

## **Intra-guild predatory behaviour of the invasive *Dikerogammarus bispinosus* on the indigenous *Gammarus roeselii* (Crustacea: Amphipoda) in laboratory experiments and the River Danube**

Comportement de prédation intraguilde de l'espèce invasive *Dikerogammarus bispinosus* de l'espèce indigène *Gammarus roeselii* (Crustacea: Amphipoda) en laboratoire et dans le Danube

Manfred Pöckl

Institute of Hydrobiology and Aquatic Ecosystem Management, University of Natural Resources and Life Sciences, Max-Emanuelstr. 17, 1180 Vienna, Austria (corresponding author: [Manfred.Poeckl@noel.gv.at](mailto:Manfred.Poeckl@noel.gv.at)).

### **RÉSUMÉ**

*Dikerogammarus bispinosus* a envahi avec succès la partie autrichienne du Danube à partir du bassin Ponto-Caspien. Cette espèce euryèce et prédatrice est un fort compétiteur pour les populations d'espèces indigènes vivant sur les mêmes sites, comme par exemple l'espèce *Gammarus roeselii*. Nous avons étudié en laboratoire le comportement prédateur (prédation intraguilde) de l'espèce invasive à différentes températures entre 2° et 22°C. Nous avons conduit des tests de prédation en microcosmes afin d'amplifier les scénarios de compétition entre les deux espèces (un *D. bispinosus* contre un *G. roeselii*). Une expérience de contrôle avec l'espèce autochtone seule a également été réalisée. La mortalité des *G. roeselii* est significativement plus importante dans les microcosmes de compétition ( $1.33 \pm 0.22$  ind.) que dans les expériences contrôles ( $0.08 \pm 0.05$  individus). La mortalité est significativement moins importante à 2°C ( $0.5 \pm 0.2$  ind.) par rapport à 12°C ( $1.6 \pm 0.3$  ind.) et 22°C ( $1.9 \pm 0.46$  ind.). De plus, des images hautes définitions ont été faites avec des *D. bispinosus* et des *G. roeselii* dans de petites cuves à 8 et à 18°C pour analyser le comportement de capture des proies. Les mouvements des gnathopodes sont significativement plus rapides à 18°C ( $69.7 \pm 42.2$  mS) qu'à 8°C ( $256.7 \pm 43.8$  mS). La durée avant la première l'attaque de la proie est également significativement plus courte à 18°C. *D. bispinosus* est donc une espèce exotique carnivore très spécialisée étant capable d'une forte prédation intraguilde sur l'espèce autochtone. Bien que la survie de l'espèce autochtone soit meilleure à basse température (2°C), il subsiste toutefois une activité de prédation significative.

### **ABSTRACT**

*Dikerogammarus bispinosus* is a successful invader into the Austrian stretch of the River Danube, originating from the Ponto-Caspian basin. This euryoecious and predatory species is a strong competitor for populations of indigenous species, IS (e.g. the sympatric species *Gammarus roeselii*). The predatory intra-guild behaviour of the non-indigenous species, NIS, was studied in the laboratory at different water temperatures, reaching from 2 to 22°C. A series of predation trials in microcosms were conducted to stimulate a competition scenario (*D. bispinosus* and *G. roeselii*, 1:1 individuals). A control scenario involved only one specimen of the IS. Mortality of *G. roeselii* was significantly higher in all competition trials ( $1.33 \pm 0.22$  individuals) compared with all control experiments ( $0.08 \pm 0.05$  individuals). Mortality was significantly lower at 2°C ( $0.5 \pm 0.2$  ind.) compared with 12°C ( $1.6 \pm 0.3$  ind.) and 22°C ( $1.9 \pm 0.46$  ind.). Moreover, highspeed-camera recordings were made of one *D. bispinosus* individual together with one *G. roeselii* individual in cuvettes at 8 and 18°C to analyse the course of actions during prey capture. Gnathopod movements took  $256.7 \pm 43.8$  mS at 8°C versus  $69.7 \pm 42.2$  mS at 18°C, being significantly different. Furthermore, the duration until the first attack on the prey was significantly shorter at 18°C. Thus, *D. bispinosus* is a highly adapted carnivorous NIS, being able of strong intra-guild predation on smaller IS. Also at very low temperatures (2°C) predatory activity is still recorded, although survival of the IS is significantly higher.

### **KEYWORDS**

Danube River, *Dikerogammarus bispinosus*, *Gammarus roeselii*, indigenous species (IS), Intra-guild feeding behaviour, invader (NIS), laboratory experiments, replacement.

## 1 INTRODUCTION

One of the most striking differences between the indigenous and the invading amphipod species is the mode of feeding. Whilst *G. roeselii* is predominantly an omnivorous shredder, feeding on live and dead plants associated with micro-organisms, the invasive *Dikerogammarus* species (*D. haemobaphes*, *D. villosus*, *D. bispinosus*) are mostly predators, feeding on other animals, including intra-guild predation. This has tremendous effects on the population of the species preyed upon and thus the composition of the benthic community. Indigenous species can get very rare, are replaced and disappear. From his aggressive behaviour noticed before, *D. villosus* has been nick-named as the “killer shrimp”.

Although *D. bispinosus*, which now dominates in the benthic samples from the Danube, has not been studied before, it is speculated that its ecological impacts are similar to those of *D. villosus*. There are a number of interesting questions to be answered, e.g. has *D. bispinosus* a similar feeding behaviour as previously studied “the killer shrimp”? Why are *Dikerogammarus* species restricted to the main river bed of the Danube, do not penetrate into tributaries, and replace also indigenous species there? In this context it is hypothesised if water temperature in the tributaries could be a factor from which indigenous species, such as *G. roeselii*, have a benefit. Thus, predation rate in warmer regions of the rivers or during summer, populations of the indigenous species should suffer under a higher predation rate of *D. bispinosus*. Under cold conditions the mortality rate of *G. roeselii* due to predation should be minimised. Furthermore, we studied the temperature dependence of the course of actions during prey capture by high-speed-camera recordings.

## 2 METHODS

### Competition trials

Forty-five microcosms (15 x 10 x 10 cm) were filled with 0.5 l of water from the sampling stations. On three corners of the container transparent glass marbled were placed in order that the thigmotactic requirements of the amphipods are met. In the fourth corner the pipe of the forced aeration was placed (c. 5 air bubbles per s). Large individuals of *D. bispinosus*, and middle-sized individuals of *G. roeselii* were chosen. In control experiments only one individual of *G. roeselii* was kept, and in the competition experiment one individual of both species were kept. The experiments were run for five days at constant temperatures of 2, 12, and 22°C and constant light conditions (l / d = 10 : 14 h). No food was added.

Tab. 1: Experimental design for competition trials, giving the number of replicates.

Temperature (°C)	Mortality experiments 1 <i>D. bispinosus</i> and 1 <i>G. roeselii</i>	Control groups 1 <i>G. roeselii</i>
2	n = 45	n = 45
12	n = 45	n = 45
22	n = 45	n = 45
	Total = 135	Total = 135

### High-speed recordings

Plexi-glass cuvettes with the dimensions of 10 x 10 cm and a width of 0.7 cm (in order to force the amphipods in a given distance from the objective) were designed and filled with 25 ml of water from the sampling stations. The water was aerated by 5 air bubbles per s. One individual of both species was added. The recordings were made at constant temperatures of 8 and 18°C, respectively, using a high-speed camera (type Photron Fastcam-X 1024 PCI) with a resolution of 1024 x 1024 pixels. 1000 frames per s were shot. The software used was Photron Fast Cam Viewer 3.0. The course of actions during prey capture of *D. bispinosus* was analysed and the speed of gnathopod movements was measured.

## 3 RESULTS AND DISCUSSIONS

### Competition trials

Mortality of *G. roeselii* was significantly higher in all competition trials (1.33±0.22 individuals) compared with all control experiments (0.08±0.05 individuals). Mortality was significantly lower at 2°C (0.5±0.2 ind.) compared with 12°C (1.6±0.3 ind.) and 22°C (1.9±0.46 ind.).

Results at 2°C were significantly different ( $p < 0.05$ ), but the difference between 12°C and 22°C was not significant ( $p > 0.05$ ).

In the control group no dead *G. roeselii* individual was observed at 2°C during 5 days. At 12°C and 22°C the mean mortality was  $0.11 \pm 0.09$  individuals. There was no significant difference between the three constant temperatures used ( $p < 0.01$ ).

### High-speed recordings

In total the prey captures and feeding behaviour of 30 male *D. bispinosus* was filmed for 23 hours and 15 minutes. The sequence of prey capture is stereotyped and extremely fast. *D. villosus* holds itself on the bottom between the front and the back glass of the cuvette. It approaches the prey with antenna 2. The speed recording begins with the first movements of the gnathopods towards the prey. Then the dactyli are hooked into the prey and antenna 2 depress downwards. The pleon rolls forward and the gnathopods are moved backwards. The prey is fixed between the gnathopods and the antenna 2 from above and from the pleon from below, and has lost the chance to escape. Now the invader begins to feed.

Gnathopod movements took  $256.7 \pm 43.8$  mS at 8°C versus  $69.7 \pm 42.2$  mS at 18°C, being significantly different. Furthermore, the duration until the first attack on the prey was significantly shorter at 18°C.

## 4 CONCLUSIONS

*Dikerogammarus bispinosus* is a highly adapted carnivorous NIS, being able of strong intra-guild predation on smaller indigenous species. Also at very low temperatures (2°C) predatory activity is still recorded, although survival of the *G. roeselii* is significantly higher. The feeding behaviour of the invasive species has tremendous effects on the population of the species preyed upon and thus the total composition of the benthic community in the Danube.

As hypothesised above, water temperature is not the key factor hindering *D. villosus* from colonising tributaries to the River Danube. Indigenous species do not benefit from the difference in water temperature between the rivers. The reason why *Dikerogammarus* species are restricted to the main river bed of the Danube and do not penetrate into tributaries, is therefore still unknown.