

## Competition for shelter sites: Testing a possible mechanism for gammarid species displacements



S. de Gelder<sup>a,b</sup>, G. van der Velde<sup>a,c,f,\*</sup>, D. Platvoet<sup>d</sup>, N. Leung<sup>a</sup>,  
M. Dorenbosch<sup>e</sup>, H.W.M. Hendriks<sup>a</sup>, R.S.E.W. Leuven<sup>a,f</sup>

<sup>a</sup>Radboud University, Institute for Water and Wetland Research, Heyendaalseweg 135, 6525 AJ Nijmegen, The Netherlands

<sup>b</sup>National Institute of Nutrition and Seafood Research, Bergen, Norway

<sup>c</sup>Naturalis Biodiversity Center, Leiden, The Netherlands

<sup>d</sup>Office Plancius, Bathmen, The Netherlands

<sup>e</sup>Bureau Waardenburg BV, Culemborg, The Netherlands

<sup>f</sup>Netherlands Centre of Expertise for Exotic Species (NEC-E), Nijmegen, The Netherlands

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### Abstract

Invasions of alien gammarid species have led to reduced abundance of many native gammarid species and earlier gammarid invaders. Intra-guild predation (IGP) has been suggested as the main mechanism causing such species displacements. This study elucidates a mechanism for species displacement that is based on competition for shelter, viz. species excluding each other from a shelter place. Preferences of *Dikerogammarus villosus* and *Gammarus roeselii* for shelter space were studied in laboratory experiments. In contrast with night conditions both species showed a strong preference for shelter sites under daytime conditions so that all experiments were carried out under permanent light conditions. Single individuals of *D. villosus* sheltered more than those of *G. roeselii*. Intraspecific competition experiments with different size classes and sexes showed that in contrast with *D. villosus*, the size class had a significant effect on the mean sheltering proportion of individuals of *G. roeselii*. When both species were brought together in a basin with a shelter site to test interspecific competition, individuals of *G. roeselii* were actively pushed out of their shelters within 24 h, while *D. villosus* showed no change in shelter. This led to a significantly increased predation risk for *G. roeselii*, which was confirmed in an experiment in which a benthic fish was added. IGP only manifested itself after 48 h, indicating that competition for shelter preceded IGP. When shelter opportunities are in short supply, shelter exclusion may be one of the initial mechanisms for gammarid species displacements.

### Zusammenfassung

Invasionen von neobiotischen Gammariden haben zu einer Reduzierung der Artenvielfalt von einheimischen und früher eingewanderten Gammariden geführt. Bisher wurde Gildeninterne Prädation (intra-guild predation, IGP) als Hauptursache für diese Artenverschiebung genannt. Diese Studie beschreibt einen anderen Mechanismus für Artenverschiebung, der auf Konkurrenz um Lebensräume basiert, wobei verschiedene Arten einander an der Nutzung von Unterschlüpfen hindern. In Laborexperimenten wurden die Präferenzen für Unterschlüpfen von *Dikerogammarus villosus* und *Gammarus roeselii* untersucht. Beide Arten bevorzugten Unterschlüpfen bei Tagesbedingungen, weswegen alle Experimente unter Dauerlichtverhältnissen durchgeführt

\*Corresponding author at: Radboud University, Institute for Water and Wetland Research, Heyendaalseweg 135, 6525 AJ Nijmegen, The Netherlands. Tel.: +31 243652471.

E-mail address: [g.vandervelde@science.ru.nl](mailto:g.vandervelde@science.ru.nl) (G. van der Velde).

wurden. Einzelne *D. villosus*-Individuen suchten mehr Schutz als *G. roeselii*-Individuen. Intraspezifische Konkurrenzexperimente mit verschiedenen Größenklassen und Geschlechtern zeigten, dass, im Gegensatz zu *D. villosus*, die Größenklasse einen signifikanten Einfluss auf den durchschnittlichen Anteil der *G. roeselii*-Individuen, die Schutz suchten, hatte. Um die Konkurrenz zwischen verschiedenen Arten zu testen wurden beide Arten zusammen in einem Becken gehalten. *G. roeselii*-Individuen wurden innerhalb von 24 Stunden aktiv aus ihren Unterschlüpfen verscheucht, während *D. villosus* keine Unterschiede zeigte. Dies führte zu einem signifikant erhöhten Prädationsrisiko für *G. roeselii*, welches in einem Experiment unter Anwesenheit einer benthische Fischart bestätigt wurde. Gildeninterne Prädation (IGP) trat erst nach 48 Stunden auf, was darauf hinweist, dass der Wettbewerb um Unterschlüpfte vor IGP stattfindet. Wenn nur wenige Unterschlüpfte vorhanden sind, kann die Verdrängung aus diesen Schutzorten einer der Mechanismen für Artenverschiebungen bei Gammariden sein.  
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## Introduction

Various gammarids are omnivorous invaders of freshwater ecosystems (MacNeil, Dick, & Elwood 1997; Felten, Tixier, Guerold, De Billy, & Dangles 2008; Platvoet, Van der Velde, Dick, & Li 2009b) impacting macro-invertebrate communities at high population density (Kelly, Bailey, MacNeil, Dick, & McDonald 2006; Van Riel et al. 2006; Noordhuis, Van Schie, & Jaarsma 2009). Invasions by alien gammarids lead to competition with native and earlier established alien species, which can result in habitat shifts, niche reduction or even exclusion and displacement of the competitively weaker species (Leuven et al. 2009; Platvoet, Dick, MacNeil, Van Riel, & Van der Velde, 2009a; Van der Velde et al. 2009; Van Riel, Van der Velde, & Bij de Vaate, 2009). Gammarid species can be arranged from highly to less predatory (Platvoet et al. 2009a; Stoffels et al. 2011; Bacela-Spychalska & Van der Velde, 2013; Dodd et al. 2014) with the Ponto-Caspian invader *Dikerogammarus villosus* being the most predaceous compared with native and earlier invasive gammarid species. This species is a threat to freshwater ecosystems owing to its predatory behaviour (Dick & Platvoet 2000; Van Riel et al. 2006; Platvoet et al. 2009b).

Based on laboratory experiments Intra-guild predation (IGP) is considered to be the main mechanism underlying gammarid species displacement processes (Dick, Montgomery, & Elwood 1993; Dick & Platvoet 1996, 2000; MacNeil, Bigsby, Dick, Hatcher, & Dunn 2003; Kinzler & Maier 2003; Dick 2008). To reduce predation risks, prey can try to escape from a predator and search for a shelter site. Shelter sites are a valuable resource and serve as protection, breeding or resting sites and in the case of predators also as a hiding place to ambush their prey. Availability of shelter sites has profound effects on IGP (Hacker & Steneck 1990; Garvey, Stein, & Thomas 1994; Kley & Maier 2005; MacNeil, Platvoet, & Dick 2008; Platvoet et al. 2009a). Furthermore, to avoid predation gammarids use shelter sites during the daytime and leave them at night, when the risk of predation by day active fish is reduced (Elliott, 2005; Lagrue et al. 2011; Van Riel, Van der Velde, & Bij de Vaate 2011). Devin, Piscart, Beisel, and Moreteau (2003) demonstrated

that shelters appear to be size-dependent for *D. villosus* as a relationship exists between substratum particle size and the length of individuals and a partial segregation was observed according to length of the gammarids. *D. villosus* dominates the stone environment of regulated rivers, lakes and canals for shipping and avoids sand and leaf litter while *Gammarus roeselii* hardly differentiates between substrates. In the presence of *D. villosus*, *G. roeselii* coexisted by occurring in submerged vegetation in a Rhine tributary (Kley & Maier 2005) and was displaced on hard substrates in Lake Constance (Hesselschwerdt, Necker, & Wantzen 2008). The preference for and domination of *D. villosus* in the stony littoral resulted in displacement of other gammarid species that needed to find alternative shelters such as sand, clay, leaf litter or vegetation when present. In this way a zonation of gammarid species can develop by which the species coexist in the same waterbody through habitat segregation (Hesselschwerdt et al. 2008; Platvoet et al. 2009a).

We studied the sheltering of *D. villosus* and *G. roeselii* per species and in interspecific competition experiments to find out if there are shelter differences and/or competition for shelter during the daytime, which may lead to replacement of one species by the other. The hypothesis was tested by a series of laboratory experiments with both species to find out, (a) if single individuals of these species show shelter behaviour when a shelter site is offered without interaction with other individuals, (b) if different sexes and size classes of each species show differences in sheltering behaviour when a number of individuals are introduced together in a microcosm (possible interactions between size classes and sexes), (c) if there is intra- or interspecific competition for shelter when the numbers introduced are varied, (d) if there is competitive exclusion in the presence or absence of a fish predator.

## Materials and methods

### Animal collection and handling

*G. roeselii*, was collected during the period May–September in 2008 in the Schipbeek brook

(52°15'13.55"N, 6°20'9.23"E), while *D. villosus* was collected in the same period from Lake Gouwzee (52°26'20.64"N, 5°3'4.93"E), from the Pannerdensch canal (51°53'33.23"N, 6°1'24.75"E) and the Waal river (51°52'56.49"N, 6°0'7.80"E), all in the Netherlands. To prevent adaptation to earlier encounters of these species both species were sampled from sample sites where they do not coexist. Both species were kept separately at natural densities of 200–600 individuals m<sup>-2</sup> (Kley 2001) in shallow aerated basins (35.5 × 30.0 × 6.5 cm) with constant light at 20 °C (the conditions of all experiments). These basins contained water, plants and organic matter from the sampling sites. The complete stock was replenished after three days by repeated field sampling. During the experiments the animals were not fed. No gravid females were used in the experiments.

## Experimental set-up

### Sheltering behaviour of single individuals

The first experiment tested the animals' sheltering behaviour. Single males and females of *D. villosus* and single males and females of *G. roeselii* were picked at random from the stock and placed in a translucent plastic basin (29.0 × 16.0 × 2.0 cm), which was filled with a layer of water (1.25 cm) from the sampling sites. In these plastic basins a glass strip (28.0 × 2.0 × 0.6 cm) was placed that leaned with one side on a small support (0.45 cm), to create space underneath the glass strip where gammarids could seek shelter (Fig. 1A and B). The advantage of using glass strips as shelter is that the position of the animals and their behaviour can be observed. Before each experiment, the sex of gammarids was determined from the size of the gnathopods for both species and on setation of the 1st antenna for *D. villosus*. Table 1 shows the lengths of individuals used in the single shelter experiment of *D. villosus* and *G. roeselii*. After 24 h, it was observed whether the gammarid sheltered underneath

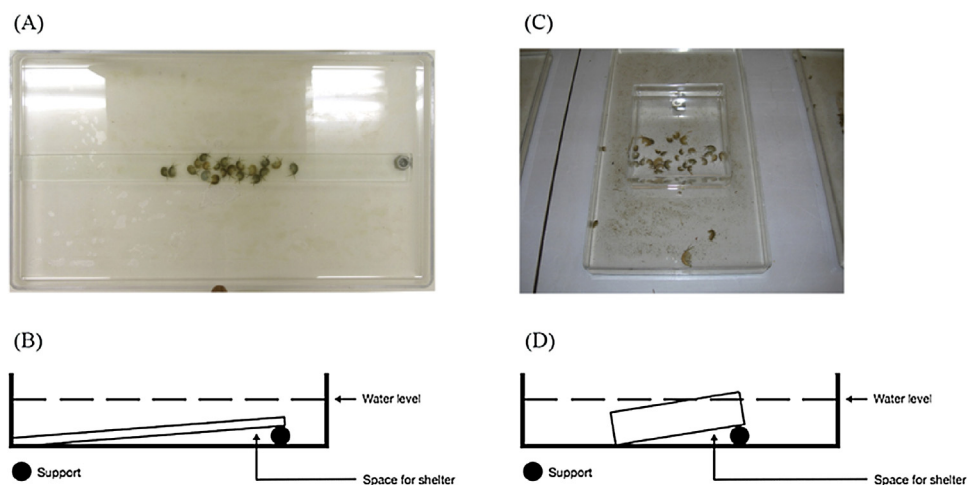
the glass strip. This was repeated for 30 single males and 30 single females of each species.

### Intraspecific sheltering behaviour of different sexes and size classes

The second experiment investigated intraspecific competition for shelter between sex and size classes of the same species. For each test, twenty individuals (per species) were placed in an experimental set-up (similar as the one used for the single individual experiments) for 24 h. Each experiment was carried out in twofold and repeated six times (using new animals for each test). Individuals were visually sorted into the following categories: (1) large males, (2) large females, (3) small males and (4) small females. Subsequently, ten small and ten large males or ten small and ten large females of the same species were selected and mixed, which gave two additional categories: (5) large males plus small males and (6) large females plus small females (Table 1). The results of these competition experiments between gammarids of different size classes and sexes were compared with results of similar experiments using only one size class.

### Intra- and interspecific competition for shelter

Intra- and interspecific competition for shelter between the two species was assessed in a set-up with varying numbers of *D. villosus* and *G. roeselii*. This experiment used the same set-up as described for the competitive exclusion experiment (see next sub-section), to mimic competition for shelter without a predator. Of each species 20 or 40 individuals were picked at random from the stock to study whether intraspecific competition was influenced by population density. Interspecific competition was assessed by adding 20 individuals of *D. villosus* and 20 of *G. roeselii* together in one experimental basin. Each experiment was carried out in twofold and lasted 24 h. The experiment was repeated six times.



**Fig. 1.** Experimental set-up with the glass strip under which the gammarids sheltered (A). Side-view of the experimental set-up with the glass strip (B). Design with the translucent plastic boxes under which the gammarids sheltered (C). Side-view of the set-up of the experiment with the translucent plastic boxes (D). The black dot indicates the position of the support (0.45 cm) of the glass strip (B) and plastic box (D).

**Table 1.** Lengths of *Dikerogammarus villosus* and *Gammarus roeselii* individuals (in mm) in shelter behaviour experiments (SD: standard deviation).

|                            | <i>D. villosus</i> |           | <i>G. roeselii</i> |           |
|----------------------------|--------------------|-----------|--------------------|-----------|
|                            | Mean $\pm$ SD      | Range     | Mean $\pm$ SD      | Range     |
| Single specimens           |                    |           |                    |           |
| Males                      | 16.3 $\pm$ 3.1     | 10.6–22.3 | 15.2 $\pm$ 2.3     | 8.4–18.3  |
| Females                    | 14.6 $\pm$ 2.4     | 10.1–18.8 | 13.5 $\pm$ 1.9     | 9.4–17.4  |
| Intraspecific size classes |                    |           |                    |           |
| Large males                | 17.2 $\pm$ 3.1     | 12.8–27.0 | 15.9 $\pm$ 1.4     | 12.3–20.4 |
| Large females              | 15.3 $\pm$ 1.7     | 11.9–23.6 | 13.2 $\pm$ 1.4     | 8.3–18.4  |
| Small males                | 12.0 $\pm$ 1.2     | 7.4–15.0  | 10.8 $\pm$ 1.5     | 6.1–15.5  |
| Small females              | 10.6 $\pm$ 1.2     | 6.2–13.5  | 10.0 $\pm$ 1.3     | 5.8–12.7  |

### Competitive exclusion with and without top predator pressure

In another series of experiments, competitive exclusion of gammarids with and without top predator pressure was assessed. Because the fish predator could easily reach sheltering gammarids under the glass strip and disturb the set-up we used translucent plastic basins, viz. a plastic box (10.5  $\times$  9.5  $\times$  3.0 cm), under which the gammarids could safely shelter (also used in the intra- and interspecific competition experiment). The box was filled with water to prevent it from floating, and was raised by 0.45 cm at one end, so the animals had the opportunity to shelter under it (Fig. 1C and D). Of *D. villosus* and *G. roeselii* 20 individuals of each species were picked at random from the stock and placed together in one experimental basin. Both species were tested for competitive exclusion in the absence and presence of a fish predator (one individual of the Stone loach, *Barbatula barbatula*). Each experiment lasted 48 h and was repeated nine times.

### Statistical analysis

All statistical analyses were performed in R version 2.15.0 (R Development Core Team) (Crawley, 2007). The effects of sex, size class, species and intra- or interspecific competition on sheltering was compared by fitting linear mixed-effect models with a log-link function based on either binomial or normal distributions.

Sheltering behaviour in the single gammarid experiments had a binomial distribution. Presence or absence of a gammarid in the shelter site was set as dependent variable, whereas sex, species, and their interaction were set as fixed factors. The effect of the experimental set-up in which each replicate experiment was conducted was included as a random factor. In the subsequent experiments, sheltering behaviour showed a normal distribution. For the intraspecific experiment, the proportion of individuals that sheltered was set as dependent variable, whereas competition condition (i.e., competition between size classes versus no competition between size classes), size class (large and small; males

and females) and their interaction were set as fixed factors. The effect of the experimental unit and the day on which the replicate tests were conducted (date) were included as random factors. For the interspecific experiment, the dependent variable and random effects were similar to those in the above experiment. Species and condition (i.e., competition based on 40 individuals, no competition based on 20 and 40 individuals) were set as fixed factors.

To test the effect of a fish predator and the presence and absence of a shelter site, the proportion of surviving individuals was set as the dependent variable, whereas species, presence of a fish predator and their interaction were set as fixed factors. Since the experiments were conducted on the same day, only the effect of the experimental unit was included as a random factor.

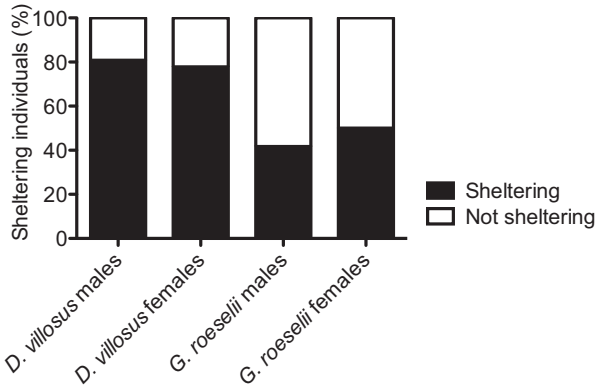
Assumptions of normality were checked by residual analysis and by a Shapiro test of normality. Where necessary, data was log-transformed by  $y = \log^{10}(x + 1)$ . Models were fitted with the lmer function in the lme4 package. To determine the effects of fixed factors, a likelihood ratio test was used to compare models with and without the variable of interest (Crawley, 2007). For variables with three or more levels, significant differences were analysed by Tukey post-hoc comparisons, available in the multcomp package.

## Results

### Sheltering behaviour of single individuals

Single individuals of both species sheltered under the glass strip after 24 h (Fig. 2). *D. villosus* sheltered significantly more than *G. roeselii* ( $\chi^2 = 15.08$ ;  $df = 2$ ;  $P = 0.001$ ). Single males as well as single females of *D. villosus* were more likely to shelter than those of *G. roeselii*, viz. 80.8% of *D. villosus* males sheltered and 41.7% of *G. roeselii* males. The sheltering percentage for females of *D. villosus* and *G. roeselii* were 75.0% and 50.0%, respectively. However, the effect of sex was not significant ( $\chi^2 = 0.37$ ;  $df = 2$ ;  $P = 0.832$ ) and no statistical interaction between sex and species ( $\chi^2 = 0.33$ ;  $df = 1$ ;  $P = 0.564$ ) was detected. The average shelter





**Fig. 2.** Mean shelter percentages of single male and female individuals of *Dikerogammarus villosus* and *Gammarus roeselii*, after 24 h.

percentage of single individuals per species was 80.0% for *D. villosus* and 46.0% for *G. roeselii*.

### Intraspecific sheltering behaviour of different sexes and size classes

The shelter behaviour of both species for both sexes and four size classes (large males, large females, small males and small females) showed that after 24 h neither the size class of *D. villosus* ( $\chi^2 = 2.87$ ;  $df = 3$ ;  $P = 0.412$ ), nor the competition condition ( $\chi^2 = 2.51$ ;  $df = 1$ ;  $P = 0.113$ ) nor their statistical interaction ( $\chi^2 = 0.75$ ;  $df = 3$ ;  $P = 0.861$ ) had a significant effect on the mean proportion of individuals taking shelter (Fig. 3A). Although the effect of the competition condition ( $\chi^2 = 1.51$ ;  $df = 1$ ;  $P = 0.219$ ) and the statistical interaction ( $\chi^2 = 5.81$ ;  $df = 3$ ;  $P = 0.121$ ) were not significant for *G. roeselii*, the size classes of smaller individuals

sheltered significantly more ( $\chi^2 = 39.20$ ;  $df = 3$ ;  $P < 0.001$ ) than larger size classes (Fig. 3B).

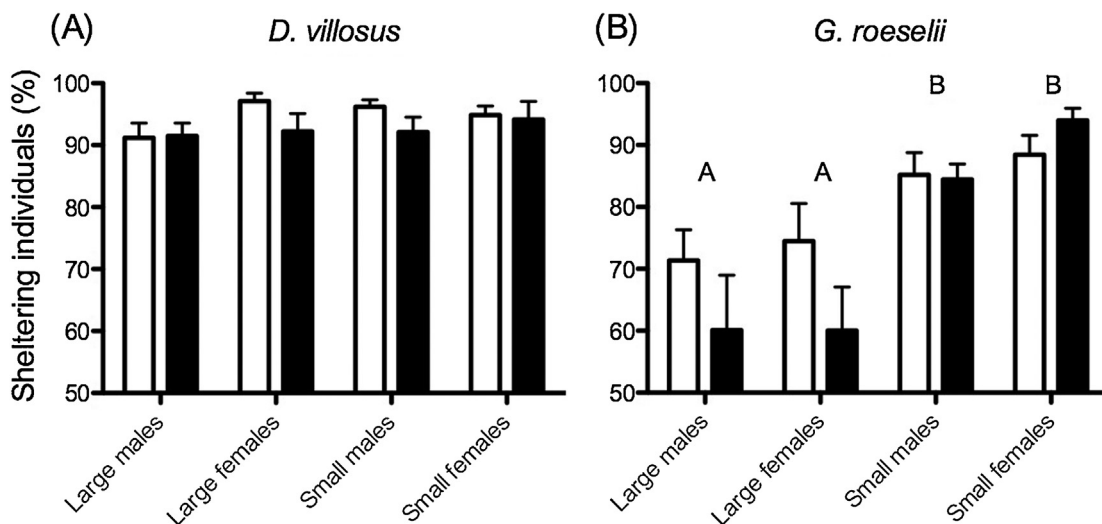
### Intra- and interspecific competition for shelter

Both species showed significant different sheltering behaviour in the intra- and interspecific competition experiments ( $\chi^2 = 114.86$ ;  $df = 1$ ;  $P < 0.001$ ; Fig. 4). Intraspecific competition led to significantly more sheltering individuals of *D. villosus* when 40 individuals were present instead of 20 individuals (*Post-hoc* analysis:  $P = 0.01$ ). A significantly lower number of *G. roeselii* sheltered in the experiment with 40 individuals in comparison with 20 individuals ( $P = 0.042$ ).

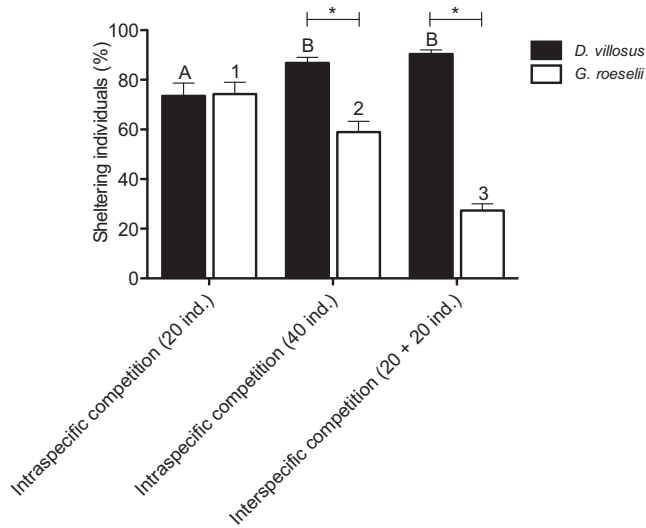
A significant interaction was observed in sheltering individuals for both species by intra- and interspecific competition ( $\chi^2 = 79.76$ ;  $df = 2$ ;  $P < 0.001$ ). In the condition with 20 individuals of a single species, neither of the two species appeared to show differences in sheltering (i.e., the intraspecific competition 20 individuals). However, in the condition with 40 individuals, there was a significant difference between the mean sheltering of the two species ( $\chi^2 = 20.94$ ;  $df = 1$ ;  $P < 0.001$ , the intraspecific competition 40 individuals). When both species were placed together, the presence of *D. villosus* significantly decreased sheltering individuals of *G. roeselii* ( $\chi^2 = 79.44$ ;  $df = 1$ ;  $P < 0.001$ ) while sheltering individuals of *D. villosus* were not affected by the presence of *G. roeselii*.

### Competitive exclusion with and without top predator pressure

The average survival percentage of *D. villosus* with a shelter site was unaffected by the presence or absence of the



**Fig. 3.** Mean shelter rates ( $\pm$ SEM) of 20 individuals of *Dikerogammarus villosus* (A) and *Gammarus roeselii* (B) for intraspecific competition with separately tested size classes of large males, large females, small males and small females (white bars). In the mixed groups, ten small and ten large males or ten small and ten large females of the same species were placed together (black bars). Different letters above the columns indicate significant differences in mean group percentages of size classes that sheltered under the glass strip ( $P < 0.001$ ).



**Fig. 4.** Mean shelter rates ( $\pm$ SEM) of *Dikerogammarus villosus* (black bars) and *Gammarus roeselii* (white bars) that sheltered during intra- and interspecific competition. Three different conditions were tested with: (1) a group of 20 individuals of each species separately; (2) a group with 40 individuals of each species separately and (3) a group of 20 individuals of *D. villosus* and 20 individuals of *G. roeselii* placed together. An asterisk above the columns shows significant differences in average shelter rates between the two species (\*  $P < 0.001$ ). Different letters above the columns indicate significant differences in average percentages of individuals that sheltered ( $P < 0.05$ ).

fish predator ( $\chi^2 = 0.002$ ,  $df = 1$ ,  $P = 0.969$ ; Fig. 5A), whereas only half of the individuals of *G. roeselii* survived in the presence of the fish predator ( $\chi^2 = 10.27$ ,  $df = 1$ ,  $P = 0.001$ ).

In the presence of a shelter site, the presence of a fish predator interacted significantly ( $\chi^2 = 22.59$ ;  $df = 1$ ;  $P < 0.001$ ), which resulted in an even stronger reduction of surviving individuals of *G. roeselii*.

When there was no shelter site, the survival of *G. roeselii* was more affected by the absence of shelter than that of

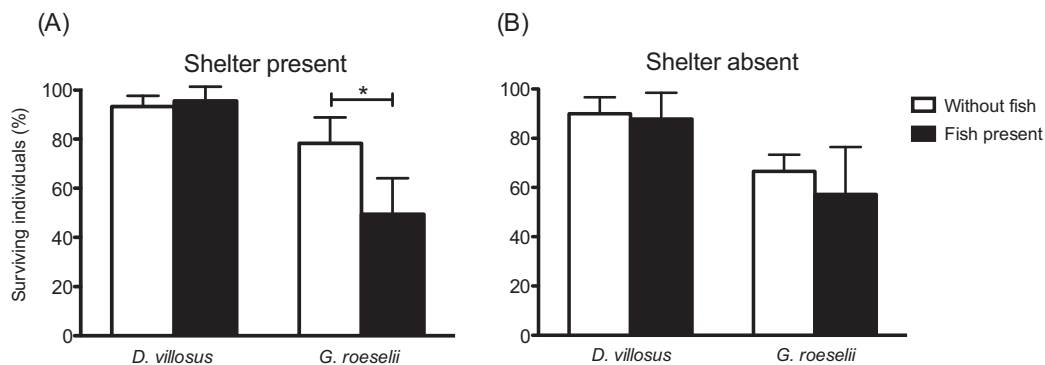
*D. villosus* ( $\chi^2 = 31.35$ ;  $df = 1$ ;  $P < 0.001$ ; Fig. 5B). Both species showed no significant interactions in the presence of the fish ( $\chi^2 = 0.89$ ;  $df = 1$ ;  $P = 0.345$ ); likewise, the presence of the fish had no effect on survival ( $\chi^2 = 2.18$ ;  $df = 1$ ;  $P = 0.140$ ). In the absence of the fish, *G. roeselii* showed a significant decrease in mean survival without a shelter site ( $\chi^2 = 6.73$ ;  $df = 1$ ;  $P = 0.009$ ).

## Discussion

The experimental shelter sites were often used by both gammarid species under continuous light conditions. *D. villosus* was the stronger competitor for shelter sites, which eventually resulted in an increased mortality of *G. roeselii* in the presence of a predatory fish.

Single individuals of *D. villosus* sheltered more than single individuals of *G. roeselii*. These results match those of the intraspecific experiment, where individuals of *D. villosus* also sheltered more than large males and large females of *G. roeselii*. It seems that *D. villosus* is not affected by intraspecific competition, in contrast to *G. roeselii*, where small individuals showed higher sheltering rates than large individuals in intraspecific competition. Since smaller individuals are more vulnerable to cannibalism and predation by (larger) predators, they hide more likely in shelters (Platvoet et al. 2009a). The small males and small females of *G. roeselii* sheltered significantly more than large males and large females in the separate as well as the mixed size classes. The sheltering individuals in our single individual experiments were not subject to competition by other individuals or species. Probably because shelter sites are only efficiently used when there is an increased predation risk (Magalhaes, Janssen, Hanna, & Sabelis, 2002), fewer single individuals of *G. roeselii* sheltered in comparison with sheltering individuals of *G. roeselii* in the intraspecific experiment.

When both species were placed together (interspecific competition) into the same set-up, *G. roeselii* sheltered



**Fig. 5.** Effect of shelter exclusion on mean survival percentages ( $\pm$ SEM) of *Dikerogammarus villosus* and *Gammarus roeselii* in the presence or absence of a predatory fish (one individual of Stone loach, *Barbatula barbatula*). Twenty individuals of each species were added together to the experimental basin where either a shelter site (Fig. 1C and D) was present (A) or no shelter was present (B). The survival percentages are shown in the absence (white bars) and presence (black bars) of the fish predator. An asterisk above the columns shows significant differences in average survival percentages between the same classes of both species (\*  $P < 0.001$ ).

considerably less in the presence of *D. villosus*, than single sheltering individuals of *G. roeselii* or with interspecific competition. We observed *D. villosus* actively pushing *G. roeselii* out of the shelter, which illustrates that shelter sites are resources for interference competition. That competition for shelter sites had a high effect on survival was furthermore observed when a fish predator was added to the two gammarids. The highest mortality of *G. roeselii* was recorded in the presence of the fish predator, *D. villosus* and a shelter site. The increased predation and mortality of *G. roeselii* is most likely related to the exclusion from the shelter site by *D. villosus* and the, overall, lower mean sheltering of *G. roeselii* which both increased predation risk. Furthermore, densities of *G. roeselii* decreased in the basins where no fish predator was present, which illustrates that IGP became relevant when the experiment was extended to 48 h (Van Riel, Healy, Van der Velde, & Bij de Vaate, 2007). *D. villosus* is very voracious and could prey on *G. roeselii* and other small individuals especially when food was not supplemented ad libitum or alternative shelter possibilities were available. Therefore, we could assume there is not only predation by fish but also cannibalism and predation by *D. villosus* as superior predator.

Multiple predators can suppress prey density when they are present at the same time, whereas prey release can occur at higher prey densities in the presence of both predators. When both species met in the same basin without a shelter site, IGP had the same effect as the presence of multiple predators. This was most likely the effect of the shelter site, since the survival rate of *G. roeselii* was highest in the presence of a shelter site when no predatory fish was present. When a shelter site and a fish were present, *D. villosus* suffered less mortality than *G. roeselii*, which is probably due to the greater competitiveness for shelter of *D. villosus*. Since *G. roeselii* was driven from the shelter site by *D. villosus*, it was more likely to encounter the fish predator. Thus, coexistence of species can be achieved by differential use of suitable habitat or shelter sites, as this reduces the risk to encounter a predator and increase their chances of survival (Platvoet et al. 2009a).

This study has provided insight into one of the ecological mechanisms involved in species displacement by non-native gammarids. Although, more sophisticated experimental setups could be used following the behaviour of the individuals and their interactions by camera and video techniques to monitor movements, hiding and displacement in the future. By using a gradient of differently spaced shelters all specimens irrespective of their size could find space for sheltering which was in reality also observed for both species.

If species rely on similar shelter sites, they will compete for this resource. When shelter opportunities are in short supply, competition will lead to increased predation risk for the weaker competitor that is excluded from its shelter. The role of the IGP mechanism is reduced when there is a diversity of physical shelters that can sustain various species. This emphasizes the importance of shelter sites in determining species displacements, irrespective of IGP. Although the importance of both mechanisms may depend on local

environmental conditions (e.g., differences in shelter availability, types of shelter, species interactions and disturbance), shelter exclusion is very likely to be an initial mechanism in species displacements. Our findings may explain species displacement processes in the field, particularly in situations where there is not enough variation in shelter opportunities. This is the case in many regulated rivers, shipping canals and man-made lakes where artificial hard substrates such as riprap were added to prevent bank erosion by current and wave action. Moreover, shipping induced waves caused decline of vegetation in rivers. This means that stones provide the only shelter options making interactions with respect to shelter between species very likely. Coexistence of native and invasive gammarid species may be facilitated by creation a variety of shelter in hydraulic engineering and restoration projects (e.g., through increasing habitat heterogeneity and complexity).

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## References

- Bacela-Spychalska, K., & Van der Velde, G. (2013). There is more than one 'killer shrimp': Trophic positions and predatory abilities of invasive amphipods of Ponto-Caspian origin. *Freshwater Biology*, 58, 730–741.
- Crawley, M. J. (2007). *The R book*. Chichester, UK: John Wiley & Sons, Ltd.
- Devin, S., Piscart, C., Beisel, J. N., & Moreteau, J. C. (2003). Ecological traits of the amphipod invader *Dikerogammarus villosus* on a mesohabitat scale. *Archiv für Hydrobiologie*, 158, 43–56.
- Dick, J., & Platvoet, D. (1996). Intraguild predation and species exclusions in amphipods: The interaction of behaviour, physiology and environment. *Freshwater Biology*, 36, 375–383.
- Dick, J., & Platvoet, D. (2000). Invading predatory crustacean *Dikerogammarus villosus* eliminates both native and exotic species. *Proceedings of the Royal Society of London. Series B: Biological Sciences*, 267, 977–983.
- Dick, J., Montgomery, I., & Elwood, R. W. (1993). Replacement of the indigenous amphipod *Gammarus duebeni celticus* by the introduced *Gammarus pulex* – Differential cannibalism and mutual predation. *Journal of Animal Ecology*, 62, 79–88.
- Dick, J. T. A. (2008). Role of behaviour in biological invasions and species distributions; lessons from interactions between the invasive *Gammarus pulex* and the native *Gammarus duebeni* (Crustacea: Amphipoda). *Contributions to Zoology*, 77, 91–98.
- Dodd, J. A., Dick, J. T. A., Alexander, M. E., MacNeil, C., Dunn, A. M., & Aldridge, D. C. (2014). Predicting the ecological impacts of a new invader: Functional responses and prey selectivity of

- the 'killer shrimp', *Dikerogammarus villosus* compared to the native *Gammarus pulex*. *Freshwater Biology*, 59, 337–352.
- Elliott, J. M. (2005). Day-night changes in the spatial distribution and habitat preferences of freshwater shrimps, *Gammarus pulex*, in a stony stream. *Freshwater Biology*, 50, 552–566.
- Felten, V., Tixier, G., Guerold, F., De Billy, V. D. C., & Dangles, O. (2008). Quantification of diet variability in a stream amphipod: Implications for ecosystem functioning. *Fundamental and Applied Limnology*, 170, 303–313.
- Garvey, J. E., Stein, R. A., & Thomas, H. M. (1994). Assessing how fish predation and interspecific prey competition influence a crayfish assemblage. *Ecology*, 75, 532–547.
- Hacker, S. D., & Steneck, R. S. (1990). Habitat architecture and the abundance and body-size dependent habitat selection of a phytal amphipod. *Ecology*, 71, 2269–2285.
- Hesselschwerdt, J., Necker, J., & Wantzen, K. M. (2008). Gammarids in Lake Constance: Habitat segregation between the invasive *Dikerogammarus villosus* and the indigenous *Gammarus roeselii*. *Fundamental and Applied Limnology (Archiv für Hydrobiologie)*, 173, 177–186.
- Kelly, D. W., Bailey, R. J., MacNeil, C., Dick, J. T. A., & McDonald, R. A. (2006). Invasion by the amphipod *Gammarus pulex* alters community composition of native freshwater macroinvertebrates. *Diversity and Distributions*, 12, 525–534.
- Kinzler, W., & Maier, G. (2003). Asymmetry in mutual predation: Possible reason for the replacement of native gammarids by invasives. *Archiv für Hydrobiologie*, 157, 473–481.
- Kley, A. (2001). *Populationsökologie der neozoischen Gammariiden Dikerogammarus villosus und Echinogammarus ischnus im Main und Main-Donau Kanal*. Germany: University of Ulm. Diploma Thesis in Biology.
- Kley, A., & Maier, G. (2005). An example of niche partitioning between *Dikerogammarus villosus* and other invasive and native gammarids: A field study. *Journal of Limnology*, 64, 85–88.
- Lagrange, C., Kaldonski, N., Motreuil, S., Lefevre, T., Blatter, O., Giraud, P., et al. (2011). Interspecific differences in drift behaviour between the native *Gammarus pulex* and the exotic *Gammarus roeselii* and possible implications for the invader's success. *Biological Invasions*, 13, 1409–1421.
- Leuven, R. S. E. W., Van der Velde, G., Baijens, I., Snijders, J., Van der Zwart, C., Lenders, H. J. R., et al. (2009). The river Rhine: A global highway for dispersal of aquatic invasive species. *Biological Invasions*, 11, 1989–2008.
- MacNeil, C., Bigsby, E., Dick, J., Hatcher, M. J., & Dunn, A. M. (2003). Differential physico-chemical tolerances and intraguild predation among native and invasive amphipods (Crustacea): a field study. *Archiv für Hydrobiologie*, 156, 165–179.
- MacNeil, C., Dick, J. T., & Elwood, R. W. (1997). The trophic ecology of freshwater *Gammarus* spp. (Crustacea: Amphipoda): Problems and perspectives concerning the functional feeding group concept. *Biological Reviews of the Cambridge Philosophical Society*, 72, 349–364.
- MacNeil, C., Platvoet, D., & Dick, J. T. A. (2008). Potential roles for differential body size and microhabitat complexity in mediating biotic interactions within invasive freshwater amphipod assemblages. *Fundamental and Applied Limnology*, 172, 175–182.
- Magalhaes, S., Janssen, A., Hanna, R., & Sabelis, M. W. (2002). Flexible antipredator behavior in herbivorous mites through vertical migration in a plant. *Oecologia*, 132, 143–149.
- Noordhuis, R., Van Schie, J., & Jaarsma, N. (2009). Colonization patterns and impacts of the invasive amphipods *Chelicorophium curvispinum* and *Dikerogammarus villosus* in the IJsselmeer area, The Netherlands. *Biological Invasions*, 11, 2067–2084.
- Platvoet, D., Dick, J. T. A., MacNeil, C., Van Riel, M. C., & Van der Velde, G. (2009). Invader-invader interactions in relation to environmental heterogeneity leads to zonation of two invasive amphipods, *Dikerogammarus villosus* (Sowinsky) and *Gammarus tigrinus* Sexton: Amphipod pilot species project (AMPIS) report 6. *Biological Invasions*, 11, 2085–2093.
- Platvoet, D., Van der Velde, G., Dick, J. T. A., & Li, S. (2009). Flexible omnivory in *Dikerogammarus villosus* (Sowinsky, 1894) (Amphipoda) – Amphipod Pilot Species Project (Ampis) Report 5. *Crustaceana*, 82, 703–720.
- Stoffels, B. E. M. W., Tummers, J. S., Van der Velde, G., Platvoet, D., Hendriks, H. W. M., & Leuven, R. S. E. W. (2011). Assessment of predatory ability of native and non-native freshwater gammaridean species: A rapid test with water fleas as prey. *Current Zoology*, 57, 836–843.
- Van der Velde, G., Leuven, R. S. E. W., Platvoet, D., Bacela, K., Huijbregts, M. A. J., Hendriks, H. W. M., & Kruijt, D. (2009). Environmental and morphological factors influencing predatory behaviour by invasive non-indigenous gammaridean species. *Biological Invasions*, 11, 2043–2054.
- Van Riel, M. C., Healy, E. P., Van der Velde, G., & Bij de Vaate, A. (2007). Interference competition among native and invader amphipods. *Acta Oecologica – International Journal of Ecology*, 31, 282–289.
- Van Riel, M. C., Van der Velde, G., & Bij de Vaate, A. (2009). Interference competition between alien invasive gammaridean species. *Biological Invasions*, 11, 2119–2132.
- Van Riel, M. C., Van der Velde, G., & Bij de Vaate, A. (2011). Dispersal of invasive species by drifting. *Current Zoology*, 57, 818–827.
- Van Riel, M. C., Van der Velde, G., Rajagopal, S., Marguillier, S., Dehairs, F., & Bij de Vaate, A. (2006). Trophic relationships in the Rhine food web during invasion and after establishment of the Ponto-Caspian invader *Dikerogammarus villosus*. *Hydrobiologia*, 565, 39–58.