DE GRUYTER

Biologia **72**/8: 877—885, 2017
Section Zoology
DOI: 10.1515/biolog-2017-0094

Experimental evidence of the successful invader *Orconectes limosus* outcompeting the native *Astacus leptodactylus* in acquiring shelter and food

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Abstract: Successful invasive species compete for the same available resources with related native species, frequently driving the latter to the cusp of extirpation because of a lack of adaptive response. In this paper we analysed the behavioural relationships between two species of crayfish, the native *Astacus leptodactylus* and the invasive *Orconectes limosus* in an ongoing invasion process in the Lower Danube, in Eastern Europe. We tested the species' ability to acquire shelter and food in laboratory experiments in both intra- and interspecific confrontations. The dominant behaviour of the invasive species is obvious even towards its own congeners, while the native species display a more tolerant conspecific behaviour. With respect to interspecific confrontation, the invasive crayfish males and females were inclined to sex-specific dominance regarding shelters. A roughly balanced behaviour was noted for intersexual confrontations. The results of this study also highlight that the occupancy of a shelter is more disputed than food resources, which appear to be opportunistically acquired. In the context of the current invasion process, we hypothesised that the effect caused by interference competition might lead to a decline of the native species. Further investigations may reveal if there is any hope for recovery of the native species.

Key words: aggression; behaviour; competition; invasive species; lower Danube

Introduction

Established relationships between related species occupying the same ecological niche are set by complex competition processes (Morse 1974), their long-term stability depending on intrinsic or extrinsic traits (Moore & Bergman 2005; Václavík & Meentemeyer 2012). A species arriving in a new environment creates a novel situation in which the rearrangement of the existent relationships becomes mandatory (Broennimann et al. 2007). In time, the resulting assemblage should lead to either the consolidation of these relationships (Chuang & Peterson 2016) or the extinction of one of the species (Gherardi 2006). Laboratory experiments involving specimens of different kind of competitors (e.g., invasive and native species) may be expected to reveal insights of their mechanisms behind the relationships establishment.

Many native species have been displaced since the introduction of invasive species (Imhoff et al. 2011; Lodge et al. 2012; Leon et al. 2016), thus leading to a critical need for an understanding of the mechanisms behind successful invasions. There are species that have been documented as not being socially dominant or having negative effects in their native habitats (Leon et al. 2016), but become better competitors once they are translocated into a new environment (Guan 1994).

The competition for natural resources usually involves aggression in order to intimidate weaker competitors (Rahel & Stein 1988; Preisser et al. 2005). By employing such behaviours, the dominant individuals acquire shelters (Groza et al. 2016), food (Stocker & Huber 2001), and increase their chances to mate (Christy 1987). Consequently, the dominated individuals lose fitness (Vorburger & Ribi 1999) and their populations decline or even become extirpated (Gherardi 2006). Studies show that the success of invasion might, among others, depend more on a species ability to respond to natural selection, rather than on vast physiological tolerance or plasticity (Lee 2002).

Crayfish represent an important component of the freshwater ecosystems. They can compete not only with related species, but other top consumers as well (Momot 1995). In general, crayfish avoid conflict, as many species usually prefer to stay hidden during daylight (Kawai et al. 2004). Crayfish use shelters for protection not only against predators (Garvey et al. 1994; Soderback 1994; Barki & Karplus 2016), but also as a refuge by berried females (Figler et al. 1997), newly moulted individuals (Ackefors 1996), or in the cold/draughty seasons (Ilhéu et al. 2003; Grow & Merchant 1980). In the last century, Europe experienced the introduction of many invasive crayfish species, through farming or even pet trading (Chucholl 2013), some of the most successful being *Pacifastacus leniusculus* (Dana, 1852), *Procambarus clarkii* (Girard, 1852), *Orconectes virilis* (Hagen, 1870), *Procambarus fallax* f. *virginalis* Martin et al., 2010 and *O. limosus* (Rafinesque, 1817) (Kouba et al. 2014). The latter was first detected in the Upper Danube in 1985 (Puky & Schád 2006), from where it spread along the river network. In 2008 this species was found in the Lower Danube, Romania (Pârvulescu 2009). This river sector currently faces the on-going spreading process of both the invasive crayfish species and the crayfish plague pathogen it carries, *Aphanomyces astaci* (Schikora, 1906). A decline of the relative abundance can be observed in native crayfish *Astacus leptodactylus* Eschscholtz, 1823 populations in the Romanian Lower Danube (Pârvulescu et al. 2012, 2015).

The continuous growing pressure of invasive crayfish populations all over the world triggered the research effort to focus on many competition issues in order to reveal the invasion mechanisms. The acute invasion of *O. limosus* in Lower Danube against the native crayfish species *A. leptodactylus* motivated us to investigate the interactions of these two competitors from behavioural perspectives in order to test whether the two species would share two very important resources, food and shelter, or would resort to compete and reject each other.

Material and methods

The crayfish were collected using fishing nets from areas where the species never overlapped to avoid experimental biases caused by previous interactions. The invasive *O. limosus* individuals were collected from the invaded sector in the Lower Danube main channel, near the village Şviniţa, on the Romanian shore (GPS 44°31′45″ N, 22°04′37″ E), and *A. leptodactylus* specimens from a pond near Bucharest (GPS 44°03′10″ N, 25°47′09″ E). Individuals were sampled outside the mating season (sexually inactive, form II) to exclude the potential influences caused by mating behaviours that might lead the males to excessive aggressiveness observed in other studies, i.e., Stebbing et al. (2003), Snedden (1990).

All captured individuals were allowed to acclimate in large tanks (100 l) to laboratory conditions prior to the beginning of experiments for at least two weeks. The individuals of both species were kept separated prior to the experiments, each species being assigned to a large tank. A

Table 1. The experimental trials, combined according to total body length, sex and species.

Intraspecific confrontations ASL vs. ASL	Individuals total length (mm) in replicates			
M > M	92.4/70.2	101.8/86.6	112/88.4	
M = M	114.2/113.7	84.4/85.1	92.9/93.2	
F > F	98.8/69.8	109.5/74.4	119.6/84.2	
F = F	74.6/75.2	100.4/101.1	72.2/72.9	
M > F	111.8/84.7	109.9/66.9	99.7/66.4	
M = F	84.2/84.7	100.6/101.2	82.9/90.1	
M < F	74.4/100.2	69.9/112.9	99.4/119.7	
OCL vs. OCL				
M > M	113.3/74.1	82.2/66.9	117.7/100.1	
M = M	117.7/116.9	88.8/89.3	101.2/99.8	
F > F	112.2/77.2	99.2/60.4	120.2/87.5	
F = F	77.3/78.3	111.9/113.1	103.3/101.4	
M > F	113.7/65.5	103.9/77.2	87.9/59.7	
M = F	75.5/77.1	86.4/88.1	109.9/108.1	
M < F	77.6/120.4	81.3/109.7	99.1/119.4	
Interspecific confrontations OCL vs. ASL	Individuals total length (mm) in replicates			
M > M	89.4/61.1	117.4/84.4	98.8/69.7	109.1/81.2
M = M	63.1/65.3	78.4/79.2	100.1/101.2	
M < M	73.4/112.4	69.9/100.7	84.2/121.8	
F > F	109.1/88.1	117.2/84.7	89.4/56.3	
F = F	77.4/79.2	109.1/111	117.7/115.9	
F < F	65.9/110.1	81.2/113.1	79.1/99.5	
M < F	55.9/99.1	74.2/119.1	81.1/119.4	
M = F	59.4/61.1	89.5/91	103.3/105	
M < F	77.4/111.2	89.2/121.3	79.9/118.7	
F > M	98.8/65.1	121.9/80.2	97.3/65.1	
F = M	77.8/79.3	89.9/87.5	100.4/102.1	
F < M	70.4/97.7	65.4/99.7	89.1/109.9	44.3/89.4
	101.1/129.1	79.9/101.2		

Explanations: ASL – *A. leptodactylus;* OCL – *O. limosus;* M – males; F – females.

total of 94 *A. leptodactylus* and 81 *O. limosus* individuals were involved in the experiments. Individuals were provided with *ad-libidum* shelter and food. The experimental recipients consisted of 20 L glass tanks, each equipped with a single ceramic shelter (approx. 80 × 150 × 50 mm) and water filtering systems. The temperature was kept around 18–20 °C under natural daylight cycle conditions. We only used intermoult adults in experimental trials, combined according to total body length and sex, provided a total of 7 combinations for intraspecific and 12 for interspecific experiments, respectively (see Table 1). Equal sized trials consisted of individuals in the pair below of 1% variation in the total length. The unbalanced confrontation was considered if one individual was smaller for at least 25% in the total length, referring to the largest opponent. Each trial lasted for at least 16 days, and for each combination we recorded the assignment of the individuals to the existing shelter and periodically measured the competitors' wet weight. After every trial, the tank, shelter and filtering pump were thoroughly cleaned in order to remove remaining pheromones which may have influenced the behaviour of the next occupants (Bergman et al. 2003). Each individual was used for a single trial timeframe, thus avoiding eventual biases caused by the learning or memory effect (Aquiloni et al. 2012). Injured or dead animals were replaced and the trial was run again in order to complete at least three replicates for each combination.

The experiment started after the acclimatization period. Each visually isolated tank contained two crayfish. Since both species are expressing similar circadian behaviour (Musil et al. 2010; Skurdal & Taugbøl 2002), the shelter occupancy was recorded every two days, two times a day, during daylight conditions (before and after the midday). We scored as either "balanced" if both individuals were found in the shelter, "winner" for the individual found in the shelter, or "loser" for the individual found outside the shelter.

Prior to the introduction in the experiment, each individual's Wet Weight was measured with the aid of a Kern analytical balance (nearest 0.01 g), and these measurements were used as baseline WW values. In the experimental trials, we weighed the crayfish every four days to determine which individual was successful in feeding against its competitor. We fed the crayfish one day before weighing, considering that the ingested food would result in an increased recorded WW (Meade & Watts 1995). The food during the experiment consisted in one piece of fish of about 10% of the combined baseline WW of the two competitors. For each weight determination, the crayfish were labelled as "successful" if their WW had increased by at least 5% compared with their baseline value, "unsuccessful" in the case of a decrease of at least 5% in WW compared with the baseline, or "constant" otherwise. An individual was scored as "winner" if it was the only one labelled "successful" in the pair, or if it remained "constant" while its competitor was "unsuccessful"; in this case the other crayfish was scored as "loser". In all the remaining cases the situation was considered "balanced".

Due to the design of the experiment and specifics of the interaction data collected, the statistical analysis of the results is primarily descriptive. Considering each combination of individuals are unique, not allowed for a more complex approach employed in other behavioural studies, i.e., Matsuzaki et al. (2012). For both shelter and food competitions, the percentages of "balanced" observations for the two species were compared, in matched pairs corresponding to sex-size experimental combinations, via Wilcoxon signed rank tests. Next, after excluding the "balanced" cases, the remaining instances were analysed to determine any dominance patterns in intra- or inter-specific confrontations. This was accomplished by comparing the proportions of "winners" in each experimental combination with 1/2, using exact binomial tests. Dominance was established when the respective proportion was significantly greater than 1/2. The statistical analyses were performed using the R software version 3.2.4 (R Core Team 2016). For the visualisation of the results the data was displayed as percentages for the given situation out of the corresponding total number of outcomes.

No protected or rare species (according to Habitats Directive and IUCN Red List) were involved in this study. After the experiment, the individuals were euthanized by freezing at –20 °C to avoid the spreading of the crayfish plague pathogen *Aphanomyces astaci* in the wild (Oidtmann et al. 2002). In this particular case, the crayfish plague pathogen was confirmed in both invasive and native crayfish species inhabiting Lower Danube (Pârvulescu et al. 2012).

Results

During the acclimatization period, the native *A. leptodactylus* specimens were found occupying a shelter in 98% of cases, and the invasive *O. limosus* in 93% of cases, respectively. We consider this data to be relevant for the interpretation of further results obtained under controlled laboratory experimental trials.

Competition for shelter

The percentages of "balanced" observations were significantly greater for *A. leptodactylus* (Wilcoxon $V = 28$, $P = 0.02225$), thus implying that *O. limosus* displays a more pronounced dominance behaviour against his congeners. In the case of *A. leptodactylus*, Fig. 1A shows that whenever two individuals were equal in size and of the same sex, it was most likely for them to be found in the same shelter. Also, when the individuals were of the same sex but different size, the proportion of instances when the larger crayfish was established as a "winner" was not significantly different from 1/2 ($P = 1$ for both males and females) showing the absence of a dominance pattern. In confrontations between crayfish of different sexes, regardless of size combinations, the proportion of "winner" males was significantly greater than 1/2 ($P < 0.05$ in all cases), therefore males can be considered dominant against their own species females. For *O. limosus,* clear dominance patterns were observed more frequently (Fig. 2A). The larger crayfish was dominant in same sex confrontations ($P < 0.01$ for both males and females). Males dominated in confrontations between males and females ($P < 0.001$ for large males versus small females; $P < 0.1$ for large females versus small males). In the case where the two opponents were equal in size, but not of the same sex, the males were always dominant ($P < 0.01$). P values were not calculated for the confrontations between crayfish in the same total body length category.

The shelter sharing behaviour revealed similar results in *A. leptodactylus* males or females confronta-

Fig. 1. Diagram showing the results (percentages of each category out of total number of observations) for shelter (A) and food (B) confrontations between individuals of native *A. leptodactylus*. Bar colours: green – balance, blue – dominance of males, red – dominance of females. Lighter blue or red bars show results associated with the smaller individual in confrontation with the larger one. M – males; F – females. *P* indicates level of statistical significance of dominance. n/a – *P* values were not computed because the crayfish were in the same category.

tions, individuals of equal sizes being observed in the same shelter in 89% of observations for males, and 83% for females, respectively. When one of the competitors was larger in size, the sharing rate slightly decreased. In male versus female confrontations, the sharing increased with female size, 33% when smaller, 50% when equal and 71% when the females were larger than males (Fig. 1A). The shelter sharing behaviour was found considerably lower in *O. limosus*, females being slightly permissive with 43% and 33% of the observations revealing the occupancy of the same shelter. None of the males of this species were found sharing a shelter with other males. The very same behaviour was observed in confrontations between males and females, only larger females being observed to share the same shelter in 25% of the observations (Fig. 2A).

In interspecific comparisons (Fig. 3A), *O. limosus* emerges as dominant in confrontations between males, confrontations between females ($P < 0.05$ in all cases) and when large *O. limosus* males faced small *A. leptodactylus* females ($P < 0.05$). In the other experimental combinations, the proportion of instances in which *O. limosus* was established as "winner" was not statistically significantly different from 1/2 ($P > 0.1$).

The shelter sharing behaviour was rarely encountered in interspecific competitions. When males or females were facing each other, we observed a slightly increased sharing behaviour when *O. limosus* were smaller (48% of observations in males, 31% in females, respectively). The same low shelter sharing behaviour was recorded also in different sex confrontations, varying between 0% and 38% of the recorded observations.

Competition for food

The percentages of "balanced" observations were not significantly different for the two species (Wilcoxon V = 22, $P = 0.2188$). In intraspecific comparisons, for both species (Figs 1B, 2B), a lack of clear dominance pat-

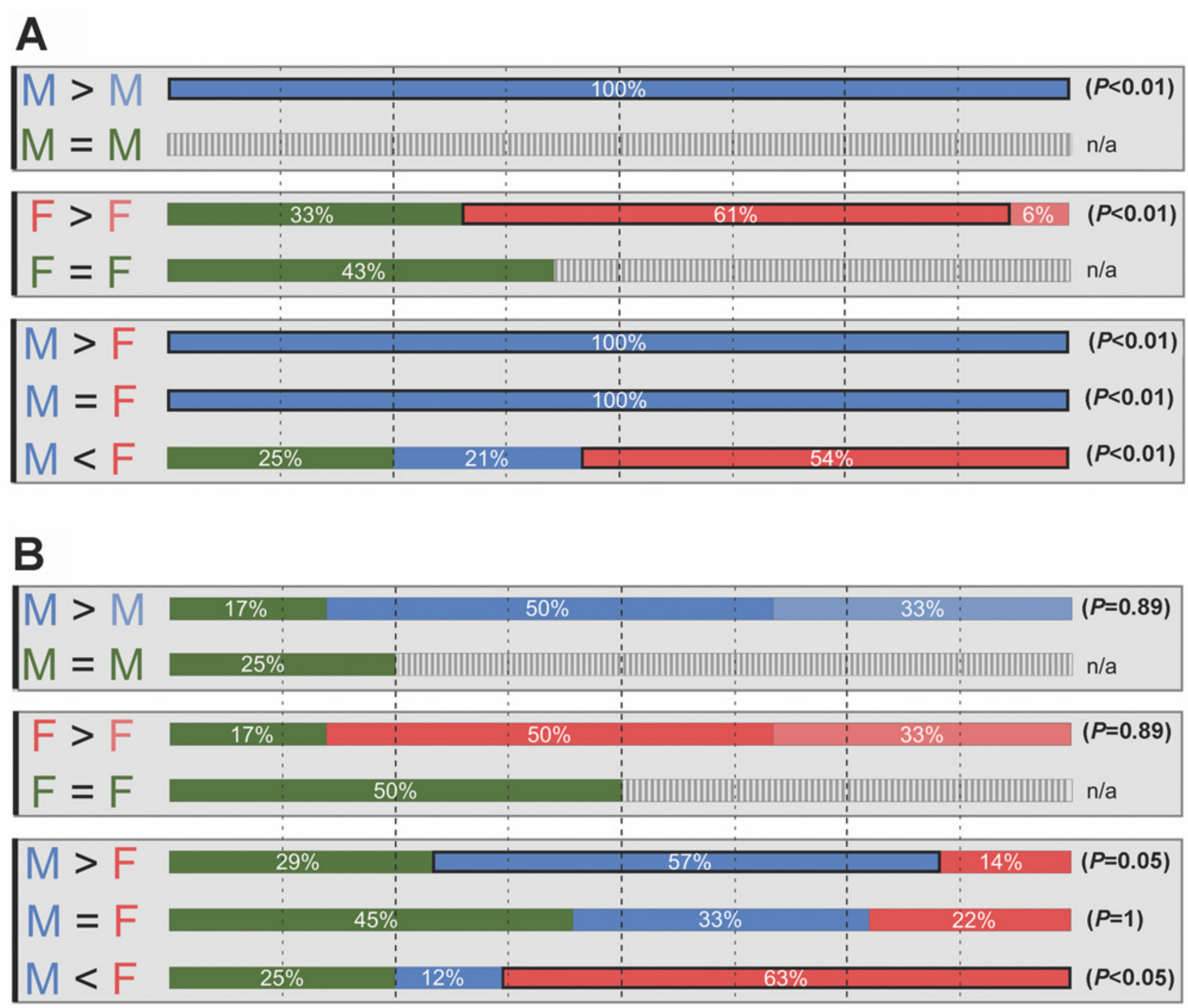

Fig. 2. Diagram showing the results (percentages of each category out of total number of observations) for shelter (A) and food (B) confrontations between individuals of *O. limosus*. Bar colours: green – balance, blue – dominance of males, red – dominance of females. Lighter blue or red bars show results associated with the smaller individual in confrontation with the larger one. M – males; F – females. *P* indicates level of statistical significance of dominance. n/a – *P* values were not computed because the crayfish were in the same category.

terns was noted for all experimental combinations ($P > 0.1$), although some were observed as during *O. limosus* experiments, larger individuals appeared to dominate the smaller ones. Also, in interspecific confrontations (Fig. 3B), the "balanced" situations constitute the majority of observations (50–87%), and the proportions of instances when *O. limosus* individuals were labelled as "winner" were not significantly different than 1/2 ($P > 0.1$). These results point to the possibility that the food is achieved opportunistically.

Other remarks

It should be pointed out that in 27% of the trials where males of different species were involved, the *O. limosus* individual killed its opponent, even when smaller in size. Deaths that showed no visible attack marks haven't been included in this observation. The aggressive behaviour of *O. limosus* was obvious in situations where the native crayfish was literally crushed between chelae or had its appendages amputated. Moreover, it was frequently observed during the experiments that the aggressive behaviour of *O. limosus* males was expressed by immobilising individuals of both of females and males of their congeners and *A. leptodactylus* opponents in pseudo-mating (Fig. 4), observed for lasting more than one day, ultimately resulting in the death of the suppressed individual.

Discussion

The results of this study reveal an innate aggressive behaviour of *O. limosus*, very visible in the case of shelter occupancy. This is shown by the dominance of invasive crayfish males and females against the same sex competitors of the native species. Dominant acts such as pseudo-mating have been observed, too. In its native environment (north-eastern USA), *O. limosus* does not appear to be a dominant crayfish species because it has

Fig. 3. Diagram showing the results (percentages of each category out of total number of observations) for shelter (A) and food (B) confrontations between individuals of invasive and native crayfish. Bar colours: green – balance, red – dominance of *O. limosus*, yellow – dominance of *A. leptodactylus*. M – males; F – females. *P* indicates level of statistical significance of dominance.

stronger competitors, e.g., *Orconectes rusticus* (Girard, 1852) (Klocker & Strayer 2004). In contrast, in Europe this crayfish species exhibits a strong competitiveness against the native crayfish species (Vorburger & Ribi

Fig. 4. Picture showing the *O. limosus* male behaviour of opponent immobilisation in pseudo-mating.

1999). It is worth noting that the competitiveness of *O. limosus* was found being lower in comparison with *P. leniusculus* (Hudina et al. 2011), one of the most threatening invasive species in Europe (Chucholl 2016).

Usually, relationships are established after repeated confrontations between opponents (Kaczer et al. 2007). Even though the invasion of crayfish is considered an irreversible phenomenon for the receiving wider community (Mathers et al. 2016). Thereby, it is possible that in the future, the native *A. leptodactylus* will cope with this aggressive invasion. It was observed that *P. leniusculus* individuals' aggressiveness is more pronounced in older invaded areas compared to active fronts of invasion (Hudina et al. 2015). Apparently this behaviour is caused by the population's density growth which makes the competition for resources even fiercer. Rebrina et al. (2015) showed that invasive *P. leniusculus* individuals have a better body condition in the active front of invasion and a worse condition in the old invaded zone, which might suggest a higher level of both intra- and inter-specific confrontations in this area due to more numerous individuals into the population. As even one infected individual can cause an outbreak, the stress caused by the competition with *O. limosus* in the active invasion front might have increased the incidence of crayfish plague, providing a potential explanation for the decline of native *A. leptodactylus* in these regions (Pârvulescu et al. 2012). We point out that no mass mortalities were observed in regions where the invaders themselves had not yet arrived, but the presence of the pathogen *A. astaci* was confirmed (Pârvulescu et al. 2012; Schrimpf et al. 2012).

O. limosus juveniles grow and reach sexual maturity faster than *A. leptodactylus* juveniles (Buřič et al. 2013). This fact, combined with the increased fecundity in the active front (Pârvulescu et al. 2015) and dominance of *O. limosus* suggests an improvement of the species' survival chances. Our study revealed *A. leptodactylus* displaying a more pronounced shelter sharing behaviour with its own congeners, than *O. limosus*. Shelter importance in *O. limosus* invasion was highlighted in the paper of Hirsch et al. (2016), as they may even adapt to non-specific shelter types, depending on the territory they occupy. Shelter occupancy by several individuals of the same species is possible, whereas interspecific cohabitation is rarely encountered. This species has also been found to display a "homing behaviour", which urges them to use one single shelter instead of wandering between multiple ones (Buřič et al. 2009). Besides of other multiple protective measures of sensitive crayfish species populations against invasive crayfish species, the artificial supply of refuges might reduce the decline, as this method has been successfully used before for other species as well (Walles et al. 2016; Bouckaert et al. 2014).

Acknowledgements

This work was supported by a grant of the Romanian National Authority for Scientific Research and Innovation, CNCS – UEFISCDI, project number PN-II-RU-TE-2014-4-0785. The authors would like to thank Claudia Zaharia for the valuable advice and support in the writing of this manuscript and Paul Richard Nicolau for providing *A. leptodactylus* individuals involved in the experiment.

References

Ackefors H. 1996. The development of crayfish culture in Sweden during the last decade. Freshwater Crayfish **11 (1):** 627–654.

Aquiloni L., Gonçalves V., Inghilesi A.F. & Gherardi F. 2012. Who's what? Prompt recognition of social status in crayfish. Behav. Ecol. Sociobiol. **66 (5):** 785–790. DOI: 10.1007/s00265-012-1326-3

Barki A. & Karplus I. 2016. The behavioral mechanism of competition for food between tilapia (*Oreochromis hybrid*) and crayfish (*Cherax quadricarinatus*). Aquaculture **450:** 162–167. DOI: 10.1016/j.aquaculture.2015.07.031

Bergman D.A. & Moore P.A. 2003. Field observations of intraspecific agonistic behavior of two crayfish species, *Orconectes rusticus* and *Orconectes virilis*, in different habitats. Biol. Bull. **205 (1):** 26–35. DOI: 10.2307/1543442

Bouckaert E.K., Auer N.A., Roseman E.F. & Boase J. 2014. Verifying success of artificial spawning reefs in the St. Clair–Detroit River System for lake sturgeon (*Acipenser fulvescens* Rafinesque, 1817). J. Appl. Ichthyol. **30:** 1393–1401. DOI: 10.1111/jai.12603

Broennimann O., Treier U.A., Müller-Schärer H., Thuiller W., Peterson A.T. & Guisan A. 2007. Evidence of climatic niche shift during biological invasion. Ecol. Lett. **10:** 701–709. DOI: 10.1111/j.1461-0248.2007.01060.x

Buřič M., Kouba A. & Kozák P. 2013. Reproductive plasticity in freshwater invader: from long-term sperm storage to parthenogenesis. PLoS One **8 (10):** e77597. DOI: 10.1371/journal.pone.0077597

Buřič M., Kozák P. & Kouba A. 2009. Movement patterns and ranging behavior of the invasive spiny-cheek crayfish in a small reservoir tributary. Fundam. Appl. Limnol. **174 (4):** 329–337. DOI: 10.1127/1863-9135/2009/0174-0329

Christy J.H. 1987. Competitive mating, mate choice and mating associations of brachyuran crabs. Bull. Mar. Sci. **41 (2):** 177–191.

Chuang A. & Peterson C.R. 2016. Expanding population edges: theories, traits, and trade-offs. Glob. Change Biol. **22 (2):** 494–512. DOI: 10.1111/gcb.13107

Chucholl C. 2013. Invaders for sale: trade and determinants of introduction of ornamental freshwater crayfish. Biol. Invasions **15 (1):** 125–141. DOI: 10.1007/s10530-012-0273-2

Chucholl C. 2016. The bad and the super-bad: prioritising the threat of six invasive alien to three imperilled native crayfishes. Biol. Invasions **18 (7):** 1967–1988. DOI: 10.1007/s10530-016-1141-2

Figler M.H., Blank G.S. & Peeke H.V. 1997. Maternal aggression and post-hatch care in red swamp crayfish, *Procambarus clarkii* (Girard): The influences of presence of offspring, fostering, and maternal molting. Mar. Freshwater Behav. Physiol. **30 (3):** 173–194. DOI: 10.1080/10236249709379023

Garvey J.E., Stein R.A. & Thomas H.M. 1994. Assessing how fish predation and interspecific prey competition influence a crayfish assemblage. Ecology **75 (2):** 532–547. DOI: 10.2307/1939556

Gherardi F. 2006. Crayfish invading Europe: the case study of *Procambarus clarkii.* Mar. Freshwater Behav. Physiol. **39 (3):** 175–191. DOI: 10.1080/10236240600869702

Grow L. & Merchant H. 1980. The burrow habitat of the crayfish, *Cambarus diogenes diogenes* (Girard). Am. Midl. Nat. **103 (2):** 231–237. DOI: 10.2307/2424621

Groza M.I., Pop-Vancia V. & Mireţan V. 2016. Diel activity and use of multiple artificially constructed shelters in *Astacus leptodactylus* (Decapoda: Astacidae). Biologia **71 (12):** 1369–1379. DOI: 10.1515/biolog-2016-0167

Guan R.Z. 1994. Burrowing behaviour of signal crayfish, *Pacifastacus leniusculus* (Dana), in the River Great Ouse, England. Freshwater Forum **4 (3):** 155–168.

Hirsch P.E., Burkhardt-Holm P., Töpfer I. & Fischer P. 2016. Movement patterns and shelter choice of spiny-cheek crayfish (*Orconectes limosus*) in a large lake's littoral zone. Aquat. Invasions **11 (1):** 55–65. DOI: 10.3391/ai.2016.11.1.06

Hudina S., Galić N., Roessink I. & Hock K. 2011. Competitive interactions between co-occurring invaders: identifying asymmetries between two invasive crayfish species. Biol. Invasions **13 (8):** 1791–1803. DOI: 10.1007/s10530-010-9933-2

Hudina S., Žganec K. & Hock K. 2015. Differences in aggressive behaviour along the expanding range of an invasive crayfish: an important component of invasion dynamics. Biol. Invasions **17 (11):** 3101–3112. DOI: 10.1007/s10530-015-0936-x

Ilhéu M., Acquistapace P., Benvenuto C. & Gherardi F. 2003. Shelter use of the Red-Swamp Crayfish (*Procambarus clarkii*) in dry-season stream pools. Arch. Hydrobiol. **157 (4):** 535–546. DOI: 10.1127/0003-9136/2003/0157-0535

Imhoff E.M., Mortimer R.J.G., Christmas M. & Dunn A.M. 2011. Invasion progress of the signal crayfish *Pacifastacus leniusculus* (Dana) and displacement of the native white-clawed crayfish *Austropotamobius pallipes* (Lereboullet) in the River Wharfe, UK. Freshwater Crayfish **18:** 45–53. DOI: 10.5869/fc.2011.v18.45

Kaczer L., Pedetta S. & Maldonado H. 2007. Aggressiveness and memory: subordinate crabs present higher memory ability than dominants after an agonistic experience. Neurobiol. Learn. Mem. **87 (1):** 140–148. DOI: 10.1016/j.nlm.2006.08.002

Kawai N., Kono R. & Sugimoto S. 2004. Avoidance learning in the crayfish (*Procambarus clarkii*) depends on the predatory imminence of the unconditioned stimulus: a behavior systems approach to learning in invertebrates. Behav. Brain Res. **150 (1-2):** 229–237. DOI: 10.1016/S0166-4328(03)00261-4

Klocker C.A. & Strayer D.L. 2004. Interactions among an invasive crayfish (*Orconectes rusticus*), a native crayfish (*Orconectes limosus*), and native bivalves (Sphaeriidae and Unionidae). Northeast. Nat. **11 (2)**: 167–178. DOI: 10.1656/1092-6194(2004)011[0167:IAAICO]2.0.CO;2

Kouba A., Petrusek A. & Kozák P. 2014. Continental-wide distribution of crayfish species in Europe: update and maps. Knowl. Managt. Aquatic Ecosyst. **413:** article number 05, 21 pp. DOI: 10.1051/kmae/2014007

Lee C.E. 2002. Evolutionary genetics of invasive species. Trends Ecol. Evol. **17 (8):** 386–391. DOI: 10.1016/S0169-5347(02)02554-5

Leon M., Merner M.J., Dreyer A.A., Cooper A., Scott L., Berendzen P.B., McCullough D.A. & Merten E.C. 2016. Range expansion of the invasive rusty crayfish *Orconectes rusticus* (Girard, 1852) (Decapoda: Astacoidea) in northeastern Iowa (USA) rivers. J. Crust. Biol. **36 (1):** 99–104. DOI: 10.1163/1937240X-00002397

Lodge D.M., Deines A., Gherardi F., Yeo D.C., Arcella T., Baldridge A.K., Barnes M.A., Chadderton W.L., Feder J.L., Gantz C.A. & Howard G.W. 2012. Global introductions of crayfishes: Evaluating the impact of species invasions on ecosystem services. Annu. Rev. Ecol. Evol. Syst. **43:** 449–472. DOI: 10.1146/annurev-ecolsys-111511-103919

Mathers K.L., Chadd R.P., Dunbar M.J., Extence C.A., Reeds J., Rice S.P. & Wood P.J. 2016. The long-term effects of invasive signal crayfish (*Pacifastacus leniusculus*) on instream macroinvertebrate communities. Sci. Total Environ. **556:** 207–218. DOI: 10.1016/j.scitotenv.2016.01.215

Matsuzaki S.I.S., Sakamoto M., Kawabe K. & Takamura N. 2012. A laboratory study of the effects of shelter availability and invasive crayfish on the growth of native stream fish. Freshwater Biol. **57 (4):** 874–882. DOI: 10.1111/j.1365-2427.2012.02743.x

Meade M.E. & Watts S.A. 1995. Weight gain and survival of juvenile Australian crayfish *Cherax quadricarinatus* fed formulated feeds. J. World Aquacult. Soc. **26 (4):** 469–474. DOI: 10.1111/j.1749-7345.1995.tb00845.x

Momot W.T. 1995. Redefining the role of crayfish in aquatic ecosystems. Rev. Fish. Sci. **3 (1):** 33–63. DOI: 10.1080/10641269509388566

Moore P.A. & Bergman D.A. 2005. The smell of success and failure: the role of intrinsic and extrinsic chemical signals on the social behavior of crayfish. Integr. Comp. Biol. **45(4):** 650–657. DOI: 10.1093/icb/45.4.650.

Musil M., Buřič M., Policar T., Kouba A. & Kozák P. 2010. Comparison of diurnal and nocturnal activity between noble crayfish (*Astacus astacus*) and spinycheek crayfish (*Orconectes limosus*). Freshwater Crayfish **17**: 189–193.

Oidtmann B., Heitz E., Rogers D. & Hoffmann R.W. 2002. Transmission of crayfish plague. Dis. Aquat. Org. **52:** 159–167. DOI: 10.3354/dao052159

Pârvulescu L., Paloş C. & Molnar P. 2009. First record of the spiny-cheek crayfish *Orconectes limosus* (Rafinesque, 1817) (Crustacea: Decapoda: Cambaridae) in Romania. North-West. J. Zool. **5 (2):** 424–428.

Pârvulescu L., Pîrvu M., Moroşan L.G. & Zaharia C. 2015. Plasticity in fecundity highlights the females' importance in the spiny-cheek crayfish invasion mechanism. Zoology **118 (6):** 424–432. DOI: 10.1016/j.zool.2015.08.003

Pârvulescu L., Schrimpf A., Kozubíková E., Cabanillas Resino S., Vrålstad T., Petrusek A. & Schulz R. 2012. Invasive crayfish and crayfish plague on the move: first detection of the plague agent *Aphanomyces astaci* in the Romanian Danube. Dis. Aquat. Org. **98 (1):** 85–94. DOI: 10.3354/dao02432.

Preisser E.L., Bolnick D.I. & Benard M.F. 2005. Scared to death? The effects of intimidation and consumption in predator-prey interactions. Ecology **86 (2):** 501–509. DOI: 10.1890/04-0719

Puky M. & Schád P. 2006. *Orconectes limosus* colonises new areas fast along the Danube in Hungary. Bull. Fr. Pêche Piscic. **380-381:** 919–926. DOI: 10.1051/kmae:2006031

R Core Team. 2016. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL http://www.R-project.org.

Rahel F.J. & Stein R.A. 1988. Complex predator-prey interactions and predator intimidation among crayfish, piscivorous fish, and small benthic fish. Oecologia **75 (1):** 94–98. DOI: 10.1007/BF00378819

Rebrina F., Skejo J., Lucic A. & Hudina S. 2015. Trait variability of the signal crayfish (*Pacifastacus leniusculus*) in a recently invaded region reflects potential benefits and trade-offs during dispersal. Aquat. Invasions **10 (1):** 41–50. DOI: 10.3391/ai.2015.10.1.04

Schrimpf A., Pârvulescu L., Copilaş-Ciocianu D., Petrusek A. & Schulz R. 2012. Crayfish plague pathogen detected in the Danube Delta – a potential threat to freshwater biodiversity in southeastern Europe. Aquat. Invasions **7 (4):** 503–510. DOI: 10.3391/ai.2012.7.4.007

Skurdal J. & Taugbøl T. 2002. Crayfish of commercial importance: *Astacus,* Part II, pp. 467–510. In: Holdich D.M. (ed.), Biology of Freshwater Crayfish, Blackwell Science, Oxford and Malden, Massachusetts, 702 pp. ISBN: 1405123494, 9781405123495

Snedden W.A. 1990. Determinants of male mating success in the temperate crayfish *Orconectes rusticus*: chela size and sperm competition. Behaviour **115 (1):** 100–113. DOI: 10.1163/156853990X00301

Soderback B. 1994. Interactions among juveniles of two freshwater crayfish species and a predatory fish. Oecologia **100 (3):** 229–235. DOI: 10.1007/BF00316949.

Stebbing P.D., Bentley M.G. & Watson G.J. 2003. Mating behaviour and evidence for a female released courtship pheromone in the signal crayfish *Pacifastacus leniusculus.* J. Chem. Ecol. **29 (2):** 465–475. DOI: 10.1023/A:1022646414938

Stocker A.M. & Huber R. 2001. Fighting strategies in crayfish *Orconectes rusticus* (Decapoda, Cambaridae) differ with hunger state and the presence of food cues. Ethology **107 (8):** 727–736. DOI: 10.1046/j.1439-0310.2001.00705.x

Václavík T. & Meentemeyer R.K. 2012. Equilibrium or not? Modelling potential distribution of invasive species in different stages of invasion. Divers. Distrib. **18 (1):** 73–83. DOI: 10.1111/j.1472-4642.2011.00854.x

Vorburger C. & Ribi G. 1999. Aggression and competition for shelter between a native and an introduced crayfish in Europe. Freshwater Biol. **42 (1):** 111–119. DOI: 10.1046/j.1365-2427.1999.00465.x

Walles B., Troost K., van den Ende D., Nieuwhof S., Smaal A.C. & Ysebaert T. 2016. From artificial structures to self-sustaining oyster reefs. J. Sea Res. **108:** 1–9. DOI: 10.1016/j.seares.2015.11.007

Received January 27, 2017
Accepted June 16, 2017