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Research Article

Composition of native crayfish assemblages in southern Ontario rivers affected by rusty crayfish (*Orconectes rusticus* Girard, 1852) invasions – implications for endangered queensnake recovery

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Abstract

Orconectes rusticus (rusty crayfish) is an aggressive and prolific aquatic invader. Where introduced, it has caused dramatic ecosystem changes, including the replacement of native crayfishes. In Canada, it was first reported in Lake of the Woods in northwestern Ontario and a small number of south-central Ontario lakes during the 1960s. It has subsequently spread to other regions of the province. Its current expansion into southwestern Ontario rivers presents an unknown risk to the endangered queensnake (*Regina septemvittata*), an obligate feeder on freshly molted crayfish. We sampled 99 river sites across southern Ontario to: (i) describe crayfish assemblages within river reaches currently occupied by Queensnake; and, (ii) characterize the impact of *O. rusticus* on native crayfish assemblages. *O. rusticus* was caught at 41% of sites sampled, and was the only species at 24% of sites. The abundance, richness, and within-site distribution of native crayfishes were all significantly lower at sites with *O. rusticus*. Within the distribution of queensnake, crayfish assemblages were almost entirely dominated by *O. propinquus*, with *O. rusticus* being absent. However, *O. rusticus* was found along the lower Speed River, where only one dam separates it from Grand River queensnake populations. Given its past spread, *O. rusticus* will likely be introduced into areas used by queensnake and replace *O. propinquus* (the primary prey of queensnake). The adaptability of queensnake to prey upon non-native crayfish is unknown and requires investigation.

Key words: invasive species, monitoring, crayfish

Introduction

Rusty crayfish, *Orconectes rusticus* (Girard, 1852) is one of three crayfish species thought to be introduced into Canada from the United States (Hamr 2010). It is an aggressive species and prolific breeder. Where introduced, *O. rusticus* has caused dramatic changes to aquatic ecosystems, including the replacement of native crayfishes, damage to macrophyte beds, and shifts in macroinvertebrate and fish assemblages (Phillips et al. 2009). *O. rusticus* was first reported in Canada during the 1960s from Lake of the Woods in northwestern Ontario and a small number of south-central Ontario lakes (Crocker and Barr 1968). It has moved or been transported (via bait bucket transfers) into Canadian waters from the northern limits of its natural range in the Ohio River basin of the United States (Rosenburg et al. 2010). In other parts of the Laurentian Great Lakes basin, the major methods of rusty crayfish introduction include the release from bait buckets by recreational anglers, the intentional release by aquarium hobbyists, and their introduction by lake users to control nuisance weeds (Olden et al. 2011). *O. rusticus* has subsequently been captured from numerous lakes and rivers in other regions of the province (Berrill 1978; Momot 1996; Edwards et al. 2009).

Trends in the status of native and non-native crayfish have been monitored across hundreds of south-central Ontario lakes (Edwards et al. 2009; Somers and Reid 2010). However, a corresponding effort has not been undertaken for southern Ontario

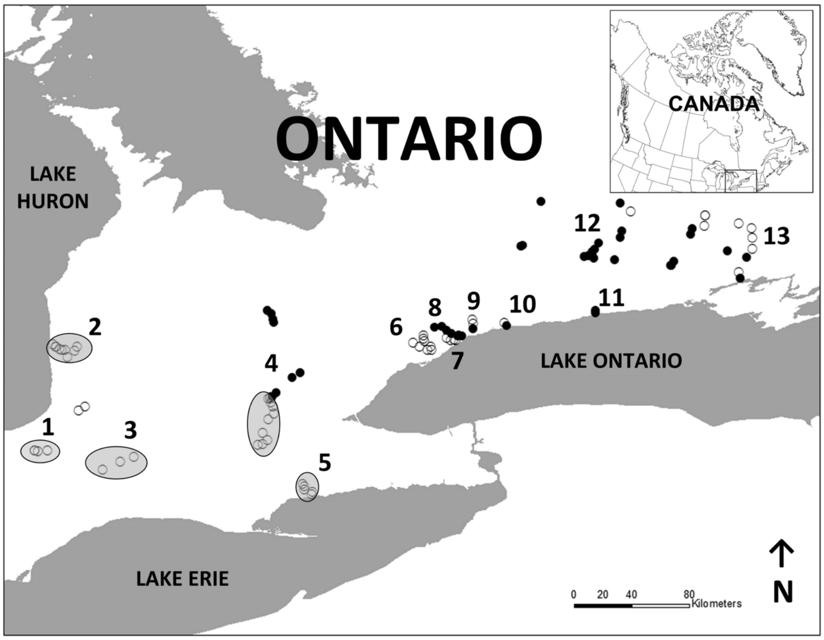

Figure 1. Distribution of crayfish sampling sites across southern Ontario (*O. rusticus* collection sites: ●). Sites associated with Queensnake habitat are within grey ellipses. Watersheds sampled are identified on the map as: 1 - Ausable R., 2 - Maitland R., 3 - Thames R., 4 - Grand R., 5 - Nanticoke Cr., 6 - Don River, 7 - Highland Cr., 8 - Rouge R., 9 - Duffins Cr., 10 - Oshawa Cr., 11 - Ganaraska R., 12 - Otonabee-Trent R., and 13 - Moira R.

streams and rivers. While the impacts of *O. rusticus* to native crayfishes in lakes have been thoroughly documented, research on declines of native crayfish populations in streams and rivers has, until lately, been less intensive (Jezerinac 1982; Jezerinac 1991; Daniels 1998). Recent stream surveys in the mid-west and eastern United States indicate a continuing spread of *O. rusticus* along with concurrent declines in native *Orconectes* species (Kuhlmann and Hazelton 2007; Kilian et al. 2010; Lieb et al. 2011a; Olden et al. 2011).

For North American freshwater fauna, alien aquatic invaders are a prevalent and often severe threat (Dextrase and Mandrak 2006). In southern Ontario rivers, the risk of indirect and direct impacts associated with invasions has been recognized for freshwater fauna at risk (Poos et al. 2010). Currently, the distribution of *O. rusticus* is expanding into the rivers of southwestern Ontario (Hamr 2010). In Canada, the federally endangered queensnake (*Regina septemvittata* Say, 1825) is only found in southwestern Ontario. Queensnake is an obligate feeder on freshly molted crayfish and is commonly associated with rock or gravel bottomed streams and rivers. Based on identifications of disgorged crayfish and crayfish collections at queensnake sites, *O. propinquus* (Girard, 1852) is assumed to be its primary prey in Ontario (Campbell and Perin 1979). Given its dependence on crayfish for survival, tracking *O. rusticus* invasions and investigating impacts to native crayfish populations are priority recovery actions identified for queensnake populations in Ontario (Gillingwater 2011).

In this study, we began to address these priorities through inventories of rivers currently occupied by queensnake, and south-central Ontario rivers within the current distribution of *O. rusticus*. The overall goal is to improve the assessment of the risk *O. rusticus* invasions pose to the recovery of the endangered queensnake. Specific objectives include: (1) describe crayfish assemblages within river reaches currently occupied by queensnake; and, (2) characterize the impact of *O. rusticus* on the composition of native crayfish assemblages in southern Ontario rivers. As declines in native crayfish species can also result from habitat degradation, we compared habitat characteristics at sites with and without *O. propinquus*.

Methods

Field sampling

Crayfish sampling occurred from July 25th to October 5th, 2011, and May 30th to October 16th, 2012. Each year, sampling was completed before *O. rusticus* initiated burrowing activity associated with winter hibernation (Hamr 2010). Ninety-nine

Table 1. Summary of habitat conditions at southern Ontario river crayfish sampling sites.

Habitat Characteristic	Mean	Median	Minimum	Maximum
Channel width (m)	28.4	17	2.8	125
Water temperature (°C)	19.4	20.7	9	27
Conductivity (µS/cm)	538	513	117	1331
Water clarity (cm)	82	87	1	120[a]
Qualitative Habitat Evaluation Index (QHEI) scores[b]				
Substrate	14.2	16	3	20
Cover	12.2	13	4	20
Channel	13.5	14	6	19
Riparian	8.4	9	3	10
Pool/Current	8.8	9	4	12
Riffle/Run	5.3	5	1	8

a: visible to bottom of the transparency tube, b: see Table 2 for descriptions of each QHEI score

sites in the following Laurentian Great Lake watersheds were sampled: Grand River and Nanticoke Creek (Lake Erie); Ausable and Maitland rivers (Lake Huron); Don River, Duffins Creek, Ganaraska River, Highland Creek, Moira River, Oshawa Creek, Otonabee-Trent River, and Rouge River (Lake Ontario); and, Thames River (Lake St. Clair) (Figure 1). These watersheds were selected as they differ in: (1) surficial geology; (2) intensity of agriculture practice and/or suburban/urban development; (3) degree of fragmentation by dams; (4) known occurrence of queensnake; and, (5) known occurrence of *O. rusticus*. Sites along each watercourse were randomly selected (including replacements for unsuitable or inaccessible sites) from a candidate site list. The list was developed using recent occurrence records of *O. rusticus* (EDDMapS Ontario 2014) and queensnake (Gillingwater 2011). The range in physical habitat characteristics is presented in Table 1.

At each site, 10 shoreline transects were evenly spaced, in alternating fashion, along both banks (Figure 2). Transects were 10 m long and 1 m wide. The length of habitat sampled at each site was defined as 15 times channel width; with a maximum of 500 m. For most sites, this permitted a variety of habitat types (*i.e.,* pool, riffle, and run) to be sampled and included at least one channel meander length. A 20-minute search was completed at each transect. Total search effort at each site was, therefore, standardized at 200 minutes. Crayfish were either caught by hand or scooped with flat bottom dip nets. Rocks were overturned to flush specimens from their refuges (Hamr 2007; Hamr and Sit 2011). All crayfish captured were identified to species (Crocker and Barr 1968), counted and released. Crayfish counts associated with each transect were recorded separately. Digital camera images and voucher specimens (preserved in 70% ethanol) were taken to confirm field identifications. Vouchers were not kept after confirmation of species identification.

Baited traps are often used to sample crayfishes in Ontario (Guiasu et al. 1996; Somers and Reid 2010). However, traps were not used as they require repeat site visits, are vulnerable to vandalism or theft (Bernardo et al. 2011), and deployment may be impractical in very shallow or fast-flowing habitats. While trapping has been an effective approach for crayfish monitoring in Ontario lakes, our approach has been effective at collecting *O. rusticus* and other native crayfishes in wadeable stream and river habitats (Hamr 2010; Hamr and Sit 2011; Reid and Devlin 2014). Hand-capture also avoided concerns related to potential harm to the queensnake associated with electrofishing.

Habitat quality was assessed at each site, as it may have a confounding effect on the impact of *O. rusticus* on native crayfish. At each site, habitat quality was scored using the Qualitative Habitat Evaluation Index (QHEI), a visual habitat index composed of seven principal metrics (Rankin 1989; Table 1). The QHEI approach was applied because it has been: (1) shown to generate scores correlated with crayfish population attributes (Burskey and Simon 2010); and, (2) successful in differentiating river habitat quality at free-flowing and impounded sites (Santucci et al. 2005) and across different physiographic regions (Reid et al. 2008). In addition to QHEI scores, channel width, water temperature, conductivity, and water clarity (using a transparency tube, Anderson and Davic 2004) were measured at each site.

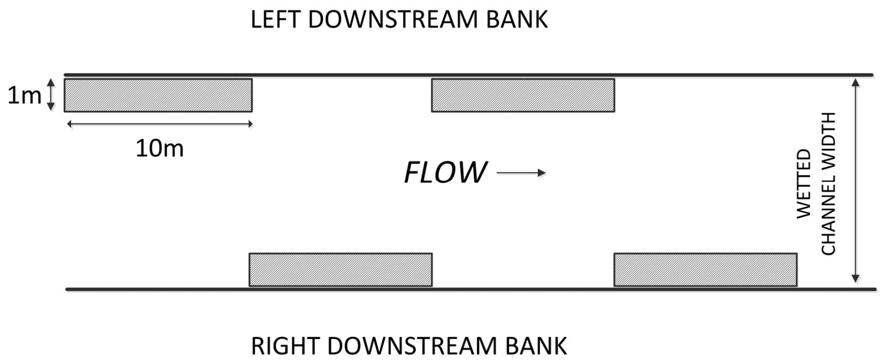

Figure 2. Plan view of generalized river channel, showing a representative distribution of sampled transects arranged in an alternating sequence along the margins of the wetted channel.

Table 2. Description of Qualitative Habitat Evaluation Index (QHEI) habitat metrics used to assess habitat quality at sampling sites. Descriptions are adapted from Rankin (1989).

Habitat metric (range of scores)	Description
Substrate (0–20)	Based on type and quality of bed material present. Sites with high scores are characterized by a greater number of particle sizes, the presence of coarse bed material (e.g. gravel and cobble) and low levels of embeddedness and silt deposition.
Cover (0–20)	Based on amount and diversity of cover present. Sites with high scores have large amounts and a variety of available cover types.
Channel (0–20)	Based on the development and stability of channel habitat. Sites with high scores have stable banks, sinuous channels and well developed riffle and pool habitats.
Riparian (0–10)	Based on the amount and quality of the riparian buffer. Sites with high scores have wide, forested riparian buffers and little bank erosion.
Pool/Current (0–12)	Based on quality of pool habitat and flow characteristics. Sites with high scores have deep, large pools and a diversity of water velocities.
Riffle/Run (0–8)	Based on quality of riffle and run habitats. Sites with high scores have deep riffle and run habitats with unembedded coarse bed material.

Data analysis

To assess the impact of *O. rusticus* on native crayfish, we compared the catches of native crayfish from sites with and without *O. rusticus*. Catch differences were tested using the non-parametric Mann-Whitney test. The following catch statistics were compared: (i) total number of crayfish; (ii) number of native crayfish; (iii) native crayfish richness (non-rarefied); (iv) number of *O. propinquus*; (v) number of transects with native crayfish; and, (vi) number of transects with *O. propinquus*. To visualize the variation in species composition across sites, we undertook a Correspondence Analysis (CA) using log-transformed species count data. As rare species can have a disproportionate effect on multivariate analysis (Jackson and Harvey 1989), *O. immunis* was excluded from the ordination (which was found at only one site).

The C-score index (Stone and Roberts 1990) was used to assess patterns of co-occurrence between *O. rusticus* and *O. propinquus*. Values significantly greater than expected by chance are interpreted to mean the two species did not tend to occur together. The test was simulated (algorithm: fixed rows-equiprobable columns, 5000 iterations) using ECOSIM 7.71. C-score was calculated using all sites sampled, and for a reduced dataset that only included those watersheds where *O. rusticus* was present.

Principal component analysis (PCA) with varimax rotation was used to reduce dimensionality and eliminate collinearity in habitat data. Because variables were measured with different units, variables were first transformed to z-scores by subtracting the mean from each observation and dividing the value by the standard deviation (Legendre and Legendre 1998). Principal components (PC) with eigenvalues greater than 1 and

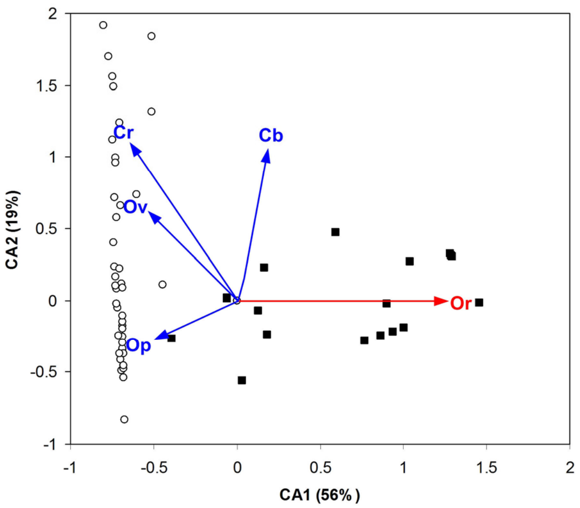

Figure 3. Biplot of site scores from Correspondence Analysis of log-transformed crayfish count data. Percent of among site variation explained by each axis is given in parentheses. Solid squares: sites with *O. rusticus*; open circles: native species only. Codes associated with species vectors: Cb – *C. bartonii*; *Cr – C. robustus*; Op – *O. propinquus*; Or – *O. rusticus*; Ov – *O. virilis*.

loadings greater than |0.6| were retained for further analysis. PC scores were used as independent variables in Multivariate Analysis of Variance (MANOVA) to test for habitat differences between sites with and without *O. propinquus*. Multivariate analyses were completed using PAST version 1.94 (Hammer et al. 2001).

Results

A total of 7596 crayfish were collected, representing six species (*Cambarus robustus* (Girard, 1852), *C. bartonii* (Fabricius, 1798), *O. immunis* (Hagen, 1870), *O. propinquus*; *O. rusticus*; and *O. virilis* (Hagen, 1870)). *O. rusticus* was collected from 41% of sites sampled, and was the only species at 24 sites. It accounted for 39% of all crayfish found during the study. *O. propinquus* was the most widespread (64% of sites sampled) and abundant (56% of crayfish collected) native species. Where either species occupied a site, they were often detected after only sampling two transects (*O. propinquus*: 83% of sites; *O. rusticus*: 93% of sites). However, at *O. rusticus* sites, *O. propinquus* represented only 12.2% (mean value, standard error (SE) = 4.0) of the crayfish caught. In contrast, at sites without *O. rusticus*, *O. propinquus* represented 72.5% (mean value, SE = 5.0) of the catch. Hybrids have been reported where the two species co-occur (Berrill 1985; Hamr 2010). We did not collect any apparent hybrids. *C. robustus*, *C. bartonii*, and *O. virilis* were relatively widespread (found at 14–39% of sites sampled), but comprised only 5.4% of all crayfish captured. *O. immunis* was found at only one site.

At sites within the known queensnake distribution, the crayfish assemblage was almost entirely dominated by *O. propinquus* (Table 3) and *O. rusticus* was not detected. However, *O. rusticus* was found along the lower reaches of Speed River, part of the Grand River watershed. In this area, a single dam separates *O. rusticus* from reaches of the Grand River used by queensnake.

Except for total number of crayfish captured, all catch statistics were significantly lower at sites where *O. rusticus* was present (Mann-Whitney Test, Table 4). The abundance, richness, and within-site distribution of native crayfishes were all substantially greater at sites without *O. rusticus*. The first two axes of CA explained 75% of the variation in crayfish assemblage composition across study sites (Figure 3). The biplot reflects differences identified with univariate comparisons. Along the first axis (CA1), there was a strong separation between sites with and without *O. rusticus*. This separation largely reflects differences in the abundance of the two numerically dominant crayfish species: *O. rusticus* and *O. propinquus*. Variation along the second CA axis reflects differences in native species abundance. C-scores for both full and reduced datasets were significantly greater than expected by chance ($p < 0.001$); further supporting the interpretation that *O. rusticus* and *O. propinquus* do not co-occur.

The first three principal components explained 58% of the variation in habitat condition among southern Ontario crayfish sampling sites (Table 5). The first axis (PC1) reflected QHEI habitat scores with strong positive loadings for Channel, Cover, and Riffle/Run. PC2 reflected Channel Width, Conductivity and Water Temperature; and PC3 had strong positive loadings for Water Clarity and Substrate. There was no separation between sites with and without *O. propinquus* along the first two PC (Figure 4). As shown by PC3, *O. propinquus* sites were characterized by a wider range of water clarity and substrate condition than sites without. PC scores were, however, not significantly different (MANOVA: Wilk's Lambda = 0.96, p = 0.27).

Table 3. Percent composition of crayfish collections from river reaches near Queensnake populations. The number of sites where the species was present is provided in parentheses. Species codes: *Op – O. propinquus*; *Ov – O. virilis; Cr – C. robustus*. Collection records for all sites sampled are provided in Supplementary material (Table S1).

Watercourse[a]	*Op*	*Ov*	*Cr*	Total Number
Ausable River (n=6)	97 (6)	0	3 (4)	813
Grand River (n=8)	90 (8)	6 (5)	4 (5)	886
Maitland River (n=9)	96 (9)	4 (8)	0	1631
Nanticoke Creek (n=5)	100 (5)	0	0	82
Thames River (n=3)	96 (3)	0	4 (3)	207

a: number of sites sampled is given in parentheses

Table 4. Comparison of mean (±SE) values of catch statistics based on the presence or absence of *O. rusticus* in southern Ontario rivers.

Catch Statistic	*O. rusticus* present	*O. rusticus* absent
Total crayfish number[n.s.]	81.3 ± 10.7	73.5 ± 10.8
Number of native crayfish[a]	9.8 ± 3.4	73.5 ± 10.8
Native species richness[a]	1.0 ± 0.5	3.9 ± 0.9
Number of *O. propinquus*[a]	8.5 ± 3.1	67.3 ± 10.6
Number of transects with native crayfish[a]	2.0 ± 0.5	7.2 ± 0.4
Number of transects with *O. propinquus*[a]	1.8 ± 0.5	6.4 ± 0.5
Index of Dispersion (95% confidence limits[b])	2.26 (1.6 - 2.8)	7.13 (4.7 - 9.2)

Significance levels for Mann-Whitney tests: n.s. ($p > 0.05$); a: ($p < 0.001$). b: bootstrap estimate ($n = 5000$).

Table 5. Principal component loadings of habitat metrics measured at southern Ontario crayfish sampling sites, showing eigenvalues and percent variance explained by each component axis. Loadings greater than |0.6| are bolded.

Habitat Metric	PC1	PC2	PC3
Channel	**0.82**	-0.16	0.20
Cover	**0.69**	-0.40	-0.29
Riffle/Run	**0.67**	0.21	0.26
Water temperature	-0.05	**0.83**	-0.008
Channel width	0.14	**0.71**	0.06
Conductivity	-0.002	**-0.76**	-0.05
Water clarity	-0.05	0.06	**0.79**
Substrate	0.45	-0.15	**0.62**
Riparian	0.59	0.15	0.05
Pool/Current	0.54	-0.09	-0.34
Eigenvalue	2.50	2.12	1.15
Percent Variance	25.0	21.2	11.5

Discussion

Almost half of the freshwater crayfish species in North America are considered imperiled. Freshwater mussels are the only other aquatic taxa at greater risk of extinction (Butler et al. 2003; Taylor et al. 2007). In Ontario, urbanization, wetland draining, acid rain, and the spread of non-native species have had a negative effect on native crayfish and their habitats (Guiasu 2007; Guiasu 2009; Edwards et al. 2009; Phillips et al. 2009). Our surveys suggest the spread and establishment of *O. rusticus* across southern Ontario rivers has resulted in substantial declines in native crayfish species abundance and diversity; often resulting in local extirpations. Our results complement recent studies in south-central Ontario streams and lakes that indicate a general, and often substantial, decline in the status of native crayfish is occurring across a large geographic area (Edwards et al. 2009; Reid and Devlin 2014). While consistent with other investigations that have surveyed over multiple decades (Daniels 1998; Olden et al. 2011), we recognize that our interpretations of *O. rusticus* impacts are based primarily on among-site

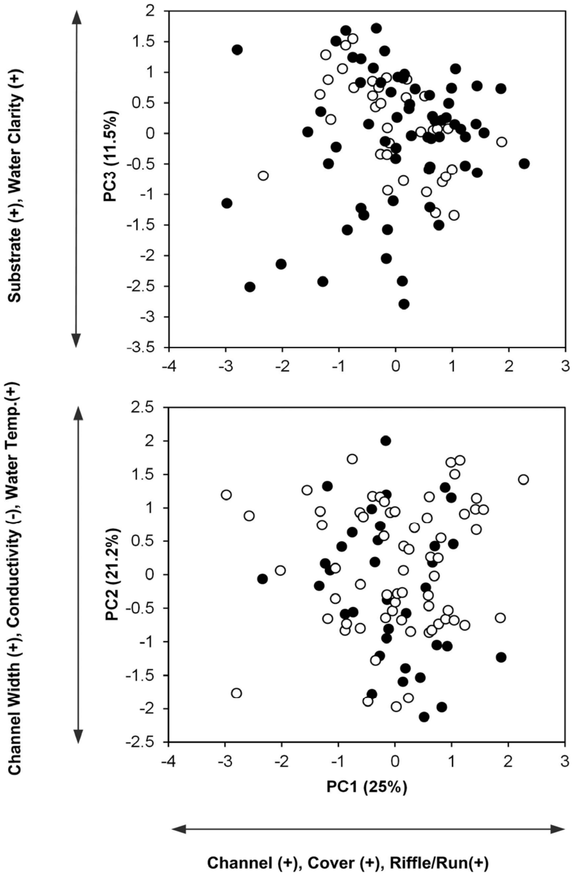

Figure 4. Biplots of scores from principal components (PCs 1-3) that summarized habitat measurements at southern Ontario river sites where *O. propinquus* was present (•) or absent (○) from 2011/2012 crayfish collections. Percent variation explained by each component is provided in parentheses. Habitat metrics with loadings >|0.6| are provided along each axis.

comparisons. Our baseline is largely restricted to Ontario distribution summaries provided by Crocker and Barr (1968); when *O. propinquus* was considered the most commonly encountered species in southern Ontario and *O. rusticus* known from only five sites. Regardless, based on the prevalence of factors known to promote introductions across the region (*i.e.,* impoundments, agricultural and urban land use: Olden et al. 2011), it is expected that the expansion of *O. rusticus* into other watersheds will continue.

Riverine habitat characteristics can influence the success and effect of non-native crayfish invasions. Examples of influential characteristics include: location within a watershed (Jezerinac 1982; Bernardo et al. 2011; Sargent et al. 2011), river gradient and hydrologic variability (Light 2003), stream temperature (Sargent et al. 2011, Momot 1984), productivity (Momot 1984), and water chemistry (Rallo and García-Arberas 2002). However, in southern Ontario, *O. rusticus* is now found in a wide range of stream and river habitats affected by varying levels of urban and agricultural land use (EDDMapS Ontario 2014, Reid and Devlin 2014, this study). This pattern supports the interpretation of Lieb et al. (2011a) that the spread of *O. rusticus* in flowing waters appears more limited by dispersal opportunities than availability of suitable environmental conditions. QHEI data across *O. propinquus* collection sites were highly variable and did not indicate that habitat quality influenced occurrence. Alternatively, presence and abundance of *O. propinquus* appears limited by that of *O. rusticus*. At the small number of sites where both species are present, it is unknown whether co-existence reflects local habitat conditions or simply that an insufficient amount of time has passed for complete species replacement.

Currently, the distribution of *O. rusticus* is within several kilometres of Grand River queensnake populations, and in watersheds adjacent to other populations. Given its spread over the past four decades in Ontario, it is likely that *O. rusticus* will become introduced along those river reaches and negatively affect *O. propinquus*. While there is a strong likelihood that *O. rusticus* introductions would have a strong, negative effect on *O. propinquus* abundance, it is unknown whether: (i) queensnake would feed on the non-native (but presumably abundant) crayfish, and (ii) freshly molted *O. rusticus* are equally vulnerable to capture. In the case of the endangered Lake Erie water snake (*Nerodia sipedon insularum* Conant and Clay, 1937), a shift in diet from native fishes and amphibians to the invasive round goby (*Neogobius melanostomus* Pallus, 1814) has resulted in rapid growth and the attainment of a larger body size (King et al. 2006). Although generally reflective of the most abundant local species, queensnake uses a variety of Orconectid and Cambarid species across its North American range (Gillingwater 2011). Chemical cues associated with molting (*e.g.* ecdysone) are critical for queensnake to locate its prey. However, other factors such as crayfish odour, appearance, and behaviour also play an important role in eliciting predatory strikes (Jackrel and Reinert 2011). While *O. rusticus* has a similar molting schedule to *O. propinquus*, it is unknown whether *O. rusticus* is equally vulnerable to capture after molting. This uncertainty requires further investigation.

Dam removal can have many ecological benefits to stream and river ecosystems. However, as dams block or limit crayfish dispersal (Momot 1996), removal of dams may facilitate invasions and negative impacts to native crayfish (Lieb et al. 2011b) and fishes (Fluker et al. 2009). Moreover, recent studies have demonstrated that dams effectively limit upstream movements of invasive crayfishes (Dana et al. 2011; Rosewarne et al. 2013). In the Grand River Fisheries Management Plan, modifying or removing existing barriers to fish passage has been identified as a habitat management strategy for the Speed River sub-watershed (OMNR and GRCA 1998). Currently, the most downstream dam (1.5 m high) along the Speed River is being considered for removal (A. Timmerman, OMNRF *personal communication*). Despite the presence of *O. rusticus* in this sub-watershed for three decades (Corey 1988), it was not found at Grand River sampling sites near the outflow of the Speed River. The large number of dams and weirs throughout this sub-watershed has likely limited the downstream dispersal of *O. rusticus*. While dam removal would provide upstream access to more than five km of habitat for Grand River fishes, it would facilitate the spread of *O. rusticus* and associated risks to native crayfishes and the endangered queensnake.

Limiting the opportunity for *O. rusticus* to be introduced into areas supporting queensnake populations is the best control option currently available (Dresser and Swanson 2013). Ontario Fishing Regulations (2007) currently limit the use of crayfish as bait to the same waterbody where collected. Further, fisheries management actions that promote healthy bass populations could control *O. rusticus* population sizes through predation (Reynolds and Souty-Grosset 2012) and reduce the risk of further dispersal. Trapping and increased fish predation has been successful at reducing *O. rusticus* abundance, but not eradicating them, within a small Wisconsin lake (Hein et al. 2006). Alternatively, intensive harvest of *O. rusticus* over a seven-month period had little impact on well-established southern Ontario river populations (Hamr 2010). Given the labour intensive nature of removing *O. rusticus*, a highly fecund species, eradication or control is only likely to succeed if it is detected early and the species is confined to a small area (Dextrase 2002; Hamr 2010). For there to be an opportunity to remove *O. rusticus* from queensnake habitats, we recommend that a regular monitoring program be implemented in "at risk" river reaches, and field protocols for *O. rusticus* removal be drafted and field-tested.

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References

Anderson P, Davic RD (2004) Use of transparency tubes for rapid assessment of total suspended solids and turbidity in streams. *Lake and Reservoir Management* 20:110–120, http://dx.doi.org/10.1080/07438140409354355

Bernardo JM, Costa AM, Bruxelas S, Teixeira A (2011) Dispersal and coexistence of two non-native crayfish species (*Pacifastacus leniusculus* and *Procambarus clarkii*) in NE Portugal over a 10-year period. *Knowledge and Management of Aquatic Ecosystems* 401: 28, http://dx.doi.org/10.1051/kmae/2011047

Berrill M (1985) Laboratory induced hybridization of two crayfish species, *Orconectes rusticus* and *O. propinquus*. *Journal of Crustacean Biology* 5: 347–349, http://dx.doi.org/10.2307/1547884

Berrill M (1978) Distribution and ecology of crayfish in the Kawartha Lakes region of southern Ontario. *Canadian Journal of Zoology* 56: 166–177, http://dx.doi.org/10.1139/z78-026

Burskey JL, Simon TP (2010) Reach- and watershed-scale associations of crayfish within an area of varying agricultural impact in west-central Indiana. *Southeastern Naturalist* 9: 199–216, http://dx.doi.org/10.1656/058.009.s310

Butler RS, DiStefano RJ, Schuster GA (2003) Crayfish: An overlooked fauna. *Endangered Species Bulletin* 28: 10–11

Campbell CA, Perrin DW (1979) A survey of the queensnake (*Regina septemvittata*) in southwestern Ontario. Ontario Ministry of Natural Resources

Corey S (1988) Comparative life histories of two populations of the introduced crayfish *Orconectes rusticus* (Girard, 1852) in Ontario. *Crustaceana* 55: 29–38, http://dx.doi.org/10.1163/156854088X00221

Crocker DW, Barr DW (1968) Handbook of the Crayfishes of Ontario. University of Toronto Press, Toronto, CAN, 158 pp

Dana ED, Garcia De Lomas J, Gonzalez R, Ortega F (2011) Effectiveness of dam construction to contain the invasive *Procambarus clarkii* in a Mediterranean mountain stream. *Ecological Engineering* 37: 1607–1613, http://dx.doi.org/10.1016/j.ecoleng.2011.06.014

Daniels RA (1998) Changes in the distribution of stream-dwelling crayfishes in the Schoharie Creek system, eastern New York State. *Northeastern Naturalist* 5: 231–248, http://dx.doi.org/10.2307/3858623

Dextrase A (2002) Preventing the introduction and spread of alien aquatic species in the Great Lakes. In: Claudi R, Nantel P, Muckle-Jeffs E (eds), Alien Invaders in Canada's waters, wetlands, and forests. Natural Resources Canada, Ottawa, ON, pp 219–232

Dextrase A, Mandrak NE (2006) Impacts of alien invasive species on freshwater fauna at risk in Canada. *Biological Invasions* 8: 13–24, http://dx.doi.org/10.1007/s10530-005-0232-2

Dresser C, Swanson B (2013) Preemptive legislation inhibits anthropogenic spread of an aquatic invasive species, the rusty crayfish (*Orconectes rusticus*). *Biological Invasions* 15: 1049–1056, http://dx.doi.org/10.1007/s10530-012-0349-z

EDDMapS Ontario (2014) Early detection and distribution mapping system for Ontario. University of Georgia. The Center for Invasive Species and Ecosystem Health. http:/www.eddmaps.org/Ontario

Edwards BA, Jackson DA, Somers KM (2009) Multispecies crayfish declines in lakes: implications for species distributions and richness. *Journal of the North American Benthological Society* 28: 719–732, http://dx.doi.org/10.1899/08-148.1

Fluker BL, Kuhajda BR, Duncan RS, Salter EL, Schulman M (2009) Impacts of a small dam removal on the endangered Watercress Darter. *Proceedings of the Annual Conference of the Southeastern Fish and Wildlife Agencies* 63:188–195

Gillingwater SD (2011) Recovery Strategy for the Queensnake (*Regina septemvittata*) in Ontario. Ontario Recovery Strategy Series. Ontario Ministry of Natural Resources, 34 pp

Guisau RC (2007) Conservation and diversity of the crayfishes of the genus *Fallicambarus hobbs*, 1969 (Decapoda, Cambaridae), with an emphasis on the status of *Fallicambarus fodiens* (Cottle, 1863) in Canada. *Crustaceana* 80: 207–223, http://dx.doi.org/10.1163/156854007780121438

Guisau RC (2009) Conservation, status, and diversity of the crayfishes of the genus *Cambarus erichson*, 1846 (Decapoda, Cambaridae). *Crustaceana* 82: 721–742, http://dx.doi.org/10.1163/156854009X407722

Guiasu RC, Barr DW, Dunham DW (1996) Distribution and status of crayfishes of the genera *Cambarus* and *Fallicambarus* (Decapoda, Cambaridae) in Ontario, Canada. *Journal of Crustacean Biology* 16: 373–383, http://dx.doi.org/10.2307/1548893

Hammer Ø, Harper DAT, Ryan PD (2001) PAST: paleontological statistics software package for education and data analysis. *Palaeontologia Electronica* 4: 1–9

Hamr P (2007) Sampling protocol for rusty crayfish *Orconectes rusticus* in Ontario. Upper Canada College Press, Toronto, ON, 33 pp

Hamr P (2010) The biology, distribution and management of the introduced rusty crayfish, *Orconectes rusticus* (Girard), in Ontario, Canada. *Freshwater Crayfish* 17: 85–90

Hamr P, Sit B (2011) Long-term monitoring of crayfish populations in the Credit River at the Upper Canada College Outdoor School. *Freshwater Crayfish* 18: 83–86, http://dx.doi.org/10.5869/fc.2011.v18.83

Hein CL, Roth BM, Ives AR, Vander Zanden MJ (2006) Fish predation and trapping for rusty crayfish (*Orconectes rusticus*) control: a whole-lake experiment. *Canadian Journal of Fisheries and Aquatic Sciences* 63: 383–393, http://dx.doi.org/10.1139/f05-229

Jackrel SL, Reinert HK (2011) Behavioral responses of a dietary specialist, the queen snake (*Regina septemvittata*), to potential chemoattractants released by its prey. *Journal of Herpetology* 45: 272–276, http://dx.doi.org/10.1670/10-047.1

Jackson DA, Harvey H (1989) Biogeographic associations in fish assemblages: local vs. regional processes. *Ecology* 70: 1472–1484, http://dx.doi.org/10.2307/1938206

Jezerinac RF (1982) Life-history notes and distributions of crayfishes (Decapoda: Cambaridae) from the Chagrin River basin, northeastern Ohio. *Ohio Journal of Science* 82: 181–192

Jezerinac RF (1991) The distribution of crayfishes (Decapoda: Cambaridae) of the Licking River watershed, east central Ohio: 1972–1977. *Ohio Journal of Science* 91: 108–111

Kilian JV, Becker AJ, Stranko SA, Ashton M, Klauda RJ, Gerber J, Hurd M (2010) The status and distribution of Maryland crayfishes. *Southeastern Naturalist* 9: 11–32, http://dx.doi.org/10.1656/058.009.s302

King RB, Ray JM, Stanford KM (2006) Gorging on gobies: beneficial effects of alien prey on a threatened vertebrate. *Canadian Journal of Zoology* 84: 108–115, http://dx.doi.org/10.1139/z05-182

Kuhlmann ML Hazelton PD (2007) Invasion of the upper Susquehanna River watershed by rusty crayfish (*Orconectes rusticus*). *Northeastern Naturalist* 14: 507–518, http://dx.doi.org/10.1656/1092-6194(2007)14[507:IOTUSR]2.0.CO;2

Legendre P, Legendre L (1998) Numerical Ecology. Elsevier Science BV, Amsterdam, The Netherlands, 853 pp

Lieb DA, Bouchard RW, Carline RF (2011a) Crayfish fauna of southeastern Pennsylvania: distributions, ecology and changes over the last century. *Journal of Crustacean Biology* 31: 166–178, http://dx.doi.org/10.1651/10-3287.1

Lieb, DA, Bouchard RW, Carline RF, Nuttall TR, Wallace JR, Burkholder CL (2011b) Conservation and management of crayfishes: lessons from Pennsylvania. *Fisheries* 36: 489–507, http://dx.doi.org/10.1080/03632415.2011.607080

Light T (2003) Success and failure in a lotic crayfish invasion: the roles of hydrologic variability and habitat alteration. *Freshwater Biology* 48: 1886–1897, http://dx.doi.org/10.1046/j.1365-2427.2003.01122.x

Momot WT (1984) Crayfish production – a reflection of community energetics. *Journal of Crustacean Biology* 4: 35–54, http://dx.doi.org/10.2307/1547894

Momot WT (1996) History of range extensions of *Orconectes rusticus* into northwestern Ontario and Lake Superior. *Freshwater Crayfish* 11: 61–72

Olden JD, Vander Zanden MJ, Johnson PTJ (2011) Assessing ecosystem vulnerability to invasive rusty crayfish (*Orconectes rusticus*). *Ecological Applications* 21: 2587–2599, http://dx.doi.org/10.1890/10-2051.1

Ontario Fishery Regulations (2007) http://laws-lois.justice.gc.ca/eng/regulations/SOR-2007-237

Ontario Ministry of Natural Resources and Grand River Conservation Authority (OMNR and GRCA) (1998) Grand River Fisheries Management Plan, 105 pp

Phillips ID, Vinebrooke RD, Turner MA (2009) Ecosystem consequences of potential range expansions of *Orconectes virilis* and *Orconectes rusticus* crayfish in Canada – a review. *Environmental Reviews* 17: 235–248, http://dx.doi.org/10.1139/A09-011

Poos MS, Dextrase AJ, Schwalb AN, Ackerman J (2010) The secondary invasion of the round goby into high diversity Great Lakes tributaries and species at risk hotspots: Potential new concerns for endangered freshwater species. *Biological Invasions* 12: 1269–1284, http://dx.doi.org/10.1007/s10530-009-9545-x

Rallo A, García-Arberas L (2002) Differences in abiotic water conditions between fluvial reaches and crayfish fauna in some northern rivers of the Iberian Peninsula. *Aquatic Living Resources* 15: 119–128, http://dx.doi.org/10.1016/S0990-7440(02)01156-7

Rankin ET (1989) The qualitative habitat evaluation index (QHEI): rationale, methods, and application. Ohio Environmental Protection Agency, Columbus, OH, 89 pp

Reid SM, Devlin J (2014) Effectiveness of stream sampling gear to capture non-native rusty crayfish (*Orconectes rusticus*) in Ontario. *Canadian Field-Naturalist* 128: 111–118

Reid SM, Carl LM, Wilson CC, Mandrak NE (2008) Influence of dams and habitat condition on *Moxostoma* species distribution in the Grand River, Ontario. *Environmental Biology of Fishes* 81: 111–125, http://dx.doi.org/10.1007/s10641-006-9179-0

Reynolds J, Souty-Grosset C (2012) Management of freshwater biodiversity: crayfish as indicators. Cambridge University Press, Cambridge, UK, 374 pp

Rosenburg DM, Turner MA, Jansen W, Mosindy T, Watkinson DA (2010) Threats to Lake of the Woods and the Winnipeg River by the Rusty Crayfish (*Orconectes rusticus*), an Aquatic Invader. Ontario Ministry of Natural Resources, Northwest Science and Information Technical Workshop Report. TWR-005, 54 pp

Rosewarne PJ, Piper AT, Wright RM, Dunn AM (2013) Do low-head riverine structures hinder the spread of invasive crayfish? Case study of signal crayfish (*Pacifastacus leniusculus*) movements at a flow gauging weir. *Management of Biological Invasions* 4: 273–282, http://dx.doi.org/10.3391/mbi.2013.4.4.02

Santucci VJ, Gephard SR, Pescitelli SM (2005) Effects of multiple low-head dams on fish, macroinvertebrates, habitat, and water quality in the Fox River, Illinois. *North American Journal of Fisheries Management* 25: 975–992, http://dx.doi.org/10.1577/M03-216.1

Sargent LW, Golladay SW, Covich AP, Opsahl SP (2011) Physiochemical habitat associations of a native and a non-native crayfish in the lower Flint River, Georgia: implications for invasion success. *Biological Invasions* 13: 499–511, http://dx.doi.org/10.1007/s10530-010-9844-2

Somers KM, Reid RA (2010) The Dorset Environmental Science Centre crayfish sampling protocol. In: Rosenburg DM, Turner MA, Jansen W, Mosindy T, Watkinson DA (eds), Threats to Lake of the Woods and the Winnipeg River by the Rusty Crayfish (*Orconectes rusticus*), an Aquatic Invader. Ontario Ministry of Natural Resources, Northwest Science and Information Technical Workshop Report. TWR-005, pp 14–24

Stone L, Roberts A (1990) The checkerboard score and species distributions. *Oecologia* 85: 74–79, http://dx.doi.org/10.1007/BF00317345

Taylor CA, Schuster GA, Cooper JE, DiStefano RJ, Eversole AG, Hamr P, Hobbs III HH, Robison HW, Skelton CE, Thoma RF (2007) A reassessment of the conservation status of crayfishes of the United States and Canada after 10+ years of increased awareness. *Fisheries* 32: 372–389, http://dx.doi.org/10.1577/1548-8446(2007)32[372:AROTCS]2.0.CO;2

The following supplementary material is available for this article:

Table S1. Crayfish hand-capture collection data associated with 99 southern Ontario river sites sampled in 2011 and 2012.

This material is available as part of online article from: http://www.aquaticinvasions.net/2015/Supplements/AI_2015_Reid_Nocera_Supplement.xls