

Collembola in the hyporheos of a karstic river: an overlooked habitat for Collembola containing a new genus for the UK

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Abstract

Collembola are well known to occur in soils and litters, as well as inter-tidal habitats, caves and tree canopies, but are not normally thought of as members of the groundwater community. Here we report on studies of the colonisation of gravels in the hyporheic zone under the river Skirfare, a karstic river in Yorkshire. Unexpectedly, flooding during the second experimental run allowed a comparison between low- and high-flow conditions. Five species of Collembola were found in permanently saturated habitats 30 cm below a river bed. One of these species, *Hymenaphorura nova*, is the first record of its genus in the UK, and appears to be a groundwater specialist. The other named species (*Anurida granaria* and *Mucrosomia* (= *Cryptopygus*) *garretti*), *Deuteraphorura cebennaria* (= *D. inermis*) have wider distributions but are often associated with mines and caves; *D. cebennaria* is known to occur in the Scoska cave, whose waters feed into the Skirfare shortly upstream of the experimental location. There was some evidence of differences between dates (more first instars and fewer adults after the flood) and between sediments (adults found mainly in coarser materials, first instars in finer sediments), but the statistical significance was weak ($0.1 > p > 0.05$). The distribution of animals was not clumped (as is typical for Collembola) but followed a Poisson distribution, suggesting it to be primarily random. These results agree with previous authors who suggested that several species of Collembola may live for prolonged periods wholly submerged. The random distribution and their water-repellent cuticles suggest a model of eggs or early instars being washed underground following groundwater, surviving underwater for prolonged periods. We do not have evidence that these Collembola are able to complete their life cycle underwater.

Keywords: Collembola, hyporheic, interstitial fauna

1. Introduction

The biodiversity of ground-water has received little attention in the UK, but the introduction of the Water Framework Directive (WFD) and Groundwater Directive (GD) has refocused attention on this overlooked system (Holzwarth 2002). The WFD requires EU member states to achieve groundwaters of good ecological and chemical status by the year 2015; to achieve this we need to understand these systems. The interface between rivers and groundwater is a volume called the hyporheic zone (HZ). Here we present some data from an experimental study on the fauna of the hyporheic zone of a karstic river in Yorkshire. The aim of this

study was to test experimentally how different grain sizes influence faunal assemblages in the hyporheic zone, by studying the colonisation of defaunated microcosms containing gravels of differing grain size.

Elsewhere in the UK the hyporheic fauna has been found to be dominated by microcrustaceans, along with insect larvae, nematodes and oligochaetes. These groups were recorded from this experiment and will be reported separately (Dunscombe 2011). Additionally, submerged microcosms were found to acquire a sparse but predictable community of Collembola, clearly distinct from the well-known community of water-surface Collembola (e.g. *Podura*, *Sminthurides*). Deharveng et al. (2008) reviewed freshwater Collembola, defining 4 groups: Epigean hydrophilous species (water-surface species), cryophilic (snow/ice associated) species, cave hydrophilous species, and a predicted but unconfirmed fourth category: interstitial hydrophilous species, living their lives submerged in groundwater. In fact Collembola have been recovered from submerged sediments, but the species concerned have not been considered capable of living to reproduction underwater.

Bketschko & Christian (1989) recorded 42 species of Collembola from the water-filled pore spaces under the stream bed in about 10% of collections from an alpine stream. Surprisingly, the majority were mainly epigeous rather than aquatic species (dominated by *Lepidocyrtus lanuginosus* and *Folsomia quadrioculata*). However their third most common species, *Agrenia bidenticulata*, is typically found in streamside habitats under stones. They concluded that epigean species are regularly washed down to considerable depth in sediments without apparent damage. Remarkably, there are records of a dry land springtail *Seira ferrarii* from sediments underneath the Mediterranean and Black seas (Da Gama 1966, Jacquesmart & Jacques 1980).

It is known that non-aquatic Collembola can hatch and survive underwater when compelled to do so: Britt (1951) immersed *Ceratophysella armata* eggs into water and found that the eggs hatched normally. The first instars survived under water for up to 40 days, but with a water-repellent cuticle that prevented them from re-entering water should they leave it.

Despite these observations, there remains no evidence that any Collembolan can complete its life cycle under water. Here we characterise the Collembola found in the saturated hyporheic zone under a river bed, and discuss whether these are better considered as an interstitial hydrophilic community, or merely an accidental assemblage.

2. Sites and Methods

Work was undertaken in the River Skirfare, Littondale, Yorkshire, UK, OS grid reference SD923724 (54° 8' 50" N, 2° 7' 4" W; Fig. 1), a karstic area known for caves. The Skirfare valley was glaciated during the Devensian period and is regarded as the UK's best example of glaciokarst. At the study site the river is perennial and fed by numerous springs, but above this point flow becomes seasonal. The weather station (location: 54° 8' 30" N, 2° 6' 18" W; Elevation: 260 m) located in the village of Arncliffe (1.5 km downstream from the study site) provided data on air temperature and rainfall in real time. The rainfall in the area averages approximately 90 mm per calendar month, but during the second experimental run the site received an unprecedented 390 mm of rain in one calendar month.

The microcosms used were plastic cylinders (volume 1 litre) sealed top and bottom but with lateral holes for ingress and collection of water. These were filled with one of four sediment treatments (differing in grain sizes) and inserted 30 cm under the river bed. The sediments used were either original riverbed substrate (control) or artificial substrates in the following



Fig. 1 Map outlining the location of the field site.

size categories; sand ('fine'), gravel ('medium') and pebbles ('coarse'). (Fig. 2). The control sediment control sediment was similar to 'coarse' but with fewer large stones. To defaunate the control treatment, the riverbed sediment was elutriated in a plastic tray to remove organic material, and the sediment then rinsed using a pressure hose.

Experimental microcosms were inserted into riffles (gravelly shallow regions between deeper pools) in holes in the river bed that were randomly located (subject to the constraint of being approximately mid-stream, and at least 1 m from the neighbouring experiments). Each experimental unit comprised a hole 30 cm deep in the river bed containing 4 microcosms, one per treatment, in random order. The design was replicated within each riffle (4 units per riffle), in space (3 riffles) and time (2 visits: September 2009 and November 2009). This gave a factorial design of 4 substrates * 4 replicates * 3 riffles * 2 visits = 96 microcosms. The spate on November 2009 removed all 16 replicates from riffle 3 for visit 2, leaving a final total of 80 microcosms.

The microcosms were left in the riverbed for 4 weeks, then harvested: first a water sample was taken from within each chamber for chemical analyses, then their contents were removed for faunal identification. Water was collected by pump, then transported in ice to the lab then frozen while awaiting chemical analysis. Ions were determined by HPLC, organic matter

by mass loss on ignition. Sediment was removed directly into a plastic bag, preserved with ethanol, sieved onto 63 μm mesh sieve then sorted by eye under a dissecting microscope to isolate all fauna. Collembola were initially keyed with Hopkin (2007), then Fjellberg (1998) and Pomorski (1998).

As this is a rather small dataset describing unusual observations, a lower level of statistical significance than usual was used, with $p < 0.1$ being defined as 'weakly significant'. Chemical variables received a standard two way ANOVA, Collembola were tested by the non-parametric Kruskal Wallis test using SPSS 16.

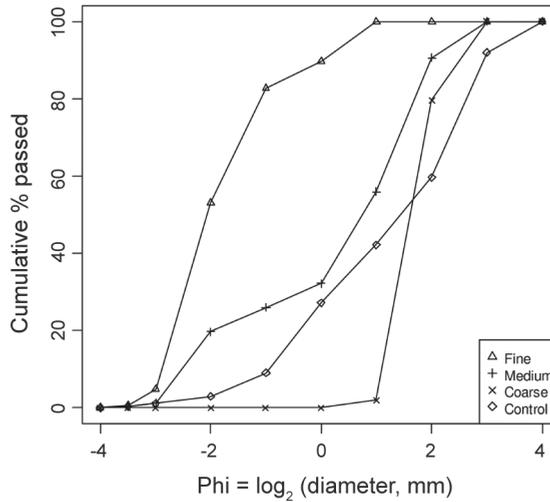


Fig. 2 Granulometry of the sediments used in the experiment.

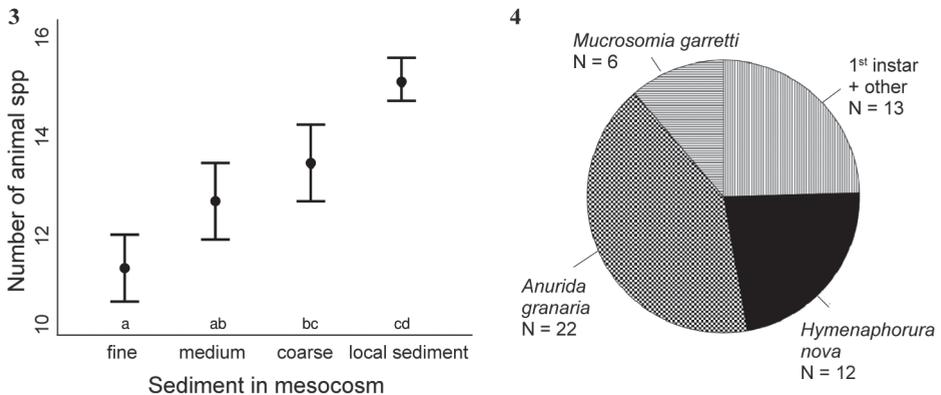


Fig. 3 Faunal species richness as a function of sediment grain size, mean +/- standard error. Means followed by the same letter do not differ by Duncan's post-hoc test.

Fig. 4 Composition of the Collembolan fauna of the Littondale hyporheos.

3. Results

Water chemistry within the microcosms (Tab. 1) did not differ between gravel size classes, except for residual organic matter which was highest in coarsest gravels ($p < 0.01$). The interstitial assemblage within each microcosm was dominated by micro-crustacea, of which there were 49 species. Micro-crustacean species richness varied significantly ($p < 0.01$) with grain size, being lowest in the finest sediment (Fig. 3).

Tab. 1 The water chemistry within the submerged microcosms. SD = Standard deviation.

Parameter	pH	Dissolved Oxygen	Conductivity	Residual OM	Ca ⁺⁺	Cl ⁻
Units	-	mg l ⁻¹	μSm cm ⁻¹	% mass	mg l ⁻¹	mg l ⁻¹
Mean	7.9	9.7	280.6	7.1	41.4	6.6
SD	0.2	1.3	6.0	4.6	11.0	0.8
Parameter	K ⁺	Mg ⁺⁺	Na ⁺	NH ₄ ⁺	NO ₃ ⁻	SO ₄ ⁼
Units	mg l ⁻¹	mg l ⁻¹	mg l ⁻¹	mg l ⁻¹	mg l ⁻¹	mg l ⁻¹
Mean	0.29	2.35	5.8	<0.1	0.71	4.06
SD	0.57	1.11	3.7	NA	1.18	1.03

From 37 out of these 80 microcosms, a total of 53 separate springtails (Collembola) from 5 species (Fig. 4) were collected. The commonest were *Anurida granaria* (Nicolet, 1847), *Hymenaphorura nova* Pomorski 1990, and *Mucrosomia* (= *Cryptopygus*) *garrettii* (Bagnall, 1939). Twelve out of the 53 Collembola collected (23%) were first instars that were probably *Anurida granaria* but were too small to be confidently identified; we also found one *Deuteraphorura cebennaria* (= *D. inermis*) (Gisin, 1956). A *Folsomia* collected resembled *F. agrelli* (Gisin, 1944). These latter 3 have been pooled under 'other' in Figure 4. The majority of records were single individuals, with three records of 2 and one of 3 individuals. These figures agree almost exactly with predicted values from a Poisson distribution (Tab. 2). Several of these individuals (principally the *Anurida*) had a silvery sheen caused by an air bubble trapped next to their cuticle. These densities correspond to a mean value of 0.60 (SE 0.08) animals per litre of hyporheos (not per litre water). If they were confined to the upper 30 cm of the hyporheos this would give a vertical projected density of approximately 180 Collembola per m², but this value could be considerably higher if they continue to occur deeper into the groundwater.

Tab. 2 Observed frequency distribution of Collembola numbers compared to a fitted Poisson distribution.

Number of animals per sample:	0	1	2	3
Observed frequency	352	44	3	1
Expected from Poisson model	350	46	3	<1

The heavy rain prior to the second sampling date reduced the ionic strength of groundwater, e.g. sodium fell from 9.3 to 1.6 mg l⁻¹ ($p < 0.001$). There were corresponding differences between sampling dates in Collembola numbers, although the significance levels were weaker. Post-spate there were fewer adult *Anurida* ($p < 0.05$), but (weakly) more *Mucrosoma* ($p < 0.1$) and first instars ($p < 0.1$) (Fig. 5). There were no statistically significant differences between Collembola densities in the four gravel sizes, although the adults were mainly found in the coarser sediments, while first instars were commoner in the finer sediments.

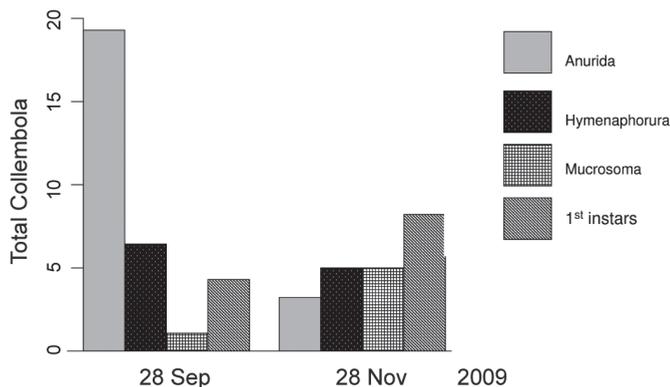


Fig. 5 Collembola totals by date.

4. Discussion

These results confirm that Collembola can be consistently collected from permanently waterlogged sediments, a largely overlooked habitat for this group. These results may be compared with Bketchko & Christian (1989), who collected fauna from under the Oberseerbach stream bed by suction pumps. It is impossible to compare densities due to this methodological difference, but the species found were quite different. Under the Oberseerbach only ten percent of samples held Collembola, which were dominated by the surface-dwelling genera *Lepidocyrtus* and *Entomobrya* and almost lacking eu-edaphic species. By contrast under the Skirfare, 46% of samples held Collembola, and these were entirely eu-edaphic species, principally *Anurida* and *Hymenaphorura*. This difference may reflect the upwelling of groundwater in the Skirfare compared to more superficial flow in the Oberseerbach, but more systems yielding submerged Collembola need to be studied before any general conclusions may be formulated. Beyond the discovery of *Deuteraphorura cebennaria* in the Skoska cave, we have no data on the distribution of Collembola in the Skirfare catchment, but there is no reason to doubt that epi-edaphic species such as *Lepidocyrtus* and *Entomobrya* occur widely.

Two observations emerge from these data which argue against these Collembola having a wholly submerged lifestyle. The first is statistical, in that the data show no sign of clumping or overdispersion, fitting instead accurately to a Poisson distribution. Wherever Collembola have been studied elsewhere they exhibit highly clumped distributional data (Usher 1969, Shaw & Usher 1996, Hopkin 1997, Negri 2004), attributed to (among other things) aggregation pheromones (Joose et al. 1977). A Poisson distribution implies one dominated by randomness, which suggests that the animals were washed passively and haphazardly through the system.

The second is the laboratory observation that these animals (especially but not exclusively *H. nova*) had water-repellent cuticles. This is commonly observed in Collembola, even when they hatch under water (Britt 1951), but is anomalous for fully aquatic invertebrates. The layer of trapped air around some *Anurida* is reminiscent of 'physical gills' used to respire by aquatic Coleoptera etc. (e.g. Popham 1954).

Deharveng et al (2008) predicted submerged interstitial habitats as a possible fourth community of freshwater Collembola. The extent to which this can be considered a 'normal' habitat for them is however debateable, since there remains no proof that life cycles are completed during submergence. The simplest model compatible with our results is that the accidental submergence and downwashing of surface-dwelling Collembola is a routine if uncommon occurrence, and that they can survive underwater for species-specific lengths of time. The composition of this immersed community will depend on the hydrology, with riparian/eu-edaphic species being especially associated with upwellings of deep groundwater.

5. Acknowledgements

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6. References

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