



Telemetry of co-occurring noble crayfish (*Astacus astacus*) and stone crayfish (*Austropotamobius torrentium*): diel changes in movement and local activity

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With 7 figures and 1 table

Abstract: Knowledge of movement patterns is essential for the effective conservation of noble crayfish (*Astacus astacus*) and stone crayfish (*Austropotamobius torrentium*), two endangered species indigenous to Central Europe. Radiotelemetry was used to evaluate diel changes in ranging behaviour and local activity of both species at a locality where they occur sympatrically. Both species exhibited similar behaviour, migrating mostly in the upstream direction. Movement probability differed significantly within both species between phases of the diel cycle. Movement probability within 3-h tracking intervals was highest at dawn (18.18 %) and night (15.32 %) in *A. astacus*, and at dawn (37.25 %) in *A. torrentium*. *Astacus astacus* moved the longest distances during the dawn interval (3.70 m) and the shortest during the day (1.41 m) and night (1.61 m) interval. The distances moved by *A. torrentium* did not differ significantly in different phases of the diel cycle. Local activity was highest at dusk in *A. astacus* (40.6 % likelihood of activity) and during dusk and night hours in *A. torrentium* (30.8 % and 24.2 % likelihood of activity). Body size and environmental parameters (light intensity, weather, atmospheric pressure, water temperature and turbidity) also influenced crayfish behaviour. Both species exhibited a post-handling fright response, which (e.g. in mark-recapture studies) can lead to the overestimation of movement rates and the extent of natural migrations.

Keywords: ranging behavior; crayfish; diurnal cycle; conservation; Czech Republic

Introduction

Crayfish are known to play an important role in freshwater ecosystems. They are among the largest and most mobile invertebrate animals in streams (Sint & Füreder 2004). Crayfish strongly affect trophic interactions in many types of freshwater environments, where they are able to significantly alter habitat conditions

by burrowing and disturbing the substrate (Statzner et al. 2000). Being popular animals with the public, crayfish are also frequently used as ‘flagship species’ in conservation and freshwater protection projects, and some species are considered suitable bioindicators of environmental quality (Füreder & Reynolds 2003).

Despite their key role in ecosystems, indigenous crayfish (IC) species in Europe are heavily affected

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by alterations of habitats, pollution, stream fragmentation and also by the spread of non-indigenous crayfish (NIC) species (Schulz & Schulz 2004; Holdich et al. 2009; Patoka et al. 2014), which threaten IC species not only through interspecific competition (Bubb et al. 2006), but also by transmitting the crayfish plague pathogen (Kozubíková et al. 2009; Filipová et al. 2013). All these circumstances have led to a decrease of populations of IC species, both in number and in size (Taugbøl & Skurdal 1999; Holdich et al. 2009).

The spatial and temporal heterogeneity of the environment determines the movement patterns of animals, affecting their dispersal rates and foraging behaviours (Forman & Gordon 1986; Johnson et al. 1992; Bowler & Benton 2005). Animal movements are motivated by internal as well as external cues, and are related mainly to finding food, escaping predators, fighting, reproduction (Colgan 1993) or avoiding environmental extremes (Davenport 1985; see also paper by Daněk et al. 2014). As all these aspects directly influence the survival of individuals and populations, knowledge of animal movement patterns is essential for their management and conservation.

Ranging behaviour affects the ability of species to colonize new areas and to recolonize depopulated places. The range and directions of crayfish movement also influence the extent and probability of crayfish plague transfer. Activity patterns of individuals in the 24h cycle can indicate intra- and interspecific interactions (Sutherland 1996), which play an important role in the protection of native populations and may significantly influence the success of restocking activities.

Approaches to monitoring the movement of crayfish individuals in nature include direct (visual) observation (e.g. Sint & Füreder 2004), mark-recapture methods (e.g. Gherardi et al. 2000; Kadlecová et al. 2012) and tracking of animals equipped with electronic tags (e.g. Bohl 1999; Bubb et al. 2002; Loughman et al. 2013; Anastácio et al. 2015). Direct observation is usually limited only to a specific set of circumstances (e.g. narrow shallow waters with low turbidity, observing only individuals wandering out of shelter). Mark-recapture methods may sufficiently cover questions about meso- and microhabitat preferences, but ranging behaviour assessed by this method can be strongly influenced by 'fright responses' associated with capture and tagging (Robinson et al. 2000; Barbaresi et al. 2004). The inclusion of fright responses in data resulting from mark-recapture studies is expected to overestimate rates of movement and the extent of natural migration (Robinson et al. 2000).

Radiotelemetry is a widely used method enabling detailed monitoring of the behaviour of marked individuals in a natural environment without the need of further recapture and disturbance after tagging. This approach allows the monitoring of the movement of individuals with high spatial and temporal resolution, and even to detect levels of local activity not connected with relocation by measuring fluctuations of transmitter signal strength (Bubb et al. 2002; Thiem et al. 2010).

Whereas numerous radiotelemetric studies have examined the movement patterns of NIC species in nature (e.g. Bubb et al. 2002; Bubb et al. 2004; Gherardi et al. 2002; Barbaresi et al. 2004; Aquiloni et al. 2005; Buřič et al. 2009a; Buřič et al. 2009b; Hirsch et al. 2016), only a few have dealt with noble crayfish (*Astacus astacus*) and, to the best of our knowledge, not a single one has been carried out on stone crayfish (*Austropotamobius torrentium*).

Moreover, two previously published radiotelemetric studies on *A. astacus* behaviour in nature were focused only on the largest individuals exceeding 90 mm in total length and about 20 g in weight (Bohl 1999), and with a total length of 120–135 mm (Schütze et al. 1999). The use of these particularly large individuals was necessary in these studies due to the size and weight of previous-generation radio tags not allowing their attachment to smaller individuals. In fact, the results obtained from these two studies may be influenced by the size of the tracked animals. Smaller individuals are usually much more frequent in populations and may behave substantially differently, for example, because they are at a greater risk of predation (Paine 1976; Nilsson & Brönmark 2000). Moreover, Schütze et al. (1999) deliberately studied the movement of stocked crayfish, so their results are difficult to apply to wild individuals.

In our study, we dealt with wild crayfish only, and used particularly small radiotelemetric transmitters, which enabled us to track much smaller individuals than previous studies (down to the total length of 48 mm and weight of 3.5 g), while retaining a similar or better tag to body weight ratio (e.g. Robinson et al. 2000; Bubb et al. 2006; Gherardi et al. 2002). Our main goal was to evaluate (1) diel changes in ranging behaviour and local activity of *A. torrentium* and *A. astacus* captured and tracked in a stream where the two species occur in sympatry and syntopy. We also tested (2) if and how the sex and size of crayfish influence their behaviour, and (3) how environmental variables influence the movement patterns and local activity of both species.

Material and methods

Study site

The study was undertaken along the Stroupínský stream, which is a 20.9 km long fourth-order watercourse located roughly 50 km west of Prague (Elbe river basin, Central Europe). The section of the stream under study (downstream from the village of Hředle, 49° 53' 54" N, 13° 56' 17" E) flows through agricultural land. It has an average width of around 3.5 m and is composed mostly of shallow riffles interspersed with pools that rarely exceed 1 m in depth. The substrate varies considerably from boulders to mud in deeper sections of the stream. The banks are covered by dense vegetation composed of grasses, nettles (*Urtica dioica*), shrubs (*Rosa canina*, *Prunus spinosa*, *Salix* sp.) and trees (*Alnus glutinosa*, *Populus* sp.). The section studied is part of the 'Stroupínský potok' Special Area of Conservation (Svobodová et al. 2017) inhabited by two indigenous crayfish species, *A. astacus* and *A. torrentium*, which occur there in sympatry and syntopy (Vlach et al. 2009b; Kadlecová et al. 2012). The fish stock consists of *Salmo trutta* m. *fario*, *Squalius cephalus*, *Barbatula barbatula*, *Phoxinus phoxinus*, *Gobio gobio* and *Anguilla anguilla* (Musil et al., unpublished data).

Capture and radiotagging

Astacus astacus and *A. torrentium* for our radiotelemetry study were caught by hand. Special care was taken to put stones back into their original position to minimize habitat disruption. Sixteen individuals (8 *A. torrentium* and 8 *A. astacus*) were caught and tagged on 2 August 2016. The mean total lengths of *A. torrentium* and *A. astacus* were 56 mm (range 48–64 mm) and 64 mm (56–75), respectively, and their mean weights were 6.7 g (3.5–10.6) and 8.3 g (5.2–12.0), respectively. Crayfish of both sexes were tagged (4 females and 4 males of *A. torrentium*, and 5 females and 3 males of *A. astacus*). The tag to body weight ratio ranged from 2.2 to 7.4%, which was lower (better) than in previous studies (up to 10% in Robinson et al. 2000, 13.4% in Bubb et al. 2006, 10% in Gherardi et al. 2002). We assume that transmitters with this tag to body weight ratio do not significantly hinder the movement or alter the survival of tagged crayfish (Bohl 1999; Robinson et al. 2000).

Coded radio transmitters NTQ 1 (Lotek Engineering Inc., New Market, Ontario, Canada; 0.26 g in air, 10 × 5 × 3 mm with a c. 15 cm long external whip antenna) were glued using cyanoacrylate adhesive on to the carapace of individual crayfish. The expected life span of each radio transmitter was 21 days (according to the manufacturer). Individual crayfish were identified by unique numeric codes displayed by a SRX 800 receiver (Lotek Engineering Inc.). All transmitters operated at the same frequency of 138.3 MHz. Tagged individuals were released exactly at the places where they were captured. The tagging procedure did not exceed 10 min.

Our research on *A. astacus* and *A. torrentium*, both of which are critically endangered according to Czech legislature, was authorized by the Regional Office of the Central Bohemian Region, Department of Environment and Agriculture (permit No. 102856/2016/KUSK-3).

Collection of radiotelemetric data

Tagged crayfish were not tracked for the first four days after tagging due to possible artificial behaviour (fright response)

associated with the capture and tagging procedure (Robinson et al. 2000; Barbaresi et al. 2004; Loughman et al. 2013). We, however, measured the total position shift between the position of the release of each individual and its first position detected by radiotracking. Telemetric data were collected from 6 August to 14 August 2016, in eight 3-h tracking intervals per day: one at dawn (light intensity, measured in exposure values (EV), ranged between 2 and 6), four during the day (>6 EV), one at dusk (2–6 EV) and two at night (<2 EV).

The positions of all crayfish and the distances they moved during each time interval were recorded. Individuals that changed position up, down or across the stream by at least 0.5 m were considered moving crayfish within a particular time interval. Metal markers (iron pegs), each with the number of an individual crayfish, placed on the stream bank, were used to measure distances moved up- or downstream (hereafter 'distances moved'). Distances from each marker (previous position) to the point representing the current position of each individual were measured with a 30 m long measuring tape laid parallel to the longitudinal axis of the watercourse to account for its slight curvature. This approach enabled us to avoid the inaccuracy of the GPS (± 4 m).

Local activity levels of individual crayfish were determined for each tracking interval during 1-min time periods by recording the number of variations in signal strength as an index of activity. Crayfish motion may alter the orientation of a transmitter's whip antenna in relation to the fixed Yagi antenna of the receiver and thus generate variation in the strength of the signal received, which can be used to assess the activity level of the individual (Bubb et al. 2002; Thiem et al. 2010). High levels of activity often occurred without the crayfish relocating by more than 0.5 m. This local activity may be connected, for example, to feeding, burrowing or fighting for shelter (Bubb et al. 2002).

Prior to the attachment of transmitters to crayfish, tests of baseline signal strength variability were carried out with each transmitter laid on the bottom of the stream. With the antenna of the receiver held motionless for one minute at a distance of approximately 3 m, the signal of each transmitter was always stable and did not vary in strength by more than 2%. The telemetric receiver used displays exact signal strength values. During the actual tracking of individual crayfish, the local activity of each individual was determined after its localization in the same manner as during the testing of individual transmitters. Variations in signal strength exceeding the threshold value of 5% were regarded as signs of local activity. Numbers of signal strength variations were classified according to Robinson et al. (2000) into three categories: 0 (0 changes), 1 (one or two changes) and 2 (> two changes).

Tagged crayfish with a still operating transmitter were recaptured after the end of the telemetry study (for confirmation that all telemetric data on individual represented the behaviour of a live and healthy crayfish). If recapture was not possible, the crayfish was considered to be alive until its last measurable position shift or until its last measurable local activity. Data after the last movement (or last local activity) of the individual were excluded from further analyses.

Environmental measurements

Light intensity levels were measured with a Gossen Digisix photographic light meter (GOSSEN Foto- und Lichtmesstechnik GmbH) at an unshaded reference point during each 3-h interval. The water level was determined using a water level

pole (cm), while the turbidity was measured with a portable turbidimeter TURB 355 IR/T (Wissenschaftlich-Technische Werkstätten GmbH). The water temperature was measured continuously using a HOBO Pendant Temperature/Light 8K Data Logger (Onset Computer Corporation). Weather conditions were categorized into four categories: 1 – clear sky, 2 – cloudy, 3 – overcast and 4 – rain. The atmospheric pressure tendency was determined using the internal barometer of a GPSmap 60CSx device (Garmin Ltd.) and categorized into three categories: ‘decreasing’ if atmospheric pressure decreased by at least 2 mbar within a 3-h interval, ‘increasing’ if atmospheric pressure increased by at least 2 mbar within a 3-h interval, and ‘stable’ if the difference during a 3-h interval was less than 2 mbar.

Statistical analyses

To examine the influence of the time of day (dawn, day, dusk, night), crayfish sex and size, and environmental variables within and between the two study species, we used generalized linear mixed models (GLMM). We analysed the *movement probability* (using data on whether crayfish did or did not move; binomial distribution, logit link), the *distance moved* (data with gamma distribution, log link) and the *local activity* (data on whether crayfish were active or not during measurement; binomial distribution, logit link). Local activity levels 1 and 2 were grouped together for the GLMM analysis. Because we observed the behaviour of each individual crayfish repeatedly, we included the identity of individuals as a random factor. We used the ‘lme4’ package in R v. 3.0.3 (R Development Core Team 2008). The resulting models were compared against matching null models using likelihood ratio tests to assess model significance. To compare the fright responses of the two species after capture and tagging, we used *t*-tests (for distances moved and the directions of their movement) and tests for the equality of proportions (for numbers of moving crayfish). When applicable, data are presented with standard errors of the mean (SE).

Results

Values of water temperature and water level over the course of the study are presented in Fig. 1. In total, 864 valid positions of crayfish were obtained. Based on the recapture of tracked individuals at the end of the study period, the moulting of one individual of *A. torrentium* (ATOR 7) was proven (a moulted carapace with a transmitter was found). One individual of *A. astacus* (AAST 4) was probably predated by a muskrat (*Ondatra zibethica*) – the signal of the transmitter had moved from the stream to a terrestrial burrow in the bank on the last day of the experiment. The signals of two individuals of *A. torrentium* (ATOR 3 and ATOR 8) disappeared half a day and six days before the end of the study, respectively, possibly due to a transmitter failure or predation. One individual of *A. astacus* (AAST 2) was found stuck after the antenna of its transmitter got entangled in grass. Because this individual showed very limited movement and local activity after 6 August, all data obtained by the radiotelemetric tracking of this individual were omitted from further analyses.

Fright response after capture and tagging

In the first four days after tagging, individuals of both crayfish species often left their previous locations and in many cases covered longer distances than during the entire subsequent telemetry study (3 individuals

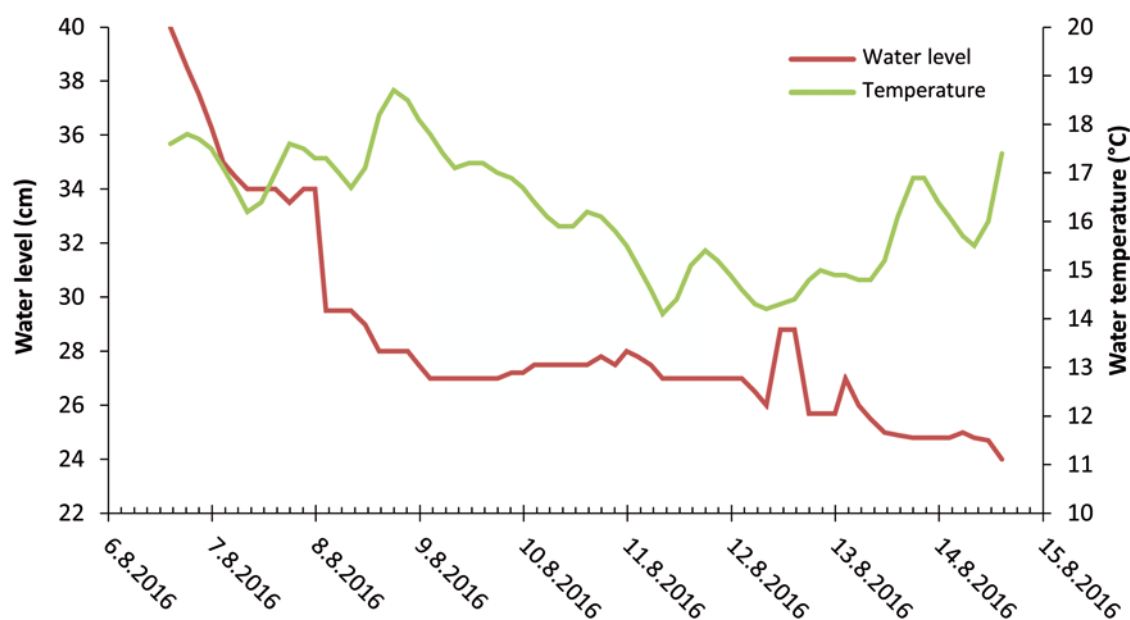


Fig. 1. Values of water level and water temperature over the duration of the study.

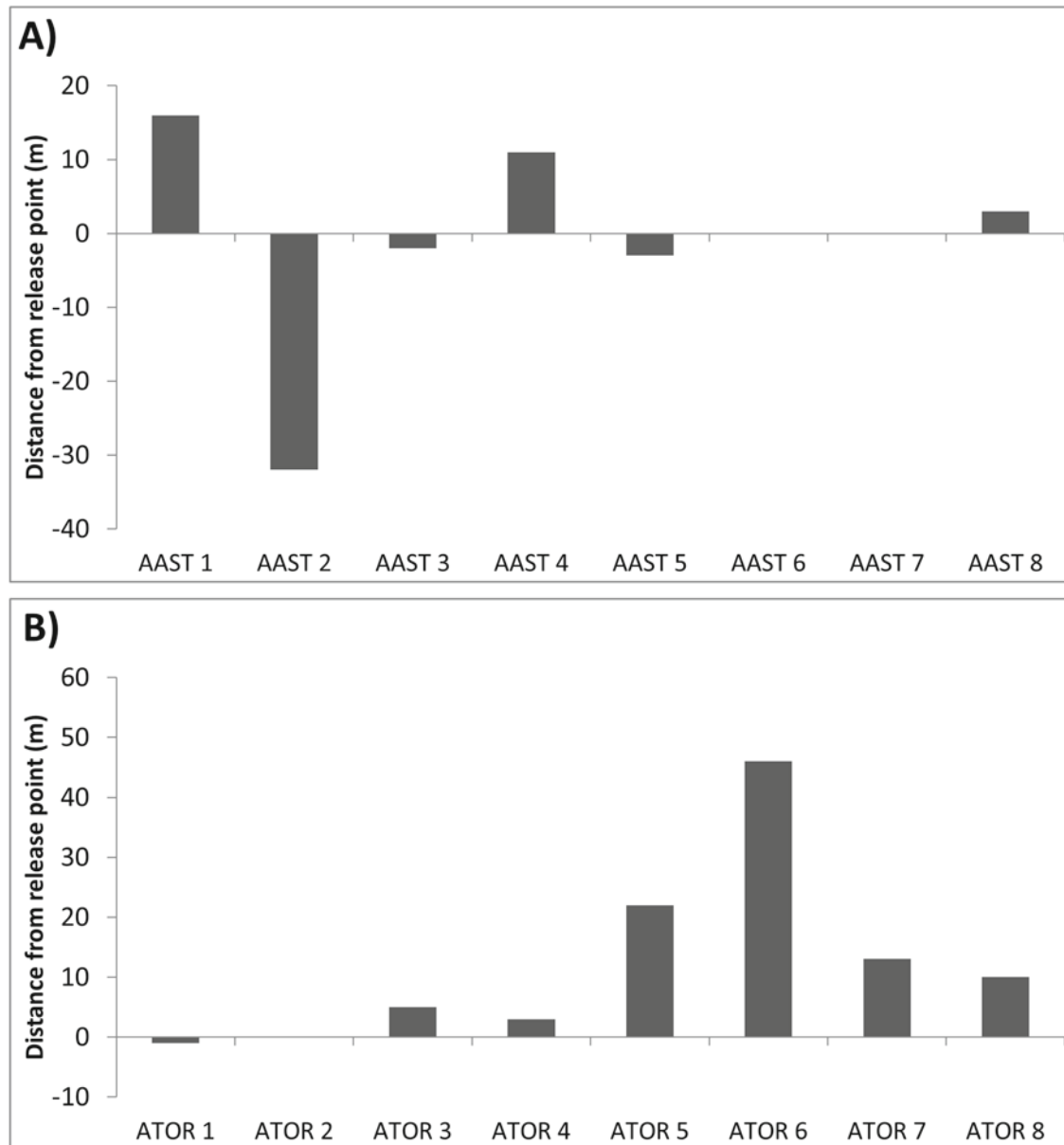


Fig. 2. Emigration response of individual crayfish after capture and tagging: **A)** *Astacus astacus*, **B)** *Austropotamobius torrentium* expressed as the distance moved from the original location in the first four days after tagging. Positive values refer to locations upstream and negative values to locations downstream from the release point. 'AAST' with number = individuals of *A. astacus*, 'ATOR' with number = individuals of *A. torrentium*.

of *A. astacus*, 4 individuals of *A. torrentium*). There was no obvious tendency to migrate up- or downstream among the *A. astacus* group (Fig. 2A). *Austropotamobius torrentium* migrated predominantly in the upstream direction (Fig. 2B). However, interspecific differences (movement probability: $\chi^2 = 0.4103$, d.f. = 1, $p = 0.5218$; distance moved: $t = -0.6154$, d.f. = 14, $p = 0.5482$; direction of movement: $t = -1.7583$, d.f. = 14, $p = 0.1005$) were non-significant.

Ranging behaviour and local activity during the telemetric survey

The relocation of individual *A. astacus* and *A. torrentium* in the stream during our telemetric study is presented in Fig. 3. The two species exhibited similar displacement patterns, with a predominantly upstream direction of position shifts. The movement was usually limited to relatively short time periods followed by longer periods of staying at a particular shelter. Af-

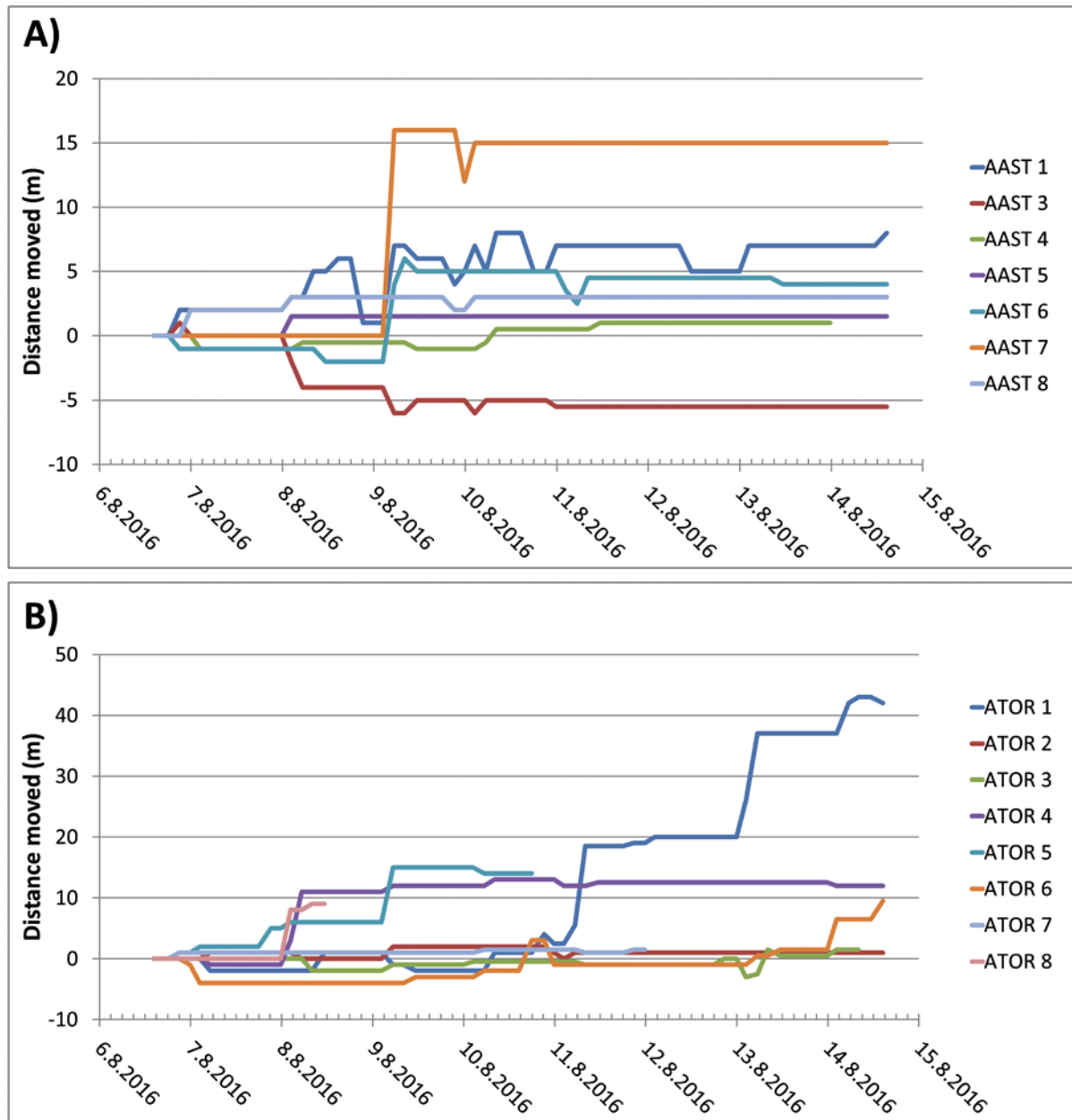


Fig. 3. Relocation of individuals **A)** *Astacus astacus* and **B)** *Austropotamobius torrentium* during our telemetric survey (6 Aug –14 Aug 2016). Positive values refer to locations upstream and negative values to locations downstream from the initial location detected by radiotelemetry. ‘AAST’ with number = individuals of *A. astacus*, ‘ATOR’ with number = individuals of *A. torrentium*.

ter migration, the individuals usually did not return to their previously occupied shelter.

The average values representing spatial behaviour within 3-h intervals and the average numbers of active *A. astacus* and *A. torrentium* are presented in Fig. 4. The overall movement probability of *A. astacus* and *A. torrentium* were 11.1% (± 3.1) and 15.3% (± 1.8), respectively, and the distances moved by relocating individuals were 2.02 m (± 0.34) and 2.40 m (± 0.33),

respectively. The overall likelihood of local activity of *A. astacus* and *A. torrentium* was 16.4% (Activity 1: 6.4%; Activity 2: 10.0%) and 17.8% (10.0%; 7.8%), respectively. There were no significant differences between the two species in movement probability ($z=1.319$, d.f. = 3, 845, $p=0.187$), distance moved ($t=0.378$, d.f. = 4, 107, $p=0.7057$) or local activity ($z=0.473$, d.f. = 3, 858, $p=0.636$).

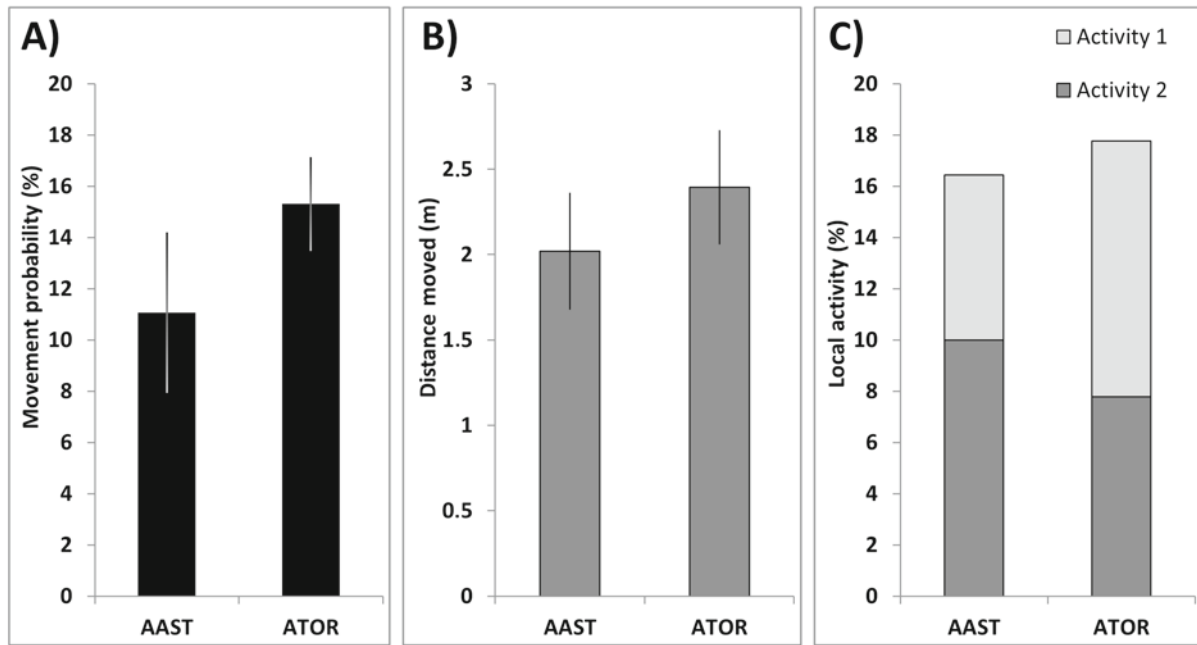


Fig. 4. Comparison of average movement and local activity of *Astacus astacus* (AAST) and *Austropotamobius torrentium* (ATOR) within 3-h intervals: **A)** movement probability; **B)** distances moved by relocating individuals; and **C)** local activity. Local activity levels were determined by measuring the number of variations in signal strength during one-minute time periods as an index of activity. The numbers of signal strength variations were classified into three categories: 0 (0 changes), 1 (one or two changes) and 2 (> two changes).

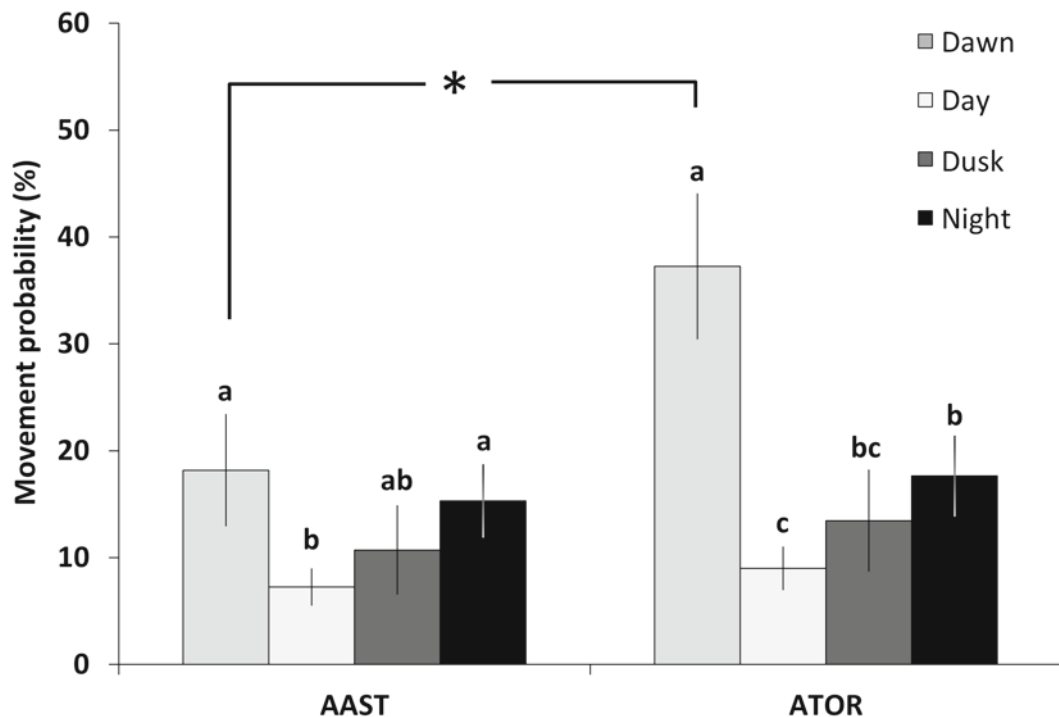


Fig. 5. Movement probability (%) of crayfish within 3-h tracking intervals at dawn, during the day, at dusk and at night. Different letters indicate significant differences ($p < 0.05$) between dawn, day, dusk and night intervals within one species. The asterisk indicates a difference between the species during the dawn tracking interval. 'AAST' = *Astacus astacus*, 'ATOR' = *Austropotamobius torrentium*.

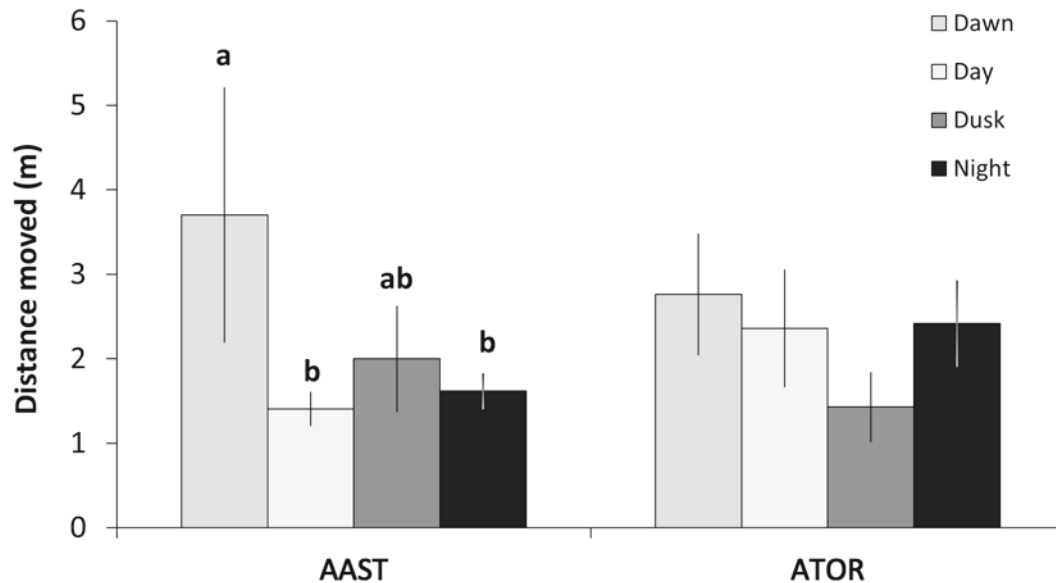


Fig. 6. Distances moved (m) by relocating crayfish during dawn, day, dusk and night in 3-h tracking intervals. Different letters indicate significant differences ($p < 0.05$) between dawn, day, dusk and night intervals for *Astacus astacus* (AAST). *Austropotamobius torrentium* (ATOR) did not exhibit any significant differences between different times of day in distance moved. No significant differences between the species were found for the same times of day.

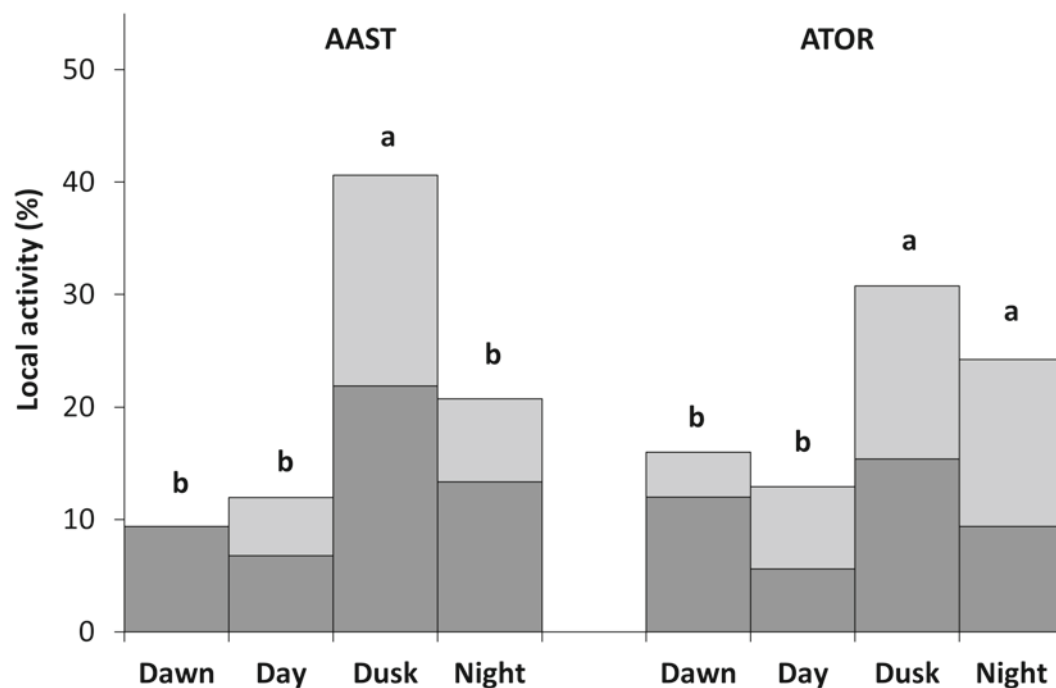


Fig. 7. Comparison of local activity of *Astacus astacus* (AAST) and *Austropotamobius torrentium* (ATOR) at dawn, during the day, at dusk and at night. Different letters indicate significant intraspecific differences ($p < 0.05$) between these phases of the diel cycle. Local activity levels were determined during 1-min time periods by measuring the number of variations in signal strength as an index of activity. Numbers of signal strength variations were classified into three categories: 0 (0 changes), 1 (one or two changes) and 2 (> two changes). Dark parts of bars indicate the activity level 2 and their light parts indicate the activity level 1. No significant differences were found between the species for the same times of day.

Diel changes in ranging behaviour and local activity

The differences in movement probability between dawn, day, dusk and night intervals in both species are shown in Fig. 5. Movement probability differed during different phases of the diel cycle significantly within *A. astacus* ($\chi^2=8.3804$, d.f. = 3, $p=0.03877$) as well as in *A. torrentium* ($\chi^2=22.280$, d.f. = 3, $p<0.001$). *Astacus astacus* exhibited the highest values at dawn ($18.18\% \pm 5.25$) and at night ($15.32\% \pm 3.43$) and *A. torrentium* moved most frequently at dawn ($37.25\% \pm 6.84$). The lowest values occurred during the day in *A. astacus* ($7.24\% \pm 1.75$) as well as in *A. torrentium* ($9.00\% \pm 2.03$). The movement probability of *A. astacus* during dawn was significantly lower than that of *A. torrentium* at the same times of day phase ($z=2.165$, d.f. = 3, 103, $p=0.0304$). All other interspecific differences were non-significant.

The distances moved by relocating crayfish within dawn, day, dusk and night during 3-h tracking intervals between the two species are summarized in Fig. 6. The distance covered differed during different phases of the diel cycle significantly in *A. astacus* ($\chi^2=13.608$, d.f. = 3, $p=0.00349$), whose individuals moved the longest distances at dawn ($3.70\text{ m} \pm 1.51$) and the shortest distances during the day ($1.41\text{ m} \pm 0.21$) and at night ($1.61\text{ m} \pm 0.21$). The distances covered by *A. torrentium* did not differ significantly between different phases of the diel cycle ($\chi^2=2.744$, d.f. = 3, $p=0.4328$). All interspecific differences were non-significant.

A comparison of local activity of *A. astacus* and *A. torrentium* at dawn, during the day, at dusk and at night is shown in Fig. 7. Local activity differed during different phases of the diel cycle significantly within

A. astacus ($\chi^2=18.112$, d.f. = 3, $p<0.001$) as well as in *A. torrentium* ($\chi^2=9.9578$, d.f. = 3, $p=0.01893$). The local activity differed significantly within both species during different phases of the diel cycle, reaching the highest values during dusk in *A. astacus* (40.6 % likelihood of activity) and during dusk and night hours in *A. torrentium* (30.8 % and 24.2 % likelihood of activity), respectively. All interspecific differences in local activity were non-significant.

Impact of sex, size and environmental parameters

Over the course of the study period, no sex-related differences were observed in either species regarding movement probability, distances moved or in local activity. The effects of body size and selected environmental variables are summarized in Table 1. We found that movement probability correlated negatively with the length of individuals. On the other hand, larger *A. torrentium* exhibited higher local activity.

Light intensity, when significant, had the same impact in all cases. Higher light intensity resulted in lower movement probability and in lower local activity. Stable, sunny weather with a clear sky had a positive effect on distances moved (by both crayfish species) and on their local activity (*A. torrentium*). Lower turbidity was connected to a higher movement probability in *A. torrentium*.

Stable atmospheric pressure was found to have a positive effect on distances moved by *A. astacus*. Higher water temperature had a positive effect on movement probability as well as on distances moved by *A. astacus*. Water level did not have any effect on crayfish behaviour observed during the study period.

Table 1. Impact of sex, size and environmental variables on the behaviour of *Astacus astacus* (AAS) and *Austropotamobius torrentium* (ATOR). Significance: *** = $p<0.001$; ** = $p<0.01$; * = $p<0.05$; . = $p<0.1$; ns = no significant difference.

	Movement probability		Distance moved		Local activity	
	AAS	ATOR	AAS	ATOR	AAS	ATOR
Sex	ns	ns	ns	ns	ns	ns
Carapace length	**	ns	ns	ns	ns	*
Light intensity	**	**	.	ns	*	.
Weather	ns	ns	**	*	ns	*
Turbidity	ns	**	ns	ns	ns	ns
Atmospheric pressure	ns	ns	***	ns	ns	ns
Water level	ns	ns	ns	ns	ns	ns
Water temperature	**	.	*	ns	ns	ns

Discussion

The main goal of our study was to assess diel changes in ranging behaviour and the local activity of *A. torrentium* and *A. astacus* in a stream where the two species co-occur in sympatry. Moreover, we examined the effects of sex, size and selected environmental parameters on crayfish movement and activity, and the extent of the fright response of the two species to being captured and tagged. The use of some of the smallest active radio transmitters currently on the market allowed us to track very small individuals.

In our study, we used radio tags with the same or lower tag to body weight ratio as previous authors (Gherardi et al. 2002; Bubb et al. 2006), and assume that the transmitters generally did not hinder the movement of individuals which we tracked (Robinson et al. 2000). The only exception was individual AAST 2, which moved to a patch of the stream with otherwise rare underwater grass, where it got trapped. We found and freed this individual after the end of the telemetric study; the flexible antenna of its transmitter became entangled among stems and roots, creating a knot. The antenna became noticeably deformed into a spiral as a result of the animal's struggle to free itself. After the study, we re-captured most of the other tracked individuals, and none of their antennas showed any signs of getting tangled or trapped. We therefore assume that the movement of all other individuals was not affected by the transmitters' antennas.

It must be noted that the data on which we base our conclusions were gathered from a limited number of individuals over a comparatively short period. Our results should, therefore, be interpreted with caution and further studies are needed to devise more general conclusions. Nevertheless, our study brings many new insights into the behaviour of both species in their natural environment.

Our results indicate that the two species behave very similarly, and statistically significant differences between the two species were rare. Over the course of the study, individuals of both species moved mostly in the upstream direction, usually in a sudden manner, followed by a longer period remaining at a particular shelter. Prevailing migrations in the upstream direction in the stream under study have also been recorded by Kadlecová et al. (2012). Many previous studies dealing with the ranging behaviour of crayfish, however, report predominantly downstream migrations.

For example, in a study focused on spiny-cheek crayfish, Buřič et al. (2009b) tentatively interpreted the downstream migrations of this species by its sea-

sonal relocation to winter refuge. Bohl (1999) remarks that, while natural behavioural patterns of *A. astacus* include downstream as well as upstream migrations, long-distance downstream migrations are associated with disturbance events. Schütze et al. (1999) also describe predominantly downstream migrations in *A. astacus*; however, as in the previous case, a combination of stress factors (i.e., the release of crayfish raised at a hatchery into a new environment, large variations of the water level caused by a hydropower station and the presence of crayfish plague) may have influenced the results of that study. The migrations in the upstream direction observed by us could, therefore, imply that the conditions in the stream were favourable at the time of our study. The objective of these upstream migrations might, therefore, be the species' expansion higher up the stream, compensation of a previous downstream migration of individuals caused by a stressful event, or the compensation of passive downstream drift of individuals caused by a high water discharge event (Momot 1966; Bohl 1999).

In our study, we did not observe any homing behaviour of individuals. After the crayfish migrated, they usually did not return to their previously occupied shelter. Loughman et al. (2013) also mentioned that individuals of *Cambarus chasmodactylus* did not return to previously used daily retreats. By contrast, Webb & Richardson (2004) observed frequent returns of individuals of *Astacopsis gouldi* to areas they had previously occupied, and a similar behaviour was described by Barbaresi et al. (2004) for *Procambarus clarkii*. For crayfish, it is usual that individuals use a patch surrounding their shelter for a few days, and then move to another (Bubb et al. 2004; Loughman et al. 2013; Anastácio et al. 2015). As a result of this behaviour, many crayfish species do not have true home ranges, but only 'ephemeral home ranges' between which they migrate (Robinson et al. 2000). We hold the opinion that the absence of homing in our study is connected to the high number of shelters available at the study site, which did not force individuals to use the same retreats repeatedly after migrations.

In our study, a comparison of average values over the entire 24-h cycle did not reveal any statistically significant differences between the study species in movement probability, distances moved or in local activity. Notably, however, the values recorded for *A. torrentium* were slightly greater than those recorded for *A. astacus*. This is surprising, as Kadlecová et al. (2012) found *A. torrentium* to be more sedentary compared with *A. astacus*.

Crayfish are generally regarded as predominantly nocturnal animals (Abrahamsson 1981; Lozán 2000; Musil et al. 2010). In our stream, the lowest movement probability of crayfish was also recorded within day-time intervals when the levels of light intensity were high. In both species, however, we found the highest movement probability not at night, but around dawn. In *A. astacus*, moreover, we recorded greater distances moved at dawn compared with the day and night. The possible explanations for this are twofold: the first is that individuals left the safety of their shelter gradually at different times during dusk and at night, and that the impending dawn forced them to find a suitable shelter. Buřič et al. (2009b) also report the greatest numbers of moving spiny-cheek crayfish within a time interval that included dawn.

The shift of the peak of migrations to morning hours at our locality may also be influenced by the increased predation pressure during the night hours. Up to 400 mm long individuals of brown trout (*Salmo trutta*) are common in the stream under study, and these trout are mostly active at night (Musil et al., unpublished radiotelemetric data), which increases the probability of encounters with crayfish at night. Many authors have reported that a temporarily higher predation pressure may alter the behaviour of prey, including temporal shifts in movement (e.g. Stein & Magnuson 1976; Fraser et al. 1993; Heggenes et al. 1993; Valdimarsson et al. 1997; Bonnot et al. 2013).

Local activity is another measure for evaluating animal behaviour. Whereas movement by definition entails relocation, local activity also covers motion that is confined to a single place (Lucas & Batley 1996; Bubb et al. 2002; Thiem et al. 2010). This motion could be associated with feeding, shelter upkeep or, for example, defence against invaders (Bubb et al. 2002; Robinson et al. 2000).

The temporal distribution of local activity during the diel cycle differs from that of movement in our study. Local activity reached its maximum values at dusk in *A. astacus* and at dusk and at night in *A. torrentium*. This distribution of activity supports the assumption that both crayfish species under study are nocturnal and also lends support for our suggestion that the highest rates of relocation of individuals in morning hours may be boosted by predation pressure. As local activity may be realized within the confines of a shelter, it can be assumed to be less risky than movement, which requires venturing from safety.

Over the course of our study, we found no sex-related differences in behaviour. We assume, however, that this is connected to the period in which we con-

ducted our monitoring, during which crayfish behaviour was not affected by migrations connected to mating or by the non-activity of females carrying eggs. Bohl (1999) points out that females move extensively after the release of juveniles in summer and very little during the egg-bearing period (particularly in November), as well as during hatching, which takes place in May and June. Males may move over long distances in the mating period, which is in October.

We found that smaller *A. astacus* moved more frequently, perhaps because larger individuals are usually stronger in intraspecific competition, for example, when fighting over a shelter (Nakata & Goshima 2003). Larger individuals are, therefore, more likely to displace smaller ones from their shelters (Webb & Richardson 2004; Loughman et al. 2013). Smaller individuals are also usually forced to occupy suboptimal microhabitats with inferior foraging conditions, which may result in more extensive spatial exploration in search of food (Nakano 1995). By contrast, larger individuals of *A. torrentium* in our study exhibited greater local activity. *Austropotamobius torrentium* prefer to shelter under stones and boulders (Vlach et al. 2009a). Presumably, smaller individuals are able to make use of existing gaps between stones, whereas larger individuals have to create and maintain a sufficiently large cavity under stones. In addition, while smaller individuals may be forced to search for food further away from their shelters, larger individuals may reside in more suitable places with better availability of food, which may be the reason behind their lower movement rates and higher local foraging-related activity.

Of the environmental variables analysed in our study, we are not surprised by the statistically significant relation between light intensity, movement probability and local activity. This variable, of course, is directly linked to the time of day. Over the duration of our study, we found a positive effect of temperature on movement probability and distances moved by *A. astacus*, which is a common phenomenon in poikilothermic organisms, in which higher temperatures (unless they are excessive) increase the rate of metabolism and energy requirements (Ultsch 1989). In crayfish, a positive correlation between the extent of movements and temperature has been found, for example, in *Pacifastacus leniusculus* (Bubb et al. 2004).

The behaviour of crayfish was also influenced by the weather. Under a clear sky, the individuals of both species moved over longer distances. *Astacus astacus* moved significantly over longer distances during periods of stable atmospheric pressure, and *A. torrentium* moved more frequently under low turbidity conditions.

These three environmental variables are correlated with each other because the higher variability of atmospheric pressure usually indicates less stable, rainy weather, higher flow rates and higher turbidity. These results suggest that crayfish movement during unstable and rainy weather is low. It is reasonable to assume that in rainy weather, crayfish are more likely to stay hidden in their shelters to avoid the risk of a sudden increase in discharge (e.g. during a flash flood), which can cause involuntary downstream drift (Momot 1966) accompanied by high mortality of individuals being swept away (Royo et al. 2002). Low movement of *A. astacus* during high water levels is also described by Bohl (1999).

The effect of the water level by itself did not turn out to be significant in our study. We assume, however, that this was caused by the absence of high discharge events in the monitored period, and by a more or less constantly decreasing water-level trend over its entire duration.

Our results also highlight the extent of migrations taking place in the first days after capture and tagging. Although our results point to possible differences in the behaviour of the two species, the interspecific differences detected are statistically non-significant, probably as a result of high intraspecific variation and the relatively low number of tracked individuals.

Robinson et al. (2000) point to the fact that the capture and tagging of individuals can elicit a fright response manifested by an elevated tendency to move and migrate away from the original place of occurrence in the first few days after tagging. Despite the limited extent of our dataset, our results indicate that a fright response does occur, and that the extent of fright-induced migrations can be considerable. We took great care not to disturb individual crayfish shelters beyond the minimum extent necessary, tagged the crayfish carefully, and tried not to cause unnecessary stress before releasing them back into the place of capture. However, they still often relocated far away after their release back into their original position, and the extent of migrations in the first four days following capture frequently exceeded that recorded over the entire telemetric study. This confirms how difficult it is to interpret data on the ranging behaviour of crayfish in traditional mark-recapture studies, in which recorded values can be overestimated compared with natural behaviour. In telemetric studies, it is advisable not to include the first few days after tagging in analyses of regular behaviour.

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Author contributions

Conceived and designed the experiments: JM TD JS. Performed the experiments: TD JM JS TB DŠ MB EB. Analyzed the data: PV TD JM. Wrote the paper: TD PV JM JS SIJ OA.

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