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A COMPARISON OF TWO TIMED SEARCH METHODS FOR COLLECTING FRESHWATER MUSSELS IN GREAT LAKES COASTAL WETLANDS

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ABSTRACT

Given the catastrophic losses of freshwater mussel diversity across the Laurentian Great Lakes, the identification and protection of remnant assemblages are priority conservation actions. In contrast to riverine mussels, there has been little evaluation of different sampling gear and strategies to support the design of coastal wetland inventory or monitoring programs. We compared timed-search (qualitative) collections from 21 Lake Ontario coastal wetlands using clam rake and visual/tactile methods. Live mussels were collected with visual/tactile searches from 90% of wetlands sampled, and from 71% with the clam-rake. A total of 756 live mussels (representing nine species) were collected. Collections included three mussel species at risk: *Ligumia nasuta, Quadrula quadrula*, and *Toxolasma parvum*. Compared to clam-raking, visual/tactile searches collected more than twice as many live individuals and fresh shells, a broader range of sizes and significantly more species (and at a faster rate). Estimates of live mussel abundance and species number associated with each method were imprecise (CV > 0.35). The concordance of variation in mussel assemblage structure among wetlands (as described by each method) was not consistent or in strong agreement. Based on our findings, we recommend visual/tactile searches for future coastal wetland sampling efforts.

KEY WORDS Unionid, Dreissenids, Clam rake, Visual/tactile, Wetlands, Monitoring

INTRODUCTION

A third of freshwater mussel species in the province of Ontario (Canada) have been assessed as either federally threatened or endangered (COSEWIC 2012). Initial declines in native unionid populations have been related to the degradation of riverine habitats (Metcalfe-Smith et al., 1998). More recent and rapid declines followed the invasion of North America by driessenid mussels: Zebra Mussel (*Dreissena polymorpha*) and Quagga Mussel (*D. bugensis*) (Schloesser & Nalepa, 1994). By the early 1990s, native mussels were nearly extirpated from the offshore waters of Lakes Erie and St. Clair (McGoldrick et al., 2009). However, remnant mussel assemblages have persisted in nearshore and coastal wetland areas of Lakes Erie, Huron and St. Clair (Nichols & Amberg 1997; Zanatta et al., 2002; Bowers & Szalay, 2003; Crail et al., 2011; Sherman et al., 2013). Compared to adjacent open water habitats, wetlands are less suitable for driessenid colonization and survival (Sherman et al., 2013), thereby providing a refuge for native mussels. Given that dreissenid mussel removal may not be practical and brood-stock is required for reintroductions, recovery depends on identifying and protecting remnant native mussel assemblages.

Actions undertaken to protect and recover Ontario's mussels at risk include the identification of protected habitats, and ongoing assessment of species status. To meet these commitments, the following information is required: (1) the locations of individuals and populations, (2) descriptions of the biophysical attributes of habitat for different life-stages, (3) the state of populations (i.e. density, size and age structure, sex-ratio), and (4) the presence of invasive species (Cudmore et al.,

2006; DFO, 2011a). Outside of the Lake St. Clair delta, these activities have focused on populations in southwestern Ontario rivers. Riverine mussel assemblages are sampled with standardized time-search (Metcalfe et al., 2000) and quadrat methods (Metcalfe-Smith et al., 2007). In contrast to riverine mussels, little research has been undertaken on the design of wetland inventory or monitoring programs. The evaluation of alternative gear and sampling strategies is required for species found in inland lakes and coastal wetlands (e.g. *Ligumia nasuta*).

In this study, we compared timed-search (qualitative) collections from Lake Ontario coastal wetlands using clam rake and visual/tactile methods. Both methods have been used in previous wetland mussel surveys (Bowers & Szalay, 2003; Sherman et al., 2013). Other sampling approaches have included snorkeling surveys and opportunistic surveys of temporarily dewatered habitats (Nichols & Amberg, 1999; McGoldrick et al., 2009; Crail et al., 2011). The two methods were chosen based on the logistics of sampling coastal wetlands (soft sediments, poor water clarity and dense aquatic vegetation), and because they are not time-intensive (Bowers & Szalay, 2003). Also, it was expected that the clam-rake would be able to sample habitats too deep for visual/tactile methods. Comparisons were based on the: (1) number of species detected, (2) number and sizes of individuals collected, and (3) precision of mussel species and abundance estimates. We also assessed the concordance of mussel assemblage patterns described using clam-rake and visual/tactile data.

METHODS

Field Sampling

During the summers of 2011 and 2012, 21 coastal wetlands along the Canadian (Ontario) shoreline of Lake Ontario were sampled. Sites were located between the cities of St. Catherines (43o11'14" N; 79o16'52" W) and Kingston (44o14'17" N; 76o32'55" W). Wetlands represented a mix of types (barrier beach, flooded river mouth and large embayments) and were 13 to 2093 (median: 86) hectares in size (Environment Canada and Ontario Ministry of Natural Resources, 2003). Aquatic macrophyte coverage ranged from absent to extensive, and water clarity (as measured with a transparency tube, Anderson & Davic, 2004) was poor (< 0.2 m) to excellent (> 1.2 m). Water depths sampled ranged 0.05 to 1.5 m.

Mussel collection methods were compared using a paired-sample approach. At each wetland, 12 sampling points were randomly selected. No *a priori* information on sediment characteristics, water depth or spatial variation in mussel densities was available to stratify each wetland before points were selected. At each sampling point, one hour of search effort with each method was completed concurrently. Sampling was limited to within 50 m of the start point, and areas sampled by either method did not overlap. Assignment of a method to an area was ad hoc, but not based on any criteria. Visual/tactile searching involved either floating on air mattresses and hand searching the sediment for mussels (on the surface and probing through sediment for burrowed mussels), or searching for mussels with an underwater viewer (Plastimo® Round Underwater Viewer, 0.33 m diameter) or polarized lenses. In wetlands with clear water, mussels could be visually detected by spotting siphons or small clusters of dreissenids. It is estimated that tactile searches of soft sediments sampled up to a depth of 0.1 m. For the clam-rake method, an Eagle Claw® Clam Rake (0.84 m long handle, with a 0.26 x 0.15 m metal basket and ten 0.15 m long steel teeth) was dragged through the sediment and wetland vegetation. Spacing of wire mesh within the basket was 2.5 cm x 5 cm.

Live individuals and fresh shells were identified to species (Metcalfe-Smith et al., 2005). Shell length (mm) of live individuals was measured with a dial caliper (±0.1 mm). Live mussels and the total mass of attached dreissenids were weighed separately (±0.1 g). After processing and removal of dreissenids, live mussels were returned to the sediment close to their area of collection.

Data Analysis

Differences between the two methods were tested using the following data: (1) number of species detected, (2) number of individuals collected, (3) number of sampling points containing mussels, (4) precision of parameter estimates (mussel abundance and species number), and (5) shell length (minimum, mean and maximum). For datasets 1-3, separate comparisons were done for live individuals and fresh shells. Precision (calculated for each wetland) was based on the coefficient of variation (CV = Standard Error/Mean) (Thompson, 2002). Except for precision and shell length, significant differences between the sampling methods were tested with the paired t-test. Due to differences in mussel data, an unequal number of CVs was calculated for each sampling method. Therefore, tests for significant differences were undertaken with the unpaired t-test. Differences in shell length were tested using the Sign-test (Zar, 1984). Species detection rates for each method were compared by calculating the mean (across wetlands): (1) time till the first species was detected at a wetland, and (2) cumulative number of species detected after each hour of searching.

Two approaches were applied to assess the concordance of variation in mussel assemblages among wetlands, as described using clam rake and tactile/visu-

al data on live individuals. To assess whether estimates of live mussel abundance and species richness across wetlands agreed, the Spearman Rank Correlation was calculated. Secondly, distance matrices were constructed from site-by-species matrices of species presence/absence (Jaccard) and log-transformed species abundance (Bray-Curtis) data (Legendre & Legendre, 1998). The relationship between matrices constructed from clam rake and tactile/visual data was evaluated using the Mantel test. Significance was assessed with a Monte Carlo randomisation method, using 9999 permutations (Manly, 2007). Statistical analyses were completed using PAST version 1.94 (Hammer et al., 2001).

RESULTS

A total of 756 live mussels (representing nine species) were collected from Lake Ontario coastal wetlands (Table 1). Between one and five species were detected as live individuals from each wetland. At least one live mussel was collected from most wetlands (except Big Island Marsh, Bay of Quinte). *Pyganodon grandis* was the most widespread (collected from all wetlands where live mussels were found) and abundant species (65% of visual/tactile and 40% of clam rake collections). Other species were encountered at five or fewer wetlands, and typically represented <10% of the total collection. Three at-risk mussel species (COSEWIC 2012) were collected: *L. nasuta*, *Quadrula quadrula*, and *Toxolasma parvum*, of which *L. nasuta* was the most widespread. For both methods, live individuals were more often collected than fresh shells (Figure 1). In total, 870 shells (either halves or whole) were collected. Shells of all species except *T. parvum* were found. At half of the wetlands, some species (range: one to three species) were detected only as fresh shells (Table 1). At four wetlands, the presence of shells was the only indicator of the occurrence of *Elliptio complanata*, *Leptodea fragilis* and *Utterbackia imbecillis*. Shells were also the only evidence of *L. nasuta* within Presqu'ile Bay.

TABLE 1

Comparison of relative abundance (% of total collection) and frequency of occurrence (% of wetlands sampled) of wetland mussel species collected using clam-rake and visual/tactile methods. Absolute numbers are provided in parentheses. Summary statistics are based on live individuals.

	Visual/Tactile		Clam Rake	
Species	Relative Abundance	Frequency of Occurrence	Relative Abundance	Frequency of Occurrence
Elliptio complanata	11.6 (65)	14.3 (3)	19.7 (38)	14.3 (3)
Lampsilis cardium	2.3 (13)	9.5 (2)	14.0 (27)	4.8 (1)
Lampsilis siliquoidea	0.5 (3)	9.5 (2)	5.2 (10)	14.3 (3)
Leptodea fragilis	0.7 (4)	4.8 (1)	2.0 (4)	4.8 (1)
Ligumia nasuta	3.2 (18)	23.8 (5)	5.2 (10)	14.3 (3)
Pyganodon grandis	65.2 (367)	81.0 (17)	40.4 (78)	47.6 (10)
Quadrula quadrula	13.0 (73)	4.8 (1)	11.4 (22)	4.8 (1)
Toxolasma parvum	1.2 (7)	4.8 (1)	1.0 (2)	4.8 (1)
Utterbackia imbecillis	2.3 (13)	19.1 (4)	1.0 (2)	9.5 (2)

Abundance and Number of Mussel Species

Live mussels were collected with visual/tactile searches from 90% (19 of 21) of wetlands sampled, and from 71% (15 of 21) with the clam-rake. Visual/tactile searches collected three times as many live mussels ($t = 2.35$; $p = 0.02$) and twice as many fresh shells ($t = 2.81$; $p = 0.01$) as clam-raking (Figure 1). They also produced significantly more (>35%) sampling points at each wetland with live individuals ($t = 4.1$; $p < 0.001$), and more species from live individuals than clam-raking ($t = 2.95$; $p = 0.008$). Alternately, there was no difference between methods in the number of sampling points with shells at each wetland or species detected with shells (t-test; $p > 0.10$). Clam-raking only detected species not present in visual/tactile collections at five wetlands. There were no significant differences between search methods in the precision of live mussel abundance and species number estimates (t-test, $p > 0.25$). Overall, estimates were typically imprecise (CV>0.35).

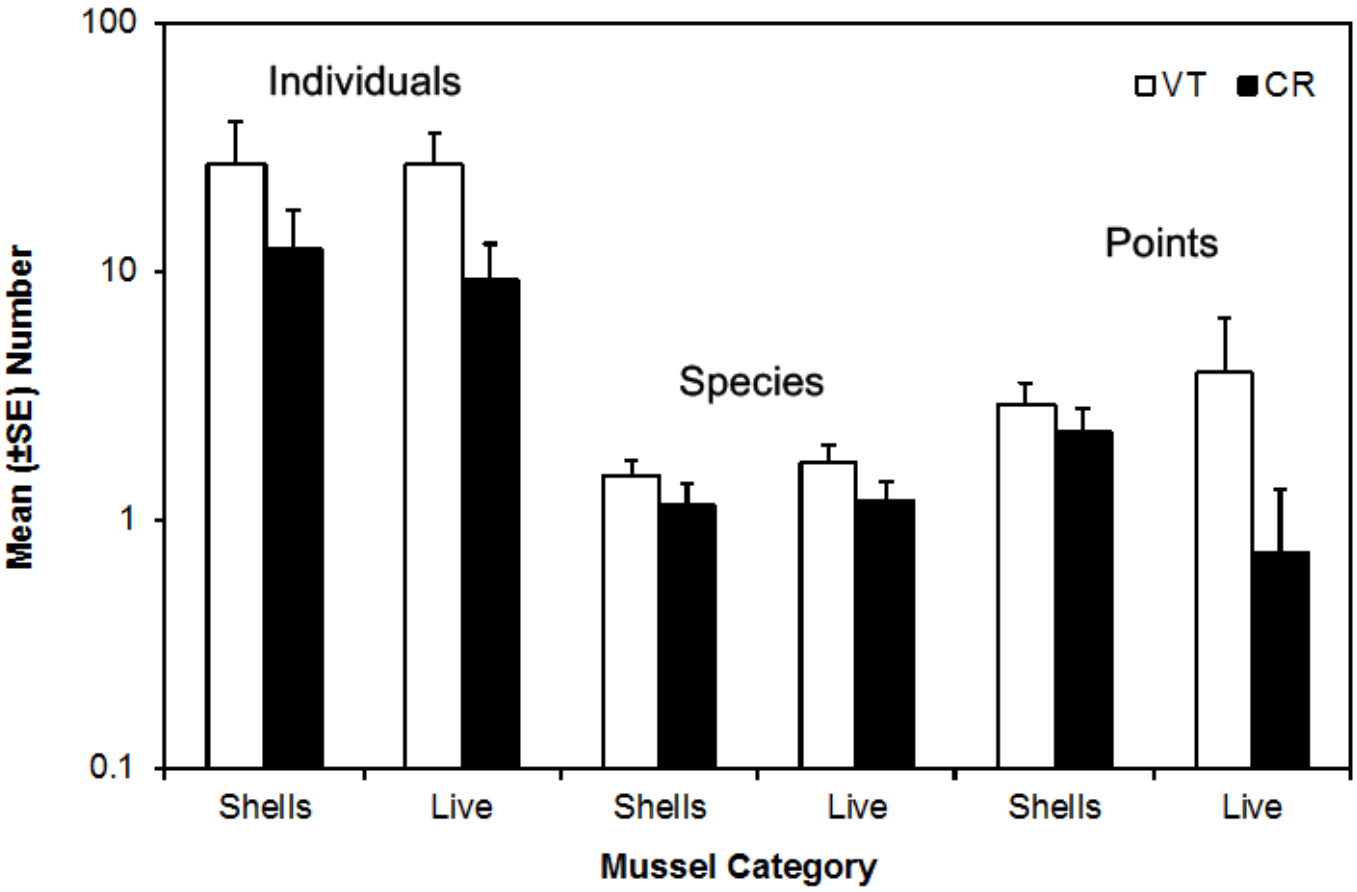

FIGURE 1

Comparison of mussel catch rates in Lake Ontario coastal wetlands using visual/tactile (VT) and clam-rake (CR) sampling methods. "Points" refers to the number of sampling points within a wetland where a fresh shell or live individual was collected.

Over the sampling period, detection of new species within wetlands was more rapid with visual/tactile searches than clam-raking (Figure 2). Although mean (±SE) time spent searching until the first mussel was collected at a wetland was slightly longer (VT: 3.5 hr ± 0.9. CR: 2.7 hr ± 0.7), there was little improvement in the detection of new species after 10 hours of visual/tactile searching or six hours of clam-raking. When compared to combined species lists (both methods) for each wetland, the mean percentage of mussel species detected using the visual/tactile method was greater than 80%. By contrast, clam-raking detected (on average) 23% fewer species known from each wetland (mean = 57.5%).

Shell Length

A greater range of shell lengths was associated with visual/tactile collections of live individuals than clam-rake samples (VT: standard deviation (SD) = 26.6, CR: SD = 24.9) (Figure 3). Compared to clam-rake, mean lengths of visual-tactile collections from each wetland were significantly greater (Sign Test: $p < 0.05$). Differences in mean length ranged from 1.0 and 39.6 mm (mean = 13.5). There were no significant differences between the lengths of the smallest or the largest individuals collected from each wetland (Sign Test: $p > 0.18$).

Variation Among Coastal Wetland Mussel Assemblages

The number of live individuals ($r_s = 0.81$, $p < 0.001$) and species ($r_s = 0.78$, $p < 0.001$) collected by each method was strongly correlated across wetlands. However, distance matrices constructed from species presence-absence data (Jaccard) were not correlated ($r = -0.04$, $p = 0.60$). At 10 of the 21 wetlands, there was no overlap in the species composition of visual-tactile and clam-rake collections. Most of these cases reflect the failure of a sampling method to collect any mussels. Using species abundance data (Bray-Curtis), there was a weak correlation ($r = -0.23$, $p = 0.007$) between distance matrices associated with each method. The relative abundances of individual species were equal at only 10% (2 of 21) of wetlands sampled.

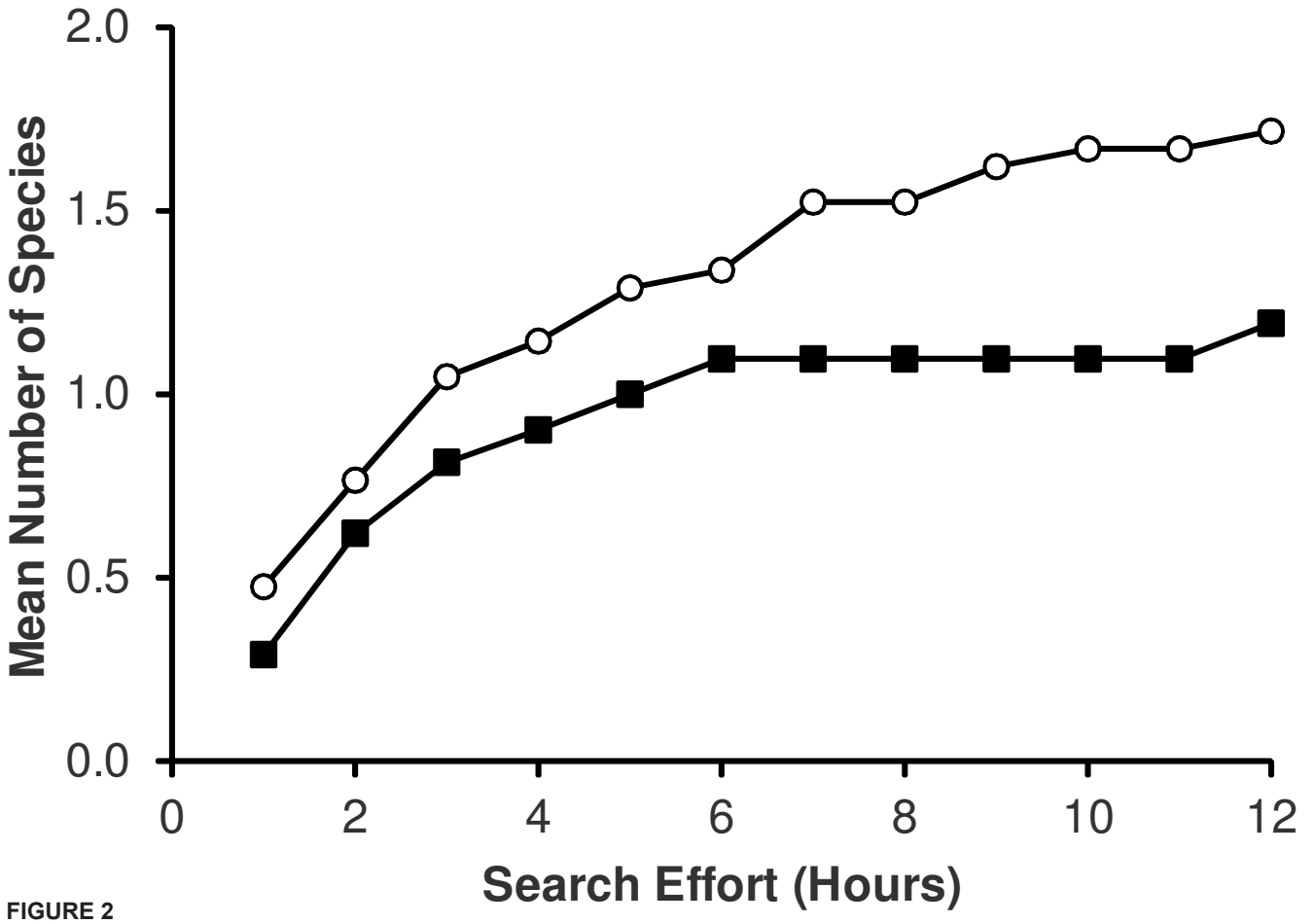

FIGURE 2

Comparison of increase in mussel species richness with increase in effort, during visual/tactile (○) and clam-rake (■) surveys of Lake Ontario coastal wetlands. Mean species richness represents the average calculated across all wetlands sampled.

DISCUSSION

Sampling gear and strategy evaluations for North American freshwater mussels have largely focused on riverine habitats. Given the catastrophic losses of freshwater mussel diversity across the Great Lakes, the inventory and population monitoring of remnant populations in coastal wetlands are priority recovery actions. Our study shows that visual/tactile surveys are more efficient at collecting mussels and detecting species than clam-raking. This result is consistent with Sherman et al. (2013) who reported that visual searches of a Lake St. Clair site collected four times more mussels than clam-raking. We also found a broader range of shell lengths to be associated with visual/tactile collections. It is not known whether this result reflects differences in the likelihood of capture between methods, or that the probability of detecting the smallest and largest sizes increases as one collects more mussels.

Compared to visual/tactile sampling, the clam-rake permits sampling at deeper water depths and further into soft sediments. However, it was less effective at collecting mussels and often labour-intensive. When sampling heavily vegetated habitats or soft sediments, the basket required continuous cleaning to remove plant and/or organic material and careful searching to find mussels. Unionids in Lake Erie wetlands have been observed to burrow 2-40 cm into the substrate for at least part of the day (Nichols & Wilcox, 1997). In rivers, the excavation of bed material during quadrat sampling improves the likelihood of collecting juveniles and small-bodied species (Obermeyer ,1998). In our study, there was no evidence that dragging the clam-rake through sediments improved the detection of small individuals. This may reflect the loss of small individuals through the basket mesh, or that tactile sampling was effective at detecting burrowed individuals by probing the sediments. Certain wetland characteristics (deep water or dense aquatic

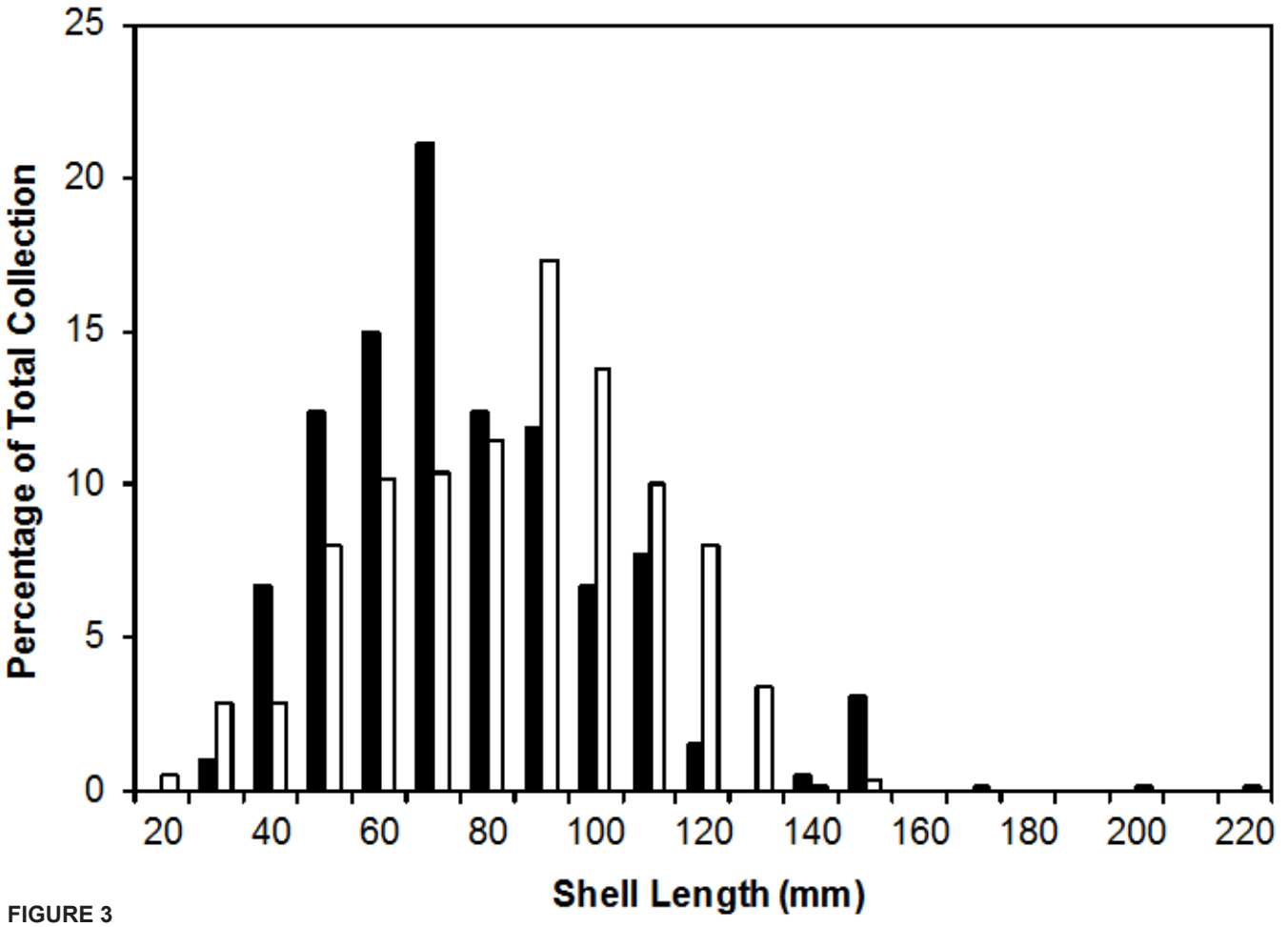

FIGURE 3

Length-frequency distributions of mussels collected from Lake Ontario coastal wetlands using visual/tactile (white) and clam-rake sampling methods (black).

vegetation) may prevent visual/tactile searches. In these cases, the completeness of species lists may be improved by increasing the time spent clam-raking and/or shoreline searches for fresh shells.

While the qualitative sampling approach applied in this study was appropriate for gear comparison and species inventory, quantitative sampling strategies are recommended for population monitoring (Strayer & Smith, 2003). Both methods evaluated in this study provided imprecise parameter estimates and were unsuitable for long-term monitoring. Future research could test whether stratified sampling designs, and/or large increases in search effort would improve precision. Alternatively, the ability of quantitative approaches developed for riverine mussel populations (e.g. systematic quadrat sampling with random starts) to provide abundance estimates could be evaluated for these low-density populations. If it is not necessary to track the number of individuals (or it is deemed impractical), repeat survey designs could be implemented across lower Great Lakes coastal wetlands to monitor species distributions instead (MacKenzie et al., 2012).

The overall objective of our study was to inform the design of native freshwater mussel collections in Great Lakes coastal wetlands. However, recovery plans for mussels at risk also identify the need to monitor dreissenid distribution and abundance. This information is used to interpret threat risks for individual populations (DFO 2011b). Live dreissenids were collected from nine of the wetlands we sampled. At these sites, dreissenid shells were present at 30% of sampling points. Across a variety of European and North American waterbodies, zebra mussel infestation rates (number or mass attached to native unionids) are correlated with zebra mussel densities (Lucy et al., 2013). We found the mass ratio of attached dreissenids to live unionids ranged from 0.0006 to 2.0 (mean = 0.15). Counts or weights of zebra mussels (and presence of byssal threads) on live mussels may

therefore provide a surrogate abundance index for monitoring and risk assessments.

Great Lakes coastal wetlands are important habitat for amphibians and reptiles, birds, fishes and mammals (Sierzen et al., 2012). Over the past 15 years, an increasing number of studies have demonstrated that, as refuge habitats, coastal wetlands are also important for unionid conservation throughout the Great Lakes basin. We found Lake Ontario wetland mussel assemblages to be less diverse than Lake Erie and Lake St. Clair wetland assemblages (Bowers & Szalay, 2003; Zanatta et al., 2002; Crail et al., 2011) but more diverse than those recently sampled in Lake Huron and Lake Michigan (Sherman et al., 2013). *Ligumia nasuta* (formerly considered one of the most common species of the lower Great Lakes) was believed extirpated from the Canadian waters of Lake Ontario (COSEWIC 2007). We detected small remnant populations of this endangered species at five wetlands. Additionally, undocumented populations of *Q. quadrula* (Threatened) and *T. parvum* (Endangered) were identified at another (Jordan Harbour). These findings highlight the need for additional inventories of coastal wetlands in the lower Great Lakes and upper St. Lawrence River to properly delineate critical habitats and identify provincially significant wetlands (OMNR 2013). We recommend that these surveys be implemented using visual/tactile methods.

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The Freshwater Mollusk Conservation Society (FMCS) is dedicated to the conservation of and advocacy of freshwater mollusks, North America's most imperiled animals. Membership in the society is open to anyone interested infreshwater mollusks who supports the stated purposes of the Society which are as follows:

1) Advocate conservation of freshwater molluscan resources;

2) Serve as a conduit for information about freshwater mollusks;

3) Promote science-based management of freshwater mollusks;

4) Promote and facilitate education and awareness about freshwater mollusks and their function in freshwater ecosystems;

5) Assist with the facilitation of the National Strategy for the Conservation of Native Freshwater Mussels (Journal of Shellfish Research, 1999, Volume 17, Number 5), and a similar strategy under development for freshwater gastropods.

OUR HISTORY

The FMCS traces it's origins to 1992 when a symposium sponsored by the Upper Mississippi River Conservation Committee, USFWS, Mussel Mitigation Trust, and Tennessee Shell Company brought concerned people to St. Louis, Missouri to discuss the status, conservation, and management of freshwater mussels. This meeting resulted in the formation of a working group to develop the National Strategy for the Conservation of Native Freshwater Mussels and set the ground work for another freshwater mussel symposium. In 1995, the next symposium was also held in St. Louis, and both the 1992 and 1995 symposia had published proceedings. Then in March 1996, the Mississippi Interstate Cooperative Research Association (MICRA) formed a mussel committee. It was this committee (National Native Mussel Conservation Committee) whose function it was to implement the National Strategy for the Conservation of Native Freshwater Mussels by organizing a group of state, federal, and academic biologists, along with individuals from the commercial mussel industry. In March 1998, the NNMCC and attendees of the Conservation, Captive Care and Propagation of Freshwater Mussels Symposium held in Columbus, OH, voted to form the Freshwater Mollusk Conservation Society. In November 1998, the executive board drafted a society constitution and voted to incorporate the FMCS as a not-for-profit society. In March 1999, the FMCS held it's first symposium "Musseling in on Biodiversity" in Chattanooga, Tennessee. The symposium attracted 280 attendees; proceedings from that meeting are available for purchase. The second symposium was held in March 2001 in Pittsburgh, Pennsylvania, the third in March 2003 in Raleigh, North Carolina, the fourth in St. Paul, Minnesota in May 2005, the fifth in Little Rock, Arkansas in March 2007, the sixth in Baltimore, Maryland in April 2009, the seventh in Louisville, Kentucky in 2011, and the eighth in Guntersville, Alabama in 2013. The society also holds workshops on alternating years, and produces a newsletter four times a year.

TO JOIN FMCS OR SUBMIT A PAPER

Please visit our website for more information at http://www.molluskconservation.org

Or contact any of our board members or editors of WALKERANA to talk to someone of your needs. You'll find contact information on the inside back cover of this publication.