (MC)
Site #1 - Site #2	1.9857	0.1020	38	0.1041
Site #1 - Control	1.4658	0.2038	38	0.2004
Site #2 - Control	1.3679	0.2324	38	0.2360

Pair-wise tests – Presence/Absence data including copepod nauplii

Groups (Tanks)	t	P(perm)	Unique perms	P (MC)
Site #1 - Site #2	1.0943	0.4179	37	0.3833
Site #1 - Control	2.3990	0.0989	38	0.0778
Site #2 - Control	2.1635	0.0975	38	0.0842

1.4. DISCUSSION

1.4.1. OBJ. #1: EVALUATE SUMMER MONTHLY VARIABILITY IN MZP COMMUNITIES PRESENT IN THE BALLAST WATER TANKS OF THE *M/V ARCTIC* IN THE PORT OF ORIGIN (QUEBEC CITY HARBOR)

General freshwater MZP community of water ballasted in Quebec City harbor

The Quebec City harbor MZP community found in ballast water tanks was composed mostly of small-sized zooplankters, typical of riverine systems, with limited presence of large cladocerans and calanoids (Basu *et al.*, 1999). All taxa found in our samples (**Appendices 3-4**) were previously recorded in the Great Lakes, as reported by Pinel-Alloul *et al.* (2011) in the St. Lawrence fluvial estuary. The species observed in Quebec City harbor are currently considered nonindigenous to Deception Bay based on their known distribution patterns (Lesko *et al.*, 2003ab; WoRMS, 2016; Global Biodiversity Information Facility, 2016; Ocean Biogeographic Information System, 2016). These taxa represent the port of origin community that would be discharged and potentially introduced to Deception Bay in the case of the *M/V Arctic* omitting ballast water exchange in the Gulf of St. Lawrence.

Summer monthly evolution of MZP communities and total density in the three tanks filled in Quebec City harbor

Total MZP density was lower in June and July, peaked in August and then decreased in September and October. We observed a similar density pattern as Loubier (1984), showing lower zooplankton density in spring and fall (< 5 000 ind. m⁻³) in the fluvial section of the St. Lawrence River. Differences between August and all the other months were expected, as zooplankton density in the freshwater section of St. Lawrence River

usually peaks in midsummer, due to low river discharge promoting plankton development. Weaker currents are generally associated with lower turbidity and increased zooplankton productivity (Basu *et al.*, 1999). However, the density peak observed in August (mean: $\approx 87\ 000$ ind. m^{-3}) was higher than reported by Loubier (1984) ($\approx 30\ 000$ ind. m^{-3}). This is explained by the fact that Loubier described productivity in the non-tidal fluvial zone of the St. Lawrence. Quebec City harbor is part of the fluvial estuary, with large tidal influence. Planktonic productivity is usually higher in tidal sections of rivers due to lower downstream advection and higher residence time, minimizing advective removal of populations (Pace *et al.*, 1992). The significant difference in total MZP density between June (mean: 1153 ind. m^{-3}) and September (mean: 7969 ind. m^{-3}) may be due to the fact that initial sampling in June was conducted exceptionally before exchange at Site 1, rather than in Quebec City harbor. The added mortality due to this delay could have decreased the initial density in the June tanks. Alternatively, this very low density could much more likely be due to secondary production still being very low in the beginning of June, at the end of spring (Basu *et al.*, 1999).

The strong dissimilarity in species composition based on densities between June and all the other months is due to lower densities in June of *Eurytemora carolleae*, copepod nauplii, mussel veligers and *Bosmina longirostris*. June was distinct because of very low total MZP density and also due to the importance of the cyclopoid *Diacyclops thomasi*, which represented between 14 and 44% of the community, while being absent or underrepresented during all the other months. Pinel-Alloul *et al.* (2011) also described the importance of *Diacyclops thomasi* in May and its absence in August in the fluvial estuary of the St. Lawrence River. The strong dissimilarity between August and all other months is explained by much higher total MZP density, composed almost exclusively of *Eurytemora carolleae* and its nauplii, the cladoceran *Bosmina longirostris* and mussel veligers. These results are in accordance with the findings of Basu *et al.* (1999) and Pinel-Alloul *et al.* (2011), who reported high MZP density dominated by *Eurytemora carolleae* (formerly *Eurytemora affinis*) in the fluvial estuary of the St. Lawrence in August.

In biology of zooplanktonic invasives, a propagule is considered to be an individual of a species (egg, larva, juvenile or adult) capable of surviving and reproducing in an environment to which it is non-native (Shanks, 2003; Colautti *et al.*, 2006b). It is the minimal part of a population required for the colonization of a new environment. Nonindigenous taxa introduced in large numbers are more likely to reproduce and survive permanently in a new environment. Furthermore, the amount and frequency of discharged NIS propagules have been positively correlated with the establishment success of a species (Lockwood *et al.*, 2005). Thus, we based our evaluation of the monthly evolution of introduction risks of NIS from Quebec City as function of total MZP density in the *M/V Arctic*'s ballast water tanks at the start of each trip. *Eurytemora carolleae* was the taxon in which we were the most interested, due to its invasion potential (see Section 1.4.2), its dominance in Quebec City harbor and its current nonindigenous status in Deception Bay. We evaluated the risks of introduction based on when this species was the most abundant. The month of August was characterized not only by the highest total MZP density, but also by the highest density of this taxon and its nauplii, which represented 53-73% of the whole community. Based on this fact, we observed a monthly trend in the risks of introduction, with August showing the highest density of the taxon with the highest known potential of invasion, *Eurytemora carolleae* (**see Section 1.4.2 – pp. 68-71 on its invasion potential**). Thus, we know that Quebec City harbor in August will provide large amounts of propagules of this taxon, already described in the literature as a prolific invader of freshwater environments (Lee, 1999; Winkler *et al.*, 2008; Pinel-Alloul *et al.*, 2011). If the ballast water tanks of the *M/V Arctic* remained unexchanged throughout a trip in August, larger amounts of this aquatic invader would be discharged in Deception Bay. This confirms our initial hypothesis stating that Quebec City harbor will provide maximal NIS density during the midsummer peak in total MZP density.

Summer monthly evolution of MZP diversity and species richness in the three tanks filled in Quebec City harbor

There was no monthly trend in MZP diversity. Generally, diversity was low ($0.5 \leq H' \leq 2$) due to the inequality of species density (McDonald, 2003). The high importance of three main species, *Eurytemora carolleae* and its nauplii, mussel veligers and *Bosmina longirostris*, tended to mask the importance of other underrepresented or rare taxa throughout all months. A similar case was described by Fransz and Gonzalez (2001) in the North Sea. This conclusion rejects our initial hypothesis stating that diversity would be minimal during the peak of MZP density in midsummer. Although August was characterized by high densities of these three dominant species, diversity was not statistically higher or lower during this period. These results contrast with the literature, as zooplankton density peaks are usually associated with minimum diversity (Irigoinen *et al.*, 2004). Marine and freshwater zooplankton diversity is typically negatively correlated with biomass, with maximum diversity at intermediate levels of biomass and minimum diversity during density peaks (Ghilarov and Timonin, 1972; Irigoien *et al.*, 2004). However, we observed high variability between replicates, possibly masking the signal in difference among months. This may be explained in part by the fact that ballast water tanks were filled up at different times of day or even on separate dates. Quebec City harbor is known to have high tide amplitude (4-5m) and strong currents (3-4 knots) due to the narrowness of the river in this area (Fisheries and Oceans Canada, 2016; Table de Concertation Régionale Zone de Québec, 2015). This could have caused the strong variability in MZP diversity and density between our replicates. Davies and Ugwumba (2013) reported an important effect of tides on zooplankton diversity and density in estuaries.

We also observed a seasonal trend in MZP species richness for Quebec City harbor. The August peak in total MZP density was associated with minimum species richness. Zooplankton density peaks often coincide with the predominance of a few species (Irigoinen *et al.*, 2004), which in our study consisted of *Eurytemora carolleae*, *Bosmina longirostris* and mussel veligers. This conclusion confirms our initial hypothesis stating that species richness would be minimal during the midsummer MZP density peak.

1.4.2. OBJ. #2: EVALUATE THE EFFECT AND EFFICACY OF VOLUNTARY BALLAST WATER EXCHANGE (BWE) PERFORMED BY THE *M/V ARCTIC* IN THE GULF OF ST. LAWRENCE

Effect of ballast water age on MZP diversity and total density

Our results showing a negative impact of ballast water age on MZP density are in accordance with general findings in the literature (Wonham *et al.*, 2001; Humphrey, 2008; Chan *et al.*, 2014). Predation, limited food supply due to the absence of photosynthesis, decreasing nutrient and oxygen concentrations, toxicity of anticorrosion/antifouling ballast water tank coatings and hydrocarbon residues, as well as other factors, have been shown to cause plankton mortality with increasing voyage duration (IMO, 1997; Ware *et al.*, 2015). Upon arrival in Deception Bay, we observed between 65 and 76% of mortality in the control tanks. June and July trips were associated with higher mortality than August, which can be explained by longer voyage duration (11 and 10 days, compared to 5 days, respectively) (Humphrey, 2008). However, mortality was lower than other studies. Wonham *et al.* 2001, showed a 99% decrease in density after a 16-day trip. Gollasch *et al.* (2000) found a 90% reduction in zooplankton density after just four days of a 23-day trip. However, in these two studies, ships departed from subtropical (Israel) and equatorial (Singapore) ports of origin, heading to temperate ports of destination (USA and Germany). Environmental dissimilarity between connected ports may have had a stronger impact on density than ballast water age. The *M/V Arctic*'s route lies within a narrow latitudinal range (46 to 62 °N), connecting a temperate port of origin to a subarctic port of destination. In this case, temperature changes in summer may have played a weaker role than in the previous studies on transoceanic pathways (Chan *et al.*, 2014). Taylor *et al.* (2007) observed that mortality was stronger in ballast water of ships using routes within subtropical to temperate zones, compared to those sailing within narrower latitudinal range. In these cases, high temperatures would also lead to lower dissolved oxygen and possibly increased mortality.

On the other hand, the drastic increase in total MZP density observed in July between Quebec City harbor and Site #1 before exchange may be explained by the hatching of *E. carolleae* eggs under favorable conditions. Ballast water tanks can sometimes act as incubators where ballasted organisms may flourish (Jing *et al.*, 2012). Indeed, several adults and ovigerous females of *Eurytemora carolleae* were observed in the control tank throughout the July trip. These individuals may have caused a massive hatching of copepod nauplii inside the tanks, importantly increasing total MZP density. In fact, copepod nauplii increased from 3055 to 19 774 ind. m⁻³ and *E. carolleae* stage I copepodids, closely related in development time to the final naupliar stages, also increased from 204 to 3195 ind. m⁻³, supporting the hypothesis of a massive hatching. Development time from egg to adult for *E. carolleae* is short, taking 10 days at 25 °C (Heinle and Flemer, 1975). This could explain the increase of both nauplii and stage 1 copepodids over a short time period. Between the stations where the hatching was observed, temperatures varied between 16 and 23 °C, thus promoting larval development.

Finally, even though ballast water age was negatively correlated with density, it did not have a significant impact on diversity, although we observed a close to significant trend toward decreasing diversity during trips. Our results, albeit non-significant, agree with another study showing a negative effect of ballast water age on MZP diversity in ballast waters of several ships (Burkholder *et al.*, 2007). This non-significant result could possibly be explained by low freshwater diversity (H' always lower than 1.6) due to the dominance of *E. carolleae* and its nauplii, *B. longirostris* and mussel veligers. Even though high mortality was observed, it did not significantly decrease diversity, which was already low at the beginning of each trip.

Risks of NIS introduction through deballasting of unexchanged (control) tanks

Our results suggest that the MZP communities in the control tanks did not change significantly between the beginning and end of each trip, and also that the species ballasted in the port of origin are still present upon arrival in the port of destination. This is in accordance with our earlier results showing a lack of strong correlation between ballast water age and MZP diversity. Ballast water age seemed to mainly impact total MZP density, but not species composition based on densities or species occurrence. This conclusion disagrees with the work of Burkholder *et al.* (2007), who observed an effect of ballast water age on total density but also on diversity. However, our analysis was unreplicated and the variability of initial MZP communities between trips may have masked the differences between stations. Furthermore, MDS representation based on species occurrence suggests that mortality during the trip caused some rare taxa to be absent upon arrival in Deception Bay in June and July, also supporting our results showing a negative, albeit non-significant effect of ballast water age on diversity.

Since the organisms present in the control tanks originate from Quebec City harbor, they form a MZP community of species that can all be found in the Laurentian Great Lakes. Most are described as either oligohaline (rotifera, cladocera, mussel veligers, *Bosmina longirostris*, *Leptodiaptomus sicilis*) or stenohaline (cyclopoida), so there is limited concern about their probability of introduction through deballasting in Deception Bay (Lesko *et al.*, 2003ab; WoRMS, 2016; Global Biodiversity Information Facility, 2016; Ocean Biogeographic Information System, 2016). Since Deception Bay harbor is a marine environment, with average salinity around 29 psu, these stenohaline or euryhaline taxa should not be able to survive in such conditions (salinity based on summer 2015 thermosalinometer measurements and CAISN, unpublished data). Hudson Strait annual subsurface (40-60m) salinities range between 29 and 33 psu (Straneo and Saucier, 2008). Some freshwater inlets create brackish conditions in limited areas of the bay in summertime, however these conditions may not be large enough or sustained over long enough time periods to support the permanent establishment of populations. Osmotic shock at deballasting, due to the drastic change in salinity (osmotic pressure), should kill most

deballasted organisms (Transport Canada, 2011). Furthermore, most of these taxa were found in very low density in tanks, since mussel veligers, *Eurytemora carolleae* and its nauplii, and *Bosmina longirostris* usually represented between 80 and 100% of the MZP community. Low density (low propagule pressure) decreases the risks of introduction of a species (Lockwood *et al.*, 2005). The month of June presented an exception, as the cyclopoid *Diacyclops thomasi* was an important component of the community (10-45%). This taxon, however, is strictly limited to freshwater environments (WoRMS, 2016). *Bosmina longirostris*, the dominant cladoceran observed in Quebec City, has a salinity tolerance of 4-5 psu (Effler, 1996). The species observed in Quebec City harbor are currently considered nonindigenous to Deception Bay (Lesko *et al.*, 2003ab; WoRMS, 2016; Global Biodiversity Information Facility, 2016; Ocean Biogeographic Information System, 2016). The only taxon observed both in Quebec City and Deception Bay harbors was *Kellicottia longispina*, a common and widespread freshwater rotifer that was also reported in lakes of Arctic Russia and Alaska (Edmondson and Litt, 1989; WoRMS, 2016; Ocean Biogeographic Information System, 2016). Its presence in the marine waters of Deception Bay may be explained by the high summer freshwater input from Deception River. Freshwater organisms from the river may drift in the brackish surface layer of the fjord and get caught in the plankton net. We observed this taxon only in June, as the surface salinity was of 6.1 psu at 1m depth, compared to 24.1 psu at 3m depth, supporting the hypothesis a of a brackish surface layer.

However, there are concerns about the invasion potential of *Eurytemora carolleae*, a dominant calanoid copepod in the fluvial estuary of the St. Lawrence River (Winkler *et al.*, 2003; Dufour and Ouellet, 2007; Winkler *et al.*, 2008; Cusson, 2011; Favier and Winkler, 2014; Cabrol *et al.* 2015). *E. carolleae* has recently been described as a separate species within the *Eurytemora affinis* cryptic species complex (Alekseev and Soussi, 2011). It was known earlier in the literature as the Atlantic clade of the *E. affinis* complex and is now being referred to as a sibling species of *E. affinis*. This estuarine taxon is euryhaline, able to withstand salinities ranging from 0 to 40 psu and temperatures ranging from 0 to 30 °C (Bradley, 1978; Lee and Petersen, 2003; Lee *et al.*, 2013). Having invaded several

freshwater, brackish and marine environments, this species was originally native to marine environments (Lee, 1999; Winkler *et al.*, 2008). It was introduced on the Pacific Coast of the United States in San Francisco Bay (California) and in Grays Harbor (Washington) (Fofonoff *et al.*, 2003). It was also reported as nonindigenous to the Laurentian Great Lakes and the Baltic Sea (Lee, 2000; Lee and Petersen, 2002; Winkler *et al.*, 2008; Alekseev *et al.*, 2009). Its invasion in the Great Lakes was explained by the opening of the St. Lawrence Seaway, followed by its introduction via ballast water (Winkler *et al.*, 2008; Pinel-Alloul *et al.*, 2011; Vasquez *et al.*, 2016). This copepod is able to survive and reproduce in freshwater, brackish, as well as in marine habitats (Lee, 2000). The first reported sighting was in Lake Ontario in 1958, probably via ballast water in the St. Lawrence Seaway (Mills *et al.*, 1993). However, its impacts on recipient ecosystems are not yet understood (Fofonoff *et al.*, 2003). It is known to be a dominant component of the mesozooplankton in estuarine environments (Laprise et Dodson, 1994; Winkler *et al.*, 2003; Devreker *et al.*, 2009). Due to their high density and their omnivorous opportunistic diet, both *E. carolleeae* and *E. affinis* (North Atlantic clade) play a crucial role in estuarine food webs by transferring energy from the primary producers to higher trophic levels, like fish larvae and mysid shrimps (Winkler *et al.*, 2003; Winkler *et al.*, 2007). Cabrol (2013) described *E. carolleeae* as a generalist species after observing no significant decrease in reproductive performance under sudden changes in environmental conditions.

We observed *E. carolleeae* in high density in the *M/V Arctic*'s tanks in Quebec City harbor (maximum in August at 15 774 ind. m⁻³) and it was still present in the control tanks of each trip upon arrival in Deception Bay (maximum in August at 1585 ind. m⁻³). It is currently considered nonindigenous to the Canadian Arctic (**Figure 26**) (Fofonoff *et al.*, 2003; Ware *et al.*, 2015; Global Biodiversity Information Facility, 2016). Also, there has not been any records based on genetic sequencing for this species in the Canadian Arctic (BOLD Systems, 2017^a). This taxon (including *E. affinis* and *E. hirundooides*, its former synonyms) was also absent from the lists of species found in other studies on Canadian Arctic waters (Harvey *et al.*, 2011; Hudon *et al.*, 1993; Grainger, 1962; Grainger, 1965). Since experiments in the literature on *Eurytemora affinis*'s (unspecified clades) tolerance

thresholds included only high temperatures and a broad range of salinities, we do not know the lowest temperature to which it could survive. There are no records of this taxon observed at temperatures below 0 °C (Global Biodiversity Information Facility, 2016). However, the genus *Eurytemora* sp. has recently been observed in port surveys from Hudson Bay, where temperatures drop below 0 °C (Kimberly Howland, personal communication). The lowest tested reproductive temperature was of 5.5 °C (Heinle and Flemer, 1975). However, the *E. affinis* species complex is known to lay, under unfavorable conditions, diapausing eggs that can withstand low temperatures, dessication, anoxia as well as hypersalinity, which can stay viable for up to 10-18 years in the sediment (Ban and Minoda, 1992; Ban and Minoda, 1994; Great Lakes Aquatic Nonindigenous Species Information System, 2013). There is a chance that diapausing eggs could survive arctic winter conditions and hatch in the summertime, under milder conditions. Summer temperatures in the Hudson Strait close to Deception Bay vary in the water column between -1 and 5 °C (Estrada *et al.*, 2012). Hudson Strait annual subsurface (40-60 m) temperatures range between -1,8 and 4 °C, with negative temperatures ($< 0 \geq -1,8$ °C) occurring from November to August (Straneo et Saucier, 2008). Water temperature of Deception Bay in August, between 4 and 10 °C (CAISN, unpublished data, 2016) is suitable for the survival of this taxon. Thus, there might be a chance that *E. carolleae* could survive and reproduce if discharged in sufficient amounts in the port of destination. However, since there is no literature describing its tolerance to cold temperatures (as well as for its diapausing eggs), we cannot assume its survival in sub-zero temperatures (≈ -1.8 °C) usually occurring in Deception Bay between December 4th and July 2nd (Stewart *et al.*, 2015; NSIDC, 2016b). Although there is currently no literature describing the presence of *Eurytemora carolleae* in the Canadian Arctic and that it has been absent from our recent port survey samples, *Eurytemora affinis* has recently been found by metabarcoding in the ports of Churchill, Deception Bay and Iqaluit (CAISN, unpublished data). Also, Zrum (2000) found *Eurytemora affinis* in the Nelson River estuary, in western Hudson Bay. Thus, further phylogenetic investigation is required to uncover what clade or sibling species of *Eurytemora affinis* occurred. There is a possibility that it might be *Eurytemora carolleae*.



Figure 26. Current and projected distribution of calanoid *E. affinis* (unspecified clades) (left to right: actual, 2050, 2100). Red areas indicate suitable habitat and the blue ones indicate unsuitable habitat. Black points indicate reported sightings of the species. Figure is based on limited data (Ware *et al.*, 2015)

The mussel veligers found in the ballast water tanks filled in Quebec City harbor often formed a dominant part of the freshwater MZP community. They were sometimes found in great densities, peaking at 16 604 ind. m⁻³ in August. They are most likely veligers of the bivalve molluscs *Dreissena polymorpha* or *Dreissena bugensis*, two highly invasive taxa commonly known as the zebra and quagga mussels, respectively (Fisheries and Oceans Canada, 2016). These mussels are both present in Quebec City harbor and are known to be an important nonindigenous component of the species assemblage of the fluvial estuary of the St. Lawrence River (Environnement Canada, 2000; Winkler *et al.*, 2005). However, these stenohaline taxa are invaders of freshwater and oligohaline environments only (WoRMS, 2016), so there is a low chance for their establishment in Deception Bay, even if discharged by the *M/V Arctic* in large amounts. Salinities greater than 6 psu are lethal to the larvae of both *Dreissena polymorpha* and *Dreissena bugensis* (Wright *et al.*, 1996). In the St. Lawrence River, the distribution of zebra mussel larvae is limited downstream to the estuarine transition zone, where a sharp decrease in density occurs at salinities of 2 psu, although some individuals have been found up to 10 psu (Barnard, 2006). No colonies of mature individuals have been reported downstream of Montmagny (Environment Canada, 2000). Also, the zebra mussel does not tolerate temperatures below 0 °C and needs a minimal temperature of 10 °C for reproduction. Similarly, *D. bugensis* cannot survive

below 4 °C and cannot reproduce below 7 °C (Mackie, 2004). Thus, under-ice negative temperatures occurring in Deception Bay between October and June are expected to be unsuitable for the establishment of both species (MFFP, 2016). There could be a potential for the survival of *D. polymorpha* if larvae could reach the Deception River, where under-ice temperature could possibly be over 0 °C. However, such a hypothesis would imply larvae getting in the river counter-current, which is improbable.

Risks of NIS introduction through ballast water exchange in the Gulf of St. Lawrence

Exchanging ballast water had a significant effect on MZP species occurrence, since the port of origin was located in a freshwater environment and the exchange zones in a marine one. Thus, unexchanged/control tanks had no species in common with the exchanged tanks, except for the scarce residual freshwater taxa left in exchanged tanks. This was expected, as the flushing of freshwater organisms and their replacement by marine species are the main purpose of ballast water exchange (Transport Canada, 2011). The repeated measures (trips) in our experimental design had to be treated as a factor of variance, thus adding error to the model. Even though BWE had a significant effect on MZP communities (raw and fourth root transformed density data), several interactions between factors were also significant (see Section 1.3.2.D). Thus, only results of presence/absence transformed data will be discussed here. SIMPER analysis revealed that ballast water exchange had a major effect on the important taxa of the freshwater community, by flushing out *Bosmina longirostris*, mussel veligers and *Eurytemora carolleae*, which were replaced mostly by *Oithona similis* and *Microsetella norvegica*, two small-sized copepods known to be dominant in the northwestern Gulf of St. Lawrence (Harvey *et al.*, 2000; St. Lawrence Global Observatory, 2016). Other frequently encountered taxa were the calanoids *Pseudocalanus* sp., *Centropages hamatus* and *Temora longicornis*. All of these taxa typically represent 50% and more of the total copepod density in the Gulf (Dufour and Ouellet, 2007). Thus, freshwater organisms were almost completely replaced by marine taxa (exception made for occasional residual freshwater taxa).

The MZP community in the Gulf of St. Lawrence is dominated by a few calanoid copepod species (MDDELCC, 2014). Copepod eggs, juveniles and adults account for more than 80% of the zooplankton community year-round in the Anticosti Gyre, consistent with communities observed in ballast water from Site #1 and Site #2 immediately after exchange (Fisheries and Oceans Canada Science, 2002). The copepod assemblage in the northeastern gulf is dominated by small copepod species, *Oithona* sp., *Pseudocalanus* sp. and *Temora longicornis*, which represented from 50 to 65% of the total copepod density in late spring

and between 60 and 85% in fall of 2000-2005. Larger copepod species, such as *Calanus finmarchicus* and *Calanus hyperboreus*, are usually found in deep waters (Dufour and Ouellet, 2007). Since the *M/V Arctic*'s ballast water in exchanged tanks originates from seawater surrounding the ship at hull depth, it is in fact surface water from the Gulf, explaining why very few of these deepwater larger copepod species were observed. Salinity in the exchanged tanks ($\geq 20 \leq 25$ psu) was typical of Gulf of St. Lawrence surface waters. The brackish surface layer of the Gulf (22-30 psu, 0-30 m depth) is influenced by the freshwater output of the St. Lawrence River and its tributaries (Plourde *et al.*, 2014). Since our exchanged ballast water comes from northern and northeastern Gulf surface waters, it is not surprising that a large part of the typical Gulf MZP community (larger copepods) was missing. MZP communities from both exchange sites at all trips were dominated by *Oithona similis* and *Microsetella norvegica*. The smaller-sized cyclopoid *O. similis* is known to have limited vertical migration patterns (non-migrant aggregated species) due to its omnivorous regime and lesser dependance on surface phytoplankton, which is compensated for by feeding on copepod nauplii and fecal pellets (Gonzalez and Smetacek, 1994; Nakamura and Turner, 1997). Also, its smaller size and erratic movement makes it less vulnerable to visual predation than larger herbivorous calanoids, possibly explaining why it does not have to avoid the euphotic zone (Brooks and Dodson, 1965; Hays *et al.*, 1994). Lagadeuc *et al.* (1997) reported constant concentrations of *Oithona similis* in the surface layer (7-9 m) in Baie des Chaleurs. *O. similis* was also observed in the surface layer (0-25 m) of the Jacques Cartier Strait in 1992 (Pierre Joly, personal communication). Vertical migration patterns could explain why our exchanged ballast water, originating from the surface layer, contained mainly the small-sized copepods *O. similis* and *M. norvegica*. A large number of vertically migrating zooplankters, such as krill, larger copepods, amphipods or mysid shrimps, live in deeper waters during the day, away from ballast uptake depth (Sainte-Marie and Brunel, 1985; Harvey *et al.*, 2002; Dufour and Ouellet, 2007; Plourde *et al.*, 2014). The deep water smaller copepod genus *Microcalanus* (Johnson *et al.*, 2010) was also rarely observed, with only single occurrences. All of these organisms have few chances of being ballasted, so a large part of the gulf zooplankton

community was absent from our samples. However, since *Microsetella norvegica* is known as a pelagic/benthic species (one of the 0.5% of Harpacticoida species not primarily benthic) with diel vertical migration, a different reason is required to explain its dominance in ballast waters originating from the surface layer (Huys and Boxshall, 1991; Uye *et al.*, 2002). *Microsetella norvegica* is often found close to the surface in the summer because of its omnivorous diet consisting mainly of sinking particles (Uye *et al.*, 2002; Koski and Kiørboe, 2005). Its high growth rate and omnivorous regime make it an important part of secondary production in a wide range of environments (Uye *et al.*, 2002). It has been shown to be a dominant species in coastal subarctic environments (Arendt *et al.*, 2012; Antonsen, 2014). Anthonsen (2014) found maximal density of this taxon in the surface layer in June and August in a fjord of northern Norway. *M. norvegica* reproduces in May in the surface layer, where it feeds and grows all summer long. Alternatively, Koski *et al.* (2014) suggested that females may detach egg sacs prior to hatching, supporting the hypothesis of surface layer egg contamination due to advection from shallow-water neritic zones (Pierre Joly, personal communication). Since both of our exchange zones were located in coastal zones, this explanation is also possible.

We attributed a nonindigenous status to the taxa observed in the exchange zones which were absent from Deception Bay. However, our knowledge of the indigenous MZP community in Deception Bay is based only on the taxonomic analysis of four harbor samples collected in June, July, August and October 2015. These samples are not necessarily representative of the whole community present year-round in the port of destination. Rarity of certain taxa or their occasional presence could explain why they were not observed in our Deception Bay harbor samples. Only year-round sampling of the harbor's MZP community would help establishing a complete list of indigenous taxa. Furthermore, species observed in Deception Bay are not necessarily indigenous and may have been introduced earlier by ballast water discharge. This is why taxonomic literature was used to confirm indigenous/NIS status. Furthermore, several species or genera of MZP ballasted in Sites 1 and 2 were not observed in Deception Bay harbor. These include *Scolecithricella minor*, *Calanus hyperboreus*, *Evadne nordmanni*, *Evadne spinifera* and

Podon sp. However, these taxa/groups are part of the typical North Atlantic MZP community and could also be present in Deception Bay. *Calanus hyperboreus* is described as an arctic species and has already been recorded in Foxe Basin and Hudson Strait (Eastern Canadian Arctic) (Grainger, 1962; Harvey *et al.*, 2001; Estrada *et al.*, 2012). The genus *Podon* has been recorded in Western Arctic (Grainger, 1965). The calanoid *Scolecithricella minor* was recorded in Hudson Strait, close to Deception Bay (Hudon *et al.*, 1993). *Evadne nordmanni* was described as being part of the zooplankton in the Chukchi Sea, in Iceland's coastal waters, in Barents Sea, in Kara Sea and in the boreal part of the White Sea (Smith and Schnak-Schiel, 1990; United States Department of the Interior, 1990; Marine Species Identification Portal, 2016a). This taxon is known to be present in arctic waters, even if its presence in Hudson Strait has yet to be reported (Global Biodiversity Information Facility, 2016). The presence of *Evadne spinifera* was documented in the North West Atlantic, but it is absent from polar and subpolar waters (WoRMS, 2015; Marine Species Identification Portal, 2016b; Global Biodiversity Information Facility, 2016). There is a possibility that *Evadne nordmanni* and *Evadne spinifera* could be nonindigenous to Deception Bay. However, the introduction risk based on our study should be low, as these two taxa were found in very low density (almost always in single occurrences) only at Site #1 (Jacques Cartier Strait). Furthermore, *Evadne spinifera* has a thermal tolerance of 3 to 28 °C, so its capacity for invading arctic waters should be limited, since temperatures in Deception Bay are likely to drop below 3 °C in the winter (Marine Species Identification Portal, 2016b). However, the Hudson Strait and Deception Bay offer warmer surface temperatures in summer, suitable for the establishment of these two taxa (around 5 °C) (**Figure 27**) (Galbraith and Larouche, 2011; Tremblay *et al.*, 2015; Hare and Montgomery, 1949). *Evadne spinifera* should have low chances of survival through wintertime, in much harsher conditions and lower temperatures (under-ice temperatures are usually around -1.8 °C) (NSIDC, 2016). *Evadne nordmanni* should also have limited chances of invading Deception Bay, since its thermal tolerance ranges from 1 to 25 °C (Marine Species Identification Portal, 2016a). No specimens were found by Onbé *et al.* (1996) in Chukchi Sea below 1 °C. However, it is also known to lay diapausing eggs,

which can stay viable during harsh conditions of the polar winter until the following season, increasing its potential of survival (Onb  , 1985; Platt and Yamamura, 1986; Onb   *et al.*, 1996; Briski *et al.*, 2011). Evans and Grainger (1980) also infrequently found *Evadne nordmanni* in the Beaufort Sea, where subzero under-ice temperatures occur, supporting the evidence of its capacity of overwintering with diapause eggs. Thus, we cannot assume that it has no potential for survival in Deception Bay. For *Evadne spinifera*, resting eggs are however currently unreported (Onb  , 1991; Marine Species Identification Portal, 2016).

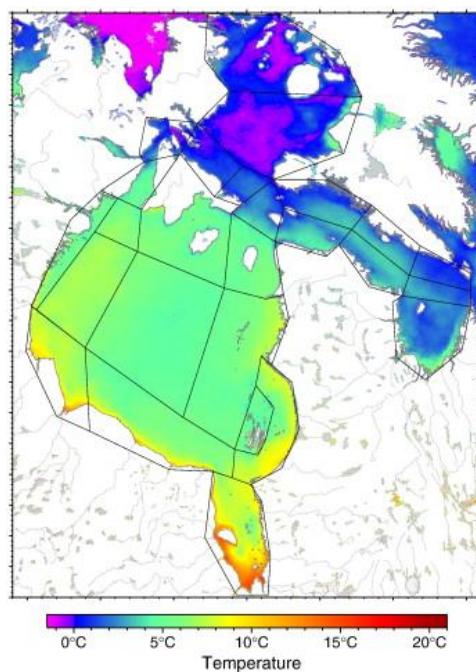


Figure 27. Average sea surface temperature for the months of July, August and September 2001 to 2005 (Galbraith and Larouche, 2011)

We also observed residual freshwater taxa from Quebec City at Sites 1 and 2 immediately after exchange. These taxa include *Lecane* sp., *Mesocyclops edax*, unidentified freshwater cyclopoids, *Eurytemora carolleae*, *Microcyclops rubellus*, *Diacyclops thomasi*, *Keratella quadrata*, *Keratella* sp. (*crassa*, *earlinae* or *cochlearis*), *Bosmina longirostris*, many of which are strictly freshwater species, indicating the residual presence of organisms ballasted in Quebec City harbor. Their presence was expected, since several studies have shown that BWE is not fully efficient in flushing out coastal organisms (Transport Canada,

2011). Residual water is always present at the bottom of the tanks after the first phase of sequential BWE and it has been found to contain abundant and diverse resting stages of invertebrate organisms (Niimi and Reid, 2003; Duggan *et al.*, 2005; Bailey *et al.*, 2005). Water from the exchange site is subsequently ballasted into the empty tank, diluting the residual water from the port of origin, with some of its organisms still remaining inside the tank. Since viability tests were not conducted, we cannot tell if these organisms were still living at the time of sampling. However, most residual freshwater taxa were absent from the exchanged tanks upon arrival in Deception Bay. In some cases, *Eurytemora* sp. juveniles were still present upon arrival at the port of destination, although in very low density. However, it is very difficult to differentiate juveniles of *Eurytemora carolleeae* (from Quebec City harbor) from juveniles of *Eurytemora herdmani* (from the Gulf of St. Lawrence, already present in Deception Bay (BOLD Systems, 2017^b)) using traditional taxonomy (WoRMS, 2016). Therefore, we cannot tell if these immature individuals were residual euryhaline *Eurytemora carolleeae* from Quebec City that survived BWE or *Eurytemora herdmani* that were potentially ballasted in low densities at Sites #1 and #2. However, the very low proportion of remaining freshwater taxa in the community immediately after ballast water exchange (< 5 % of community) shows that the sequential method (Empty/Refill) was nevertheless highly efficient in removing organisms from the port of origin. These results confirm the works of Wonham *et al.* (2001), who reported 80-100% removal of coastal organisms with the sequential method (1 tank volume).

In summary, there was clear evidence that exchanging ballast water in the Gulf of St. Lawrence greatly reduced the density of NIS in the ballast water of the *M/V Arctic*. Since the vast majority of species in the control tanks were absent from Deception Bay, and that nearly all the species present in the exchanged tanks were indigenous to Deception Bay, we can conclude that voluntary BWE conducted by the *M/V Arctic* was efficient in reducing the density of NIS. These results confirm our initial hypothesis and were expected, since there was a major environmental mismatch between the port of origin and the exchange sites. BWE has maximal efficacy for routes connecting freshwater ports of origin to marine ports of destination (Ware *et al.*, 2015). On the other hand, there was higher similarity

between the exchange sites and the port of destination, resulting in similar MZP communities and potentially lower risks of NIS introduction. We can then conclude that exchanging ballast water in the gulf flushed out freshwater rich in NIS, replacing it by seawater with higher density in organisms, but more depleted in NIS. Furthermore, omitting to conduct BWE led to the discharge of three high-impact NIS, one of which is showing high potential for survival and establishment in Deception Bay (*E. carolleae*). It is although worth noting that our exchange sites may have already established NIS, and that several larval stages of benthic species could not be identified (barnacle nauplii and cyprids, polychaet worm larvae, bryozoan cyphonauts, etc...). Also, other species may not have been ballasted due to deeper habitat behaviour. Thus, we cannot conclude that performing ballast water exchange is risk-free. Moreover, due to the absence of replication at the two exchange zones, we cannot use statistical inference to generalize our results concerning ballast water exchange in the gulf to other domestic vessels using the same route, even if our results did show a general trend and gave important and new information on potential risks for this region.

MZP communities in the two ballast water exchange zones (Sites 1 & 2)

There were no differences in MZP communities or species occurrence between the two tested exchange zones. These results were expected, since the two BWE zones are located close to one another, as part of the northern Gulf of St. Lawrence. The general circulation in the gulf is driven partly by the Labrador Current, bringing cold water inflows from the Labrador Shelf through the Strait of Belle Isle (**Figure 28**). These waters then generally follow the lower north shore and go through the Jacques Cartier Strait (Dufour and Ouellet, 2007; Canadian Coast Guard, 2013). Since waters from our two sites originate from the Labrador Shelf, they were expected to have similar MZP communities. Intrusion of Labrador Shelf waters represents a minimal fraction of 3 to 30% (1996-2005) of total gulf water input, with strong inter-annual variability (Galbraith, 2006). This input varies each year according to the fluvial input from the estuary and wind (Saucier *et al.*, 2009). This intrusion of Labrador Shelf cold waters (-1.8 to 0 °C) originating from the southbound Labrador Current brings planktonic species of Arctic origin. Since this intrusion has an

important yet variable impact on MZP diversity and productivity in our BWE zones, the species ballasted by the *M/V Arctic* in Sites 1 and 2 could differ substantially depending on the year (Dufour and Ouellet, 2007; Descroix *et al.*, 2005). Years with lesser influence of this inflow could be associated with a boreal rather than arctic MZP community and thus risks of introduction could potentially increase. Also, coastal wind-driven upwelling all along the north shore of the gulf makes the deeper layer of cold water from the Labrador Current come to the surface, explaining similarity of MZP communities in the coastal zones of Sites 1 and 2 (Saucier *et al.*, 2009). However, even if the cold water inflow of the Labrador Current can influence the presence of arctic species, it alone cannot explain the similarity of the surface MZP community between our two sites. The mixed brackish surface layer in the gulf, 10-30 m thick in spring and summer, contains the MZP communities present at ballasting depth (Saucier *et al.*, 2009). Jacques Cartier Strait is known to form a stranglehold where strong tidal currents and intense vertical mixing occur, with high densities of mesozooplankton being reported for this zone. The Strait of Belle Isle also acts as a topographic stranglehold causing strong tidal mixing and deepwater resurgence (Fisheries and Oceans Canada, 2007b).

Our results also show that communities ballasted in the two BWE zones were not significantly different between exchanges sites and upon arrival in Deception Bay. In other words, the propagules deballasted in Deception Bay were the same as when they were originally ballasted in Sites 1 and 2. Since ballast water age in the exchanged tanks was lower than in the control tanks, limited mortality was expected. Much higher densities of propagules were deballasted from the exchanged tanks upon arrival in Deception Bay. However, these results were based on a multivariate analysis without replication and must be treated with caution. The statistical power of our analysis is thus limited and our results can only be applied to the *M/V Arctic* and to the year 2015. It is impossible to certify that other vessels using this route and exchanging at the same sites would lead to the same conclusions, even if our results give a general representation for other vessels conducting exchange in this region.

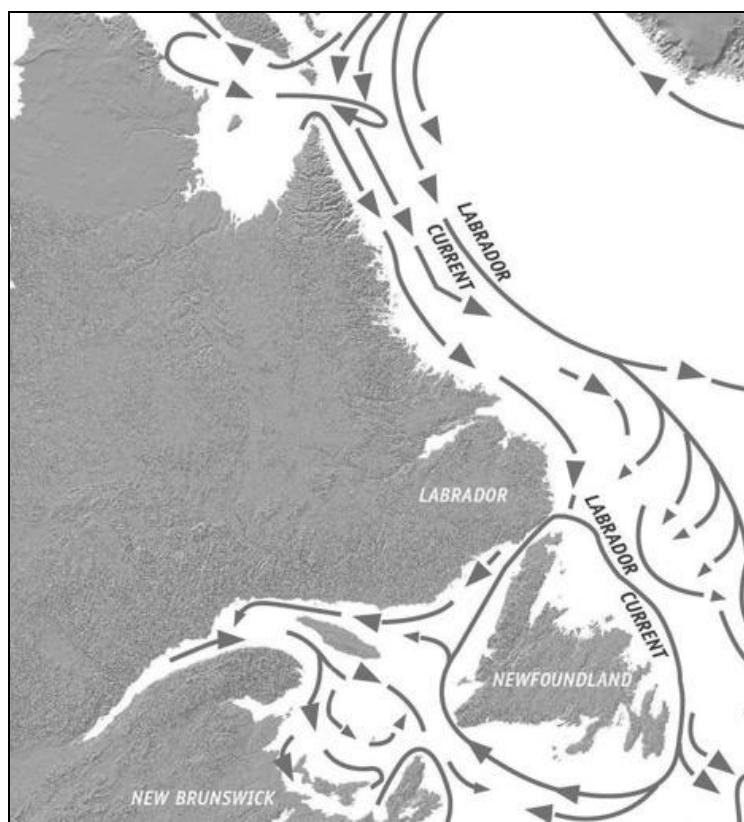


Figure 28. General circulation in the Gulf of St. Lawrence (Environment and Climate Change Canada, 2016)

Effect and efficacy of ballast water exchange in the Gulf of St. Lawrence

Our results revealed that exchanging ballast water in the Gulf of St. Lawrence during summer generally increased total MZP density. More propagules are discharged from the exchanged tanks than from the control tanks upon arrival in Deception Bay. This can be explained by the fact that “older” ballast water is deballasted from the control tanks, since they were filled up several days before the exchanged tanks, in Quebec City. Increased mortality due to higher ballast water age caused propagules to be less abundant in the controls. Ballast water exchange replaced the older freshwater with younger seawater containing higher densities of organisms. These conclusions are consistent with our earlier results showing a negative effect of ballast water age on total MZP density in the control tanks. Therefore, voluntary BWE conducted by the *M/V Arctic* failed to reduce the number of propagules deballasted in the port of destination. These results confirm our initial partial

hypothesis that voluntary BWE performed by the *M/V Arctic* would have low efficacy in reducing total MZP propagule density. The reduction of propagule density is one of the main purposes of BWE, as it is required under Canadian legislation for international vessels entering Canadian waters (Transport Canada, 2011; Chan *et al.*, 2012; Chan *et al.*, 2014; DiBacco *et al.*, 2012). Poor efficacy of the tested BWE zones in our study might be due to the fact that they were located in a coastal area. Coastal zones around the world are generally known to have high primary and secondary productivity, as they are often associated with coastal upwelling and high terrigenous nutrient influx. The north shore of the Gulf of St. Lawrence is known to have wind-induced coastal upwelling events and intense tidal mixing, which are important vectors of nutrient supply to the photic zone and make the northwestern gulf region a biological hotspot with high productivity (Fisheries and Oceans Canada, 2007a). Based on this information, Site 1 should be avoided for BWE, because it has high chances of supplying high densities of propagules and there are also risks of boreal and temperate species being present (including high-risk NIS), especially in the summertime. The north shore of Quebec is known to have high primary productivity in May and in September, which is followed in the next weeks by high secondary production, especially in Jacques Cartier Strait (Fisheries and Oceans Canada, 2007a). Also, intense tidal mixing in the Strait of Belle Isle makes this location another biological hotspot in the Gulf of St. Lawrence. In summary, there is evidence that Sites 1 and 2 should both be avoided for BWE, as well as the area between them (lower north shore of Quebec) (**Figure 29**). Both sites are known as ecologically and biologically significant areas in the Gulf of St. Lawrence. The north shore of the gulf also has high riverine input of organic matter, also promoting plankton productivity.

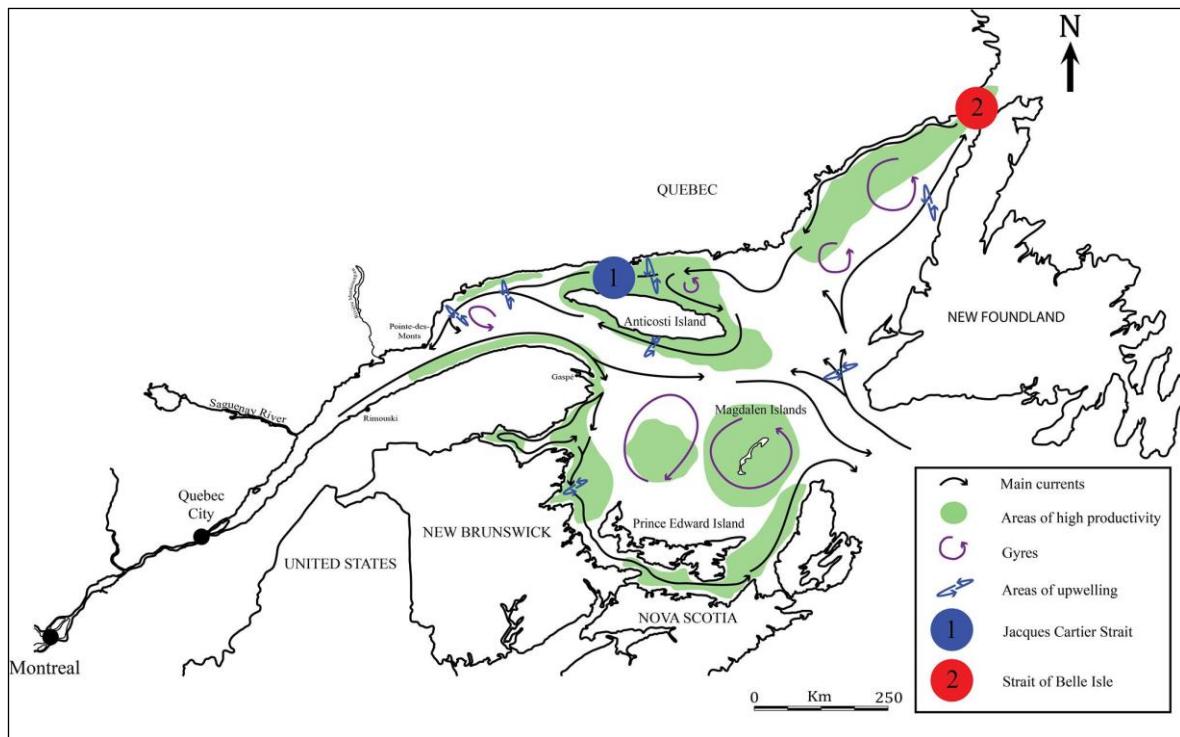


Figure 29. Zones in the Gulf of St. Lawrence associated with high productivity (modified from DFO, 2005)

The downwelling zone near the coast of Newfoundland, associated with lower surface nitrate concentrations and productivity, could possibly be used as an alternative BWE zone (Dufour and Ouellet, 2007). However, this alternative BWE zone would delay operations for the *M/V Arctic*, since it is not on the ship's direct route. On the coast of Labrador, heavy seas, fog, gales and ice make this location an inadequate alternative for BWE, since sequential BWE requires calm sea conditions in order to ensure ship safety (Transport Canada, 2011). Furthermore, Fisheries and Oceans Canada (2010) concluded that no zone could be recommended as potential alternative ballast water exchange zones for Newfoundland and Labrador. The Hudson Strait (300m minimal depth) has been suggested by Transport Canada (2010b) and Stewart *et al.* (2015) as an alternative zone for BWE, but was also considered inadequate by CAISN scientists (Kimberly Howland, personal communication) (**Figure 30**). However, performing BWE at this site may lead to excessive freezing of Quebec City freshwater in the ballast water tanks of the *M/V Arctic*.

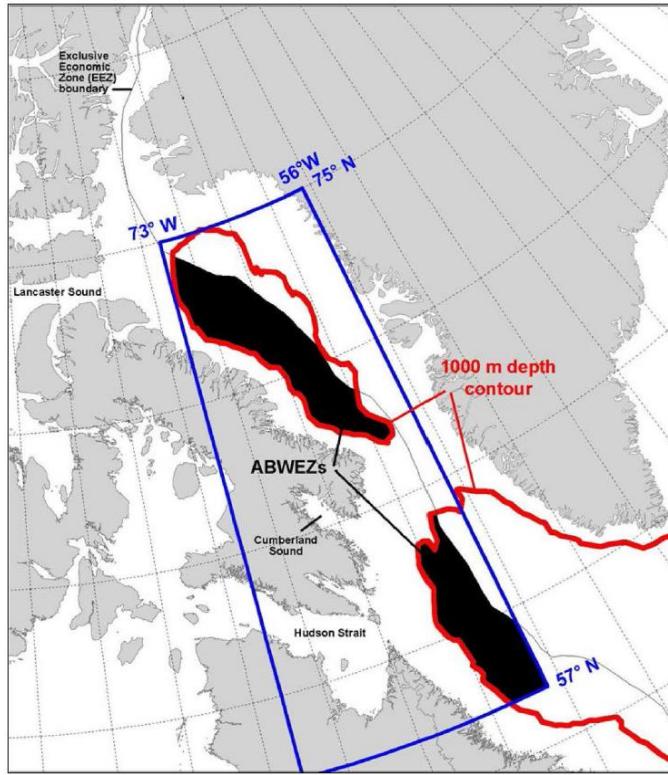


Figure 30. Recommended alternative ballast water exchange zones (ABWEZs) for the eastern Canadian Arctic, shaded in black. Red lines correspond to 1000m isobaths (Stewart *et al.*, 2015)

Conversely, BWE for international vessels is performed offshore (mid-ocean exchange), where productivity is generally lower. However, several studies have shown that mid-ocean exchange does not always decrease plankton density (MacDonald and Davidson, 1998; Wonham *et al.*, 2001; Roy *et al.*, 2012). However, marine zooplankton has also been shown to have reduced diversity offshore (Fernando *et al.*, 1990). IMO recommendations for international vessels require that BWE be conducted at least 200 nautical miles from the nearest land and in a minimal depth of 200m, in order to take on ballast waters with low organism density (International Maritime Organization, 2016). IMO recommendations also state that BWE should meet the D-2 Ballast Water Performance Standard, discharging less than 10 viable organisms of 50 µm minimal dimension by cubic meter at the port of destination (International Maritime Organization, 2016). Our two sites clearly failed to meet these standards, as densities deballasted in Deception Bay ranged from 5345 to 56 958 ind. m⁻³ ($\geq 80 \mu\text{m}$ dimension). Straits of Jacques Cartier and Belle Isle

are known to have high secondary productivity events in the summertime. However, they seem to have globally lower primary productivity than the northwestern Gulf (Anticosti gyre) and the Estuary (Starr *et al.*, 2003). These zones are used voluntarily by the vessel for operational reasons, mainly because they are located on the way to Deception Bay. These sites allow the vessel to remove freshwater on the way to Deception Bay, thus reducing ice formation in the ballast water tanks. Also, since there is currently no legislation concerning domestic ships in Canada, vessels are free to exchange ballast water in whatever location they choose.

Our results also show that exchanging ballast water in the Gulf of St. Lawrence did not significantly reduce or increase MZP diversity. Exchanged tanks were not significantly more or less diverse than control tanks, even though controls had higher ballast water age and increased mortality. Thus, voluntary BWE conducted by the *M/V Arctic* failed to reduce MZP diversity. Reduction of plankton diversity is another reason for conducting BWE under international law. These results reject our initial hypothesis stating that BWE in coastal marine areas would increase MZP diversity. Marine zooplankton communities are generally more diverse than freshwater ones, due to the higher distinction of meroplanktonic and holoplanktonic components in marine environments. In addition, since our exchange zones were located in a coastal area, they were expected to provide higher diversity than mid-ocean locations (Lehman, 1988; Fernando *et al.*, 1990). These results were unexpected and could be explained by exchanged ballast water originating from surface waters with lower diversity, with several deepwater species missing.

Perspectives on ballast water sampling difficulties and study limitations

Literature on BWE shows much uncertainty about the true efficacy of this ballast water management method. There are often considerable limitations in the interpretation of results and in the representativeness of data, due to opportunistic experimental designs and sampling techniques. The present study is no exception. The absence of replicates (ships) and pseudoreplicates (tanks) prevented us from generalizing our results to other vessels using the same route. The absence of technical replicates did not allow evaluating the error due to the sampling method in ballast water with possible organism patchiness. Several studies have described plankton patchiness in ballast waters, thus samples can sometimes be unrepresentative of real-time plankton density. Vertical migration, concentration and sedimentation of organisms can occur in the water column of ballast tanks, causing inaccurate evaluation of population estimates. It has also been suggested that light penetration during sampling, when manhole covers are open, could be sufficient to drive the vertical migration of copepods in the ballast tanks. When ballast water is held for longer periods of time, as in our control tanks, moribund or inactive individuals may also settle to lower tank strata (First *et al.*, 2013). Moreover, differential mixing of ballast water due to ship's movements on the surface may greatly alter organism distribution. Organisms are unlikely to have random distribution because of their tendency to aggregate, in response to physical parameters, predation, resources or morphotype seeking (Frazier *et al.*, 2013). Zooplankton density in ballast waters has been shown to differ with depth (Murphy *et al.*, 2002). Temporal and spatial variability of plankton dispersion may not be accurately estimated with actual sampling methods (Costa *et al.*, 2015). The collection of technical replicates (additional samples of the same ballast water tank collected with alternative sampling methods) could have allowed us to see variability due to the sampling method. Our sampling campaign did not include them because of limited sampling time (daylight time), equipment and personnel. Since we were also sampling phytoplankton, dinoflagellates and bacterioplankton, the entire day was committed to the preparation and collection of the different samples. Having collected two or three technical replicates per tank to assure representativity of whole ballast tanks would have required nightshifts,

which were not allowed aboard the ship. However, having filtered a large volume of 1000L with multiple net tows gave the best representation as possible of the water column, considering sampling limitations.

Finally, there were limitations in our taxonomic analyses. Some groups, such as barnacle nauplii and cyprids, fish eggs, crab zoea and megalope larvae were observed but left unidentified due to taxonomic limitations. These groups were observed in the exchange zones, but not in Deception Bay. Some other groups, observed both in the exchange zones and in Deception Bay, were also left unidentified, such as polychaete worm larvae, bryozoans cyphonauts and various eggs. In addition, some fish larvae, aquatic insects and insect larvae were occasionally observed in Quebec City harbor but left unidentified. Thus, we cannot comment on their invasion potential or their nonindigenous status due to the lack of taxonomic resolution. Alternative taxonomic analysis techniques, such as genetic metabarcoding, could however resolve this issue, allowing the identification of other potential benthic NIS carried in ballast water as planktonic larval stages.

CONCLUSION GÉNÉRALE

Les eaux de ballast de navires domestiques peuvent agir comme vecteur d'introduction d'espèces planctoniques non-indigènes. Cela est dû au fait qu'il peut y avoir une forte similarité environnementale entre les ports d'origine et de destination sur un trajet domestique, augmentant ainsi les chances de survie et d'établissement des espèces transportées. De plus, ces navires effectuent généralement des trajets de plus courte durée que les navires internationaux, augmentant également les chances de survie des organismes contenus dans l'eau de ballast. L'effet limité de l'âge de l'eau de ballast sur un trajet domestique augmente le nombre de propagules viables au moment du déballastage. De plus, le trafic domestique peut agir à titre de vecteur d'introduction secondaire à partir de ports d'origine où des ENI sont déjà présentes, en les transportant dans d'autres régions. Cette étude présente une évaluation des risques d'introduction d'espèces de MZP non-indigènes via les eaux de ballast du vraquier *M/V Arctic*, qui transite à l'année longue entre le port de Québec (port d'origine) et le port de Baie Déception (port de destination). L'échange des eaux de ballast a été réalisé aux détroits de Jacques-Cartier (Site #1) et de Belle-Isle (Site #2) afin d'évaluer les risques d'introduction liés à ces deux zones d'échange volontaire d'eau de ballast, régulièrement utilisées par le navire. Finalement, une évaluation du risque d'introduction a également été effectuée sur des réservoirs contrôles contenant de l'eau du port d'origine, simulant ainsi un voyage entre les ports de Québec et de Baie Déception sans ÉEB.

Obj. #1 : Évaluer la variabilité mensuelle estivale dans les communautés mészooplanctoniques présentes dans les réservoirs de ballast du M/V Arctic au port d'origine (Québec) - Commentaires et limitations du design expérimental

Pour répondre à cet objectif de recherche, nous avons utilisé les résultats des analyses réalisées sur les échantillons prélevés au port de Québec dans les trois réservoirs de ballast. Chacun des trois réservoirs échantillonnés correspondait à un réplicat pour chacun des cinq mois échantillonnés. Étant donné qu'au port d'origine, l'équipage du navire remplit les réservoirs de ballast progressivement en fonction des opérations de déchargement, l'eau contenue dans les trois réservoirs est en fait un mélange d'eaux provenant de dates et d'emplacements différents dans le port. Ceux-ci ont été remplis pour la plupart du temps en deux ou trois phases distinctes, pouvant être séparées temporellement par plusieurs jours, voire même jusqu'à une semaine. Il s'agit donc d'un mélange de plusieurs masses d'eau d'eau d'âges différents. De plus, si le navire devait changer de quai pendant les opérations de déchargement, il pouvait même arriver qu'un réservoir soit rempli à des endroits différents dans le port de Québec (estuaire de la rivière Saint-Charles, anse au Foulon). Cette contrainte, due aux opérations du navire, pourrait affecter grandement les communautés de départ présentes dans les réservoirs de ballast. Un réservoir contenant un mélange d'eaux plus vieilles pourrait donc contenir moins d'organismes en raison de l'effet du vieillissement des eaux sur la mortalité. Cela pourrait expliquer pourquoi il y a parfois autant de différence dans la densité entre les trois réservoirs au port d'origine. De plus, il est à noter que les trois réservoirs du mois de juin n'ont pu être échantillonnés au port de Québec pour des questions de logistique maritime. Le navire a dû quitter dès notre arrivée à bord et nous n'avons donc pu échantillonner que le lendemain, avant l'ÉEB au site #1. Après consultation, nous avons toutefois décidé de traiter ces échantillons comme s'ils avaient été également prélevés au port de Québec. Finalement, l'absence de pseudoréplicats aux autres stations du voyage ne nous permettait pas d'évaluer l'effet de la saisonnalité sur les risques d'introduction liés à l'ÉEB dans le golfe du Saint-Laurent.

Obj. #1 : Évaluer la variabilité mensuelle estivale dans les communautés mésozooplanctoniques présentes dans les réservoirs de ballast du M/V Arctic au port d'origine (Québec) - Conclusions

Nos résultats mettent en évidence un effet marqué du mois sur la densité totale en organismes. La densité totale en organismes, ainsi que la densité d'ENI notoires dans les réservoirs du navire étaient nettement plus élevées au mois d'août, correspondant au pic estival d'abondance du zooplancton, venant ainsi confirmer en partie notre hypothèse de départ. Le port de Québec a fourni au mois d'août une densité maximale en propagules d'ENI à haut potentiel invasif décrites dans la littérature, étant donnée l'augmentation marquée dans la densité d'*Eurytemora carolleae*. Nous considérons ce taxon comme un envahisseur potentiel en raison de son statut eurytherme/euryhalin et de son potentiel élevé d'invasion décrit dans la littérature (voir Section 1.4.2). C'est également au mois d'août que le port d'origine fournissait une densité maximale en propagules de moules zébrée/quagga, deux espèces envahissantes à fort impact écologique (voir Section 1.4.2).

Nous n'avons pu observer aucun effet du mois sur la diversité en organismes, venant infirmer notre hypothèse de départ qui stipulait que la diversité serait minimale pendant le pic d'abondance du MZP en raison d'une communauté dominée par un minimum d'espèces. Ce résultat était en désaccord avec la littérature, décrivant une diversité minimale lors des maxima d'abondance du zooplancton (Irigoien *et al.*, 2004). La communauté au mois d'août se distinguait toutefois par un plus faible nombre d'espèces (richesse) que les autres mois. Les réservoirs au mois d'août à Québec étaient également caractérisés par une prédominance quasi-totale (99,7% en moyenne) du copépode calanoïde *Eurytemora carolleae* et de ses nauplii, du cladocère *Bosmina longirostris* et des véligères (moules zébrées/quagga, *Dreissena polymorpha/bugensis*). Bien que ces deux espèces de la famille des Dreissenidae soient reconnues internationalement comme des espèces envahissantes parmi les plus dévastatrices, leur potentiel d'invasion se limite aux eaux douces et oligohalines. Ainsi, il n'y aurait pas de possibilité pour cette espèce d'envahir de manière permanente l'environnement polyhalin du fjord de Baie Déception.

Obj. #2 : Commentaires et limitations du design expérimental

Nous ne disposions que d'un unique pseudoréplicat pour cet objectif (un réservoir pour chaque site à chaque voyage), ainsi la puissance de nos analyses statistiques est limitée. Pour plus de puissance statistique, il aurait fallu disposer à chaque voyage de trois réservoirs contrôles, de trois réservoirs échangés au Site #1 et de trois réservoirs échangés au Site #2 pour disposer tout au moins de pseudoréplication. Évidemment, un tel plan d'échantillonnage n'aurait pas été réaliste, étant donné que nous ne pouvions monopoliser neuf réservoirs de ballast à des fins expérimentales, compte tenu des contraintes de chargement et d'ÉEB qui doivent être respectées par l'équipage du navire. Un autre moyen d'augmenter la puissance statistique de nos résultats aurait été de disposer simultanément de trois navires pour réaliser l'expérience, chacun d'entre eux agissant comme réplicat pour chacun des voyages mensuels. Bien-sûr, un tel design n'est pas réaliste dans le cadre d'une étude sur les eaux de ballast. Nos résultats ne peuvent donc qu'être appliqués au *M/V Arctic* pour l'année 2015, sans possibilité de généraliser nos résultats à tous les navires domestiques canadiens empruntant cette même route et échangeant dans les mêmes zones. Néanmoins, il s'agit là de nouveaux résultats pour cette région et ils pourraient être utilisés à titre consultatifs par les navires pratiquant l'échange d'eau de ballast dans ces zones.

Obj. #2 : Risques d'introduction liés au déballastage de réservoirs inchangés (contrôles)

Selon nos résultats, les eaux de ballast transportées par le *M/V Arctic* représentent un vecteur potentiel d'introduction d'ENI si elles n'ont pas été échangées. L'eau prélevée dans le port d'origine contenait un grand nombre d'espèces dulcicoles qui n'ont pas été retrouvées dans les échantillons du port de Baie Déception. Ces espèces, bien que non-indigènes, sont pour la plupart sténohalines ; le copépode calanoïde *Eurytemora carolleae* faisant toutefois exception. Ces espèces dulcicoles sténohalines ne pourraient survivre au choc osmotique important lors du déballastage dans l'environnement marin de Baie Déception ($S \approx 29$ psu). Seul *Eurytemora carolleae* représente un risque réel pour l'établissement à Baie Déception. Il a été démontré que cette espèce, dominante dans nos

réservoirs contrôles, peut survivre à une gamme de salinité variant entre 0 et 40 grâce à une grande capacité d'osmorégulation (Lee *et al.*, 2013). De plus, ce taxon est déjà reconnu pour avoir une forte capacité d'invasion, ayant été introduit dans les Grands Lacs dans les années 1950 (Vasquez *et al.*, 2016). À la fin des cinq trajets réalisés par le navire, l'eau contenue dans le réservoir contrôle contenait toujours des densités importantes de propagules de zooplancton, dont notamment *Eurytemora carolleeae*. Celà témoigne de leur survie pendant le trajet, malgré la pression de mortalité exercée sur la communauté mésozooplanctonique par le vieillissement des eaux de ballast.

Le fait d'avoir ajouté un réservoir contrôle au design expérimental nous a permis de simuler les risques d'introduction associés à un trajet du *M/V Arctic* sans ÉEB. Toutefois, en réalité, ce navire réalise en tout temps l'échange de ses réservoirs dans le golfe du Saint-Laurent pour des questions de logistique maritime. Le fait d'effectuer l'échange de ses réservoirs permet d'y purger l'eau douce et de la remplacer par de l'eau de mer, dont le point de congélation est plus bas. Cette pratique permet de réduire la formation de glace à l'intérieur des réservoirs, qui pourrait endommager les pompes à ballast et les cloisons des réservoirs. Cela est crucial pour un navire opérant sur un trajet en zone subarctique, où les températures sont très basses la majeure partie de l'année. Le navire n'a déballasté de l'eau douce à Baie Déception uniquement que dans le contexte de l'expérience que nous avons réalisée. Ainsi, il est donc théoriquement impossible pour le navire d'introduire, via les eaux de ballast, des organismes du port de Québec à Baie Déception. De plus, l'analyse des échantillons prélevés dans les réservoirs fraîchement échangés dans le golfe a révélé que ceux-ci ne contenaient pratiquement plus d'organismes dulcicoles, témoignant de l'efficacité de l'échange à détruire ou purger les organismes du port d'origine. Aucune expérience de viabilité n'a toutefois été réalisée sur le peu d'organismes dulcicoles résiduels trouvés dans les réservoirs échangés, par conséquent il n'est pas possible de dire si ces organismes avaient survécu ou non à l'ÉEB.

Obj. #2 : Effet de l'âge d'eau de ballast sur la densité totale et la diversité du MZP

Il était difficile d'attribuer un âge d'eau de ballast initial à l'eau contenue dans les réservoirs contrôles lorsque nous avons échantillonné ceux-ci au port de Québec. Étant donné qu'au port d'origine, l'équipage du navire remplit les réservoirs de ballast progressivement en fonction des opérations de déchargement, l'eau contenue dans les réservoirs contrôles est en fait un mélange d'eaux provenant de dates et d'emplacements différents dans le port de Québec. Ceux-ci ont été remplis pour la plupart du temps en trois phases distinctes, pouvant être séparées temporellement par plusieurs jours, voire même jusqu'à une semaine. Il s'agit donc d'un mélange de plusieurs masses d'eau d'âges différents. Il était ainsi complexe d'attribuer un âge d'eau de ballast d'origine aux réservoirs en fonction de leur historique de remplissage. Ainsi, pour éviter ce problème, nous avons considéré que l'âge d'eau de ballast lors de l'échantillonnage initial à Québec était de 0 jour. Nous n'avons donc évalué que l'effet du vieillissement des eaux de ballast pendant le voyage entre le port d'origine et le port de destination. Les résultats renseignent donc plutôt sur l'effet du vieillissement des eaux de ballast sur la mortalité du MZP dans des réservoirs où la densité initiale en organismes était connue, mais pas l'âge initial véritable. Ainsi, il s'agit donc de l'effet de la durée de la traversée sur la densité totale en organismes initiale.

Nous avons observé un effet négatif de l'âge d'eau de ballast sur la densité totale en organismes, établissant un lien direct entre le vieillissement de l'eau de ballast et la mortalité chez le MZP. Cependant, il arrivait parfois que la densité totale en organismes puisse augmenter pendant le trajet, ce qui peut s'expliquer par deux raisons. Le réservoir de ballast pourrait avoir agi comme incubateur pour le MZP. Plusieurs femelles ovigères (*Bosmina longirostris*, *Eurytemora carolleae*) ont été retrouvées dans les échantillons et il est donc possible qu'une éclosion massive de larves pendant la traversée ait fait augmenter la densité totale en organismes. Cette augmentation pourrait également être due, moins probablement, à la répartition hétérogène des organismes au sein du réservoir. L'absence de replicats techniques (techniques d'échantillonnage) dans notre plan d'expérience, pour des raisons de contraintes de temps et de matériel, ne nous permet toutefois pas d'évaluer

l'erreur due à cette hétérogénéité. Des traits de filet à plancton successifs ont toutefois été utilisés pour avoir une représentation du réservoir la plus fidèle possible. Finalement, nous avons également observé un effet négatif, quoique non-significatif, de l'âge d'eau de ballast sur la diversité du MZP.

Obj. #2 : Perspectives et commentaires sur les risques liés à l'échange d'eau de ballast (ÉEB) dans le golfe du Saint-Laurent

Les analyses de taxonomie classique réalisées dans le cadre de ce projet ne nous permettent pas de faire la différence entre de potentielles sous-populations différentes au sein d'une même espèce (possibilité de complexes d'espèces cryptiques morphologiquement semblables). Seules des analyses génétiques permettraient de voir si l'ÉEB dans le golfe du Saint-Laurent serait un vecteur d'introduction de populations de la même espèce (sous-espèces, souches) qui soient non-indigènes. Finalement, les cinq voyages pendant lesquels nous avons procédé à l'échantillonnage des eaux de ballast ne nous permettent pas de certifier hors de tout doute qu'il n'existe pas, à d'autres périodes de l'année, d'autres espèces n'ayant pas été observées dans les zones d'échange qui pourraient être non-indigènes à Baie Déception. L'échantillonnage ayant été réalisé dans une fenêtre de temps très restreinte (cinq voyages répartis entre l'été et l'automne 2015), cette expérience ne nous renseigne donc pas sur la totalité de la communauté mésozooplanctonique présente pendant toute l'année dans le golfe du Saint-Laurent. Il n'est pas exclu que d'autres espèces, ainsi que des ENI déjà établies dans le golfe soient présentes à d'autres moments. Il n'est donc pas possible de certifier hors de tout doute que de réaliser l'échange d'eau de ballast dans le golfe du Saint-Laurent ne soit pas un vecteur potentiel d'introduction d'ENI. Seul un monitorage complet de la succession zooplanctonique du golfe et des analyses génétiques réalisées sur les communautés indigènes de Baie Déception et dans les zones d'échange pourraient permettre de telles conclusions.

Le fait de procéder à l'échange d'eau de ballast dans le golfe du Saint-Laurent évoque toutefois l'éventualité d'introductions potentielles d'espèces au sein des zones d'échange. Le *M/V Arctic* échange régulièrement ses eaux de ballast dans le nord du golfe depuis plusieurs années, en y déversant une grande quantité de propagules d'*Eurytemora carolleae*. Toutefois, cette espèce n'a pas encore été rapportée dans la littérature comme ENI introduite dans le golfe (Dufour and Ouellet, 2007). Bien que tolérante à des variations drastiques de la salinité et de la température, cette espèce est pourtant confinée à la zone fluviale et aux eaux oligohalines de la zone de transition estuarienne de l'estuaire du Saint-Laurent. Elle a également été observée dans les marelles hypersalines de l'Île Verte ($S \approx 40$ psu). La section mésohaline de la zone de transition estuarienne de l'estuaire (entre l'île d'Orléans et l'île-aux-Coudres) est plutôt occupée par *Eurytemora affinis* (clade Nord-Atlantique), tandis que les eaux polyhalines de l'estuaire supérieur et du golfe sont occupées par *Eurytemora herdmani*. Les différentes espèces au sein du genre *Eurytemora* sont soumises à une forte ségrégation spatiale limitant leur coexistence (Fofonoff *et al.*, 2003 ; Winkler *et al.*, 2016). Il est possible qu'*Eurytemora carolleae* ne puisse être introduit dans le golfe par exclusion compétitive. Les interactions et la compétition avec les autres espèces occupant une niche trophique semblable pourraient expliquer l'absence d'*Eurytemora carolleae* dans l'estuaire moyen et le golfe du Saint-Laurent, malgré plusieurs années de déversement dans ces régions. De plus, la forte tolérance de cette espèce euryhaline et eurytherme n'a été documentée de manière expérimentale que sur des individus matures. La tolérance à des changements importants des facteurs abiotiques chez les stades juvéniles et les nauplii n'a pas encore été documentée. Ainsi, l'échange d'eau de ballast dans le golfe ne semble pas présenter de risques sérieux, car aucun des taxons dulcicoles prélevés par le *M/V Arctic* dans l'estuaire fluvial n'a encore été rapporté comme ENI dans le golfe. Pourtant, le ministère des Pêches et Océans du Canada (MPO) effectue chaque année un suivi rigoureux des espèces envahissantes présentes dans le golfe.

Cependant, nous pouvons conclure que l'échange d'eau de ballast a permis de purger l'eau douce riche en ENI, la remplaçant par une eau de mer plus riche en organismes, mais contenant très peu d'ENI. De plus, omettre d'échanger l'eau de ballast du *M/V Arctic* a mené au déversement, en quantités variables, de trois ENI notoires, dont l'une d'elles présentait un potentiel élevé de survie et d'établissement à Baie Déception (*Eurytemora carolleae*). Toutefois, l'absence de pseudoréplication aux zones d'échange ne nous permet pas de généraliser ces résultats concernant l'échange d'eau de ballast dans le golfe à d'autres navires domestiques empruntant la même route, même ceux-ci montraient un patron similaire à chacun des voyages et ont fourni d'importantes et nouvelles connaissances sur le sujet pour l'Arctique canadien. Ces résultats ont apporté une contribution significative dans le domaine pour cette région, même s'ils doivent être utilisés avec précaution.

Perspectives sur la gestion des eaux de ballast en Arctique

L'Organisation Maritime Internationale (OMI) a adopté en 1991 une série de recommandations effectuées sur base volontaire pour minimiser les effets néfastes du déversement d'eau de ballast à travers l'ÉEB. Or, l'efficacité des ÉEB est très variable, selon le trajet emprunté, l'emplacement de la zone d'échange, la saison, la structure et la capacité du navire, la latitude, la durée du trajet, l'emplacement où se situent les ports d'origine et de destination, les conditions météorologiques ou encore le type d'organisme ciblé (Jing *et al.*, 2012). Bien que le procédé d'ÉEB ait certes contribué à ralentir la fréquence d'introduction d'ENI par rapport à l'époque où aucune réglementation n'était encore en vigueur, leur mise en place n'a pu empêcher complètement le transfert d'organismes aquatiques envahissants. Il s'agissait plutôt d'une solution transitoire à un problème qui était à l'époque encore peu étudié. Maintenant que la complexité de ce vecteur d'introduction a été démontrée, il apparaît clairement que l'ÉEB ne suffit pas à éradiquer complètement les risques d'introduction dans un grand nombre de situations. Une importante revue de littérature sur les traitements d'eau de ballast, réalisée par Jing *et al.* (2012), a permis de démontrer la nécessité d'utiliser l'ÉEB conjointement avec une autre méthode de gestion des eaux de ballast, dans le but de répondre aux standards de densité en organismes établis par l'OMI. Plusieurs traitements d'eau de ballast ont été testés au cours des dernières années, chacun ayant ses avantages et inconvénients. De plus, les coûts, limitations et aspects logistiques liés à chacun des traitements varient considérablement. Un procédé doit répondre à trois critères déterminant son applicabilité : risques environnementaux, capacité à détruire les organismes de tailles variables (virus au macrozooplancton) dans des conditions physicochimiques variables (température, salinité et nutriments), ainsi que coûts et contraintes techniques (le traitement doit être facilement réalisable et sécuritaire pour l'industrie maritime). Certains auteurs ont démontré qu'aucun traitement unique ne pouvait répondre simultanément à tous ces critères (Silva *et al.*, 2004 ; Gregg *et al.*, 2009).

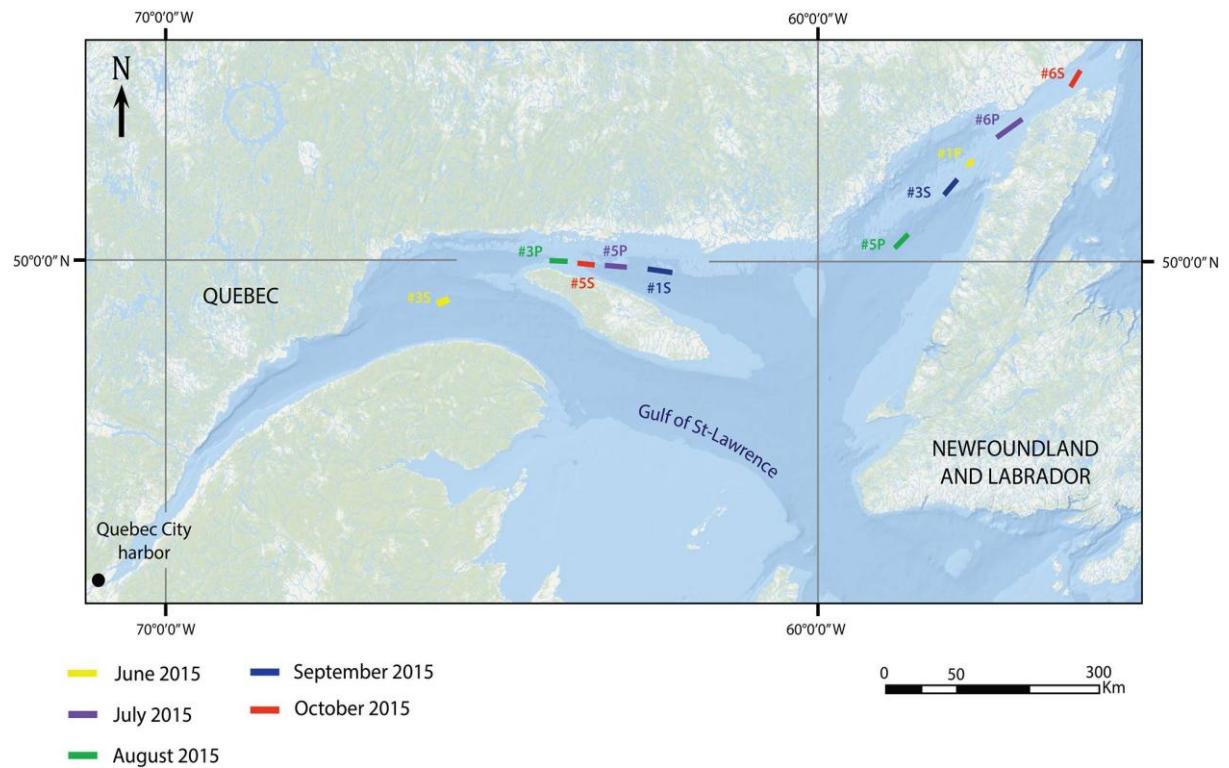
Ces traitements incluent filtration, traitement thermique, traitement par hydrocyclone, traitement aux rayons UV, ultrasons, ozonation, biocides, oxydation, impulsion électrique, désoxygénéation et traitement magnétique (Jing *et al.*, 2012). Ils peuvent être combinés et/ou utilisés conjointement avec l'ÉEB. Or, la rudesse et l'imprévisibilité du climat dans l'Arctique et dans l'Atlantique Nord peuvent compromettre grandement l'efficacité et même l'applicabilité de ces méthodes de gestion d'eau de ballast. L'applicabilité de ces traitements à la navigation arctique a été peu étudiée, ainsi il est impossible pour l'instant de proposer une combinaison de traitements qui serait adéquate pour cette région du monde. Le traitement thermique ne serait pas applicable en zone arctique en raison des températures extrêmes, qui peuvent également réduire l'efficacité de plusieurs biocides nécessitant une gamme de températures spécifiques pour les réactions chimiques (Jing *et al.*, 2012). De plus, les brise-glaces naviguant dans l'Arctique doivent pomper de l'eau de ballast régulièrement et rapidement durant le trajet lorsque le navire est pris dans les glaces. Le déplacement contrôlé des eaux dans la cale peut aider à libérer le navire de l'emprise des glaces. Pour être implantés sur les navires naviguant en zone arctique, les traitements d'eau de ballast doivent donc être rapides d'utilisation et ne pas compromettre la capacité de pompage (Tim Keane, capitaine du *M/V Arctic*, communication personnelle). Ainsi, le traitement physique des eaux de ballast par hydrocyclone serait un procédé intéressant pour les navires naviguant dans l'Arctique, car il permet de séparer les organismes de l'eau à de fortes pressions (débit $\approx 3000 \text{ m}^3 \text{ h}^{-1}$). Ce procédé peut également séparer les sédiments de l'eau au moment du ballastage, minimisant ainsi l'accumulation de sédiments dans les réservoirs de ballast, qui agissent comme un autre vecteur important d'introduction d'ENI (Jing *et al.*, 2012). De plus, cette méthode ne pose aucun risque environnemental puisque les organismes filtrés sont retournés immédiatement dans l'eau du port d'origine au moment du ballastage. Or, ce traitement n'est pas totalement efficace et ne peut retirer efficacement le zooplancton de petite taille, de même que le phytoplancton et le bactérioplancton. Il sert donc à préfiltrer l'eau et doit être utilisé préalablement à un deuxième traitement. L'inclusion de la phase microbienne du plancton dans les standards de traitement d'eau de ballast de l'OMI limite l'applicabilité des procédés physiques sains

pour l'environnement. Vu les conditions climatiques extrêmes en zone arctique, l'utilisation de biocides est problématique en raison des risques de déversements. De surcroît, l'agitation de l'eau à l'intérieur des réservoirs de ballast, par mer agitée, peut endommager les installations nécessaires au traitement d'eau de ballast, telles que les lampes UV et les hydrocyclones (Jing *et al.*, 2012). La mer du Labrador est empruntée par le *M/V Arctic* pour se rendre dans le détroit d'Hudson et est fréquemment agitée par des vents forts et des tempêtes, ce qui compromettrait l'efficacité et l'intégrité des dispositifs de traitement d'eau de ballast. Celle-ci doit pourtant être échangée rapidement, pour minimiser les risques de bris des pompes dus au gel de l'eau douce. Le détroit d'Hudson est couvert de glace la majeure partie de l'année et le navire doit déjà avoir échangé son eau de ballast avant d'y entrer, rendant impossible les traitements d'eau de ballast dans cette région. L'armateur Fednav a récemment annoncé l'acquisition de systèmes de traitement par filtration et chlorination pour 16 de ses vraquiers transocéaniques destinés aux Grands Lacs (Marine Link, 2016). La mise en service de navires semblables pour l'Arctique canadien cependant représente un défi de taille en raison des contraintes opérationnelles.

Perspectives sur le rôle des eaux de ballast dans le contexte des changements climatiques

Le transport d'ENI via les eaux de ballast risque d'être profondément affecté par les changements climatiques dans les années à venir. Les scénarios climatiques pour les prochaines décennies suggèrent des changements dans les courants marins, la température et la salinité de l'eau, en plus de modifications dans la distribution et la phénologie des espèces indigènes. On prévoit donc des interactions complexes entre les ENI et les changements climatiques. Avec la diminution du couvert de glace dans l'océan Arctique, une augmentation du rayonnement solaire pourrait avoir des effets sur la capacité d'invasion de phytoplancton non-indigène. La fréquence des épisodes d'hypoxie hivernale pourrait aussi diminuer en raison d'une plus grande interaction atmosphère-océan dans le contexte de banquise fragmentée (Jing *et al.*, 2012). Ainsi, des espèces boréales ou tempérées, présentement inaptes à coloniser les eaux arctiques, pourraient voir leur aire de répartition ou leur potentiel d'invasion augmenter. Et bien que le développement minier arctique soit incertain pour les prochaines années, les ressources du Nord seront inévitablement de plus en plus convoitées par l'industrie minière. Une augmentation potentielle de la quantité d'eau de ballast déversée doit donc être considérée à long terme, même si les conditions de navigation resteront fortement hasardeuses, malgré la diminution de l'étendue de la glace de mer et l'ouverture de nouvelles routes maritimes. L'augmentation de la fréquence d'événements climatiques extrêmes dans l'Arctique et dans l'Atlantique nord risque également de compromettre l'efficacité de la plupart des mesures de gestion des eaux de ballast. Les réseaux trophiques de l'Arctique, peu résilients aux perturbations, seront également particulièrement vulnérables à l'introduction de nouvelles espèces (Jing *et al.*, 2012). Des introductions ayant eu des conséquences dévastatrices ont déjà été documentées dans d'autres régions subarctiques et tempérées (Grands Lacs, mer Baltique, Côte Est américaine, mer Noire), où les réseaux trophiques sont généralement plus résilients. Celles-ci pourraient donc avoir des impacts irréversibles sur les réseaux trophiques de l'Arctique et sur les communautés humaines qui y sont établies et dont le mode de vie est indissociable de la chasse et la pêche de subsistance.

ANNEXES



Annexe I. Localisation des échanges d'eau de ballast réalisés par le *M/V Arctic* pendant la campagne d'échantillonnage 2015. Le numéro du réservoir de ballast employé est indiqué pour chaque échange. S = Tribord ; P = Bâbord (Laget, 2016)

		June			
Sampling zones Treatments		Quebec City	Site 1	Site 2	Deception Bay
Start		H			
Before exchange			C S1 S2	S2	
After exchange			C S1	C S2	
End				C S1 S2 H	

		July			
Sampling zones Treatments		Quebec City	Site 1	Site 2	Deception Bay
Start		C S1 S2 H			
Before exchange			C S1	S2	
After exchange			C S1	C S2	
End				C S1 S2 H	

		August			
Sampling zones Treatments		Quebec City	Site 1	Site 2	Deception Bay
Start		C S1 S2 H			
Before exchange			C S1	S2	
After exchange			C S1	C S2	
End				C S1 S2 H	

		September			
Sampling zones Treatments		Quebec City	Site 1	Site 2	Deception Bay
Start		C S1 S2 H			
Before exchange					
After exchange					
End					

		October			
Sampling zones Treatments		Quebec City	Site 1	Site 2	Deception Bay
Start		C S1 S2 H			
Before exchange					
After exchange					
End					S1 S2

Annexe II. Schéma du design expérimental lors de l'échantillonnage réalisé à bord du *M/V Arctic*. Site #1 = détroit de Jacques-Cartier, Site #2 = détroit de Belle-Isle. S1 = Réservoir échangé au Site #1, S2 = Réservoir échangé au Site #2, C = Réservoir contrôle, H = Échantillon de port (Harbor). Les cases vertes correspondent à des réservoirs contenant de l'eau douce (non-échangeés), les cases bleues à des réservoirs contenant de l'eau de mer (échangés) et les cases rouges à des échantillons de port (Laget, 2016)

Annexe III. Listes des taxons trouvés en fonction du site et leur statut à Baie Déception

Taxa	Quebec City harbor	Site 1	Site 2	NIS to Deception Bay
Acari	X	-	-	NIS
<i>Acartia</i> sp. (♀ ♂ longiremis)	-	X	X	-
<i>Alona rustica</i>	X	-	-	NIS
<i>Alona</i> spp.	X	-	-	NIS
Appendicularia	-	-	X	NIS
<i>Ascomorpha</i> spp.	X	-	-	NIS
<i>Asplanchna</i> spp.	X	-	-	NIS
<i>Bosmina longirostris</i>	X	-	-	NIS
<i>Brachionus bidentata</i>	X	-	-	NIS
<i>Brachionus calyciflorus</i>	X	-	-	NIS
<i>Brachionus havanaensis</i>	X	-	-	NIS
<i>Brachionus quadridentatus</i>	X	-	-	NIS
<i>Brachionus rubens</i>	X	-	-	NIS
Bryozoa cyphonauts	-	X	X	NIS
<i>Calanus finmarchicus</i>	-	X	X	-
<i>Calanus glacialis</i>	-	-	-	-
<i>Calanus hyperboreus</i>	-	-	X	-
<i>Centropages</i> sp. (♀ ♂ hamatus)	-	X	X	-
<i>Ceriodaphnia dubia</i>	X	-	-	NIS
<i>Ceriodaphnia</i> spp.	X	-	-	NIS
Chaetognatha	-	-	X	NIS
<i>Chydorus sphaericus</i>	X	-	-	NIS
Cirriped nauplii	-	-	X	-
Cirripedia cyprid	-	X	-	NIS
Cladoceran eggs	X	-	-	NIS
Cnidaria	-	-	-	-
Copepod nauplii	X	X	X	-
Crab megalopes	-	-	X	NIS
Crab zoea	-	-	X	NIS
<i>Daphnia dubia</i>	X	-	-	NIS
<i>Daphnia magna</i>	X	-	-	NIS
<i>Daphnia mendotae</i>	X	-	-	NIS
<i>Daphnia</i> spp.	X	-	-	NIS
<i>Diacyclops thomasi</i>	X	-	-	NIS
Echinodermata larvae	-	X	-	-
Egg sacs	X	X	X	-
<i>Epischura lacustris</i>	X	-	-	NIS
<i>Eucyclops agilis</i>	X	-	-	NIS
<i>Eucyclops prionophorus</i>	X	-	-	NIS
<i>Eukrohnia hamata</i>	-	-	X	-
<i>Eurytemora carolleeae</i>	X	-	-	NIS
<i>Eurytemora</i> spp. (♀ affinis)	-	X	X	NIS
<i>Evadne nordmanni</i>	-	X	-	NIS
<i>Evadne spinifera</i>	-	X	-	NIS
Fish eggs	-	-	X	NIS
Fish larvae	X	-	-	NIS
Freshwater calanoida	X	-	-	NIS
Freshwater cyclopoida	X	-	-	NIS
<i>Fritillaria</i> c.f. borealis	-	-	-	-
Gammaridae	X	-	-	NIS
Gastropoda	-	X	X	-
<i>Halicyclops fosteri</i>	X	-	-	NIS
Harpacticoida	X	-	X	-
Hydrozoa juveniles	-	X	X	-

Annexe IV. Listes des taxons trouvés en fonction du site et leur statut à Baie Déception (suite)

Taxa	Quebec City harbor	Site 1	Site 2	NIS to Deception Bay
Hyperiidae amphipods	-	-	-	-
Insect larvae	X	-	-	NIS
Insects	X	-	-	NIS
<i>Kellicottia longispina</i>	X	-	-	-
<i>Keratella c.f. crassa/earlinae/cochlearis</i>	X	-	-	NIS
<i>Keratella crassa</i>	X	-	-	NIS
<i>Keratella quadrata</i>	X	-	-	NIS
<i>Keratella serrulata</i>	X	-	-	NIS
<i>Keratella taurocephala</i>	X	-	-	NIS
<i>Keratella tecta</i>	X	-	-	NIS
<i>Keratella testudo</i>	X	-	-	NIS
<i>Lecane crepida</i>	X	-	-	NIS
<i>Lecane inermis</i>	X	-	-	NIS
<i>Lecane mira</i>	X	-	-	NIS
<i>Lecane spp.</i>	X	-	-	NIS
<i>Lecane unguilata</i>	X	-	-	NIS
<i>Lepadella patella</i>	X	-	-	NIS
<i>Leptodiaptomus sicilis</i>	X	-	-	NIS
Marine calanoida	-	X	X	-
<i>Mesochra alaskana</i>	X	-	-	NIS
<i>Mesocyclops edax</i>	X	-	-	NIS
<i>Microcalanus</i> spp.	-	X	-	-
<i>Microcyclops rubellus</i>	X	-	-	NIS
<i>Microsetella norvegica</i>	-	X	X	-
<i>Monostyla lunaris</i>	X	-	-	NIS
<i>Monostyla</i> spp.	X	-	-	NIS
Mussel veligers	X	X	X	-
<i>Nitokra hibernica</i>	X	-	-	NIS
<i>Notholca acuminata</i>	X	-	-	NIS
<i>Notholca labis</i>	X	-	-	NIS
<i>Oikopleura c.f. vanhoeffeni</i>	-	X	-	-
<i>Oithona similis</i>	-	X	X	-
<i>Oncaea borealis</i>	-	-	-	-
<i>Oncaea conifera</i>	-	-	-	-
<i>Oncaea</i> spp.	-	-	-	-
<i>Onychocamptus mohammed</i>	X	-	-	NIS
Ophiuridae	-	-	-	-
<i>Platyias quadricornis</i>	X	-	-	NIS
<i>Ploesoma lenticulare</i>	X	-	-	NIS
<i>Ploesoma truncatum</i>	X	-	-	NIS
<i>Podon</i> sp.	-	X	-	NIS
<i>Polyarthra dolichoptera</i>	X	-	-	NIS
Polychaeta larvae	X	X	-	-
<i>Pseudocalanus</i> spp. (<i>Enewmani</i>)	-	X	X	-
Rotiféra	X	X	X	-
<i>Schizopera borutzkyi</i>	X	-	-	NIS
<i>Scolecithricella minor</i>	-	X	X	NIS
<i>Sida crystallina</i>	X	-	-	NIS
<i>Temora longicornis</i>	-	X	X	-
<i>Trichocerca</i> spp.	X	-	-	NIS
<i>Tropocyclops prasinus</i>	X	-	-	NIS
Unidentified eggs	X	X	X	-
Unknown organisms	X	X	X	-

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