NIWA
Taihoro Nukurangi

Field guide to New Zealand phreatoicid isopods

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WHAT ARE PHREATOICIDS AND WHY ARE THEY IMPORTANT?

If these freshwater crustaceans were as large as tuatara or even wetas, they would be a well-known, iconic group within New Zealand's special biota because of their ancient lineage. Phreatoicid (free-ato-ik-id) isopods (= like foot) are truly living fossils: the suborder Phreatoicidea is an ancient gondwanan group that originated at least 325 million years ago (mya) in the Palaeozoic, and have changed little morphologically since that time (http://www.personal.usyd.edu.au/~buz/popular.html; Wilson & Edgecombe 2003; Wilson in press). For a simple comparison, phreatoicids have been around for at least 1.5 times longer than tuatara and 100 million years longer than wetas (tuatara and wetas are known from 220-250 million years ago (Russell 1998)). Thus, they are modern-day peculiarities, occurring on the remnants of ancient Gondwana: Australia (apparently their evolutionary epicentre, with freshwater fossils dating from the Triassic (200-250 mya)), New Zealand, South Africa and India. They occurred in Antarctica is the past (freshwater fossils from the Jurassic (145-200 mya)), but are not yet known from South America.

Figure 1. Like all New Zealand phreatoicids, species of the endemic genus *Notamphisopus* are unpigmented, lack obvious eyes and are similar in general shape and body plan. The large first pereopods or legs show that this is a male specimen.

Figure 2. Phreatoicids walk upright over debris, flex their bodies to the side when turning, and cannot swim.

Since New Zealand separated from Gondwana c. 80 million years ago, our species have evolved independently. The fauna, as we currently know it, comprises just nine species in three genera, all endemic and restricted to freshwater habitats. However, our knowledge of New Zealand phreatoicids is sketchy because they live in habitats that are usually not collected by ecologists, the species are difficult to distinguish, the guides for their identification are largely inadequate, and few sightings of these animals have been reported since first discovered more than 60-100 years ago. Thus, although there are almost certainly several new, undescribed species in New Zealand, their presence adds to the confusion when trying to identify specimens using the available keys.

Phreatoicids belong to the Isopoda, a group with hugely diverse body forms and representatives inhabiting the ocean's depths to subalpine zones. Few common names for isopods are available, but the group's most familiar members include sand lice, fish lice, wood-boring gribbles, and terrestrial pill bugs and slaters or woodlice (all unrelated to true lice). Although an ancient group, phreatoicids are unremarkable in appearance (Fig. 1) and relatively small (up to c. 20 mm long). New Zealand species appear to lack eyes and any significant body pigmentation (Figs 1-2), and live in cryptic habitats, so they are easily overlooked. Phreatoicids are slow moving, cannot swim and lack dispersal and resistant stages in their life histories, making them especially vulnerable to habitat modification and land-use activities.

PURPOSE OF THIS GUIDE AND THE STUDY

As a result of these knowledge gaps and the human pressures on their habitats, the conservation status of our phreatoicid fauna remains largely unknown. This guide is one step in determining the present-day distribution and conservation status of epigean (on the surface, not subterranean) phreatoicids, the distribution and nature of significant phreatoicid habitat, and the vulnerability of each species and its habitat to human pressures.

Funded by the Department of Conservation's TFBIS programme, the overall study will achieve this through several stages. Stage 1 gathered all available records of phreatoicids and developed a preliminary field-guide to all known phreatoicids based on these data. During Stage 2, the known distribution ranges of each species were searched intensively to gather detailed information on location, habitat, other species, land-use and vegetation. The resultant information was used to produce this guide for fieldworkers (DoC staff, unitary authorities, universities, research organisations, Fish & Game officers), so that a more comprehensive understanding of phreatoicid diversity, distribution and conservation status can be developed by encouraging you and others to collect and report specimens of these animals during the 2007-08 field season.

Data from all collections will be added to the Freshwater Biodiversity Information System (FBIS), a large, public-access database managed by NIWA, to make this valuable biodiversity information available for better decision-making by resource and biodiversity managers. In the future, links to FBIS will provide easy access to detailed distribution mapping of records for all species. These data will be supported by identified voucher specimens lodged in New Zealand's museum holdings and available for present and future national and international researchers.

In addition, dedicated pages on NIWA's biodiversity website will outline the resulting knowledge of phreatoicids and why they are important in New Zealand. Habitats for each New Zealand species will be specifically described and threats to these habitats identified. The web pages will also present photographs and illustrations of local species, their habitats, general geographic distributions, abundances and conservation status. An improved illustrated key for species identifications will also be accessible via these web pages

How you can help

Your help in searching for and collecting phreatoicids can make a significant contribution to our knowledge of this fascinating, ancient group of native crustaceans. By co-ordinating and focussing the searching efforts of several people working in the field over this summer, we

hope to develop a comprehensive understanding of these animals. Here's what you can do to help:

- Read this guide.
- Pass it on to others who might also be interested and/or have the opportunity to look for phreatoicids.
- Use the information in the guide to identify likely habitats for phreatoicids.
- Try collecting phreatoicids and record what you do and don't find.
- Keep any specimens, preserve and label them, and send them to NIWA along with a record of everywhere that you searched.

The NIWA team will examine all specimens that you send to us and advise you of the results (there may be some delays in getting the results back to you if we receive lots of specimens). We will enter the data, including your name as the collector, into the FBIS database that will appear on the web. Our team may also re-visit the site of your collections to gather extra specimens for further analysis.

What do we know about phreatoicids?

GENERAL

New Zealand's phreatoicids require permanent water for all of their life stages (some species appear capable of surviving in with damp moss or soil for considerable periods) and are mostly associated with groundwaters, either inhabiting aquifers or living in springs, seeps, spring-fed streams and wetlands. Within New Zealand, phreatoicids appear confined to the South Island, Stewart Island and to Ruapuke Island, the large island at the eastern end of Foveaux Strait. NIWA has specimens of apparently new species from northwest Nelson, the West Coast and inland Canterbury, and has completed some more intensive collecting in parts of Otago and Southland (Fig. 3).

EPIGEAN (SURFACE-DWELLING) SPECIES

Six of our nine known phreatoicid species inhabit epigean (above ground) habitats. Two species occur only on Stewart and Ruapuke Islands, apparently one restricted to each island. The remaining four described epigean species are known from coastal and inland Otago and Southland. All six belong to the genus *Notamphisopus* (note-am-fie-so-pus)(George Nicholls named the genus, which translates to "southern like-foot"). Another possibly undescribed species occurs in the central Canterbury foothills and we have additional unidentified specimens from the central to northern West Coast.

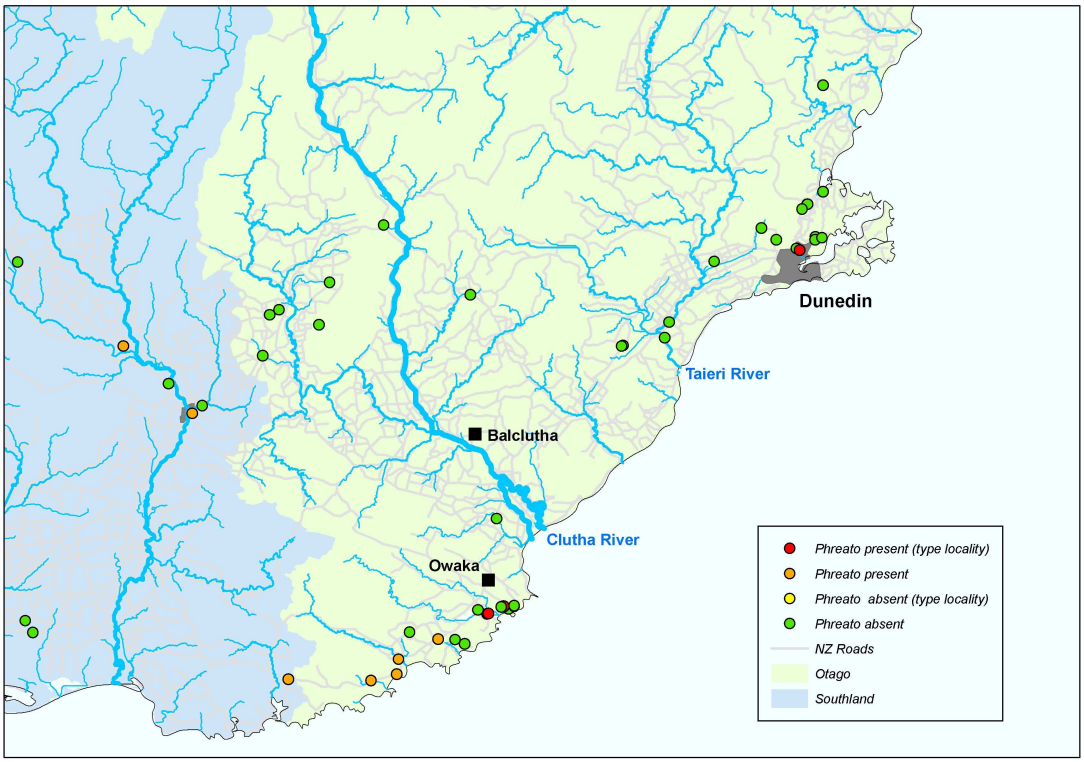

Figure 3. Positive and negative records for phreatoicids in Otago and Southland, based on recent collections, mainly by NIWA.

Almost nothing is known about the biology or ecology of these species. Like all isopods, female phreatoicids brood eggs and small juveniles in their ventral brood pouch. We know nothing about the growth rates of New Zealand species, but, assuming that they're similar to our groundwater species (Wilson & Fenwick 1999), a life-span of c. three years seems likely.

Our recent collecting indicates that epigean phreatoicids are associated with groundwaters, living in seeps, springs, spring streams, Sphagnum bogs and wetlands. They cannot swim, so seek out sheltered habitats amongst the roots of macrophytes and surrounding trees and ferns, or mats of decaying grasses hanging into streams. Within heavily shaded, spring-fed forest streams, phreatoicids live amongst dense leaf and twig litter and plant roots close up against the banks. One species also seems much at home living in the drains of a peat bog, where conditions are very acid (pH = 4.2), whilst another was found in slightly anoxic conditions (DO = -0.1mg/l), as well as amongst saturated sphagnum mosses. Because phreatoicids appear consistently associated with fine sediments, dead plant matter, tree roots exposed in stream beds or sphagnum, they are probably detritivores and/or root herbivores.

Figure 4. Pheatoicids occur in native beech forest, here found in a roadside seep near Lake Hauroko (detail of the habitat shown below).

Figure 5. Closer view of the roadside seep on the beech forest margin, showing the very small, shallow nature of the seep and its association with forest litter and tree roots.

Figure 6. View of a shallow seep on the floor of lowland forest near Purakanui Falls (Catlins) that was inhabited by phreatoicids.

Figure 7. Phreatoicids were present in these small seeps from the heavily forested banks of the Purakanui River in the Catlins.

Figure 8. A small perennial stream in this forested valley in the coastal hills near Pounawea drains shallow groundwater from surrounding land and is inhabited by populations of phreatoicids (see Fig. 9 for detail of the habitat).

Figure 9. The small, heavily shaded stream with its mixed clay and gravel bed and banks matted with dense plant roots near Pounawea is inhabited by abundant phreatoicids living amongst roots and gravel and under cobbles.

Figure 10. Abundant phretoicids occurred in this small tributary to the Tahakopa River draining forested hillsides inland from Papotowai (Catlins) (see Fig. 11 for detail of the habitat).

Figure 11. Phreatoicids were common amongst the roots of dense regenerating forest on the banks of this heavily shaded, muddy stream bed near Papotowai (Catlins).

Figure 12. Water seeping through the water-logged soils below this small valley near Lumsden appears to sustain this densely vegetated, perennial stream, providing excellent habitat for phreatoicids (see Fig. 13 for detail of the habitat).

Figure 13. A successful hunt for phreatoicids under grasses, other vegetation and amongst their roots beside a culvert over this small stream near Lumsden.

Figure 14. A groundwater fed stream on the Waimea Plains near Riversdale (upper Mataura River valley) that provided good phreatoicid habitat amongst the deeper stems and roots of grasses and macrophytes (notably monkey musk).

Figure 15. A similar perennial groundwater fed stream inhabited by phreatoicids.

Figure 16. Drainage channels cut deep into the peat just downstream of Southland's remarkable Bayswater Wetland Reserve (near Otautau) contained very dense populations of phreatoicids.

Figure 17. A live, but apparently inactive phreatoicid found in damp moss at the Bayswater Wetland Reserve, Southland.

HYPOGEAN (SUB-SURFACE DWELLING) SPECIES

The three known hypogean (underground, including caves and alluvial aquifers) species occur in mid Canterbury's underground aquifers. One of these, *Phreatoicus typicus* (Fig. 18), the first species ever discovered in this suborder (so unusual was it at that time, that leading

taxonomists initial debated whether it was an amphipod or an isopod), is moderately well known (Wilson & Fenwick 1999), although details of its distribution remain sketchy. First described by Charles Chilton in 1883 from Eyreton, near the large Waimakariri River just north of Christchurch, this phreatoicid grows to almost 20 mm long and inhabits Canterbury's coarse alluvial aquifers from immediately beneath riverbeds to aquifers >20 m below soil surface and to >20 km from the nearest river. Eyreton is its northern-most record and Burnham its southern-most known location. Two studies (Sinton, 1984; Scarsbrook & Fenwick 2003) found large numbers of this deposit-feeding phreatoicid immediately downstream of a wastewater disposal site, indicating that, although recorded infrequently, this species is probably not threatened at present.

The other two known subterranean species appear to live in equivalent habitats in the vicinity of Orari and Temuka, but there are very few records. *Phreatoicus orarii* was taken from shallow groundwater in the Orari River valley, while *Neophreatoicus assimilis* lives in the same habitat and a spring just a few kilometres to the south at Winchester and Temuka (c. 5 km apart and <30 km from the Orari River valley).

WHAT DO THEY LOOK LIKE?

Phreatoicids look like a hybrid between a sand hopper (amphipod) and a woodlouse (isopod), having the lateral compression more typical of an amphipod's body shape, and relatively similar legs of an isopod. When alive, they lack body pigment, ranging in colour from creamy white to light pinkish brown, and have no obvious eyes, although some reflective remnant of eyes seem to remain (Figs 1, 19). Often, parts of their bodies will be stained rusty brown, especially the margins of segments and areas covered with setae and spines. One half of an individual may appear cleaner or lighter than the other, because these animals shed the anterior and posterior halves of their exoskeleton at different times as they grow. Large specimens of both epigean and hypogean species reach almost 20 mm in length, and we could find unknown species that may grow to no more than 5 mm as adults, as in some Australian species.

When disturbed, specimens lie passively on their sides in a slightly curled posture (C-shaped). When is water, they soon recover to stand upright and walk about moderately slowly (they cannot swim), flexing laterally as they turn from one direction to another.

Figure 18. *Phreatoicus typicus*, a 20 mm long hypogean phreatoicid from 20 m below ground surface in the Canterbury aquifer taken from Templeton, near Christchurch.

Figure 19. Characteristic curled posture of disturbed phreatoicid either in water or out of water. After a minute or so undisturbed in water, individuals usually right themselves and begin to walk about.

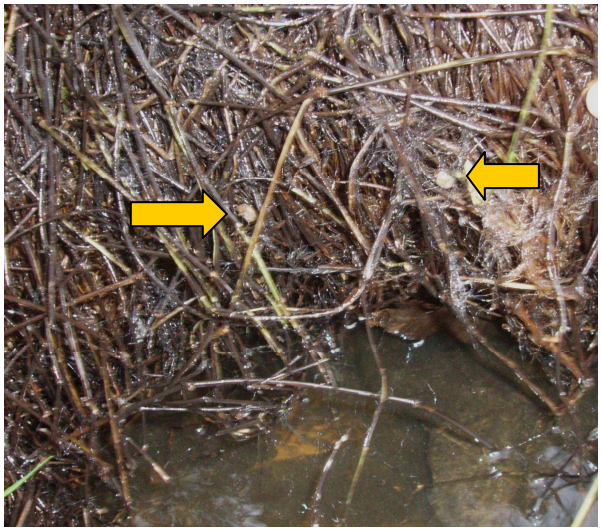

Figure 20. Curled phreatoicids exposed when dead grass mats were lifted from the water.

COLLECTING PHREATOICIDS

WHERE TO LOOK FOR PHREATOICIDS

We know little about where New Zealand phreatoicids live, both in terms of their habitats and geographically. They seem to inhabit permanent waters, but we know little else. What we do know of phreatoicids indicates the following characteristics of their habitats:

- Most common in Southland and Otago, but occur on the West Cost to northwest Nelson, and north along the eastern side of the foothills to at least the Rangitata River.
- Occur from near sea level to least 250 m above sea level (but may live at higher altitudes).
- Always in water or rarely in damp mosses when water levels have dropped (they appear to be able to survive dry periods by resting in damp moss, soils, plant matter).
- Appear to inhabit surface waters where groundwater influences are strong: springs, wetlands, spring-fed streams (road-side drains to larger spring-fed streams), even shallow (1-2 cm deep) depressions in saturated soils on the forest floor, streams in caves. Some streams inhabited are small (<20 cm wide, scarcely flowing), whereas others are 4-5 m wide, 1-2 m deep and flowing quite swiftly (but here the phreatoicids occur within sheltered, muddy marginal microhabitats amongst roots of macrophytes, grasses and adjacent trees).

- Live in shallow water (1-80 cm depth).
- Appear to avoid streams susceptible to flooding (they cannot swim) or frequent drying.
- In faster flowing streams, associated with roots of aquatic plants.
- Usually in habitats that are heavily shaded (i.e., full canopy cover), but also found under dense grass or aquatic plant cover in streams flowing through open farm land.
- Associated with plant or organic matter (mosses in bogs, peat deposits, leaf and twig litter in forest streams, fern and punga roots, roots of non-native trees such as Macrocarpa).
- Tolerate widely differing water conditions (e.g., pH 4-7; anoxic to fully oxygenated).

Figure 21. Phreatoicids were abundant amongst Macrocarpa (tree) roots in this heavily shaded, man-made drain. Note, the exposed dense gravel in the banks indicates close connectivity with groundwater.

Figure 22. The soft mud and rooted vegetation in this man-made drain was another good habitat for phreatoicids in Southland.

Figure 23. You might not think to try, but phreatoicids were here! This very muddy remnant of a small stream during a very dry period on Ruapuke Island yielded several phreatoicids, especially from amongst the roots of surrounding grasses.

HOW TO COLLECT EPIGEAN PHREATOICIDS

Dip nets and kitchen sieves are great for collecting these sluggish, bottom dwellers. Extracting the phreatoicids from the plant material is the difficult part, however. This is best

done using two square, 2 litre ice cream boxes. White ones are by far the best because this colour makes it easier to distinguish the movements, characteristic shapes and dull brown colour of these animals from the darker and lighter plant debris and rubble. The light colour is also helpful in the poor light (dense bush, under road culverts) where we often search for these beasts.

1. Start by working a sieve or net through the submerged stems roots of aquatic plants, along the bottom and amongst the roots of terrestrial plants or by catching the finer plant debris from small streams, springs, etc. Quite large quantities of material are easily collected.

2. Dump a small amount (half a cup) of the collected material into one ice cream box and add clean water (muddy water makes seeing anything difficult) to half fill the box. It's usually best to work in bright light to make it easier to spot these dull coloured animals. Pick out any twigs, leaves, roots, check them quickly for adhering phreatoicids, then discard them.

3. Briskly pour most of the remaining material into the second box (plant debris and phreatoicids are less dense), leaving behind any sand and gravel. Inspect the retained sediment carefully (best with c. 1 cm of clean water) for any phreatoicids, before discarding it. Repeat as necessary until your have a box of clean litter and animals.

4. Next, add more clean water, swirl the contents to re-suspend most of its debris, allow it to settle for 1-2 seconds, then pour enough of the suspension into the empty box to cover the bottom by c. 1-2 cm, but ensure that the debris is sufficiently sparse that it covers no more than c. half of the bottom. Any phreatoicids will either sink to the bottom or cling to twigs and leaves, so pick out, inspect and discard larger leaves, twigs, etc., before pouring off (discarding) the floating finer debris.

5. More clean water and agitating/stirring might be necessary to winnow the debris to a low density of finer particles (Fig. 25), so that phreatoicids (slightly denser than most plant debris) become visible on the box's bottom. Any specimens found can be placed in a screw top jar of water as you sort.

6. Repeat this procedure until you have searched each collection of debris. Phreatoicids are not easy to spot, so spend a few minutes carefully scanning the bottom of each box for both large (up to 15-20 mm long) and small specimens (3-4 mm long) before discarding the debris.

Figure 24. Step 1 in processing a sample: dump the contents of your sieve into a white ice cream box. Here, in dense bush in Dunedin, the samples were taken closer to abundant clean water and brighter light for subsequent steps.

Figure 25. The result of Step 5 should be a sparse layer of debris in c. 1 cm of clean water, so that any phreatoicids can be spotted. One of the two medium-sized specimens in this sample is obvious, the other will show itself when it moves.

Figure 26. A concentrated sample from several sieve scoops and multiple washings.

Figure 27. Another concentrated sample, this time containing phreatoicids (light coloured), similarly sized idoteid isopods (brown with triangular posterior) and amphipods (<half as long as the isopods).

If you don't get any from your first collection, make another from a slightly different habitat. If, after checking 3-4 collections, no specimens are found, they may still be present at the site, but either at low densities or in a slightly different habitat. Where they are present, they are often quite abundant, so that 5-10 specimens may be found in a single net collection. It also takes a bit of experience to develop a search image and to become good at seeing phreatoicids

in your collection, so be patient and try a few more habitats and locations. We just don't know where they'll be found, nor how many new species will turn up.

PRESERVING YOUR CATCH

For our purposes, any specimen is better than none, so long as it is labelled with the collection location, collector, date of collection, habitat, position and altitude, and any other information. If possible, please provide a precise GPS position (NZ grid reference) with your specimens. Write this information with pencil on good quality (tough) paper or light cardboard and place the label in the container (plastic vial, jar, plastic bag, etc.); marker pen inevitably rubs or washes off the outside of containers holding the most interesting specimens. Ideally, specimens should be dropped straight into 70-95% ethanol (the volume of preservative should be at least three times that of the specimens), but 5 % formalin, 70 % meths, 100 % of your favourite spirit or even freezing will do as a temporary means of preserving them until they reach us. Pure ethanol is best because we hope to study the genetics of these animals someday. Alternatively, you can place a few live specimens in loosely packed, damp *Sphagnum* moss or clean, damp tissue paper in a plastic container, ideally with small holes in the lid, and mail them to us in a handy bag, or similar packaging. If kept cool (not frozen) and damp, the animals will survive for several days. Naturalists early in the previous century used to send these isopods to each other this way (nowadays it is illegal to send living or unpreserved animals between countries without special permits).

IDENTIFYING PHREATOICIDS

New Zealand phreatoicids can be identified to genus and some species relatively easily using the keys on the NIWA website (http://www.niwascience.co.nz/__data/assets/pdf_file/0003/59277/Isopoda.pdf). Note that species identifications are difficult and unreliable because the morphology of each species is not well known; intraspecific morphological variability with life-history stages and between locations has not been examined, especially with respect to the characters used in the key. Also, species may occur together and there are almost certainly some undescribed species in New Zealand, making the task of recognising each species more complicated.

Identification does require examination and careful checking of some fine characteristics using a stereomicroscope and assessments of relative dimensions or sizes of some characteristics. Some species identifications depend on detailed counts and/or the structure of spines, so some familiarity with crustacean anatomy is helpful. Despite its limitations, this key is the best available at present. Although not part of this project, we hope to produce a more reliable key from examining the collections resulting from the project.

PLEASE HELP

You can greatly assist this project by sending any collections and records of your collecting, successful or unsuccessful, to Graham Fenwick, at NIWA, PO Box 8602, Riccarton, Christchurch (email g.fenwick@niwa.co.nz). The last page of this document comprises a data sheet that we've used for every site investigated. You might like to use this sheet, including for recording any negative locations, and compete as many of the fields as practical (it may be impractical for you to collect water quality measurements, but any information is helpful). We will add your data to the project data base and identify any specimens. We will also send you identifications of any specimens provided, and send you a summary of our final results. Contributions from anyone working in the field over the 2007-08 summer will increase the geographic coverage of this project and result in a much better understanding of the conservation status of these fascinating beasts.

ACKNOWLEDGEMENTS

This work is supported by funding from the Department of Conservation's Terrestrial and Freshwater Biodiversity Information System (TFBIS) Programme. Dr GDF (Buz) Wilson (The Australian Museum, Sydney) contributed substantially to this project and the information contained in this document, as well as taking most of the phreatoicid photos and several of the site photos. Buz continues to contribute, especially in providing his indispensible taxonomic expertise to the project. Julian Sykes prepared the maps and Anna John helped prepare the data files and manage specimens.

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PHREATOICID SAMPLING

Date: ……..… 2008. **Location:** ...……

Location detail: ……..……...

GPS: Northing: ……. Easting: ……..

Habitat type: Seep spring drain stream wetland other

Stream width: …….m Water depth: ……..cm Velocity: ……..… cm/s

Vegetation: in-stream: …….. Surrounding land: ……...

Vegetation on banks: …… Canopy/shading: …… %

Stream bottom substrate: ……

Habitat detail: …….

……

Water depth (m)	Conductivity µS/cm	Temp °C	pH	DO_2	turbidity

SAMPLES

	TAKEN	SORTED	*INTO GLASS*
PHREATOICIDS			
AMPHIPODS			
OTHER			

Other: ……

…….

…….

…….

…….

Weather: rain/ no rain River: no flow / low / medium / high