



Assessment of recreational boating as a vector for marine non-indigenous species on the Atlantic coast of Canada

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Received: 8 January 2018 / Accepted: 11 April 2019 / Published online: 4 May 2019
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Abstract We developed a new predictive approach to evaluate the relative invasion hazard posed by recreational boats as vectors for non-indigenous species (NIS) in marine ecoregions on the Atlantic coast of Canada. It combines data from behavioral boater questionnaires, surveys of boat macrofouling, and an extensive NIS monitoring program in marinas. The relative invasion hazard posed by boats in nine marine ecoregions was estimated by combining information on NIS infestation levels in source ecoregions, the probability that boats will be fouled, boat movements and environmental similarity between source and receiving marinas, and estimates of annual recreational traffic within and between ecoregions. A total of 52.1% of surveyed boats had macrofouling on hull or niche areas, suggesting a high

potential for NIS transport. High-risk boats were those that had spent extended periods in water, traveled extensively from invaded locations, and received little maintenance. Further, the high degree of connectivity between and within ecoregions, and high survival probabilities of NIS, highlight the threat of NIS introduction and spread via recreational boating in Atlantic Canadian waters. Of all Atlantic Canadian ecoregions, the Scotian Shelf ecoregion was classified as having the “Highest” Final Hazard score. However, boats with Intermediate to Highest scores travel to most ecoregions, underlining the importance of this vector to the introduction and spread of NIS in Atlantic Canadian coast. A simulated out-of-water cleaning after 3 months reduced the hazard in ecoregions with greater scores.

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Keywords Biological invasions · Biofouling · Non indigenous species · Recreational boat · Hazard assessment

Introduction

Non-indigenous species (NIS) are a growing global concern, posing significant ecological and economic threats to ecosystems around the world (Grosholz 2002; Molnar et al. 2008; Simberloff et al. 2013). For example, in Canada, the projected economic impact of the non-indigenous clubbed tunicate (*Styela clava*) on the oyster, scallop, clam and mussel productions has been estimated between 34 and 88 million Canadian dollars per year (Colautti et al. 2006). Multiple pathways allow species to be introduced to areas outside their natural ranges (Carlton 1985; Ruiz et al. 1997), highlighting the need for vector hazard assessments to better quantify, understand and limit the introduction and spread of NIS. Moreover, McGeoch et al. (2016) have stressed the importance of a science-based approach for pathway prioritization.

Many vectors transport NIS in marine and estuarine ecosystems, but ballast water carried by commercial ships and aquaculture transfers have been among the most studied (e.g., Fofonoff et al. 2003; McKindsey et al. 2007). Smaller recreational boats are also significant vectors of NIS via hull fouling (Ashton et al. 2006; Davidson et al. 2010) and may contribute to the secondary spread of NIS that have been introduced via other vectors, such as commercial shipping (Clarke Murray et al. 2011; Davidson et al. 2010; Floerl and Inglis 2003; Lacoursière-Roussel et al. 2012a). Williams et al. (2013) suggest that vessel biofouling in California accounted for as many NIS introductions as ballast water and aquaculture combined. Despite the role of recreational boat hull fouling in the introduction and spread of NIS, this vector remains largely unregulated (Ashton et al. 2012; Clarke Murray et al. 2011). Likewise, recreational

traffic patterns between marinas and ports from Atlantic coast of Canada are not well known because there is no mandatory Automated Identification System (Simard et al. 2014) as it is the case for commercial ships.

Species introduced and spread by recreational boating include diverse taxonomic groups, such as bivalves, algae, bryozoans, and ascidians (Bird et al. 1993; Farrell and Fletcher 2006; Floerl et al. 2005; Hay 1990). The probability of a boat being fouled depends on numerous factors, such as background environmental fouling pressure, time spent in marinas, boat type and boat maintenance practices (e.g., application and age of antifouling paint and cleaning of underwater structures), and travel history (Clarke Murray et al. 2011; Floerl and Inglis 2005). The latter may vary markedly among regions (Lacoursière-Roussel et al. 2012b) and knowledge of regional boat traffic patterns is thus needed to guide efficient NIS management efforts. A number of invasive ascidian introductions have been linked to recreational vessel hull fouling (Lambert and Lambert 1998; Lutzen 1999) and have been shown to cause substantial ecological and economic impacts (Aldred and Clare 2014; Zhan et al. 2015). In eastern Canada, population genetic similarity among marinas showed that recreational boats are important vectors for the spread of tunicates (Lacoursière-Roussel et al. 2012a).

The general objective of the present study was to evaluate the relative biosecurity hazard posed by recreational boats and boating with respect to the introduction and transfer of NIS among regions. Specifically, we developed a relative hazard scoring system for recreational boats among nine eastern Canada biogeographic ecoregions. Hazard scoring is based on the probability of non-resident recreational boats being biofouled by hull-fouling NIS and the probability of boats transporting those NIS that may establish in new locations, combined with estimates of boating activity.

Materials and methods

Boat hazard model

A Boat Invasion Hazard score (hereafter, “Boat Hazard score”) was calculated for each transient boat (i.e., a boat that visits a non-home marina in an Atlantic ecoregion for a minimum of one night) as the product of the probability that it is fouled by NIS (Boat Fouling Probability) and the probability that NIS arrive and survive in a non-home marina (Introduction Probability) (Fig. 1). Fouling species were assumed to survive transit, given the relatively short duration of most transits and that many fouling NIS may do so (Clarke Murray et al. 2012; Kauano et al. 2017). Boat Hazard scores were not attributed to resident boats

(i.e., boats that only undertake day trips or do not voyage at all, which represented 44% of all boats based on questionnaires), as their probability of introducing and spreading NIS to other marinas is limited.

Boat Fouling Probability for NIS was estimated as the product of regional NIS infestation levels (regional background of NIS in marinas–Regional NIS Background) and a Fouling Predictive Model which estimates the probability that a given boat will be fouled (boat and boater characteristics–Boat Infestation Probability). Introduction Probability was estimated as the product of the probability of a boat arriving (transient boat traffic data–Arrival Probability) and survival of NIS fouling species (salinity and

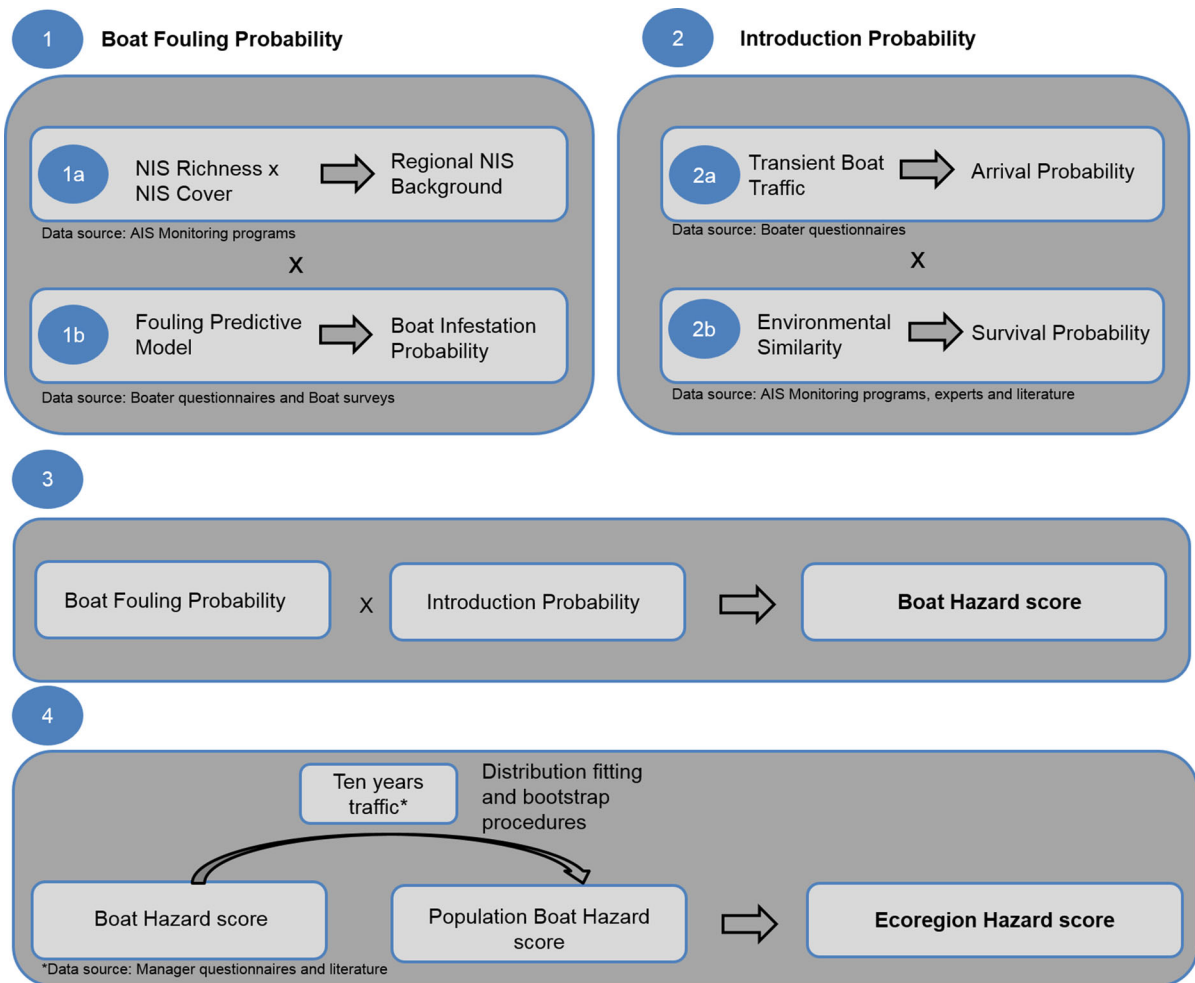


Fig. 1 Flow chart illustrating steps for recreational boating hazard assessment. Modified from Simard et al. (2017)

climate match between source and recipient marinas–Survival Probability).

$$\begin{aligned} \text{Boat Hazard score} = & \text{Regional NIS Background} \\ & \times \text{Boat Infestation Probability} \\ & \times \text{Arrival Probability} \\ & \times \text{Survival Probability} \end{aligned}$$

Boat Hazard scores were attributed to each boat visiting an ecoregion such that a single boat may have more than one score assigned to it if it visited multiple ecoregions. Each variable of the model was standardized by dividing these variables by the maximum value obtained for each variable to obtain a maximum possible value of 1. A minimum value of 0.001 instead of 0 was used because we assumed that there always remains some level of hazard, even if extremely small.

Study area

The Atlantic coast of Canada was divided into nine ecoregions using the Parks Canada biogeographic classification (Harper et al. 1993) (Fig. 2), which was chosen because it corresponds well to variations in native biological communities.

Data

Recreational boats included in this hazard assessment were generally less than 15 m in length (only 2.6% of boats > 15 m) and used for recreational purposes (sailboat, power boat) or small fishing boats (including converted fishing boats and combined fishing and recreational boats). Larger commercial fishing vessels were not included in this assessment.

Boater and marina manager questionnaires

We used boater questionnaires (a total of 1291 completed from 2009 to 2014) to evaluate boating movement patterns and maintenance history of the Canadian and foreign boaters (in 2009 by Lacoursière-Roussel et al. 2012a, b and in 2011–2014 by Fisheries and Oceans Canada, see Simard et al. 2017 for details). Boater questionnaires solicited information about boats (home marina, boat type, boat size, hull type, where it is stored or trailered), antifouling practices (type and time since last antifouling paint application,

time since last cleaning) and travel history (marinas visited, number of nights spent in each marina and types of trips undertaken). Trip types were classified as local day trips (departure and return to same marina in the same day), weekend trips (trips of a few days duration visiting 1–2 different locations), tours (long trips with multiple destinations along the way, staying in each moorage for only a few nights), and long trips (long-haul travel to destinations further away, and once there remain in a single moorage the entire time).

General information on marinas and their boating communities, including annual number of resident and transient boats, was obtained using questionnaires that were completed by 198 marina managers (details of the full questionnaires are presented in Simard et al. 2017).

Regional infestation

The regional level of NIS infestation was estimated based on a standardized regional Aquatic Invasive Species (AIS) monitoring program (McKenzie et al. 2016; Sephton et al. 2011; Simard et al. 2013). Settlement plates were used to monitor sites to assess NIS richness and percent cover, assuming that the organisms observed on plates reflect the species most likely to be transported by recreational boats. A total of 172 sites were surveyed between 2006 and 2014, most over multiple years, and sampling effort (number of plates and time of immersion) was standardized among sites (see Simard et al. 2017 for detailed methods). NIS richness at each site was defined as the total number of NIS observed on all plates in that location over the history of the monitoring program prior to and including 2014, when the recreational boater surveys were concluded. NIS cover was determined semi-quantitatively by visual examination; categories for the percentage cover were 0 (absent); < 25% coverage; 25–50% coverage; 51–75% coverage and > 75% coverage. For each plate, the median of each category (%) was used to calculate the total NIS cover (sum of all species) and then averaged per location per year. The overall NIS cover was defined as the maximum mean NIS cover observed in a given location for any given sampling year over the survey period. Maximal mean NIS cover was used as a precautionary approach (worst case scenario) and to also not underestimating NIS cover for sites where species have been recently introduced.

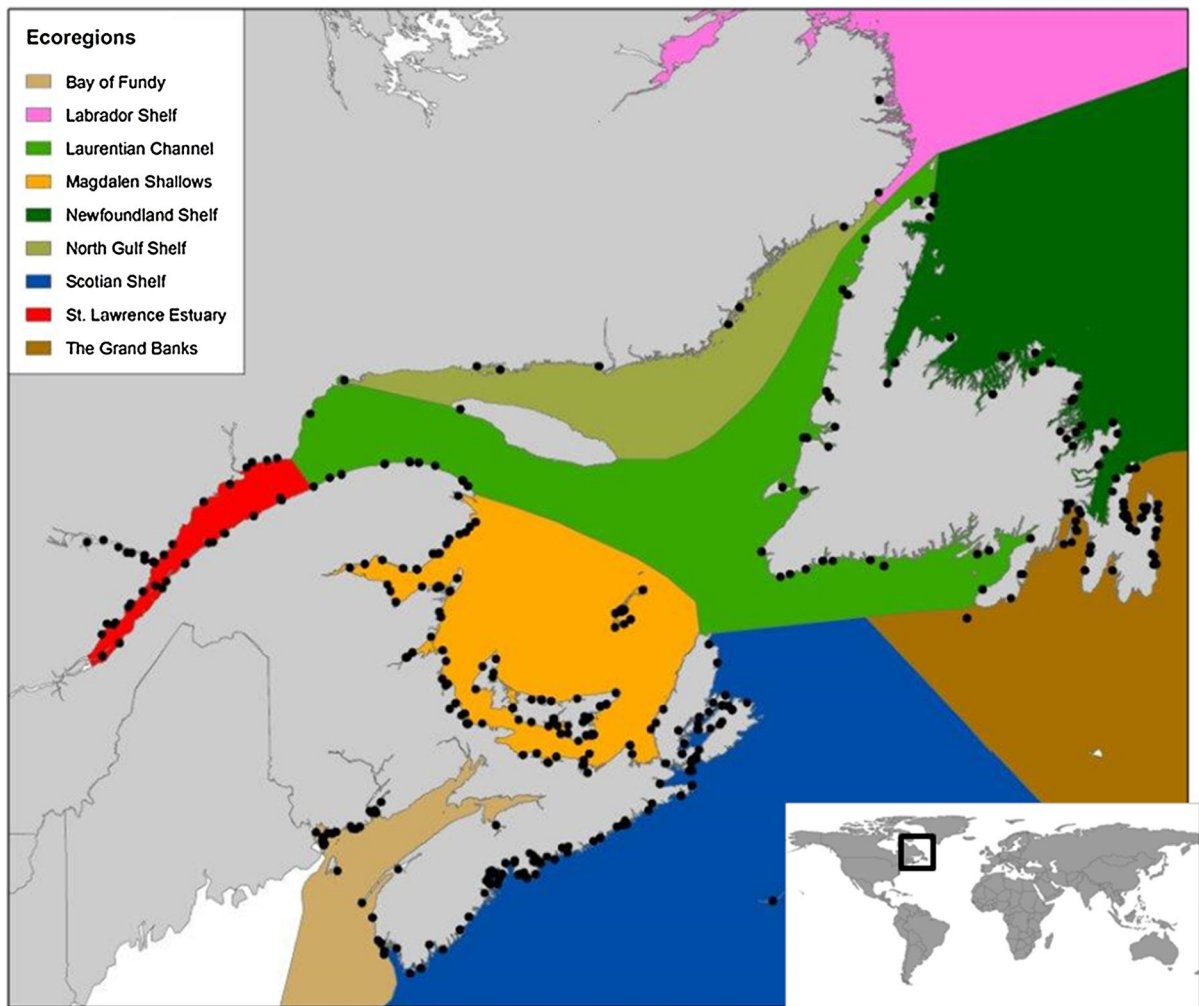


Fig. 2 Atlantic Canadian ecoregions based on the Parks Canada biogeographic classification (Harper et al. 1993) and visited and home marinas mentioned in boater questionnaires

(black dots) in these ecoregions. Freshwater portions of those ecoregions were excluded using several databases and relevant publications (see Simard et al. 2017 for details)

Boat fouling surveys

The occurrence of general macrofouling was considered as a potential indicator of NIS presence. The occurrence of macrofouling species (i.e., excluding biofilm) on boats was evaluated by examining the boats of 438 of the questionnaire survey respondents. Most boats were inspected using an underwater video camera mounted on a frame ($n = 346$), except for a few boats that were examined using a visual inspection by snorkeling ($n = 72$) or in dry-dock ($n = 20$). When using a video system, hull and niche areas (e.g., propellers, rudders, trim tabs) were examined by manoeuvring the camera with an adjustable telescopic

arm. Videos were later analyzed in the laboratory and the presence of macrofouling species noted for each boat surveyed. Surveyed boats were classified as being “fouled” or “clean”. Boats with less than 0.1% of hull and niche areas covered by fouling were classified as clean; those with greater than 0.1% cover of macrofouling on hull and niche areas were classified as fouled.

Boat fouling probability

Boat fouling probability was considered to be proportional to the relative presence of NIS in home

ecoregions and the probability of being fouled with native or non-native species.

Regional NIS background

The Regional NIS Background in a given ecoregion was calculated as the mean of the products of NIS richness and cover (%) observed for each monitored site in that ecoregion (a value of “1” was added to NIS richness and NIS cover of all monitored sites to remove the possibility of obtaining zero values to capture the possibility that survey plates may not have detected all NIS). The mean Regional NIS Background score was used for all boats from that ecoregion since the monitoring database did not include all home marinas of transient boats. The highest (1) and lowest (0.001) possible Regional NIS Background scores were attributed to foreign marine and freshwater ecoregions, respectively. Therefore, it was assumed that foreign marine ecoregions have a greater probability of having novel NIS compared to those observed in Canadian marine ecoregions and that freshwater ecoregions would have few (if any) fouling NIS that would be of concern for marine ecoregions.

Fouling on each transient boat was assumed to be associated with the biota (fouling NIS) present in their home ecoregion, and not from all the ecoregions they may have visited before arriving at a marina in a Canadian ecoregion as detailed boat voyage histories (order of visited marinas and timing of visits) were not available.

Boat infestation probability

The relative importance of boat characteristics to fouling was determined with boosted regression tree (BRT) analyses performed on MATLAB[®] r2012b programming language (Mathworks[®], Natick, Massachusetts, USA). BRT analyses are based on multiple logistic regression trees to predict binary outcomes (here fouled/clean) from predictor variables. This method allows for missing variables and can incorporate continuous and categorical predictor variables and automatically fit interactions between predictor variables (see Elith et al. 2008 for details). All BRT analyses were restrained to the same tree complexity (5) and learning rate (0.001). Boater questionnaire results were converted into several continuous and

categorical variables: boat type (sailboat, powerboat and fishing boat), hull type (wood, fiberglass, aluminium, and other), storage type (in the water for part of the year, in the water year-round, stored on land and trailered to boat launch, and in-water at principal residence), trip types (day trips, racing, weekenders, long trips and tours), days in water, use of antifouling paint (yes or no), time since last manual cleaning, location of last manual cleaning (in water, at boat launch, on land, and other), time since last antifouling paint application, total number of destinations visited, and total number of nights spent in Canadian ecoregions. BRT analyses were fit using the boats for which inspections were available (N = 438). The selected Fouling Predictive Model was then used to predict fouling of boats that were not evaluated using the video surveys (these values were used in subsequent analyses) (N = 853). BRT model predictive power was assessed by computing the predicted fouling probability for boats that had been evaluated with video surveys and comparing results using the area under the curve (AUC) from receiver operating characteristics (ROC) based on the rate of true positive and true negative predictions (note that the measured values, when available, rather than predicted values were used in subsequent analyses). The correct classification rate was calculated by the addition of true positive and negative prediction rates. Boat Infestation probabilities of 0.001 were attributed if the predicted probability of fouling was below the optimal probability threshold (the probability that best discriminated fouled and clean boats on the ROC curve) and 1 if the probability was above the threshold. A “leave-one-out cross validation” procedure was also used to obtain independent predictions for each boat with a video survey and to evaluate the AUC from the ROC curve obtained; each datum was successively removed from the data set, a model fitted using the remaining data, and the model used to obtain an independent prediction for the excluded boat.

Introduction probability

Arrival probability

Arrival Probability was estimated as a function of the number of non-home marina overnight visits, using the maximal number of non-home marina overnight

visits to scale from the minimal number of non-home marina overnight visits to 1.

Survival probability

Survival Probability was estimated between source (home) and recipient marinas for each transient boat in a given ecoregion as the product of salinity and climate match scores to yield a single environmental similarity score. Both variables are fundamental to species survival and reproduction (Barry et al. 2008). We assumed that all fouling species survive transit and may be released in recipient marinas.

Each marina was classified into a salinity category based on mean salinity surface values (June–October) using values from several databases and publications (see Simard et al. 2017 for details): salinity < 5 psu as freshwater, salinity \geq 5–20 psu as brackish, salinity > 20 psu as marine. A matrix approach was used to attribute salinity match scores (Lowest, Intermediate or Highest) for source-recipient marina pairs (Table 1). If a given boat visited multiple marinas with different salinities (marine and brackish marinas) in a given ecoregion, the highest salinity match score was attributed. Climate similarity of paired source and recipient marinas was estimated based on their geographic locations. All marinas were classified by latitude into one of four climate zones: Tropical (0°–20°N/S), Warm-Temperate (20°–40°N/S), Cold-Temperate (40°–60° N/S) and Polar (> 60° N/S) following Spalding et al. (2007) and Gollasch and Leppäkoski (2007). A matrix approach was used to attribute climate match scores for all source-recipient marina-pairs; Lowest to Highest (Table 2). Note that scores for both salinity and temperature matches range from 0.032 to 1 so that their products—Survival

Probability—range from 0.001 to 1 to be consistent with other variables used to calculate Boat Hazard scores.

Boat hazard and population boat hazard scores

Each transient boat in a given ecoregion was classified into a hazard category, as modified from Mandrak et al. (2012) (Table 3), based on their Boat Hazard scores. These hazard rankings were based on the distributions of Boat Hazard scores of all transient boats in all ecoregions. The ‘Lowest’ hazard category was set to 1% of the distribution, to capture the very high occurrence of boats with very low Boat Hazard scores (Fig. 3); all other hazard categories were divided equally (Table 3). The annual traffic in each ecoregion was then incorporated into the hazard equation by a combination of distribution fitting and bootstrap procedures performed on MATLAB® r2012b programming language. Annual traffic was estimated as the product of the average number of transient boats in a marina in a given ecoregion and year (estimated based on marina manager questionnaires; Simard et al. 2017) and the total number of marinas in that ecoregion. The total number of marinas in each ecoregion was compiled using the list of marinas in a DFO recreational boating database, supplemented with the DFO small craft list of ports and an on-line resource (<http://marinas.com/browse/marina/CA/>). Marinas, yacht clubs, fishing harbors, wharfs, and ports that may be used by recreational boaters were considered as “marinas” but large commercial ports were excluded (although individual marinas associated with these ports were included). To allow a larger number of boats in each ecoregion to be categorized and to better evaluate the occurrence of rare boats that could represent greater risk, Boat

Table 1 Matrix used to determine salinity similarity scores for source-recipient marina pairs (after Carlton 1985)

RECIPIENT marina	SOURCE marina		
	Fresh water	Brackish water	Marine water
Fresh water	Highest (1)	Intermediate (0.667)	Lowest (0.032)
Brackish water	Intermediate (0.667)	Highest (1)	Intermediate (0.667)
Marine water	Lowest (0.032)	Intermediate (0.667)	Highest (1)

Salinity zones are categorized as fresh water (< 5 PSU), brackish water (\geq 5–20 PSU) and marine water (> 20 PSU). Numbers in parentheses are used in the calculation of survival probability

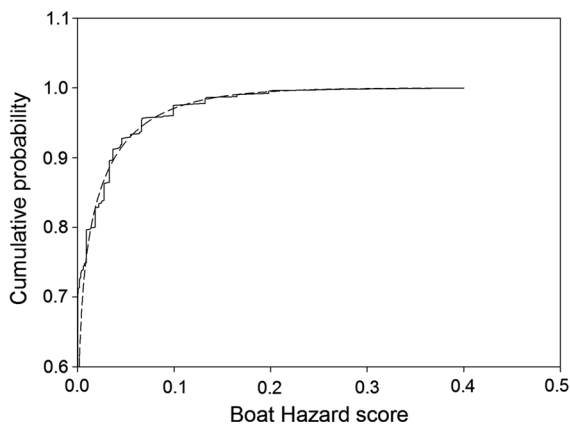
Table 2 Matrix used to determine climate similarity match scores for source-recipient marina pairs (after Gollasch and Leppäkoski 2007)

RECIPIENT marina	SOURCE marina			
	Polar	Cold-temperate	Warm-temperate	Tropical
Polar	Highest (1)	Intermediate (0.667)	Low (0.333)	Lowest (0.032)
Cold-temperate	Intermediate (0.667)	Highest (1)	Intermediate (0.667)	Low (0.333)
Warm-temperate	Low (0.333)	Intermediate (0.667)	Highest (1)	Intermediate (0.667)
Tropical	Lowest (0.032)	Low (0.333)	Intermediate (0.667)	Highest (1)

Climate zones are identified as polar (+ 60°N/S), cold-temperate (40–60°N/S), warm-temperate (20–40°N/S) and tropical (0–20°N/S). Numbers in parentheses are used in the calculation of Survival Probability

Table 3 Hazard categories as Hazard score distribution ranges. Modified from Mandrak et al. (2012)

Hazard category	Probability category
Lowest	0.000–0.010
Low	0.011–0.250
Intermediate	0.251–0.500
High	0.501–0.750
Highest	0.751–1.000

**Fig. 3** Cumulative probabilities of boat hazard scores obtained for all transient boats and beta distribution fitted on all data (dashed line). Beta distributions fitted on each ecoregion individually not shown

Hazard scores were calculated for the entire population of boats in each ecoregion over a 10 year period (i.e., ten times the annual traffic; Population Boat Hazard score). Note that the 10 year period does not include any inter-annual heterogeneity since it is ten

times the annual traffic estimated with manager questionnaires.

For each ecoregion, a distribution was fit to the Boat Hazard score distributions, all of which were best approximated by a generalized Beta distribution (Chi square = 6.61, degree of freedom = 3 and p value = 0.09; Fig. 3 for the generalized distribution of all ecoregions). For each ecoregion, a number of Boat Hazard scores (equal to ten times the Annual Traffic observed in each ecoregion) were then randomly drawn from the described distributions. Ecoregion Hazard score was calculated as the mean of all boats scores of each ecoregion over 10 years. Final Ecoregion Hazard scores were attributed based on which of the 5 bins of equal width over the range of observed values scores fell in (from Lowest to Highest).

Expected distribution of hazard scores and management simulations

A procedure similar to that used to calculate Ecoregion Hazard scores was used to estimate the range of possible annual mean Boat Hazard scores. Distributions were fit as described earlier, but we also incorporated the precision of parameter estimates in the bootstrapped data to create a range of possible hazard score distributions. We assumed a normal distribution for each Beta parameter (with mean and variance as obtained in the parameter estimate), drew a random value for each, and computed the distribution of Hazard scores. Values equal to annual traffic for a given ecoregion were generated for each Hazard score distribution and mean scores calculated; this was repeated 1000 times. Results from this bootstrap

procedure are sensitive to the quality of data used to compute Hazard scores in each ecoregion.

We used the Fouling Predictive Model to simulate the effect of different management options on Boat Hazard scores. After examining the independent variables included in the model, it was concluded that only two management actions could realistically be undertaken by boaters: frequency of antifouling paint application and frequency of boat cleaning (it was assumed that boat movement and number of destinations could not be managed easily whereas boat maintenance could be). We obtained predictions after altering the data for the time since last cleaning, and age of antifouling paint and evaluated the effect of requiring boaters to clean and repaint their boats every 1, 2, 3, 4, 5, 6, 9 or 12 months. We tested the results for cleaning frequency scenario alone, and then for cleaning frequency and antifouling paint application together. For each boat, we determined how old the paint would be and how long since the boat would have been cleaned if different rules had been applied. As an example, in a scenario requiring boat painting every 6 months, a boat reporting eight-month old paint was converted to a boat with two-month old paint. Note that paint application and cleaning actions also require removing the boat from the water; thus time in water was altered in a similar way. Hazard scores for each ecoregion were calculated under each of these different management scenarios. Variation of Ecoregion Hazard score among simulations of management options was evaluated with permutational analysis of variance (PERMANOVA, with 9999 permutations), using PRIMER 7 (v. 7.0.13) and PERMANOVA + 1, as data transformations were unable to constrain the data to meet the assumptions of ANOVA. Euclidean distances were used to construct the similarity matrix of Ecoregion Hazard score univariate measure and a fourth root transformation was used. Differences among simulations in each ecoregion were determined using a posteriori pairwise comparisons on PERMANOVA + 1.

Sensitivity analysis

Sensitivity analysis was performed to determine the influence of each variable (Regional NIS Background, Boat Infestation Probability, Arrival Probability and Survival Probability) on the outcome of the Boat Hazard model. One variable at a time was excluded

from the Hazard calculations, and the distribution fitting and bootstrap procedures were conducted again to observe the effects on final results. The magnitude of changes observed was the change in the Final Ecoregion Hazard score attributed to each ecoregion.

Results

Manager and boater questionnaires

The number of completed manager (1–47) and boater (0–589) questionnaires varied greatly among ecoregions (Table 4). Sailboats and power boats were the most abundant boat types in most ecoregions, except for the Bay of Fundy, Laurentian Channel, and Newfoundland Shelf ecoregions, where fishing boats were the most abundant boat type (Table 4).

Regional NIS background

A total of eight different NIS were observed at monitoring sites (Table 5), varying from zero (no NIS detected) to seven within a given site (Fig. 4). The Scotian Shelf had the maximal number of seven species observed (Table 5) and generally had the highest NIS richness (ten sites with five NIS, three sites with six NIS, one site with seven NIS), followed by Bay of Fundy (12 sites with five NIS). The greatest maximum NIS cover (> 75%) was only observed in the Scotian Shelf (24 sites), Magdalen Shallows (15 sites), Bay of Fundy (14 sites), Laurentian Channel (one site), and The Grand Banks (one site) ecoregions (Fig. 4). Scotian Shelf and Bay of Fundy ecoregions had the highest Regional NIS Background scores (0.495 and 0.377, respectively) (Table 5). All other Atlantic Canadian ecoregions had scores of 0.137 or lower. Because no monitoring sites were located in the Labrador Shelf and St. Lawrence Estuary ecoregions, they were assigned the lowest Background scores (0.001), based on AIS local expert knowledge and risk assessment predictions of these species (Therriault and Herborg 2007).

Boat fouling probability

Of the 438 boats surveyed for macrofouling, 228 (52.1%) had macrofouling on their hull and/or niche areas and were classified as fouled; the remainder (210

Table 4 Number of manager and boater questionnaires completed and percentage of boat types surveyed in each ecoregion

Ecoregion	Manager questionnaires	Boater questionnaires ^a	% Sailboats	% Powerboats	% Fishing boats
Bay of Fundy	10	67	36 (24)	18 (12)	40 (27)
Labrador Shelf	1	0	–	–	–
Laurentian Channel	34	50	40 (20)	18 (9)	42 (21)
Magdalen Shallows	47	589	51 (299)	34 (203)	14 (83)
Newfoundland Shelf	17	8	–	–	100 (8)
North Gulf Shelf	6	19	58 (11)	42 (8)	–
Scotian Shelf	35	344	64 (221)	28 (97)	7 (24)
St. Lawrence Estuary	15	156	69 (108)	30 (47)	1 (1)
The Grand Banks	33	58	9 (5)	53 (31)	38 (22)

Number of boats per boat type is indicated in brackets, modified from Simard et al. (2017)

^aSome boater questionnaires had missing information on boat type

Table 5 Number of monitoring sites, NIS observed (X) in monitoring sites and NIS Background score assigned to each ecoregion. Modified from Simard et al. (2017)

	Ecoregion								
	Bay of Fundy	Labrador Shelf	Laurentian Channel	Magdalen Shallows	NFLD Shelf	North Gulf Shelf	Scotian Shelf	St. Lawrence Estuary	The Grand Banks
# Monitoring sites	22	–	14	82	1	3	44	–	6
NIS									
<i>Ascidella aspersa</i>	–	NA	–	–	–	–	X	NA	–
<i>Botryllus schlosseri</i>	X	NA	–	X	–	–	X	NA	X
<i>Botrylloides violaceus</i>	X	NA	X	X	–	–	X	NA	–
<i>Caprella mutica</i>	X	NA	–	X	–	–	X	NA	–
<i>Ciona intestinalis</i>	X	NA	–	X	–	–	X	NA	–
<i>Diplosoma listerianum</i>	–	NA	–	–	–	–	X	NA	–
<i>Styela clava</i>	–	NA	–	X	–	–	X	NA	–
<i>Membranipora membranacea</i>	X	NA	X	X	–	X	X	NA	X
NIS background score	0.377	0.001	0.028	0.137	0.001	0.008	0.495	0.001	0.051

NFLD Newfoundland

boats, 47.9%) were classified as clean. Three variables were significant predictors of fouling state: time in water (41.5% of the deviance explained), time since last cleaning (16.7% of the deviance explained), and boat type (11.1% of the deviance explained). Time in

water and time since last cleaning were positively correlated with fouling, while fishing boats were less fouled compared to all other boat types. Time since last cleaning was not correlated to time in water (Pearson correlation coefficient = -0.033 ; $df = 364$;

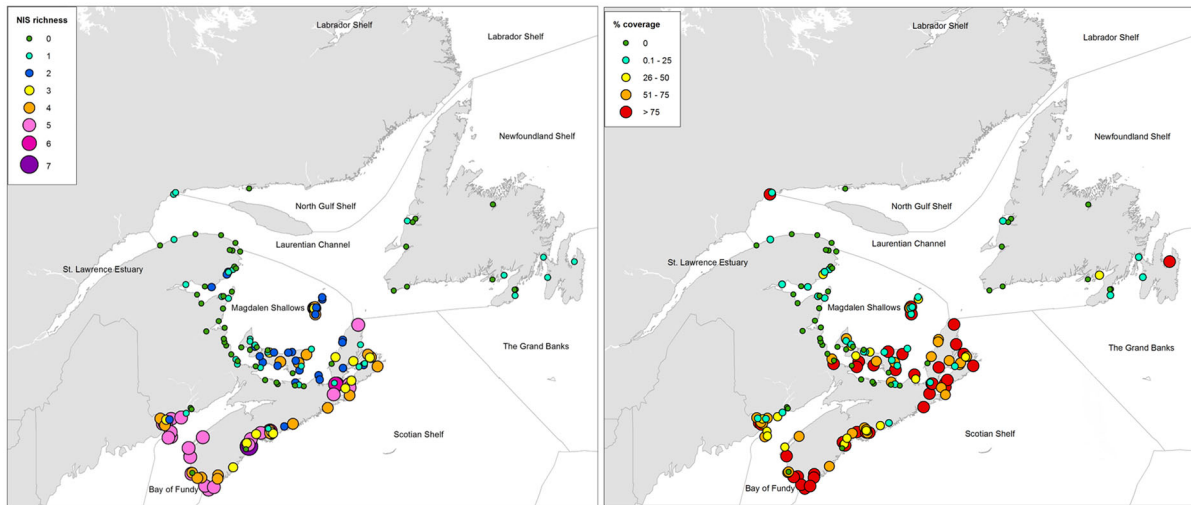


Fig. 4 NIS richness and NIS cover (%) measured at monitoring sites. Modified from Simard et al. (2017)

$p = 0.523$). All other variables included in the Fouling Predictive Model explained relatively little of the fouling classification (each $\leq 9.5\%$, Table 6). The area under the ROC curve built for the model was 0.82 when predictions were dependant, and 0.65 when the predictions were independent (Fig. 5). With the calculated optimal threshold probability of 0.52, the correct classification rate was 0.75.

For all boaters surveyed in this study, the median value for Time in water varied between 37 (North Gulf Shelf) and 97 days (Bay of Fundy; Fig. 6). Newfoundland Shelf had a median value of 152 days for

this variable, but only 2 boats had answered this question.

Arrival and survival probabilities

The sailboat was the dominant boat type for transient boats in all ecoregions (65.0–100.0%), except in the Newfoundland Shelf and The Grand Banks ecoregions, where the powerboat was the dominant boat type (40.0 and 57.1%, respectively; Table 7). Intra-ecoregional traffic was the most common traffic type in the Magdalen Shallows, Scotian Shelf, and The

Table 6 Deviance explained by each predictor (boat characteristics) included in boosted regression tree analyses

Predictors	Deviance explained (%)
Days in water	41.5
Time since last cleaning	16.7
Boat type	11.1
Total number of destinations	9.5
Time since last antifouling treatment	9.5
Storage type	6.6
Hull type	1.9
Tour trip	1.3
Cleaning location	0.9
Total number of nights	0.8
Long trip	0.1
Use of antifouling paint	0.1
Day trip	0.0
Racing trip	0.0
Weekender trip	0.0

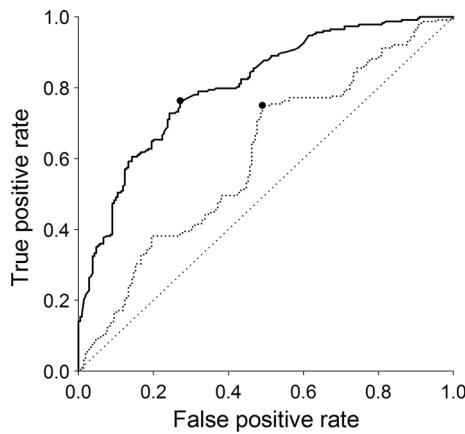


Fig. 5 Receiver operating characteristic curve for dependant (thick line) and for independent (dashed line) predictions of boat fouling probabilities based on BRT model. The diagonal dashed line represents the expected results if the model was no better than random and the black dots represent the optimal threshold values

Grand Banks ecoregions (56.7–69.1%) while inter-ecoregional traffic was the dominant traffic type in the Bay of Fundy, Newfoundland Shelf, and North Gulf

Shelf ecoregions (51.2–82.1%; Table 7). The total number of destinations per transient boat in a given ecoregion varied between one and 15. The Laurentian Channel and St. Lawrence Estuary transient traffic had the highest proportion of boaters (in terms of number of destinations) from freshwater ecoregions (47.0 and 61.6%, respectively). With the exception of Labrador Shelf, international arrivals represented a relatively low proportion of arrivals (2.6–17.1%) to most ecoregions, with Bay of Fundy having the highest proportion of international arrivals. Although international arrivals accounted for 33% of all recorded arrivals in the Labrador Shelf ecoregion, only three transient boats were sampled there, suggesting that the data in this ecoregion is not of sufficient quality to be included in subsequent analyses. Boat and Population Boat Hazard scores were not attributed for this ecoregion. Most transient boats (52.2–100%) visiting Atlantic Canadian ecoregions were given the highest Survival Probability score (1), with the exception of the St. Lawrence Estuary ecoregion, which received mainly transient boats with a Survival Probability score of 0.6 (44%) because of the large proportion of

Fig. 6 The number of days in water spend by surveyed boaters per home ecoregion. The top and bottom of each box plot are the 25th and 75th percentiles, lines extending vertically from the boxes (whiskers) indicate variability outside the 95th and 5th percentiles. The black line inside the box is the median and outliers are marked by dots. The sample sizes (i.e., number of successful answers for each ecoregion) are presented over each box

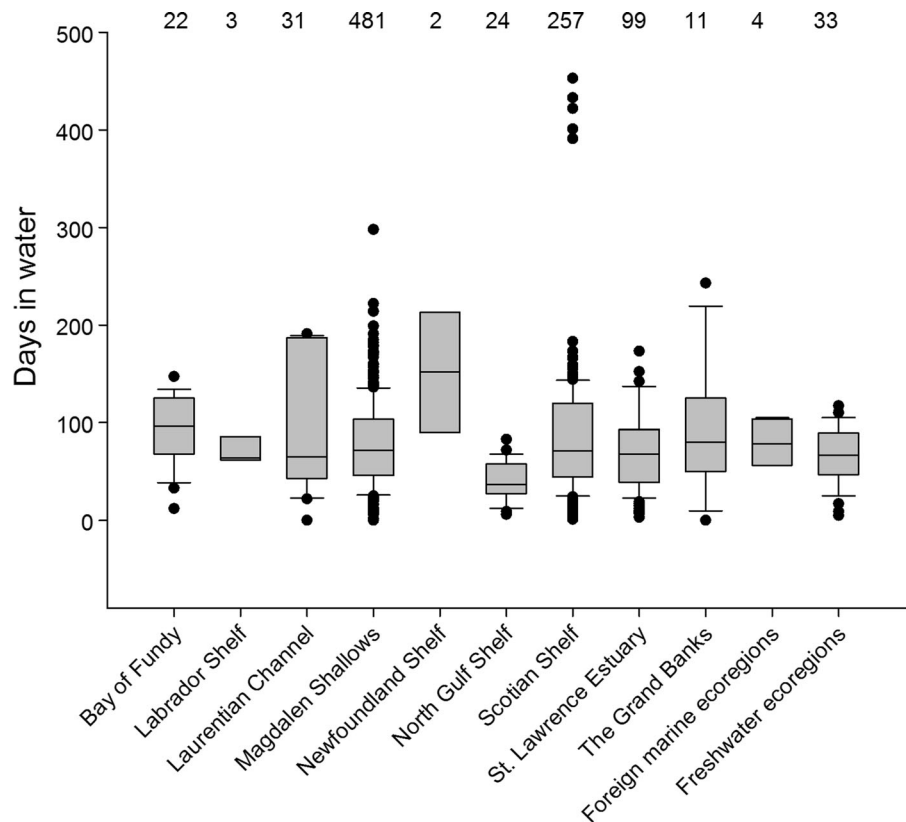


Table 7 Total number of transient boats and number of transient boats per boat type (S = sailboat, P = power boat and F = fishing boat) for which boater questionnaires were completed in each Canadian ecoregion. Modified from Simard et al. (2017)

Ecoregion	Transient boats ^a				Arrival types				Survival probability scores			Annual traffic
	# Total	# S (%)	# P (%)	# F (%)	Intra (%)	Inter (%)	Int (%)	F (%)	0.2 (%)	0.6 (%)	1.0 (%)	# Annual boats
Bay of Fundy	37	75.7	18.9	5.4	19.5	51.2	17.1	12.2	13.5	8.1	78.4	3511
Labrador Shelf	3	100.0	–	–	0.0	33.3	33.3	33.3	33.3	–	66.7	1063
Laurentian Channel	90	80.0	15.6	4.4	4.2	36.7	12.0	47.0	43.3	4.4	52.2	2713
Magdalen Shallows	374	65.0	29.7	5.3	69.1	11.2	3.8	16.0	5.6	9.6	84.8	14,552
Newfoundland Shelf	20	25.0	40.0	35.0	2.6	82.1	15.4	0.0	10.0	–	90.0	1577
North Gulf Shelf	18	83.3	5.6	11.1	0.0	52.4	14.3	33.3	38.9	–	61.1	962
Scotian Shelf	305	67.2	26.2	6.6	56.7	26.3	8.8	8.3	4.6	5.2	90.2	29,669
St. Lawrence Estuary	168	74.4	25.0	1.0	23.3	9.6	5.5	61.6	17.2	44.0	38.7	6901
The Grand Banks	21	23.8	57.1	19.1	66.7	7.7	2.6	23.1	–	4.8	95.2	754

Arrival types (intra = intra-ecoregion, inter = inter-ecoregion, int = international and F = freshwater), survival probability scores, and annual traffic (number of boats and score) are indicated for each of the transient boats in Canadian ecoregions

^aA single transient boat may have visited more than one ecoregion

boaters from freshwater marinas (Table 7). See “Appendix 1” for detailed transient traffic.

Boat hazard and population boat hazard scores

Most transient boats visiting Atlantic Canadian ecoregions were categorized in the “Lowest” and “Low” Boat Hazard score categories; few boats were classified in the “Highest” category (Fig. 7a). The Laurentian Channel, Magdalen Shallows, St. Lawrence Estuary, and Scotian Shelf ecoregions received some transient boats with “Intermediate”, “High” and “Highest” Boat Hazard scores. The Scotian Shelf ecoregion received by far the greatest number of boats on an annual basis, followed by the Magdalen Shallows ecoregion (Table 7). All the other ecoregions received less than half as many transient boats annually.

Several transient boats were classified as being of “Highest”, “High” and “Intermediate” Hazard over a 10 year period in the Bay of Fundy (0.06%; 20 boats), Laurentian Channel (0.20%; 54 boats), Magdalen Shallows (0.14%; 204 boats), and Scotian Shelf (2.77%; 8, 233 boats) ecoregions (Fig. 7b). A Final

Hazard score of “Highest” was attributed to the Scotian Shelf ecoregion. Bay of Fundy, Laurentian Channel and Magdalen Shallows ecoregions were attributed “Low” Final Hazard scores, and Newfoundland Shelf, North Gulf Shelf, St. Lawrence Estuary and The Grand Banks ecoregions “Lowest” Final Hazard scores (Fig. 7b).

Expected distribution of hazard scores

The estimated annual mean Boat Hazard scores are presented for each ecoregion in Fig. 8. The Bay of Fundy, Newfoundland Shelf, North Gulf Shelf and The Grand Banks ecoregions had the greatest range of possible mean yearly Hazard scores but the greatest mean scores were observed for the Scotian Shelf, Magdalen Shallows, and Bay of Fundy ecoregions.

Management simulations

Ecoregion Hazard scores were significantly affected by simulation scenarios (1, 2, 3, 4, 5, 6, 9 and 12 months) of a mandatory out-of-water cleaning of boat hulls (Pseudo-F = 13.331; $p = 0.0001$; Fig. 9).

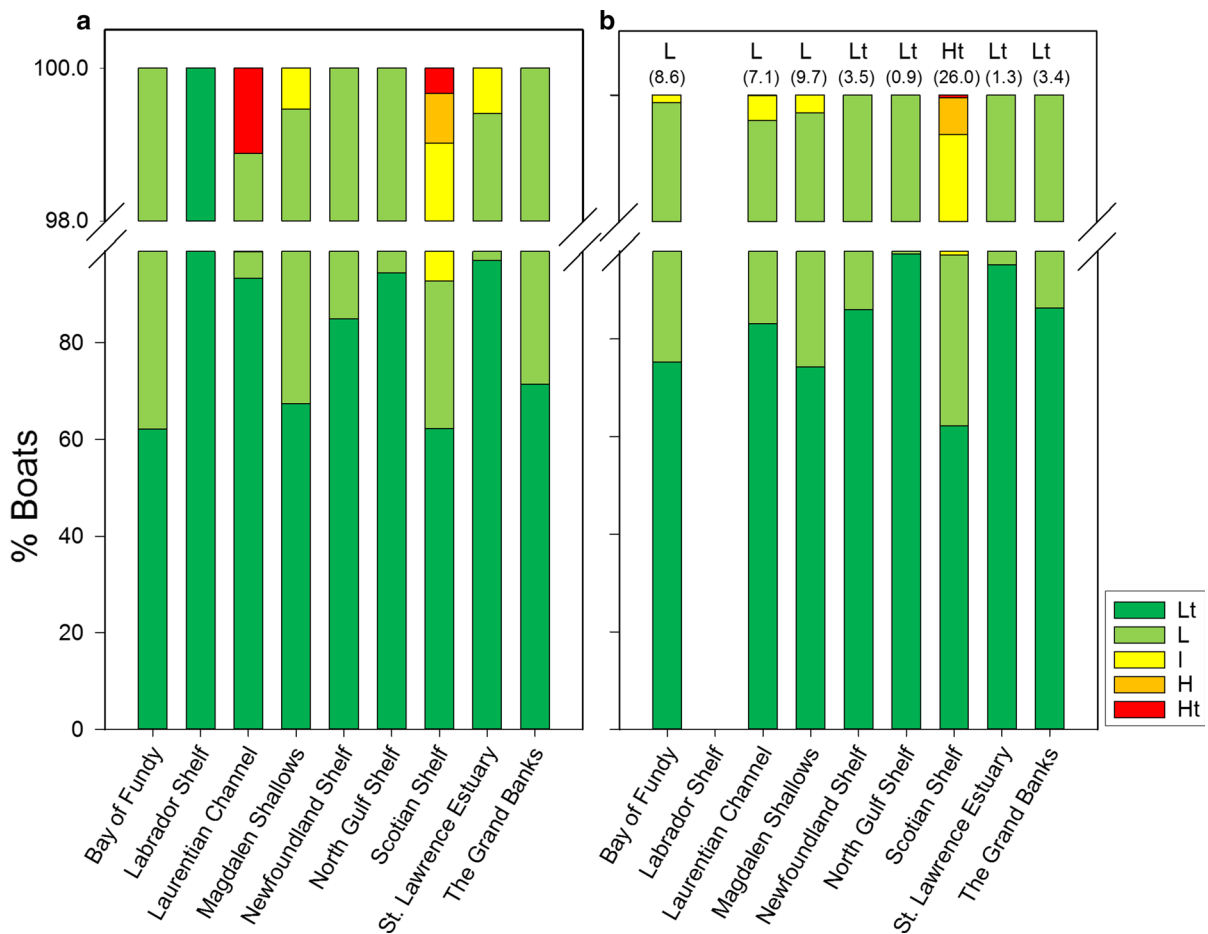


Fig. 7 Percentage of boats classified in each hazard category for the boat hazard score (a) and population boat hazard score (b). Ecoregion hazard scores ($\times 10^{-3}$, in brackets) and final

hazard scores attributed to each ecoregion are indicated above each bar (insufficient data for Labrador Shelf). Ht = highest, H = high, I = intermediate, L = low, Lt = lowest

The first scenario significantly affecting the Ecoregion Hazard scores was the cleaning after 3 months of boats being in water for Magdalen Shallows ($t = 4.1616$; $p = 0.0001$) and the Scotian Shelf ($t = 2.8067$; $p = 0.0052$) ecoregions, and after 4 months for Bay of Fundy ($t = 2.5406$; $p = 0.0126$), North Gulf Shelf ($t = 1.8568$; $p = 0.0486$) and St. Lawrence Estuary ($t = 2.7114$; $p = 0.0057$) ecoregions. The simulations of management options did not significantly affect Ecoregion Hazard scores in all the other ecoregions (see “Appendix 2” for detailed results of the statistical analysis). Simulation scenarios with a renewal of antifouling treatment following each cleaning scenario do not bring additional changes in Ecoregion Hazard scores (data not shown).

Sensitivity analysis

Regional NIS Background has the largest influence on final results. The removal of this variable changed all Final Ecoregion Hazard scores (-1 to $+4$), except for the North Gulf Shelf ecoregion (Table 8) (e.g., the Final Ecoregion Hazard score for Magdalen Shallows changed from “Low” to “Highest”, a categorical jump of $+3$ score categories from original Ecoregion Hazard score). Removal of Boat Infestation Probability and Arrival Probability had impacts on few ecoregions, with respectively one (-1) and three ecoregions (-1 to $+3$) being classified in another Final Ecoregion Hazard score. Final results were generally insensitive to the removal of the Survival

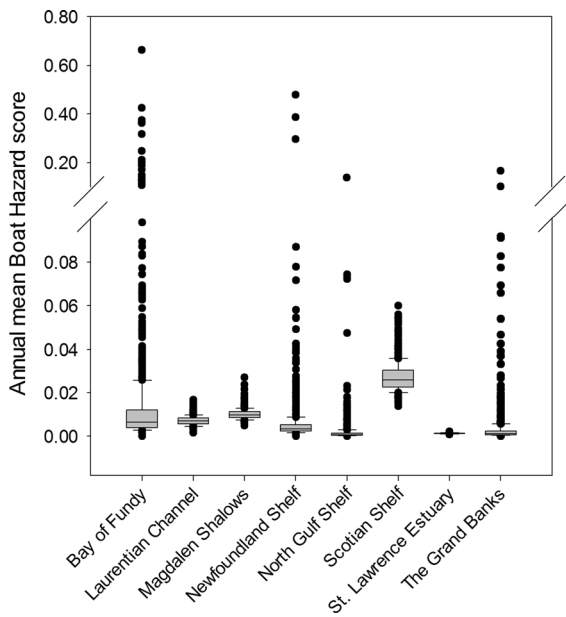


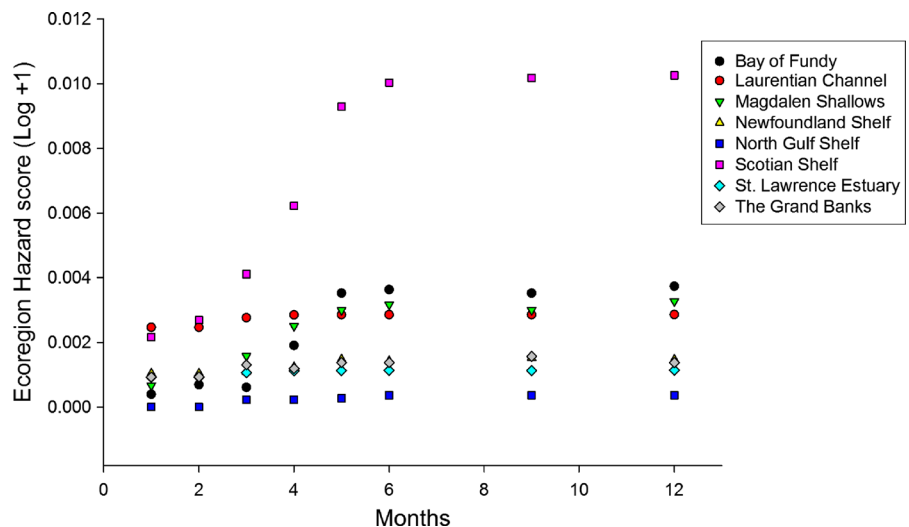
Fig. 8 Estimated range of possible annual mean boat hazard scores in each ecoregion using bootstrap analyses. Labrador Shelf not shown because of insufficient data

Probability, with no change in the Final Ecoregion Hazard score attribute to all ecoregions.

Discussion

Given the potential importance of marine recreational boating contributing to the introduction and spread of NIS there have been relatively few risk assessments

Fig. 9 Simulations of the ecoregion hazard score for boats that have been cleaned out-of-water for 1–12 months. Labrador Shelf not shown because of insufficient data



(but see Ashton et al. 2006; Acosta and Forrest 2009; Acosta et al. 2010; Clarke Murray et al. 2013; Ferrario et al. 2016; Floerl et al. 2005; Lacoursière-Roussel et al. 2012b). However, these previous assessments are either not data-driven (i.e., based on results from literature and expert opinion), focus only on the probability of being fouled, exclude important environmental linkages and traffic data, or are specific to limited types of organisms or traffic. Unlike the other modelling studies, we have put together a large dataset to do a formal quantitative hazard assessment of recreational boating activities which includes biological data from the environment and vessel hulls, a new predictive fouling model, boater behaviour data and environmental similarity between source and receiving marinas. In addition, this study presented, for the first time, a large scale picture of the transient recreational boating traffic in nearly all Atlantic Canadian ecoregions and simulation scenarios to identify management actions.

This assessment supports results from previous studies conducted at a smaller scale (Clarke Murray et al. 2014; Lacoursière-Roussel et al. 2012b, 2016) and shows the importance of recreational boating activities as potential vectors for initial introduction and secondary spread of NIS in all Atlantic Canadian ecoregions. The high proportion of fouled boats sampled in the study (52.1%), the importance of boat maintenance behaviours in boat fouling status, the high degree of connectivity between marinas within and between Atlantic Canadian ecoregions via

Table 8 Changes in the final ecoregion hazard score (number of category changed) from the original values when one variable at a time is removed from the boat hazard calculations

Ecoregion	Without regional NIS background	Without boat infestation probability	Without arrival probability	Without survival probability
Bay of Fundy	- 1	-	+ 3	-
Laurentian Channel	- 1	- 1	- 1	-
Magdalen Shallows	+ 3	-	-	-
Newfoundland Shelf	+ 4	-	-	-
North Gulf Shelf	-	-	-	-
Scotian Shelf	- 1	-	-	-
St. Lawrence Estuary	+ 2	-	-	-
The Grand Banks	+ 2	-	+ 1	-

Labrador Shelf not shown because of insufficient data

recreational traffic, and the high survival probabilities associated with most transient boat trips highlight the importance of managing this potential NIS vector in Canada and elsewhere. Results from this assessment demonstrate that the most hazardous boats (boats with “Intermediate”, “High” and “Highest” scores) are represented by a small subset of all recreational boats in eastern Canadian ecoregions (2.5%, 26 boats in the population sampled in this study), but when these boats visit a non-home marina, they may act as sources for further secondary spread via recreational traffic (Clarke Murray et al. 2014; Lacoursière-Roussel et al. 2012b, 2016).

The Scotian Shelf and Magdalen Shallows ecoregions received the greatest number of transient boats. Similar to the Scotian Shelf and Magdalen Shallows, the Laurentian Channel and the Bay of Fundy ecoregions received mainly boats that tended to visit a greater number of destinations, spent a longer period of time in water during a boating season, and for which any associated fouling organisms had a high survival probability, thus increasing Introduction probabilities. All these factors, combined with a mostly high NIS Background, contributed to the Scotian Shelf receiving the “Highest” Hazard score, and the Magdalen Shallows, Laurentian Channel and Bay of Fundy scoring the “Low” score. It is important to note that the attribution of “Lowest” Hazard scores to all the other ecoregions does not indicate that recreational

boating poses a low absolute risk for those ecoregions, but rather that their risk is lower compared to the other ecoregions where hazard scores were higher. The Bay of Fundy ecoregion received relatively less transient traffic, but this ecoregion received the highest proportion of international traffic because of its close proximity with the United States, which could introduce novel NIS, thus increasing hazards.

Marinas provide suitable environments for NIS with diverse substrates and environmental conditions that facilitate NIS establishment (Floerl and Inglis 2003). This, combined with high propagule pressure due to boating, makes marinas among the most invaded marine habitats (Lambert and Lambert 1998). Highest NIS Richness and NIS cover were observed in southern ecoregions and both metrics were typically less in more northern ecoregions. In parallel, a greater number of transient recreational boats sail in southern ecoregions (e.g., Scotian Shelf, Magdalen Shallows) compared to northern ecoregions. However, due to the high connectivity between ecoregions, nearly all northern ecoregions received traffic from those southern invaded ecoregions. As an example, the Newfoundland Shelf ecoregion, a northern ecoregion with no NIS reported in the dataset used in this study, had low transient traffic, but this traffic was primarily from other Atlantic Canadian ecoregions, including Scotian Shelf and Magdalen Shallows ecoregions, which are invaded with high-impact NIS (e.g., tunicate

species). Likewise, *Botryllus schlosseri* and *Botrylloides violaceus* first invaded southern regions of the east coast of Canada (e.g., Nova Scotia, Prince Edward Island), and then later invaded northern regions (e.g., Quebec, southern Newfoundland) (Carver et al. 2006; McKenzie et al. 2016; Simard et al. 2013). However, the hypothesis of independent introductions could not be excluded for these cases. Thus, the high connectivity among marinas within and between Canadian ecoregions via recreational boating highlighted by this study may increase the rate of NIS introduction and spread to northern ecoregions through “stepping-stone” processes (Floerl et al. 2009). The role of recreational boats in the secondary spread of invaders has been suggested in Nova Scotia, based on the relationship between population genetic similarity and the strength of boating connectivity between populations in Nova Scotia (Lacoursière-Roussel et al. 2012a).

Due to the strong sensitivity of final results to Regional NIS Background, future research is warranted to more fully determine the distribution of NIS in ecoregions, especially for areas with limited monitoring. The use of ‘Regional NIS Background’ as a measure of the probability that fouling on boats includes NIS is clearly a simplification of a more complex process as fouling communities on boats may result from different sources. Transient boats that visit multiple destinations during a trip before arriving in a given ecoregion may transport multiple NIS from different sources and transfer may arise via such “stepping-stone” processes (Floerl et al. 2009). For example, some eastern Canada boaters travel with their boats to warm-temperate (e.g., Florida) or tropical regions for winter, spending long periods of time in water, and may therefore pose a greater risk of introducing or spreading NIS when coming back to eastern Canada. Moreover, propagule exposure has been shown to be the most important predictor for boat fouling in Eastern Canada (Lacoursière-Roussel et al. 2012b) and transient boats may spend considerable time outside of their home marinas. The longer a boat stays in a marina, the more its fouling community resembles the fouling community present in that marina (Floerl and Inglis 2005) and boat fouling assemblages may consequently be more similar to those in visited than in home marinas. The inclusion of data on the directionality of recreational traffic would therefore likely improve this hazard assessment. As

already used for commercial ships (Simard et al. 2014), a mandatory Automated Identification System for recreational boats would give precious information on traffic movements and its directionality. This data acquisition is even more essential in the context of potentially increasing NIS loads and changing vessel movements and volume with changing climate (Sorte et al. 2010).

The fouling predictive model shows that hazardous boats are those that receive little maintenance (i.e., low cleaning frequency) and that have spent an extended period in water. Time in water, cleaning type, and frequency of cleaning have been shown to be linked to fouling levels on recreational boats in other studies (Ashton et al. 2006; Clarke Murray et al. 2013; Floerl et al. 2005, 2009). In countries where boats are in the water year-round, antifouling paint age has also been shown to be linked to fouling intensity (Ashton et al. 2006; Clarke Murray et al. 2011; Floerl et al. 2005). As observed at a smaller scale by Lacoursière-Roussel et al. (2012b), this study also suggests that, because of the relatively short summers on the Atlantic coast of Canada, time in water and time since last cleaning are better predictors of fouling level than antifouling paint age.

In this study, presence of fouling on hull and niche areas was not considered separately, and this may have increased variability in the model. In addition, hazard estimates are based on hull fouling organisms only and associated with non-resident boats. While the probability of hull fouling organisms being introduced by day tripper boats may be low, it is not zero. Planktonic organisms, plants, fish or invertebrate larvae/eggs have not been considered in this study, nor organisms transported via other vessel components (e.g., bilge, deck) (Acosta and Forrest 2009; Darbyson et al. 2009; Hayes 2002). In addition, the NIS dataset used in this study did not include all hull fouling organisms. As an example, *Didemnum vexillum*, a very high-impact NIS, was detected in Nova Scotia in 2013 but not observed on PVC plates used by the AIS monitoring program (Moore et al. 2014). Consequently, hazard associated with those particular organisms could have been underestimated in this assessment.

The population of boaters surveyed was represented by a relatively small proportion (3.2%) of international boaters. However, international boats may present a high hazard because of their potential to transport novel NIS. The close proximity of

northeastern US states (e.g., Maine), where many high-risk species are present (e.g., see NIS list in Pederson et al. 2006), to the Bay of Fundy and Scotian Shelf ecoregions make these ecoregions more susceptible to primary introductions to Atlantic Canada via recreational boats and boating. In the absence of ice, we assume that boaters from northeastern US states are more likely to keep their boats in the water year-round and that these boats would have older antifouling paint, thereby increasing their risk to transport and spread NIS to Atlantic Canadian destinations. The high boating connectivity within and between ecoregions suggests that ecoregions near the US border or infested locations in Canada may also act as sources for secondary spread in northern ecoregions via recreational or commercial traffic.

Simulations of Boat Fouling Probability (under the scenario that manual out-of-water cleaning of boats would be required following 3–4 months duration in water) show that Ecoregion Hazard scores would be reduced in the majority of ecoregions with higher Final Hazard scores. Management actions related to cleaning frequencies could thus be a suitable option for the Atlantic coast to reduce the hazard associated with this vector. As ice cover is reduced, vessel owners are increasingly not removing their boats (and floating docks) to reduce effort and maintenance costs. Floerl et al. (2016) modelled effects of different management strategies on fouling of recreational vessels, and showed that a strategy targeting a larger population of boaters with a lower threshold (i.e., more frequent renewal of antifouling paint) was more effective than a strategy targeting only the riskiest population of boaters. In addition to antifouling paint application, a management strategy (voluntary or mandatory basis) for recreational boating activities pertaining to cleaning frequency needs to be developed and should therefore include the majority of boaters. Based on the simulations presented in this study, a 3 months out-of-water cleaning frequency is a suitable option to reduce Ecoregion Hazard score in the ecoregions with greater hazard, such as the Bay of Fundy, Laurentian Channel, Magdalen Shallows and Scotian Shelf. This management option could reduce the NIS introduction and spread hazard associated with recreational boating in nearly all ecoregions due to the high inter-ecoregion connectivity via boating traffic, but especially in less invaded northern ecoregions. As implemented in New Zealand ([biosecurity\), Canada should develop hull cleaning guidelines and put in place protocols for the management of this unregulated vector.](http://www.biodiversity.govt.nz/seas/</p></div><div data-bbox=)

Conclusion

On the Atlantic Coast of Canada, the probability of boating-mediated NIS introduction and spread is compounded by the high number of recreational boats voyaging in each ecoregion, particularly the Scotian Shelf and Magdalen Shallows ecoregions. Boats with low cleaning maintenance and extended periods in water (more than 3 months) are high risk and act as NIS sources for secondary dispersal via the recreational boating vector. The strong connectivity between marinas both within an ecoregion and between ecoregions by recreational traffic suggests that once established, fouling NIS can spread in all Atlantic Canadian ecoregions. Research and efforts to better assess NIS distributions as well as boat traffic patterns in all ecoregions of Canada would help to refine hazard assessment associated with the recreational boating vector. A serious management of the recreational boating vector would entail the development of hull fouling cleaning guidelines and the implementation of Canadian vessel cleaning regulations. Meanwhile, a management option that encourages outreach programs to promote the habit of cleaning boats out-of-water after 3 months in water and maintain effective antifouling coatings could help reduce the risk of transporting NIS between Atlantic ecoregions.

Acknowledgements The authors wish to thank the laboratory and field assistants who helped in different aspects of this study: Isabelle Bérubé, Geneviève Perrin, Rafael Estrada, Sophie Boudreau, Benedikte Vercaemer, Murielle LeGresley, Paul McCurdy, Jocelyn Lowther, Terri Wells, Kyle Matheson, Robert O'Donnell, Andrew Perry, Mark O'Flaherty, George Bishop, Haley Lambert, Vanessa Reid, Erica Watson, André Nadeau, John Davidson, Colin Forsythe, Chantal Coomber, Janelle McLaughlin, Emily White, Siobhan Curry, and Rod Asher. We are also grateful to Jessica Paulmert and Paul Couture for database development; to Vicky Yaroshewski and Pierre Clement who provided background data from the AIS Monitoring Database, Andrea Weise who prepared numerous maps and reviewed the document, and Émilie Simard and Marie-France Lavoie for their precious advice. Finally, we are very grateful to marina managers for allowing access to their facilities and boat owners for their participation. We thank the anonymous reviewers whose contribution helped to produce a

better manuscript. This work was funded by the Fisheries and Oceans Canada Aquatic Invasive Species program. Additional funding was provided through the Canadian Aquatic Invasive Species Network (study by Lacoursière-Roussel) and the Newfoundland Provincial Department of Fisheries and Aquaculture. Lacoursière-Roussel was further supported by scholarships from McGill University, Québec-Océan and Fonds de recherche sur la nature et les technologies (FQRNT).

Appendix 1

See Fig. 10.

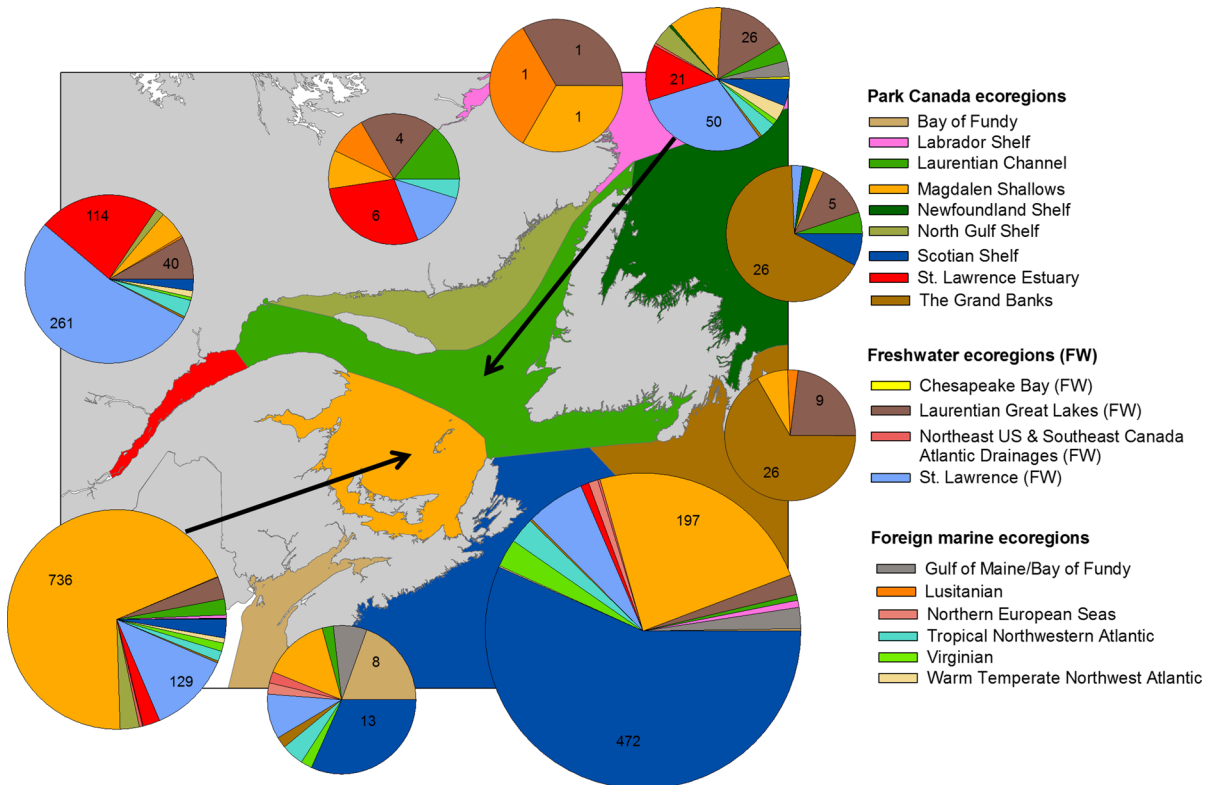


Fig. 10 Total number of destinations visited by transient boats in Atlantic ecoregions. The size of pie charts is relative to the estimated number of visitors in each ecoregion. Ecoregions are based on the Parks Canada biogeographic classification (Harper

et al. 1993), the Marine Ecoregions of the World (MEOW) classification (Spalding et al. 2007) and the Freshwater Ecoregions of the World (FEOW) classification (Abell et al. 2008). Modified from Simard et al. (2017)

Appendix 2

See Table 9.

Table 9 Results from a posteriori pairwise comparisons analysis for the simulation scenario factor performed for each ecoregion

Groups	t	P (perm)	Unique perms
<i>Bay of Fundy</i>			
OC_1, OC_2	0.49061	0.6426	9817
OC_1, OC_3	0.87872	0.4025	9825
OC_1, OC_4	2.5406	0.0126	9830
OC_1, OC_5	3.3345	0.0014	9835
OC_1, OC_6	3.6746	0.0003	9812
OC_1, OC_9	3.433	0.0007	9820
OC_1, OC_12	3.9222	0.0003	9845
OC_2, OC_3	0.34592	0.7414	9808
OC_2, OC_4	2.0491	0.0452	9825
OC_2, OC_5	2.8915	0.0037	9827
OC_2, OC_6	3.2124	0.0021	9830
OC_2, OC_9	2.9811	0.0047	9816
OC_2, OC_12	3.4487	0.0013	9822
OC_3, OC_4	1.7891	0.0743	9830
OC_3, OC_5	2.6678	0.0097	9828
OC_3, OC_6	2.9883	0.0033	9824
OC_3, OC_9	2.756	0.0066	9822
OC_3, OC_12	3.2251	0.0019	9825
OC_4, OC_5	0.97775	0.3324	9809
OC_4, OC_6	1.2466	0.2156	9842
OC_4, OC_9	1.0425	0.2915	9841
OC_4, OC_12	1.4522	0.1543	9829
OC_5, OC_6	0.24138	0.8017	9805
OC_5, OC_9	0.053992	0.9585	9819
OC_5, OC_12	0.42902	0.674	9822
OC_6, OC_9	0.18836	0.8553	9816
OC_6, OC_12	0.18837	0.8488	9830
OC_9, OC_12	0.37691	0.7093	9827
<i>Laurentian Channel</i>			
OC_1, OC_2	0.023486	0.9824	9838
OC_1, OC_3	0.84471	0.4044	9828
OC_1, OC_4	1.3221	0.1857	9853
OC_1, OC_5	1.539	0.1252	9833
OC_1, OC_6	1.6095	0.112	9845
OC_1, OC_9	1.5334	0.1296	9819
OC_1, OC_12	1.7238	0.0911	9847
OC_2, OC_3	0.82317	0.4171	9824
OC_2, OC_4	1.3012	0.1975	9837
OC_2, OC_5	1.5183	0.1333	9837

Table 9 continued

Groups	t	P (perm)	Unique perms
OC_2, OC_6	1.5887	0.115	9838
OC_2, OC_9	1.5126	0.1321	9844
OC_2, OC_12	1.703	0.0898	9802
OC_3, OC_4	0.46695	0.6461	9854
OC_3, OC_5	0.67297	0.5068	9808
OC_3, OC_6	0.73801	0.4701	9841
OC_3, OC_9	0.66744	0.5215	9837
OC_3, OC_12	0.84461	0.4008	9822
OC_4, OC_5	0.20298	0.8451	9816
OC_4, OC_6	0.26618	0.7992	9842
OC_4, OC_9	0.19745	0.8426	9840
OC_4, OC_12	0.37029	0.7163	9875
OC_5, OC_6	0.062824	0.951	9837
OC_5, OC_9	0.0055689	0.9966	9859
OC_5, OC_12	0.16656	0.8646	9838
OC_6, OC_9	0.068414	0.9474	9823
OC_6, OC_12	0.10381	0.9163	9817
OC_9, OC_12	0.17218	0.863	9825
<i>Magdalen Shallows</i>			
OC_1, OC_2	1.0245	0.3069	9840
OC_1, OC_3	4.1616	0.0001	9822
OC_1, OC_4	6.9967	0.0001	9822
OC_1, OC_5	8.5655	0.0001	9821
OC_1, OC_6	9.4623	0.0001	9853
OC_1, OC_9	8.7522	0.0001	9837
OC_1, OC_12	9.8539	0.0001	9856
OC_2, OC_3	3.1523	0.0014	9842
OC_2, OC_4	6.0285	0.0001	9847
OC_2, OC_5	7.6011	0.0001	9834
OC_2, OC_6	8.4855	0.0001	9834
OC_2, OC_9	7.7831	0.0001	9850
OC_2, OC_12	8.8713	0.0001	9833
OC_3, OC_4	2.9554	0.0033	9844
OC_3, OC_5	4.5329	0.0001	9841
OC_3, OC_6	5.385	0.0001	9817
OC_3, OC_9	4.7029	0.0001	9847
OC_3, OC_12	5.7557	0.0001	9827
OC_4, OC_5	1.5605	0.122	9834
OC_4, OC_6	2.3802	0.0182	9830
OC_4, OC_9	1.7202	0.0867	9854
OC_4, OC_12	2.7361	0.0067	9834
OC_5, OC_6	0.80961	0.4222	9806
OC_5, OC_9	0.15611	0.8706	9826
OC_5, OC_12	1.1607	0.2476	9839
OC_6, OC_9	0.65428	0.5163	9842

Table 9 continued

Groups	t	P (perm)	Unique perms
OC_6, OC_12	0.35084	0.7213	9814
OC_9, OC_12	1.0056	0.3079	9830
<i>Newfoundland Shelf</i>			
OC_1, OC_2	Negative		
OC_1, OC_3	0.26758	0.8008	9424
OC_1, OC_4	0.59888	0.5627	9585
OC_1, OC_5	1.6577	0.1104	9736
OC_1, OC_6	1.3929	0.1716	9781
OC_1, OC_9	1.9236	0.0606	9793
OC_1, OC_12	1.6487	0.1125	9741
OC_2, OC_3	0.26758	0.8043	9446
OC_2, OC_4	0.59888	0.5525	9533
OC_2, OC_5	1.6577	0.112	9723
OC_2, OC_6	1.3929	0.1811	9764
OC_2, OC_9	1.9236	0.0602	9787
OC_2, OC_12	1.6487	0.1067	9730
OC_3, OC_4	0.33587	0.7376	9654
OC_3, OC_5	1.383	0.1797	9760
OC_3, OC_6	1.1233	0.2788	9763
OC_3, OC_9	1.6422	0.1026	9756
OC_3, OC_12	1.3737	0.1795	9768
OC_4, OC_5	1.0177	0.3202	9762
OC_4, OC_6	0.76797	0.4566	9763
OC_4, OC_9	1.2647	0.2184	9794
OC_4, OC_12	1.008	0.3183	9756
OC_5, OC_6	0.24276	0.8123	9770
OC_5, OC_9	0.23496	0.8139	9748
OC_5, OC_12	0.011442	0.9899	9726
OC_6, OC_9	0.4789	0.6363	9730
OC_6, OC_12	0.23179	0.8193	9743
OC_9, OC_12	0.24693	0.8105	9683
<i>North Gulf Shelf</i>			
OC_1, OC_2	Negative		
OC_1, OC_3	1.249	0.233	7857
OC_1, OC_4	1.8568	0.0486	8233
OC_1, OC_5	1.9561	0.0414	8590
OC_1, OC_6	2.503	0.0099	8991
OC_1, OC_9	2.273	0.0227	8970
OC_1, OC_12	2.503	0.0093	8978
OC_2, OC_3	1.249	0.2331	7813
OC_2, OC_4	1.8568	0.0466	8275
OC_2, OC_5	1.9561	0.0436	8541
OC_2, OC_6	2.503	0.0098	9024
OC_2, OC_9	2.273	0.0224	8943
OC_2, OC_12	2.503	0.0103	8988

Table 9 continued

Groups	t	P (perm)	Unique perms
OC_3, OC_4	0.43415	0.6434	7796
OC_3, OC_5	0.62638	0.5484	8304
OC_3, OC_6	1.1759	0.2531	8777
OC_3, OC_9	1.0087	0.3299	8713
OC_3, OC_12	1.1759	0.2569	8727
OC_4, OC_5	0.21975	0.8327	8355
OC_4, OC_6	0.79881	0.4512	8706
OC_4, OC_9	0.63378	0.5407	8739
OC_4, OC_12	0.79881	0.433	8731
OC_5, OC_6	0.56431	0.5827	8383
OC_5, OC_9	0.40954	0.6901	8397
OC_5, OC_12	0.56431	0.5843	8346
OC_6, OC_9	0.14217	0.8955	7908
OC_6, OC_12	Negative		
OC_9, OC_12	0.14217	0.8895	7923
<i>Scotian Shelf</i>			
OC_1, OC_2	1.0813	0.2857	9839
OC_1, OC_3	2.8067	0.0052	9818
OC_1, OC_4	5.4354	0.0001	9843
OC_1, OC_5	8.2117	0.0001	9814
OC_1, OC_6	8.8582	0.0001	9825
OC_1, OC_9	9.0351	0.0001	9815
OC_1, OC_12	9.2663	0.0001	9799
OC_2, OC_3	1.7581	0.0755	9844
OC_2, OC_4	4.4216	0.0001	9851
OC_2, OC_5	7.2328	0.0001	9841
OC_2, OC_6	7.8797	0.0001	9843
OC_2, OC_9	8.054	0.0001	9840
OC_2, OC_12	8.2789	0.0001	9831
OC_3, OC_4	2.6696	0.008	9849
OC_3, OC_5	5.5027	0.0001	9843
OC_3, OC_6	6.1447	0.0001	9803
OC_3, OC_9	6.3137	0.0001	9840
OC_3, OC_12	6.5272	0.0001	9842
OC_4, OC_5	2.854	0.0031	9828
OC_4, OC_6	3.4898	0.0005	9833
OC_4, OC_9	3.6526	0.0003	9831
OC_4, OC_12	3.8527	0.0001	9846
OC_5, OC_6	0.62771	0.5295	9830
OC_5, OC_9	0.78461	0.4376	9811
OC_5, OC_12	0.97287	0.3333	9843
OC_6, OC_9	0.15617	0.8788	9798
OC_6, OC_12	0.34265	0.7302	9813
OC_9, OC_12	0.18626	0.8491	9832

Table 9 continued

Groups	t	P (perm)	Unique perms
<i>St. Lawrence Estuary</i>			
OC_1, OC_2	0.24146	0.8147	9841
OC_1, OC_3	1.9211	0.0566	9848
OC_1, OC_4	2.7114	0.0057	9848
OC_1, OC_5	3.8825	0.0001	9840
OC_1, OC_6	4.3772	0.0001	9850
OC_1, OC_9	3.6809	0.0003	9831
OC_1, OC_12	4.5113	0.0001	9847
OC_2, OC_3	1.6939	0.0904	9826
OC_2, OC_4	2.4907	0.0099	9856
OC_2, OC_5	3.6574	0.0002	9836
OC_2, OC_6	4.15	0.0001	9815
OC_2, OC_9	3.4563	0.0003	9829
OC_2, OC_12	4.2835	0.0001	9851
OC_3, OC_4	0.80558	0.4251	9853
OC_3, OC_5	1.8913	0.0568	9829
OC_3, OC_6	2.3486	0.0181	9839
OC_3, OC_9	1.7027	0.089	9855
OC_3, OC_12	2.4722	0.0124	9853
OC_4, OC_5	1.0501	0.2898	9835
OC_4, OC_6	1.4919	0.1375	9851
OC_4, OC_9	0.867	0.3817	9842
OC_4, OC_12	1.6111	0.108	9835
OC_5, OC_6	0.44129	0.6712	9837
OC_5, OC_9	0.18389	0.8577	9837
OC_5, OC_12	0.56018	0.5824	9845
OC_6, OC_9	0.6256	0.534	9832
OC_6, OC_12	0.11882	0.9121	9845
OC_9, OC_12	0.74463	0.4606	9860
<i>The Grand Banks</i>			
OC_1, OC_2		Negative	
OC_1, OC_3	0.67494	0.4998	8675
OC_1, OC_4	0.613	0.5352	8449
OC_1, OC_5	0.9416	0.3512	8647
OC_1, OC_6	0.9416	0.3506	8650
OC_1, OC_9	1.2672	0.2145	8730
OC_1, OC_12	0.9416	0.3461	8681
OC_2, OC_3	0.67494	0.5016	8637
OC_2, OC_4	0.613	0.5428	8516
OC_2, OC_5	0.9416	0.3602	8739
OC_2, OC_6	0.9416	0.3552	8658
OC_2, OC_9	1.2672	0.2085	8749
OC_2, OC_12	0.9416	0.3548	8661
OC_3, OC_4	0.071426	0.9436	8708
OC_3, OC_5	0.25333	0.8032	8688

Table 9 continued

Groups	t	P (perm)	Unique perms
OC_3, OC_6	0.25333	0.7993	8687
OC_3, OC_9	0.57178	0.5684	8506
OC_3, OC_12	0.25333	0.8049	8657
OC_4, OC_5	0.32936	0.7407	8712
OC_4, OC_6	0.32936	0.739	8670
OC_4, OC_9	0.6525	0.508	8632
OC_4, OC_12	0.32936	0.7362	8650
OC_5, OC_6	4.0229E-08	1	8658
OC_5, OC_9	0.32155	0.7501	8476
OC_5, OC_12	4.0229E-08	1	8644
OC_6, OC_9	0.32155	0.745	8432
OC_6, OC_12	3.7159E-08	1	8677
OC_9, OC_12	0.32155	0.7479	8426

OC = cleaning out-of-water, with the number associated corresponding to the months when the action is required

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