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### Dandak: a mammalian dominated cave ecosystem of India

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#### ABSTRACT

Perpetual darkness, high humidity with almost constant geophysical factors are some of the abiotic factors which make the cave ecosystem unique. For any species a high degree of adaptation is always needed to thrive in such an ecosystem. Mammals in general have never adapted to cave life but they can play a major role in the cave ecosystem. Structurally, the Dandak cave has two distinct chambers that are completely different from each other in several geophysical factors. Thus both the cave chambers offer two distinct types of ecological niche. In the present study we found that both chambers of this cave were dominated by mammals all year round. Additionally, the group of mammals using the outer chamber completely differs from the group using the inner one. Possible geophysical factors responsible for such differences are discussed.

Key words: cavernicoles, cave ecosystem, troglophiles, mammals, carnivores

#### INTRODUCTION

Subterranean habitats are always unusual and fascinating. These ecosystems are characterized by perpetual darkness, constant temperature and high humidity with limited air currents, which altogether creates an ecosystem that is difficult to support the organisms adapted to normal surface conditions. Nevertheless, some organisms are attracted to live in caves at least for part of their life cycle due to low predation pressure, easy availability of prey and less risk from natural calamities along with some of the typical geophysical factors mentioned above. Cave dwelling/adapted organisms are generally evolved from such epigean forms which had some degree of preadaptation for a hypogean lifestyle prior to invasion for cave life (Racovitza 1907; Vandel 1965; Gunn 2004). Cave dwelling organisms can be broadly divided into two categories: an organism once trapped accidentally or get intentionally inside a cave, and unable to survive in an external epigean environment is referred as an obligatory cave dwelling organism-stygobionts (aquatic) and troglobionts (terrestrial). On the other hand, organisms that use caves for a short period either for shelter, feeding, breeding, roosting, hibernating etc. but depend mostly on the external epigean environment can be referred as non-obligate cave dwellers. Further, ecologically several classifications have been forwarded to classify the cavernicoles, which are based on their respective level of bondage to the cave life. However, as per the most recent classification proposed by Sket (2008) the cavernicoles have been categorized into four types: troglobites, eutroglophiles, subtroglophiles and trogloxene. In the present work the same classification has been adopted

to interpret the cavernicolous status of the organisms observed in Dandak Cave as the same was utilized by Biswas (2010) while describing the fauna of its adjacent cave "Kotumsar" of Kanger Valley National Park, India.

Dandak cave of Kanger Valley National Park was initially discovered in 1995 by the forest staff of the National Park while chasing some of the local tribes that had been illegally hunting the Indian (crested) porcupine (Hystrix in*dica*). The episode ended as the porcupine entered a deep tunnel that turned out to be the entrance of Dandak cave. Since the cave discovery several construction projects have been undertaken inside the cave to make the cave more accessible for tourism. However in order to reach this particular cave of Kanger Valley National Park one has to climb at least 450 stairs, and due to this specific reason the tourists generally avoid it. At the present time the cave is not open for general tourism; special permission is required to enter. It is apparent that the ecosystem of this cave is not under high anthropogenic pressure, and to a large extent its inner ecosystem is still left in its in situ condition.

Dandak is a limestone cave (18°51'30" N; 81°57'00" E) situated on the top of an isolated hillock exhibiting a few patches of exposed boulders. The cave opens at the base of a rock wall situated in a north-west direction. The cave entrance is wide and opens up horizontally into a large hall; the outer chamber (Fig. 1). Despite a few side passages and downward sloping passages the approximate length of this cave is only 360m. Structurally this cave is divided into two main chambers connected with each other by two small convoluted narrow passages. Due to its wide entrance, the external light penetrates to the far end of the outer chamber, which possesses massive and beautiful dripstone structures. Some of the stalagmites and stalactites have joined each other forming

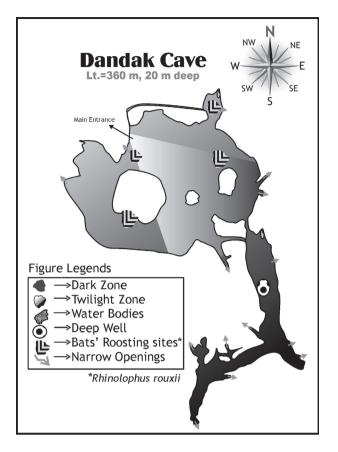


Fig. 1 - Schematic layout of Dandak Cave, showing both the chambers connected with each other by two narrow passages. Figure is also representing the luminous factors existing inside each chamber.

columns. The inner chamber is configured more like a tunnel with an unexplored deep well-like vertical pit near its entrance. In this cave, the major portion of the outer chamber can be safely referred as "twilight zone" whereas the inner chamber represents a "deep zone" cave environment. It appears that in this cave, the "intermediate/ transition zone" is almost absent and the existence of real "stagnant zone" is yet to be explored, if any; inside the well (Vermeulen and Whitten 1999; Biswas 2009, 2010).

#### **OBJECTIVES AND METHODS**

This study is intended to understand the biodiversity together with obtaining preliminary ideas about the ecosystem operating inside the cave. The study is based on qualitative collections and ecological field records that are in turn based on the direct or indirect evidence of animals making use of this cave. In the first phase of this study a tentative layout of the cave was drawn by mapping it with the help of measuring tape and compass. We noted all the possible openings and blow holes on the map where the cave connects to the outer surface. We carefully surveyed the entire cave that includes recognizing and noting all sorts of small and spacious habitats which could be used by any animal / organism. The survey included the locations, where drip water accumulates as well. In addition, several geophysical parameters viz; atmospheric temperature, humidity, water temperature and its pH value were also measured during dry (May) and wet (September) seasons in the field by using digital thermo-hyrometer (Pacer® TH 402) and pH meter.

The observations were made at every second month through a span of one year and in each field visit every nook and cranny of the cave was carefully observed to document the direct /indirect evidence(s) of any animal which used the cave for any purpose. Evidences were photographed and also other scientific techniques were employed to determine the animal species. Help from local tribes were taken to confirm the existence of suspected animals. We also attempted to determine the role of each visitor and/ or permanent inhabitants inside the cave, in their particular zone of occurrence. A speculative food pyramid has been constructed that may be operating within the cave (Fig. 4).

#### OBSERVATION AND RESULTS

A tentative map of the cave is shown in Figure 1 and which has been described in the earlier part. The geophysical factors between the two chambers apparently differ from each other while compared between dry and wet season (Fig. 2). We observed that rain water enters the cave mainly through the main entrance of the outer chamber which finally drained out via the deep well existing in the inner chamber. No standing water is apparent inside any chamber of this cave. However, few very small water pockets are present in both the cave chambers, fed by seepage which have almost constant temperature  $(18.0 - 20.0^{\circ}C)$  pH values (7.2-7.5) throughout last year. These are presumably with epikarst water fed small pools (Pipan 2005).

Though the major portion of the outer chamber of the cave is mostly lit during the day time, the organisms we observed here are mostly trogloxene. Nevertheless, the organisms we observed in the inner chamber represent all the categories of cavernicoles. Species inhabiting and/or approaching regularly in both the chambers are described here.

VERTEBRATES:

Chiroptera, Rhinolophidae Rhinolophus rouxii Temminck, 1835 (Fig. 3a)

Common name: Rufous Horseshoe bat.

A small colony of about 50-60 individuals roost in the outer chamber of Dandak Cave. We marked 4-5 different locations (small habitats) which are from time to time used by this colony. Roosting sites could be easily recognized by seeing the decomposed deposits of guano piles in the floor (Fig. 3b). Occasionally a few solitary bats were also apparent in some of the hidden cavities of this chamber. Observed evidence supports that this particular bat remains under predation pressure in this cave (Fig. 3c). However, during our few visits we failed to observe this colony, which directly indicates that it often travels to an alternative roosting site, perhaps to avoid predation pressure.

#### Chiroptera, Hipposideridae Hipposideros cineraceus Blyth, 1853 (Fig. 3d)

Common name: Ashy Leaf-nosed Bat.

This species occupy the inner chamber of this particular cave and were found to be highly sensitive to human disturbance. Whenever we entered the inner chamber this bat flew away. We confirmed the species by mist net capture.

#### Carnivora, Viverridae Viverricula indica Desmarest, 1804

Common Name: Small Indian Civet.

During our routine survey, we observed several pieces of evidences that altogether support that the small In-

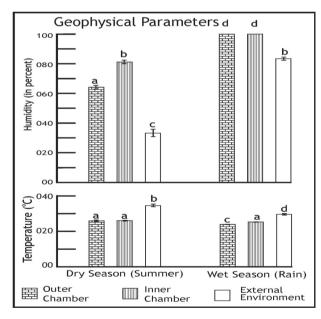


Fig. 2 - Each bar (with its respective standard error) representing mean (air temperature / percentage of water saturation in air; n=5) recorded from three different sites during dry and wet seasons. Results from two way ANOVA (Analysis of Variance) revealed significant season (p<0.0001), site (p<0.0001), and site X season interaction (p<0.001) effects for air temperature. Similarly it also revealed significant season (p<0.008) effects while comparing percent saturation of water in the air (humidity) by two way ANOVA. Further, the histograms bearing similar types of super scripts do not differ statistically from each other at minimum 5% level.

dian civet is a regular visitor for the outer chamber of this cave. Some of the major evidentiary items include: pugmarks near almost every external passage (Fig 3e); scat in various sizes usually found in the outer chamber (Fig 3f); and nail scars (claw marks) on the walls of the cave (Fig 3g). Further the bioinventory records of the park as well as the local tribes' observations also supported the existence of this species in that particular locality. This civet can climb the vertical wall very well, the claw marks on the cave wall as unconsumed bat remains directly suggests that it preys upon roosting bats (Fig. 3c).

> Rodentia, Hystricidae Hystrix indica Kerr, 1792

Common name: Indian Crested Porcupine.

We counted eighteen possible dens in the inner chamber of this cave of this particular species (Fig 3h). However out of the total number eight were abandoned as evidenced by either being blocked by debris or by being partially collapsed. It is possible that none of these dens are directly connected with the external epigean world. Some well demarcated passages exist in the inner chamber which appears to be used by this porcupine species to enter the chamber. Due to their long term regular use, the walls and floor of these passages have become textured by the quills of these porcupines. Occasionally porcupine dung was also observed, but we had never seen any other organism feeding upon it. The occurrences of quill(s) in the entrance of their passage confirmed its taxonomical status (Fig 3i).

#### Anura, Ranidae Hydrophylax malabaricus Tschudi, 1838 (Fig. 3j)

Common name: Fungoid Frog

This frog can be easily seen in the outer chamber of this cave. Its population generally diminished during rain. However, juveniles could be apparent inside the cave from August-September. Cave cricket; *Kempiola shankari* (described later) was observed to be the major prey for it.

#### Anura, Bufonidae Duttaphrynus melanostictus Schneider, 1799 (Fig. 3k)

Common name: Black-spined Toad/ Common Indian Toad

A small population of this toad species was always observed during rainy seasons in a particular hidden rocky enclosure of the outer chamber of this cave. Perhaps it remains under torpid conditions in remaining months. Possibly, the abundantly occurring cave cricket is the major food source for this particular species.

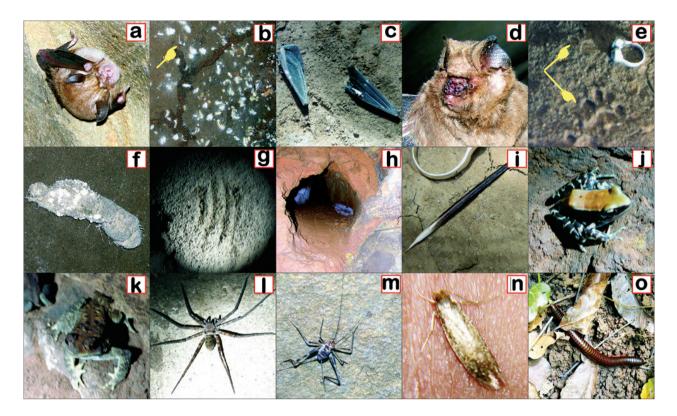


Fig. 3 - Collage of the evidences regarding the animals which generally visits/inhabits the cave. a) *Rhinolophus rouxii* roosting in a wall of the outer chamber. b) decomposing guano piles of *Rhinolophus rouxii* in the floor of outer chamber. c) wings of killed bat *Rhinolophus rouxii* found in the floor of the outer chamber. d) *Hipposideros cineraceus* caught by mist net roost in the inner chamber of the Dandak cave. e) pugmarks of *Viverricula indica* usually seen in the outer chamber. f) decomposing scat (excreta) of *Viverricula indica* in outer chamber of the cave. g) nails' scars of *Viverricula indica* on the walls of the outer chamber of the cave. h) living den of *Hystrix indica* in the inner chamber of the cave. i) quill of *Hystrix indica* found inside one of the den found in the inner chamber of the cave. j) *Hydrophylax malabaricus* always found in the outer chamber. k) *Duttaphrynus melanostictus* apparent during rain in a specific zone in the outer chamber. of the cave. l) *Heteropoda venatoria* usually seen in the outer chamber of the cave. m) *Kempiola shankari* (cave cricket) equally distributed in both the chambers of the Dandak Cave. n) Guano moth; *Kangerosithyris kotumarensis appears* in the inner chamber of the cave, possibly play a major role in the cave ecosystem. \*Some images are with a finger ring (b, e & i) i.e., to estimate the sizes.

Table 1 - List of all the observed (direct/indirect) species with their respective habitats (chamber) and cavernicolous status (as per Sket, 2008).

Name of the Species	Living Status	Observed in	Observed during		
Rhinolophus rouxii	Subtroglophile	Outer chamber	Round the year		
Hipposideros cineraceus	Subtroglophile	Inner Chamber	Round the year		
Viverricula indica	Trogloxene (habitual)	Outer chamber	Round the year.		
Hystrix indica	Subtroglophile	Inner Chamber	Round the year.		
Hydrophylax malabaricus	Subtroglophile	Outer Chamber	Round the year		
Duttaphrynus melanostictus	Eutroglophile	Outer Chamber	Rainy season		
Heteropoda venatoria	Eutroglophile	Outer Chamber	Round the year		
Kempiola shankari	Eutroglophile	Both the Chambers	Round the Year		
Kangerosithyris kotumarensis	Troglobite	Inner Chamber	Rainy Season		
Giant millipede	Trogloxene (accidental)	Outer chamber	Round the year		

INVERTEBRATES:

#### Araneae, Sparassidae/Heteropodinae Heteropoda venatoria Linnaeus, 1767 (Fig. 31)

Common name: Giant crab spider or the banana spider. Throughout the year juveniles and adults, both of this species can be easily seen in every nook and corner of the outer chamber of this cave. It has prominent eyes but generally remains unaffected from torch lights, as their eyes reflect light (Wallace 1937; Biswas 2010). This species seems a little sluggish in nature and inhabit the cave in search of easily available prey.

#### Orthoptera, Gryllidae/Phalangopsidae Kempiola shankari Sinha and Agarwal, 1977 (Fig. 3m)

Common name: Cave cricket.

It is the only species that can be easily observed directly from the entrance gate of the outer chamber through the deeper zones of the inner chamber of the Dandak cave. It is the most abundantly occurring species in this cave. The species is highly endemic for the caves of this specific zone (Biswas 2010). The insect has been observed to reproduce throughout the year and all developmental stages can be easily found inside the cave.

#### Lepidoptera, Tineidae Kangerosithyris kotumarensis Skalski, 1992 (Fig. 3n)

Common name: Guano-moth

Like *Kempiola shankari* this moth is also highly endemic for the caves of Kanger valley national park (Biswas 2010). It is guanophilic moth, only apparent during wet season when humidity is 100%. It occurs only around the guano deposits in the inner chamber of this cave. It rarely flies, usually moves in short leaps. It is truly cave adapted and never responds to light (Skalski 1992; Biswas 2010).

Few arthropods such as isopods acari, ants etc. were often seen in both the chambers of this cave. Since we failed to record any permanent population(s) of these animals we did not tabulate them as an inhabitant of this cave. Possibly they represent the accidental trogloxene categories which have been entered into the cave via air current, with certain habitual trogloxene or by flood during rain. However, we frequently observed a giant millipede in the outer ecotone zone of this cave, which sometimes often enters far end of the twilight zone of this cave. Perhaps, this millipede plays a role on the ecosystem of the outer chamber, thus we have tabulated it under the category of accidental trogloxene.

#### DISCUSSION

To date not a single mammal is reported as permanently cave adapted, though these animals regularly use the subterranean caves to somehow meet their biological requirements. In fact, caves offer a wide spectrum of microhabitats (Ginet and Decou 1977) which attract several animals either for resting, mating, nesting or hibernating. Nevertheless, easy availability of prey also attracts various nocturnal and even diurnal animals to caves. Though the Dandak cave as a whole represents a single identity, its two chambers represent two nearly distinct ecosystems and offer two completely diversified habitats. The outer chamber remains highly influenced by the epigean environmental conditions and thus its complete ecosystem also seems to be regulated by the external environmental conditions. Conversely the inner chamber is like a sealed enclave being far less influenced by the external world (Fig. 2).

In our study we found Viverricola indica as the dominant mammal, a regular visitor for the outer chamber of the cave. Observed evidence directly suggests that it comes in the outer chamber of this cave to hunt the roosting bat; Rhinolophus rouxii. Additionally, crickets, spiders, millipedes and amphibians are also known to be the favorable prey for this particular civet (Ayyadurai et al 1987; Chuang and Lee 1997; Mallick 2006) which are always available in the outer chamber of the this cave. Perhaps it is the first record suggesting that, the civet is at the topmost level of an ecological food pyramid for any cave. However, as per the notes of Kemp (1924) on the mammals of the Siju Cave of Meghalaya, India it could be speculated that the small cat (Felis sp.) which was a regular visitor of Siju cave prey upon the rat; Ratus nitudus nitidus which was very common in that cave.

Ringtail (*Bassariscus astustus*), red fox (*Vulpes vulpes*), coyote (*Canis latrans*), badger (*Meles meles*) and striped shunk (*Mephtis mephtis*) are some of the reported mammals that usually visit the temperate caves in search of young bat or birds as prey (Tvrtkovic 2005). As per Juberthie et al (1998) the temperate cave bats usually become victim during their hibernation. Occasionally various carnivores have been reported from tropical caves either in search of prey or to meet some other biological requirements (Gunn 2004, Harries et al 2008).

Porcupines use subterranean caves for their diurnal resting phase were already reported from several parts of the world (Price 2004; Moseley 2007; Harries et al 2009). In the inner chamber of the Dandak cave most of the dens are found as blind pockets, where this species remain during their resting phase. However, we speculate that in addition to the day-time resting and/or escaping predation pressure, the abundantly occurring cricket (as prey) and the water channels which feed seepage water throughout the year are some of the additional factors

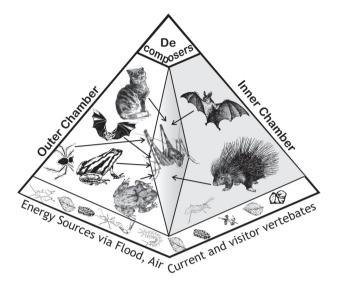


Fig. 4 - Ecological food pyramid, possibly operating inside the Dandak cave.

that altogether attracting the porcupine *Hystrix indica* to inhabit this cave.

The cricket, Kempiola shankari is the most abundantly occurring species appearing to be equally distributed in both the chambers of this cave. This species plays the key role in binding the complete ecosystem of this cave together. Two distinct insectivorous bats; Rhinolophus rouxii and Hipposideros cineraceus occupying two different chambers of this cave definitely prey upon this particular cricket. Further, two anurans; Hydrophulax malabaricus and Duttaphrynus melanostictus inhabit the outer chamber of this cave and also prefer cricket as their food. Indian (Crested) Porcupines which have been found to be a regular visitor to the inner chamber of this cave were also reported to feed occasionally on insects and small mammals (Kadhim 1997) which directly suggests that this cricket often become the victim of the porcupine. It has also been reported that in cave ecosystems giant Heteropoda sps. also feed upon the cave crickets (Harries et al 2009).

Microchiropterans are always the major energy source importer for any subterranean cave ultimately playing a major role in its ecosystem. The bat guano deposits are usually able to maintain a complete ecosystem in itself. However, in Dandak cave, bat droppings are found to be of incidental importance in the ecosystem. In both the chambers bat droppings were observed to be very haphazardly distributed on the floor and never forming substantial guano deposits which can support a guanophilic community. We speculated that the high predation pressure forced them to alter their roosting sites in a regular fashion. Though the predation pressure is less in the inner chamber and the abiotic factors are also relatively constant and distinct, a true cave adapted guanophilic species; *Kangerosithyris kotumarensis* was observed here as is common to the nearby cave "Kotumsar" (Biswas 2010).

Fossil records testify that the Viverricula indica as well as Hystrix indica were inhabitant of Asian caves even during Late Pleistocene-Holocene age (Ghosh 1991; Patnaik et al 2008; Mashkour et al 2009). However, the speculations regarding their existence in the caves were restricted to the idea that these species used the cave as a day resting phase or shelter. Our study directly suggests that both of these mammals play a major role on the complete ecosystem of the Dandak cave. Inspite of taking meager shelter these species are also getting easily available prev inside the respective chambers of occurrence of this cave. Conclusively, the two chambers of the Dandak cave possess two separate habitats, thus the ecological process operating inside each chamber are highly different from each other. The cricket; Kempiola shankari is the only factor found common for both the chamber which in fact is the most fascinating species to bind the complete ecosystem together (Fig. 4).

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# Observations on the biology of the endangered stygobiotic shrimp *Palaemonias alabamae*, with notes on *P. ganteri* (Decapoda: Atyidae)

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#### ABSTRACT

*Palaemonias alabamae* is endemic to subterranean waters in northern Alabama. Its type locality is Shelta Cave, Madison County, and ostensibly conspecific shrimps have been found in Bobcat and two other caves. Pollution and other factors may have extirpated the shrimp from the type locality. In Shelta Cave the species is smaller than the shrimp in Bobcat Cave and *P. ganteri* in Mammoth Cave, Kentucky. Adult female *P. alabamae* (s.s.) and *P. ganteri* are larger than males. Female *P. alabamae* with visible oocytes or, rarely, attached ova, were observed from July through January in Shelta Cave. Each female there produces 8 to 12 large ova, whereas females of the population in Bobcat Cave produce 20 to 24 ova, and *P. ganteri* produces 14 to 33 ova. Plankton samples taken in Shelta and Mammoth caves yielded nothing identifiable as zoea or postlarvae. *Palaemonias alabamae* and *P. ganteri* usually feed by filtering bottom sediments through their mouthparts, but both sometimes feed upside down at the water's surface. Although there is some overlap, the compositions of the aquatic communities in Shelta and Mammoth caves differ, and there are some major differences among the Alabama shrimp caves. The stygobiotic fish, *Typhlichthys subterraneus*, is a known predator on *P. alabamae* in Shelta Cave.

Key words: cave shrimps, Palaemonias alabamae, P. ganteri

#### INTRODUCTION

The shrimp family, Atyidae, is a large, cosmopolitan group of decapods that, in the Americas, contains nine described stygobiotic species (Holthuis 1956; Hobbs et al 1977). Two of them are found in caves of the United States: Palaemonias alabamae Smalley, 1961, occurs in subterranean systems in northern Alabama, where its type locality is Shelta Cave in Huntsville, and Palaemonias ganteri Hay, 1902, occurs in underground waters in the Mammoth Cave area of central Kentucky, where its type locality is Roaring River in Mammoth Cave. Unfortunately there is evidence of reductions in the numbers of the aquatic stygobionts in Shelta Cave (Lee 1987; Hobbs and Bagley 1989), which has been subjected to an influx of pollutants, including cadmium and the pesticide dieldrin (McGregor et al 1997:33, 56). Elevated levels of cadmium have also been reported for Bobcat Cave, in which Palaemonias occurs (McGregor et al 1997:33; McGregor & O'Neil 2000:9, 26). Cadmium has been shown to accumulate in much higher concentrations in the tissues of a stygobiotic crayfish species that occurs in Shelta Cave than in the tissues of a stygophilic species that also occurs there (Dickson et al 1979). This was attributed to the greater longevity of the stygobiont as reported by Cooper (1975), and Cooper and Cooper (1976, 1978a). Other studies on the

toxicity of cadmium to crayfishes were reported by Wigginton and Birge (2007).

An additional factor that has certainly had an impact on Shelta's aquatic community is abandonment of the cave by a longtime maternity colony of the grey bat, Myotis grisescens Howell, 1909, which contributed guano and occasional dead individuals to the community's trophic inputs (Cooper 1975:45-48). Grey bats were present during many of our trips to the cave, including in July 1975, but apparently vacated sometime in the later 1970s. On our final trip to the cave, during 12-17 July 1975, we spent 34 hours inventorying the aquatic fauna. In West Lake, the populations of all crayfishes were in very good shape, but no shrimp were observed. Our sighting of two shrimp on 24 November 1973 was the last reported for Shelta Cave. Thus, it is possible that P. alabamae has been extirpated from the type locality. On the other hand, P. ganteri was once assumed extinct (Lisowski and Poulson 1981; Lisowski 1982), but later intensive field work resulted in its "rediscovery" (Lisowski 1983). At any rate, because of its rarity, or possibly its extirpation from Shelta Cave, we have decided to publish our baseline observations on some aspects of the biology of P. alabamae (s.s). Both it and P. ganteri are now considered federally endangered species.

#### MATERIALS AND METHODS

During the period 24 August 1963 through 17 July 1975 we made 78 research trips into Shelta Cave, spending a total of approximately 580 hours there (Cooper 1975). Although most of our efforts were devoted to studies of the three stygobiotic crayfishes that inhabit the cave, during some of our trips we also collected information on *P. alabamae*. In addition, at times during this period we collected and made observations on the biota of two of the other caves where shrimp are now known to occur (Cooper & Cooper 1974; McGregor et al 1997; Rheams et al 1992).

We maintained a small colony of 9 to 13 P. alabamae in a large plastic pool within Shelta Cave for a little over five months during the winter of 1968-69. Measurements of preserved specimens were made to the nearest 0.1 mm with a Helios dial caliper. The standard length measurement for shrimp is postorbital carapace length (PCL), measured from the level of the posterior rim of the orbit to the center of the proximal margin of the carapace. The length of the rostrum (RL) usually is not included in such measurements, but can itself be informative. Total length (TL) of these shrimp is difficult to accurately assess, since the abdomen is very pliable and has a normal flexion. Nevertheless, TL measurements were included in this study in order that (1) potential sexual dimorphism related to comparative abdomen length could be assessed, and (2) the series of P. alabamae could meristically be compared with a series of P. ganteri, whose TLs (exclusive of RLs) were reported by Barr and Kuehne (1971), and with Palaemonias from Bobcat Cave, whose TLs (including RLs) were reported by McGregor & O'Neill (2000) and McGregor et al (1997).

#### RESULTS AND DISCUSSION

#### Occurrence and Abundance

Palaemonias alabamae was "apparently first collected by T. L. Poulson," who found a single female in Shelta Cave on 13 July 1958 (Cooper 1975:129). Biologists from Tulane University and their colleagues then made three visits to the cave and acquired the 18 specimens that constituted the type series for Smalley's (1961) description. The holotype (USNM 107028) and six paratypes (USNM 107029) are at the National Museum of Natural History, Smithsonian Institution, but the whereabouts of the rest of the Tulane collection are unknown (N. Rios, pers. comm.). The first specimens we found were two females collected on our initial trip to the cave on 24 August 1963. Table 1 is a chronological summary of our P. alabamae sightings there in 30 of our trips between 1963 and 1973. McGregor & O'Neil (2000:35, 38) provided chronological summaries for all Palaemonias observations in Bobcat Cave on the Redstone Arsenal, Madison County, from December 1990 through September 2000.

Table 1 - Chronological summary of *P. alabamae* observations in Shelta Cave, Alabama, 1963 through 1973.

10/0								
1963								
24 Aug 2 ♀. 22 Dec "plentiful"								
1964								
18-19 Oct 7. ? Dec 5. 3 Jan 8 ♂, 10 ♀ (1								
ovigerous); many others observed.								
1966								
18 Sep "plentiful." 20 Nov 3.								
1968								
27 Jul 12. 23 Sep 2. 1-3 Nov 15. 8-9								
Nov 9. 7-10 Dec 25.								
1969								
18-20 Jan 5. 10 Jul 3. (1 ovigerous ♀). 12-								
14 Sep 1.								
1970								
11 Feb 1♀. 26-28 Nov 3								
1973								
24 Nov 2								

Palaemonias was believed endemic to Shelta Cave (Cooper 1966, 1967, 1968a, 1974, 1975; Cooper & Cooper 1966, 1971) until in 1973 William Torode discovered shrimp in Bobcat Cave (Cooper & Cooper 1974). We visited the cave with him on 23 November 1973 and, in residual pools, found 10 Palaemonias, several Cambarus (Aviticambarus) jonesi Hobbs and Barr, 1960, and a number of Typhlichthys subterraneus Girard, 1860. In 1990, under terms of a cooperative agreement with the U. S. Fish and Wildlife Service, investigators from the Geological Survey of Alabama (GSA) began conducting multidisciplinary field studies in and around Shelta, Bobcat, and other caves in Madison County, with Palaemonias as their primary focus. Their work produced a wealth of data relating to the biota and other aspects of those particular caves (McGregor et al 1997; McGregor & O'Neill 2000; Moser & Rheams 1992; Rheams et al 1992, 1994). In October and November 1991, Karen Rheams, Stuart McGregor and Randall Blackwood found Palaemonias at a third locality, the extensive Hering-Glover cave system in eastern Madison County (Rheams et al 1992:66). Rheams et al (1994:58) additionally reported that Torode and others had in November 1991 observed shrimp in Brazelton Cave, identified by Graham (1969) as a possible western resurgence for the Hering-Glover system. They later confirmed the occurrence of Palaemonias in Brazelton Cave, and reported a connection between it and the Hering-Glover system (McGregor et al 1997:18). Recently, Kuhajda (2006) reported finding Palaemonias in Muddy Cave, south of Huntsville, and cave shrimps of undetermined taxonomic status have been found in three caves in northwestern Alabama (B. Kuhajda, pers. comm.).

#### Palaemonias Habitats

Shelta Cave -- The type locality of P. alabamae is located in Huntsville, Madison County, Alabama. It is an active drainage conduit of considerable volume in which large quantities of water are "stored" for many months every year. There are no incoming or outgoing streams, but.at least some of the cave's continuous input of water is of seasonal meteorological origin in the zone of aeration. In winter, heavy precipitation in the recharge areas results in rapid upwelling of water into the aquifer, the water table experiences a dramatic rise, and the result within the cave is prodigious flooding. Spring and early summer rains maintain relatively high but fluctuating and generally receding water levels, and as precipitation decreases in late summer and fall the water table recedes by lateral migration and accessible water disappears from most of the cave (Cooper 1975:29). However, in fall and early winter there is a permanent, shallow perched lake (West Lake) in the extreme western part of the cave (Miller Hall), which maintains fairly constant levels and feeds a perennial stream that sinks before reaching the main cave. When standing water was essentially absent from the rest of the cave, we found that large numbers of aquatic stygobionts "retreated" to West Lake. The lake is probably fed by movement of groundwater through the porous limestone, since there are only what seem to be minor inputs of meteorological origin at this time (Cooper 1975:102-110). The resurgence for Shelta Cave's waters appears to be Brahan Spring (Moser & Rheams 1992:31-32; Rheams et al 1994:43), which is impounded in a park northeast of Drake Avenue, about 3.5 air km south of Shelta Cave.

For descriptions of the cave's size, structure, hydrologic cycles, fauna, ecology, physicochemical characteristics, and history, with maps and photographs, see Cooper (1966, 1968a, 1974, 1975), Cooper & Cooper (1971, 1976, 1978a, b), French (1968), Hobbs & Bagley (1989), Johnston (1933), Jones & Varnedoe (1968), Moser & Rheams (1992), Peck (1989), Rheams et al (1992), Tarkington et al (1965), Torode (1973), Varnedoe & Lundquist (2005), Veitch (1967), and Wilson (1969). Bobcat Cave -- This cave is located on the Redstone Arsenal about 13 air km southwest of Shelta Cave. It is not as voluminous as Shelta, but their hydrologic cycles are virtually identical, with lentic waters that experience heavy seasonal rises and drops. During periods of low water, only small residual pools remain and, as in all of Shelta Cave except the West Lake, the water table at this time lies below the accessible levels of the cave. For additional information, including maps and photos, see McGregor et al (1997), McGregor & O'Neill (2000), Moser & Rheams (1992), Rheams et al (1992, 1994), and Varnedoe & Lundquist (2005).

The Hering-Glover System -- The caves that make up this system are located between Keel and Troy mountains in southeastern Madison County, southwest of the town of Gurley. The Hering-Glover system is very different from Shelta and Bobcat caves, being an open system that is mainly a fairly narrow, winding phreatic tube with some side passages and alcoves. During seasonal spates the entrance may be nearly sumped, and the lotic outgoing water then becomes a rocky surface stream that passes under a highway before sinking into Glover Sink Cave.

Our observations in 1965, 1967, and 1968, and published information from 1929 (Johnston 1933), 1953 (Malmberg & Downing 1957), 1968 (Graham 1969), and 1990-1992 (Rheams et al 1992), show that this system floods in late winter and early spring, the waters begin to recede in mid-summer, and lowest water levels are present from late summer through fall and into early winter. Some fluctuations occur at various times in response to heavy surface precipitation. Even during extreme drawdowns, however, there are lentic bodies of water in the caves, varying from small, shallow residual pools to base-level "lakes" (Rheams et al 1992). The water table is often broadly exposed in parts of the system and, as with Shelta and Bobcat caves, it would appear to be underlain by an extensive aquifer inaccessible to investigators.

When we made our second trip to Hering Cave, on 30 December 1965, the scoured floor of the main passage was dry and sandy, with some limestone and chert pebbles. The only accessible water there was a number of small residual pools that contained the stygobiotic fish, *T. subterraneus*, the stygobiotic crayfish, *Orconectes australis* (Rhoades, 1941), the stygobiotic isopod, *Caecidotea bicrenata bicrenata* (Steeves, 1963), the stygophilic crayfish, *Cambarus (Erebicambarus) tenebrosus* Hay, 1902; and the stygophilic fish, *Cottus carolinae* (Gill, 1861). No shrimp were observed.

Graham (1969:11, corrected) observed that the resurgence for the Hering-Glover system "is not definitely known although it may be Brazelton Cave (AL 337), 4000 feet northwest of Glover Cave and on the other side of the mountain." The waters of Brazelton Cave apparently rise and fall on the same seasonal schedule as those of the Hering-Glover system. On 8 November 1990, and 30 October and 13 November 1992, Rheams et al (1992) found pools and lakes varying from 0.3 to 4.8 m deep in the cave. On 23 July 1992, however, they found the entrance sumped, and flowing from it was a swift stream about 1.2 m deep.

Not only is the Hering-Glover system of interest because of its basically lotic habitat , but also because the shrimp's occurrence there places it in a different physiographic and geologic setting. Shelta and Bobcat caves are within the eastern periphery of the Highland Rim, and both are developed in the Warsaw Limestone and the underlying Fort Payne Chert (Cooper 1975, Rheams et al 1994). Keel and Troy mountains, though, are outliers of the Jackson County Mountains, thus part of the dissected rim of the Cumberland Plateau, and the caves there are developed in the Monteagle Limestone. For additional information on the Hering-Glover system, including maps of the caves, see Graham (1969), Johnston (1933), Jones & Varnedoe (1968), McGregor & O'Neil (2000), McGregor et al 1997), Rheams et al (1992, 1994), Tarkington et al (1965), and Wilson (1969).

#### Molting and Potential Longevity

No direct data are available on growth or longevity in *P. alabamae*, but some information on molting has been obtained, and limited inferences on longevity can be made from our studies of Shelta's other stygobiotic decapods.

On 3 November 1968, a female with a postorbital carapace length (PCL) of 5.4 mm and very early oocytes visible through the carapace was found in the process of molting in the shrimp pool, but the animal neither completed nor survived the process. The exuvium of another individual was found on 10 December 1968, along with seven live animals. This is the only direct molting information we obtained, and it suggests at least a winter molt for some adults. Indirect evidence of a summer molt was obtained from an adult female (PCL 5.3 mm) collected on 10 July 1962, which had obvious calcareous concretions on many parts of the body. These appeared to be similar to the molting nodules that Jefferies (1964) described on the epigean palaemonid shrimp, Palaemonetes varians, and could indicate relative molting condition. These nodules are most numerous immediately following molting, after which they decrease in number.

We initially were tempted to conclude that P. ala*bamae* has a longevity of one year, with the adult class diminishing in abundance from later winter into early summer. This, however, would be inconsistent with our current knowledge concerning the longevity of the other stygobiotic decapods in Shelta Cave (Cooper 1975; Cooper and Cooper 1978a). Additionally, it would require relatively rapid growth, which is also at odds with what we now know of the rate of growth of the other stygobiotic decapods (Cooper 1975; Cooper & Cooper 1976, 1978a; Culver 1982). It is more likely that the broad habitat expansion that occurs during high water results in greatly decreased population density, which renders these small, largely transparent animals less conspicuous than they are during low water levels (Cooper 1975). When habitats were compressed, population densities increased and shrimp were found in shallow residual basins.

One of the stygobiotic crayfish found in Shelta Cave, *Orconectes (Orconectes) sheltae* Cooper & Cooper, 1997, is a diminutive species endemic to the cave. This crayfish has a seasonal and spatial occurrence almost identical to that of *P. alabamae* (Cooper 1975; Cooper & Cooper 1997). Like the shrimp, the crayfish produces only 8 to 12 ova, and in proportion to adult female body length its hatchlings, which are about the size of an adult shrimp, are suspected to be the largest found in any crayfish species (Cooper 1975; Cooper and Cooper 1997). From direct growth and size data, we estimated the lifespan of *O*. (*O*.) sheltae to be from 10 to 20 years (Cooper 1975:314; Cooper and Cooper 1976; Cooper and Cooper 1978a). One specimen that we had permanently marked was recaptured more than four years after its initial capture, and still had not approached the maximum total carapace length known for the species. In our opinion, a longevity of 10 to 15 years would not be unrealistic for *P. alabamae*.

Leitheuser & Holsinger (1983:2-30) reported for *P. ganteri* that "Information has also been obtained on age and growth rates through observations on aquarium specimens. Growth is slow and variable among individual shrimp. Based upon information such as this, the life span of the shrimp has been estimated to be 10 to 15 years." Based on data from laboratory cultures, Dickson & Holsinger (1981:45) suggested that the life span of the stygobiotic amphipod, *Crangonyx antennatus*, "may extend from 8 to 10 years."

#### Comparative Sizes, Sexual Dimorphism, and Sex Ratios

Smalley (1961:129) said that P. alabamae may be distinguished from P. ganteri by features that included its "smaller size" and "shorter rostrum," but provided no data other than noting that the RL of P. alabamae constitutes two-thirds of its PCL. Our measurements of both species showed that the abdomen averages about 71 percent of total length, which enabled us to extrapolate probable PCLs for the P. ganteri data. Smalley (1961:129) reported a range of 4.2 to 5.9 mm PCL for 19 adult P. alabamae. The range in our sample of 34 adults from Shelta Cave was 4.1 to 5.9 mm (mean = 5.0 mm) PCL. Abdomen lengths of our adults ranged from 10.1 to 14.2 mm (mean = 12.1 mm), and total lengths (excluding rostrum) from 14.5 to 20.0 mm (mean = 17.1 mm). Our measurements, and those we obtained or extrapolated from Barr and Kuehne (1971), support Smalley's statement for the Shelta Cave population of Palaemonias (Table 2), which was the only population known at the time the species was described. Cooper and Cooper (1974) reported that specimens from Shelta Cave were smaller than those from Bobcat, but provided no comparative measurements. PCL measurements of Bobcat Cave specimens are not available, which is also true of specimens from the Hering-Glover population. McGregor et al (1997:46) listed total lengths, including rostrum, of ca. 83 presumed adults or subadults, ranging up to 29.4 mm. Considering that the greatest total length, including rostrum, in 34 adult P. alabamae from Shelta Cave was 24.5 mm, these data provide some quantitative support for the idea that the Shelta shrimp are considerably smaller than those in Bobcat Cave.

The PCL of *P. ganteri* averages only 0.6 mm greater than that of *P. alabamae* from Shelta, but its TL averages 2.2 mm greater. Male *P. ganteri* average 0.5 mm greater in PCL and 1.3 mm greater in TL than male *P. alabamae*, while females of the former average 0.7 mm greater in PCL and 2.9 mm greater in TL than females of the latter.

P. alabamae	P. ganteri							
Postorbital ca	rapace length							
males: $R = 4.1-5.9$ (mean = 4.5; $n = 13$ )	R = 4.1-5.8 (mean = 5.0; $n = 7$ )							
females: $R = 4.1-5.9$ (mean = 5.3; $n = 19$ )	R = 5.2-6.7 (mean = 6.0; $n = 14$ )							
totals: $R = 4.1-5.9$ (mean = 5.0; $n = 34$ )	R = 4.1-6.7 (mean = 5.6; $n = 22$ )							
Total length w	vithout rostrum							
males: $R = 14.8-18.3$ (mean = 16.0; $n = 13$ )	R = 14.0-23.0  (mean = 17.3; n = 7)							
females: $R = 15.5-20.0$ (mean = 17.8; $n = 19$ )	R = 17.9-23.0  (mean = 20.7; n = 14)							
totals: $R = 14.5-20.0$ (mean = 17.1; $n = 34$ )	R = 14.0-23.0  (mean = 19.3; n = 22)							
Abdome	en length							
males: $R = 10.1-13.0$ (mean = 11.4; $n = 13$ )	N/A							
females: R = 10.9-14.2 (mean = 12.6; <i>n</i> = 19)	R = 12.7-14.9 (mean = 13.8; $n = 4$ )							
totals: $R = 10.1-14.2$ (mean = 12.1; $n = 34$	R = 12.7-14.9 (mean = 13.8; $n = 4$ )							
Rostrur	n length							
males: $R = 2.9-4.3$ (mean = 3.4; $n = 10$ )	N/A							
females: $R = 3.1-4.5$ (mean = 3.8; $n = 17$ )	R = 4.4-5.3 (mean = 4.7; $n = 4$ )							
totals: $R = 2.9-4.5$ (mean = 3.6; $n = 29$ )	R = 4.4-5.3 (mean = 4.7; $n = 4$ )							
Percent rostrum length of	postorbital carapace length							
males: $R = 62.0-86.0$ (mean = 74.2; $n = 10$ )	N/A							
females: $R = 57.4-78.4$ (mean = 69.4; $n = 17$ )	R = 75.4-102.0  (mean = 85.0; n = 4)							

Table 2 - Comparative measurements (mm) of adult *P. alabamae* from Shelta Cave and *P. ganteri* from Mammoth Cave; some totals include unsexed individuals. N/A = data not available.

Thus, *P. alabamae* is measurably smaller than *P. ganteri*, and at least the females of the latter (and probably the males) have a longer rostrum than all *P. alabamae*.

totals: R = 57.4-86.0 (mean = 70.7; n = 29)

Sexual dimorphism in size is evident in both species. Adult female P. alabamae average 1.8 mm longer in TL and 0.8 mm longer in PCL than males. Abdomen length averages 1.2 mm longer in females than in males. Adult female P. ganteri average 3.4 mm longer in TL and 1.0 mm longer in PCL than males. Palaemonias alabamae also shows slight sexual dimorphism in absolute length of the rostrum. The female rostrum in our sample averages 0.4 mm longer than the male rostrum. Expressing RL as percent of PCL, however, shows that males average about 4.8% greater than females, which is due to the greater average PCL of females ( $3^{\circ}$  mean = 4.5,  $9^{\circ}$  mean = 5.3). These differences are not an allometric effect, since the males at the low and high extremes of the range both measured 5.0 mm PCL, and the female extremes differed only slightly. In a total of 34 P. alabamae examined from Shelta Cave, there were 21 females, 13 males. The only sample large enough (n = 18) to provide significant information on the sex ratios at any given time is the one made on 3 January 1964, which consisted of 10 females and 8 males. Smalley (1961) reported that the same number of shrimp in an 18 August 1960 sample consisted of 11 males and 7 females. In both these collections combined, there are 19 males, 17 females. Barr & Kuehne (1971) reported that two collections of P. ganteri from Mammoth Cave contained 12 females, 7 males.

#### Reproduction

In Shelta Cave, P. alabamae with highly visible internal oocytes, or on rare occasions with externally attached ova, were found in every month in which numbers of shrimp were seen, which was from July through January. A female measuring 9.8 mm PCL, found on 24 August 1963, contained 8 oocytes that measured from 0.6 to 0.8 mm long, and on 3 November 1968 an uncounted number of oocytes measuring about 1.0 mm long was observed in a female measuring 5.2 mm PCL. Such medium-to-large oocytes, considered late stage or nearly so, were also seen in a female on 13 September 1968, in four on 18 and 19 October 1969, in three on 3 and 8 November 1968, and in one on 8 December 1968. Smaller, earlier stage oocytes were observed in a female on 3 November 1968. Despite the not infrequent presence of advanced oocytes, only two specimens bearing extruded pleopodal ova were found in Shelta Cave. A female measuring 5.8 mm PCL taken on 3 January 1964 had 9 such ova, measuring 1.0 X 1.2 mm, along with several remnant chorions. A second female, measuring 5.0 mm PCL, taken on 10 July 1969 and placed in the shrimp pool, had 4 attached ova measuring 0.9 X 1.4 mm when examined alive several days after capture (see Cooper 1975:146, Fig. 22). An equal number of ova had been present, however, when the animal was captured, but were lost in the habitat. It thus appears that P. alabamae in Shelta Cave produces from 8 to 12 ova per female. Rheams et al (1994:63) inadvertently reported that Cooper (1975) found individual

R = 75.4-102.0 (mean = 85.0; n = 4)

shrimp in Shelta Cave that "carried from four to 30 ova each," and this error was repeated in the recovery plan (USFWS 1997) and in the five-year followup report (USFWS 2006). It would further appear, considering that the female collected on 24 August 1963 contained 8 large oocytes in the ovary, that little if any resorption of additional oocytes occurs in females that actually extrude eggs. We know little, however, about the number of females in the population that complete the process, although late-stage oocytes were observed from late August through December. Our research on the stygobiotic cravfishes of the cave produced clear evidence that, while many females contain late stage internal oocytes, only a very few of the largest females actually produce successful clutches of ova or young (Cooper 1975; Cooper & Cooper 1978a, b). Resorption in these cravfishes is a common phenomenon.

For Bobcat Cave, Cooper & Cooper (1974) reported that ovigerous females had been found in September and October, and noted that in Shelta "females appear to produce fewer ova" than do females in Bobcat, but provided no data. Rheams et al (1992:68, 70) reported that "Cave shrimp with attached ova were observed in Bobcat Cave during May (a single, very large specimen with very large eggs observed May 25, 1992), July (an unknown number of the 51 individuals observed on July 22, 1991), August (3 of 13 individuals observed on August 21, 1991), and October (4 of 30 individuals observed on October 28, 1991). These individuals were noted to carry 20 to 24 eggs each." Except for the "very large specimen with very large eggs" observed in May, this general reproduction schedule appears to be similar to that observed in Shelta.

However, the notable fecundity of Palaemonias in the Bobcat Cave population far exceeds that in the Shelta Cave population. Not only is the number of ova per female more than twice the number of advanced oocytes (and attached ova) observed in or on Shelta females, but ostensibly far more females were observed carrying ova. The greater number of ova could be attributable to the larger size of the Bobcat Cave shrimp, but this does not explain the greater number of females apparently completing the reproductive process. Analysis of factors that could account for this putative disparity in fecundity, between two geographically close populations of what has been determined to be the same species, will have to await the results of future studies. It should be noted that only a few of the GSA studies distinguished between visible internal oocytes or ovarian ova, and extruded pleopodal ova. Thus, potential resorption of oocytes cannot be evaluated.

In the Mammoth-Flint Ridge system, ovigerous *P. ganteri* have been collected or observed in every month except February, March, and July (Barr & Kuehne 1971; Cooper 1975; Holsinger & Leitheuser 1982; Leitheuser et al 1985; Poulson 1964; Poulson & Smith 1969). One of the four specimens collected by Barr in August 1965 (Cooper 1975) measured 5.2 mm PCL and had 14 at-

tached ova, which measured about 1.0 X 1.4 mm each. A series of seven ovigerous females reported by Barr and Kuehne (1971), taken on 13 May 1962, measured from 20 to 22 mm TL (mean = 20.9 mm), which translates to ca. 5.2 to 6.4 mm PCL (mean = 6.1 mm), and had from 14 to 33 attached ova.

An additional female reported by the same authors, collected on 13 October 1961, measured 23 mm TL (6.7 mm PCL) and had 15 attached ova. Barr & Kuehne (1971) did not include ova measurements, but did provide weights of each of the egg masses on the eight ovigerous females. The much greater weight of October eggs led Barr & Kuehne (1971:86) to suggest "that peak egg production occurs in later winter or early spring and eggs are carried into or through summer." They added, "We do not know when the eggs hatch and young shrimps join the population, but presumably this occurs between later summer and winter." It should be observed that a period of larval development of unknown duration may well separate the time of eclosion and the time of appearance of "young shrimps" in the population (see following "Development" section).

In the two ovigerous *P. alabamae* reported above, ovum length was 1.2 and 1.4 mm, or 20.7 and 28.0% of female PCL (6.0 and 8.1% of TL). In the single *P. ganteri* for which this datum is available ovum length was 1.4 mm, or 26.9% of female PCL (7.8% of TL). It thus appears that *P. alabamae* in Shelta Cave produces only onethird to one-half as many ova (8 to 12) as does *P. ganteri* (14 to 33), although the ova are of comparable size, and that *Palaemonias* in Bobcat Cave produces numbers of ova (20 to 24) that are nearly comparable to the numbers produced by *P. ganteri*. No measurements of ova from Bobcat Cave shrimp are available.

#### Development

The ova of the ovigerous P. alabamae taken in January exhibited midsagittal cleavage and appeared close to eclosion, and several empty chorions were adjacent to them, which would indicate hatching at that time. The slightly longer ova of the smaller July female were not noticably differentiated, being generally amorphous envelopes of pale, yellow-white yolk. Based on just these few observations, egg laying could occur during medium and falling water levels in mid-summer, with an incubation period of about six months before hatching and emergence. Juberthie-Jupeau (1974) reported that incubation in the stygobiotic European atyid, Troglocaris anophthalmus (Kollar, 1848) requires 7 to 8 months, but takes only 20 to 35 days in the larger epigean atyid, Atyaephyra demaresti (Millet, 1832). The ova of an ovigerous P. ganteri collected in August 1965 showed clear polarity and obvious development, with white, well-developed larvae at one pole, deep yellow-orange yolk at the other (Cooper 1975).

Nothing is known of the post-hatching "stages" in either *P. alabamae* or *P. ganteri*. Our plankton collections and silt samples from Shelta Cave produced nothing identifiable as shrimp zoea or postlarvae, and as far as we know there have been no collections of these early life stages in *P. ganteri*. Barr and Kuehne (1971:83-85) remarked that, in Mammoth Cave, "The most striking feature revealed by analysis of plankton tows is the low density of organisms." Juberthie-Jupeau (1969) reported that larval development in *T. anophthalmus* consisted of five intermolt stages.

In one of the few such studies yet made of an American stygobiotic shrimp, Dobkin (1971) studied the development of larvae from a single female of the palaemonid, *Palaemonetes cummingi*, from Squirrel Chimney, Florida. The results of the study, conducted in the laboratory at 22-24°C, showed that this species goes through three intermolt "stages" prior to postlarval metamorphosis. The female apparently was carrying from 30 to 35 ova. The entire development period, from early egg to postlarva, required a maximum of 42 days. Based on extensive collections of various life stages of the stygobiotic palaemonid, *Palaemonetes antrorum* Benedict, 1896, which is endemic to the Edwards Aquifer in Texas, Strenth et al. (1988) reported three zoeal and two postlarval stages.

Considering that the incubation period for P. alabamae ova appears to be similar in duration to that of T. anophthalmus, and much greater than that of the laboratory-reared P. cummingi, we consider it likely that the larvae and postlarvae of Palaemonias are considerably larger and much slower growing than those of P. cummingi. To date, however, few direct observations have been made in support of this contention. In addition to being a member of a different caridean family, the palaemonid would seem to be a less highly adapted (i.e., more recent) stygobiont than the atyids. Dobkin (1971: 296) observed that its development "is remarkably like that" of Palaemonetes paludosus (Gibbes, 1850). The latter, a large epigean palaemonid common in fresh waters of the eastern United States, is considered to be the closest relative of P. cummingi (Franz 1982: 108). Although these two species are so similar in development, including in the duration of the larval period, the smaller stygobiont still produces larger larvae and postlarvae, and generally fewer but larger ova (with respect to female size), than its epigean relative.

In summary, it appears that *Palaemonias* and the few other stygobiotic atyids for which data are available display a general reproductive strategy that has become evident across a broad spectrum of highly-adapted stygobionts, both invertebrates and vertebrates. This includes the production of fewer and larger ova (thus young), and longer incubation periods than are seen in epigean relatives. These are adaptive responses to life in relatively predictable, food-limited environments, in which a survival premium is placed on smaller populations and "fitter" offspring. The stygobiotic atyids probably also display delayed maturity and reproduction, longer larval development periods, and decreased growth rate with increased longevity, what we have termed a "Methuselah Strategy" (Cooper & Cooper 1978*a*). All these adaptations are evident in Shelta's stygobiotic crayfishes (Cooper 1975; Cooper & Cooper 1976, 1978*a*,*b*), but most remain conclusively to be demonstrated in any stygobiotic shrimp.

#### Feeding and Other Behaviors

The observations of Hay (1902:226-227), Giovannoli (1933:620), Barr and Kuehne (1971:85), and Smalley (1961) on certain aspects of the behavior of P. ganteri and P. alabamae are very similar to our own on the latter species. In Shelta Cave, P. alabamae were observed sifting through bottom and other silt. They clasp minute bolts of silt in the opposable brushes of the chelate tips of the first and second pereiopods, and rapidly shift these to be scraped off by their mouthparts. Specimens found in areas floored with bat guano, which analysis has shown to be rich in calories and nutrients (Fenolio et al 2005), had black digestive tracts. Barr & Kuehne (1971:85) remarked that "Palaemonias ganteri was observed to strain and filter the bottom sediments through its mouthparts more or less in the same manner described for the Brazilian atyid Atyoida potimirim [Müller, 1881] by Müller (1981). The food of these troglobitic shrimps probably consists of microorganisms in the silt." We made no attempt to analyze gut content, or to determine the potential bacterial or other microfloral composition of submerged substrates, but we did qualitatively sample the unicellular eukaryotes that live in the silts ingested by all of Shelta Cave's decapods (Table 3).

On 3 November 1968, a shrimp was found in the stream that flows from West Lake, "fluttering" upside down against the surface tension with its limbs. This was the only occasion on which we found *P. alabamae* in flowing water, where the substrate is low in silt. Our impression of this activity was that it involved feeding (Cooper 1975). Hartnoll (1964:78) reported identical

Table 3 - Unicellular karyotes living in silts ingested by *P. ala-bamae* in Shelta Cave. Those denoted by asterisk (\*) also occur in Mammoth Cave (Gittleson and Hoover 1969, 1970).

Flagellates									
Bodo caudata	Caecomonas crassicauda								
Bodo celer*	Gyropaigne lefevrei								
Bodo compressus	Monas elongata								
Bodo lens*	Monas obliqua*								
Bodo minimus*	Rhynchomonas nasuta*								
Ar	nebas								
Amoeba guttulata*	Heterophrys glabrescens*								
Amoeba vespertilio	Pseudodifflugia gracilis								
Ciliates									
Aspidisca andrewi	Cyclidium glaucoma								
Chilodonella caudata	Vorticella microstoma*								

behavior in the stygobiotic palaemonid, *Troglocu-banus jamaicensis* Holthuis, 1963, in a Jamaican cave, and interpreted it similarly, saying that "several were swimming upside down along the surface apparently feeding on floating material." Lisowski (1983:91) observed *P. ganteri* exhibiting the same behavior in Mammoth Cave.

Our experiences with general movement behavior of *P. alabamae* in Shelta Cave were very similar to those of Hay (1902:226-227) for *P. ganteri*. The shrimp are essentially transparent except for the central thorax, which is opaque enough to be seen. They swim slowly, generally for short distances, by continuous paddling of the pereiopods. The body is held rigid, and they "hover" through the water. We never observed the caudal elements being used for movement, since their only escape response when disturbed was to rise slowly in the water, almost to the surface, and swim off.

#### Ecological Associates and Relationships

The aquatic community composition of P. alabamae habitats differs from that of P. ganteri in its macroscopic components, despite some overlap, and there are some differences among the communities in three of the Alabama Palaemonias caves (Table 4). Typhlichthys subterraneus is the most abundant and significant predator in all four systems, and Palaemonias undoubtedly plays a role, perhaps a major one, in its energetics. Although we never observed this fish pursuing shrimp in either Shelta or Bobcat caves, a very large individual collected in Shelta on 20 November 1966 regurgitated an adult shrimp (Cooper 1974, 1975; Cooper and Cooper 1974). Mammoth, Shelta, and Bobcat caves each have a second potential stygobiotic predator -- the fish Amblyopsis spelaea DeKay, 1842, in Mammoth and the salamander Gvrinophilus palleucus McCrady, 1954, in Shelta and Bobcat -- but they are so comparatively rare (Barr 1967; Barr & Kuehne 1971; Cooper 1968b, 1975) that they are proba-

Table 4 - Macroscopic fauna of aquatic communities in which *Palaemonias* occurs. C = common; R = rare; U = present but abundance unknown; P = presence reported but unverified by voucher specimens.

	Shelta	Bobcat	Hering-Glover	Mammoth
Fishes				
Typhlichthys subterraneus	С	С	С	С
Amblyopsis spelaea				R
Chologaster agassizi				R
Cottus carolinae			С	С
"Ictaluridae"			P1	
Salamanders				
Gyrinophilus palleucus	R	R		
Eurycea lucifuga	U	U	U	С
Snails				
Antroselates spiralis				С
Crayfishes				
Cambarus jonesi	С	С	P <sup>2</sup>	
Orconectes australis	С		С	
Orconectes sheltae	R			
Orconectes pellucidus				С
<i>Cambarus tenebrosus</i> (s.l.)	R	?	С	С
Amphipods				
Stygobromus exilis				С
Stygobromus fecundus				С
<i>Stygobromus</i> n. sp.	R			
Stygobromus vitreus	R			
Crangonyx barri				С
Isopods			· · ·	
Caecidotea bicrenata bicrenata			С	
Caecidotea bicrenata whitei				С
Caecidotea stygia				С

<sup>1</sup> Reported by McGregor et al (1997:50).

<sup>2</sup> Reported by Rheams et al (1992:71, 76; 1994:66).

bly not significant shrimp predators. *Gyrinophilus palleucus*, which does not occur in the Hering-Glover system, is known to feed on crustaceans in other caves (Cooper & Cooper 1968). Leitheuser & Holsinger (1983:25-26) reported, "On several occasions, Leitheuser has observed *Amblyopsis* apparently 'stalking' *Palaemonias ganteri* for periods in excess of one hour in Mystic River."

Larvae of the terrestrial, cave-frequenting salamander, Eurycea lucifuga Rafinesque, are aquatic predators on small invertebrates, and have been reported from Bobcat Cave (McGregor et al. 1997:48). Fishes of the genus Cottus thrive in caves in the United States and are formidable predators, with a large head and mouth. Lisowski (1983:91) reported Cottus feeding on the stygobiotic isopod, Caecidotea, in Mammoth Cave. Cottus carolinae is common in the seasonally lotic waters of the open Hering-Glover system, but does not occur in the essentially closed lentic waters of Shelta or Bobcat caves. Catfishes of the genus Ictalurus are also known predators on cave crustaceans (Relyea & Sutton 1974). However, among Alabama's shrimp caves, ictalurids have been reported only in Brazelton Cave (McGregor et al 1997:50).

One species of stygobiotic crayfish, Orconectes pellucidus (Tellkampf, 1844), and the stygophilic C. (E.) tenebrosus, are found in Mammoth Cave. Three species of stygobiotic crayfishes occur in Shelta Cave -- C. (A.) jonesi, O. (O.) australis, and O. (O.) sheltae. A few individuals of C. (E.) tenebrosus have been found in Shelta, but it is not a prominent member of the community. No interactions of any kind were observed between any of these crayfishes and P. alabamae. Cambarus (A.) jonesi is the only stygobiotic crayfish that we and others have found in Bobcat Cave, and this is one of but a few known caves inhabited by this species where it does not occur with at least one other stygobiotic crayfish. Cambarus (E.) tenebrosus may occur in Bobcat Cave, but is yet to be reported from there by name, although "sighted crayfish" occur in a nearby cave (McGregor et al 1997:48). Rheams et al (1992:71, 76; 1994:66) reported the first and only known record of C. (A.) jonesi from the Hering-Glover system, collected in Hering Cave on 14 November 1990. In caves where it occurs, this species is always quite abundant and prominent, but we did not find it among the many crayfish we examined in several visits to that cave, and the putative C. jonesi specimen cannot be found.

When we collected in Hering Cave on 30 December 1965 and 15 September 1968, the aquatic community included a number of *Caecidotea b. bicrenata*, including females with ova in their marsupia. Neither *Caecidotea* nor any other aquatic isopod occurs in Shelta Cave, and they have not been vouchered from Bobcat Cave although a "white isopod" was reported there by McGregor et al (1997:80). *Caecidotea stygia* Packard, 1871, and *C. bicrenata whitei* Lewis and Bowman, 1981, are common in the Mammoth Cave system (Lewis 1982). Two species of stygobiotic amphipods, *Stygobromus vitreus* Cope, 1872, and a new species of this genus being described by J. R. Holsinger (pers. comm.), occur in Shelta Cave. Neither is common, and they appear primarily to be elements of the interstitial fauna, occurring in permanent or transient pools, including in the runoff from a bucket collecting drip water from the ceiling (Cooper 1975:56). Amphipods have not been reported from Bobcat Cave or the Hering-Glover system.

Three species of *Stygobromus* and one species of *Crangonyx* occur in the Mammoth Cave system (J. R. Holsinger, pers. comm.). Isopods and amphipods may be the preferred prey of adult *Typhlichthys* (Cooper & Beiter 1972).

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## Composition and distribution of stygobionts in the Tafna alluvial aquifer (north-western Algeria)

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#### ABSTRACT

Little is known about the hypogean fauna of Algeria, with studies mostly dating to the beginning of the twentieth century (Gurney 1908; Racovitza 1912; Monod 1924; Pesce and Tetè 1978); moreover, the knowledge varies markedly among regions. In this study, we examined the composition and distribution of the invertebrate communities in the phreatic zone of the Tafna aquifer (N-W Algeria). Twelve wells close to the Tafna wadi, ranging between 120 and 1100 m a.s.l., were studied from May 2005 to March 2006. Many specimens belonging to 37 taxa were collected, the most frequent taxa being *Typhlocirolana* sp., a stygobitic Gammaridae species, Cyclopidae and Ostracoda. Other crustacean species were relatively scarce, with discontinuous distribution, being present only in a few wells. The taxonomic richness and abundance of stygobitic crustacean communities were relatively constant over time. The spatial distribution of stygobionts was mainly related to the exchanges with surface water.

Key words: Tafna, Algeria, Stygobionts, Wells, Invertebrates

#### INTRODUCTION

Interest in groundwater habitats increased in the last century, particularly due to the attention given by biospeleologists and hydrobiologists. The overall image obtained is that of an unseen "ocean" beneath our feet with spatial continuity (Danielopol et al 2000). Due to the size and diversity of habitats, groundwater hosts a very diverse assemblage of adapted taxa (stygobites *sensu* Botosaneanu 1986; Rouch and Danielopol 1987), with high rates of endemism and reduced dispersal capabilities (Gibert et al 1994). Studies and assessments show that, in spite of the severity of the underground environment (Danielopol 1997), stygobitic communities present an unexpected richness.

Stygobitic communities normally consist of a majority of crustacean taxa with a very restricted distribution (high endemicity). The species diversity is locally low but can be very high at the regional scale (Ferreira et al 2003; Dole-Olivier et al 2005). In North Africa, research has been progressing in the last century (Monod 1924; Henry and Magniez 1972; Pesce et al 1981), with recent studies in Tunisia (Ghlala et al 2009) and Morocco (Boutin and Boulanouar 1983; Mathieu et al 1999; Berrady et al 2000; Boutin et al 2002). However, the groundwater domain in Algeria and its organisms are still largely unknown (Gurney 1908; Nourisson 1956; Pesce and Tete 1978; Henry and Magniez 1981; Gagneur and Chaoui-Boudghane 1991; Belaidi et al 2004). The Algerian stygofauna includes a small number of stygobitic species, which are only known through the description of a few specimens. A recent synopsis revealed the presence of nearly 34 stygobitic species, six of which are listed for the north-western part of Algeria (Decu et al 2001), reflecting a low sampling effort in the groundwater area.

This study makes up for this insufficiency by providing a first database on the Algerian stygofauna collected in the wells drilled in the alluvial aquifer of the Tafna wadi. Indeed, wells are the simplest way to access groundwater in the porous aquifer (Boutin 1984). The Tafna alluvial aquifer is a phreatic zone within a Plio-Quaternary alluvial aquifer, characterized by a deposit of stone cemented by silt and clay. Therefore, this study, aims to (1) assess the diversity of the stygobitic fauna and (2) verify the spatial distribution of the populations that have colonized the Tafna aquifer.

#### **METHODS**

#### Study sites

The study area is the alluvial aquifer of the western and northern watershed of the Tafna wadi. The Tafna wadi (Fig. 1) is a stream approximately 170 km long, draining an area of 7425 km<sup>2</sup> in north-western Algeria. The difference in altitude between the source in the Tlemcen Mountains and the Mediterranean Sea is 1100 m. The geology of the drainage basin includes dolomites and limestone upstream and a Tertiary basin downstream. The terraces and the valley floor are occupied by cultivated land, while the slopes are covered with scrubland and shrub vegetation. The water regime is influenced by the Mediterranean climate (with sub-humid to semi-arid tendencies), characterized by a cold rainy season from

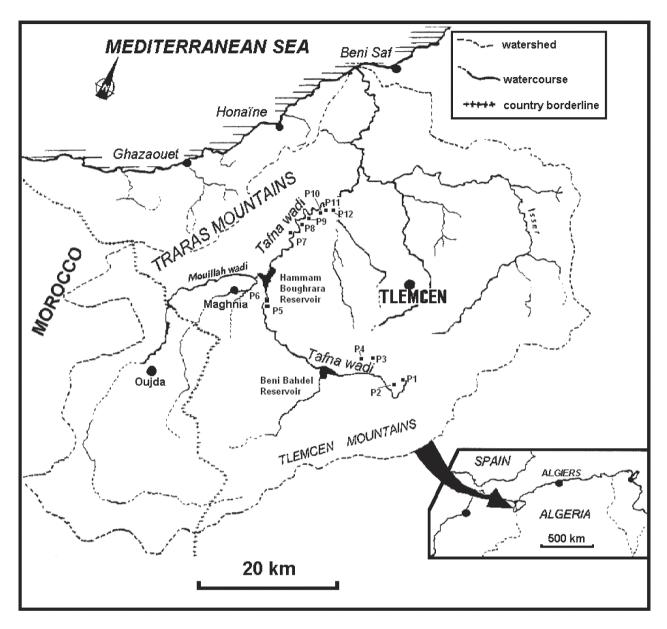


Fig. 1 - Hydrographic map of the Tafna wadi and location of the study wells.

November through April and a hot dry period from May through October. The Tafna wadi is ephemeral; it flows permanently only in winter from January to March, with discharge decreasing in the spring, and it is interrupted in the summer starting in May.

Most of the 12 study wells are used for agricultural or domestic purposes, and are located on the right (P1, P2, P3, P4, P5, P6) and left (P7, P8, P9, P10, P11, P12) banks of a 100-Km stretch of the stream starting from the Ghar Boumaza Cave. Four wells are located in the headwaters (P1, P2, P3, P4) and eight of them in the lowland (P5, P6, P7, P8, P9, P10, P11, P12). The characteristics of the wells are summarized in Table 2.

The water table flows in Jurassic limestone in the upper course of the stream and in Quaternary calcareous marl in the lower course, and it is subject to marked oscillations. The piezometric level in the study area was around 4.20 m below the ground, and it was deeper in the low plain wells with a maximum of 16.5 m.

#### Sampling methods

Temperature, pH, conductivity and dissolved oxygen were measured *in situ* with a WTW multi-parameter probe. The piezometric level (water level below the soil) was measured at each sampling date to monitor the quantity of groundwater resources. We used a sound sensor to measure the water depth in the wells. Fauna was collected in each well six times from May 2005 to March 2006 with two types of sampling equipment: a phreatobiological net sampler (Cvetkov 1968; Bou 1974) 20 cm in diameter at the opening, composed of a cone filter with a 150 µm mesh, drawn up 10 times in each well through the entire

Wells	Mean water level below the soil (m)	Distance/ wadi (m)	Mean water column (m)	Diameter (m)	Protection wells	Altitude (m a.s.l.)	
P1	-4.64	100	2	1.25	Partial	1010	
P2	-7.33	250	2	1.5	-	920	
P3	-7.31	1000	2.5	1.2	Partial	922	
P4	-9.68	1500	2.5	1.25	Partial	940	
P5	-3.77	50	3	1.60	-	308	
P6	-3.87	50	3	1.40	+	305	
P7	-8.74	150	2.5	1.25	+	194	
P8	-6.24	100	1.5	1.25	+	189	
P9	-16.25	800	2.5	1.25	+	192	
P10	-12.58	500	9	1.20	+	180	
P11	-8.50	150	2	1.30	-	179	
P12	-15	1000	3	1.25	Partial	180	

Table 1 - Characteristics of the twelve wells.

water column, which was of different depths in the various wells; a nasse-type baited trap developed by Boutin and Boulanouar (1983). The traps were set in contact with the bottom for 24 hours. The same number of traps was placed in each well throughout the sampling period, with the same time of immersion. The samples were fixed in 5% formalin in the field. After the sorting, individuals were preserved in the field in 70% ethanol before being identified.

All animals were identified to the lowest taxonomical level possible using published and informal keys, and the numbers of individuals of each taxon were recorded. Identification sometimes required dissection and examination under a compound microscope. All Ostracoda were identified by P. Marmonier, Isopoda by C. Boutin and Amphipoda Salentinellidae by M. Messouli. The Copepoda were not identified beyond the listed taxonomic level because of the unavailability of a specialist.

#### Statistical methods

Abundances were calculated for each type of sampler at each well and are expressed in number of individuals per volume (ind/m<sup>3</sup>) and number of individuals per trap (ind/trap). One-way analysis of variance (ANOVA 1) was used to compare spatial and temporal variations of the piezometric level, i.e. between the two hydrological periods of winter (high water period) and summer (low water period) (factor: season) in each well, and between the twelve wells (factor: well). ANOVA 1 was also used to compare spatial and temporal variations of the different physico-chemical variables. The abundance and richness of taxa were compared between wells and between periods (ANOVA 1). To obtain a normal data distribution, the abundance of each taxon was expressed logarithmically (Logx+1). The ADE 4 program of Thioulouse et al (1997) was used for the statistical analysis. The distribution of all crustaceans in the wells was assessed by Correspondence

Analysis (CA) of the log-transformed data. Two separate analyses were performed from a table (two matrixes) with 72 rows (each representing one well on one sampling date) and 8 columns (i.e. variables) representing the density of the taxa chosen for analysis (ind/m<sup>3</sup> and ind/traps). The spatial community distribution was determined with a Between Analysis. An inter-class discriminant analysis, with the variable well, was used to compare the density ranking among the twelve wells. All crustacean species, including the rare ones, were taken into account in the analysis.

#### RESULTS

#### *Physico-chemical variables*

The temporal variation of the physico-chemical data was low (Fig. 2). The temperature followed a normal seasonal cycle with the minimum in winter (15 °C recorded in P4) and maximum in summer (26 °C in P7). The mean temperatures recorded in the wells varied from 16 °C to 22 °C, with a gradual and significant increase from upstream to downstream (p<0.05). Mean conductivity values were high (3900  $\mu$ S/cm) and increased significantly from P1 to P12 (p<0.05). Conductivity varied from 540 to 1143  $\mu$ S/cm in the upper part of the reach and from 2610 to 7810  $\mu$ S/cm in the lower part. The pH ranged from 7.3 to 11.27, with the lowest mean value (7.3) measured in well P5.

Groundwater was generally under-saturated with dissolved oxygen, the DO concentrations ranging from 2 mg/l at P9 to 9 mg/l at P2; the highest values occurred in the upstream wells (more than 70% saturation) (Fig. 2).

#### Piezometric level

The depth of groundwater in the wells generally ranged from a mean of -3.77 m in P5 to -16.25 m in P9.

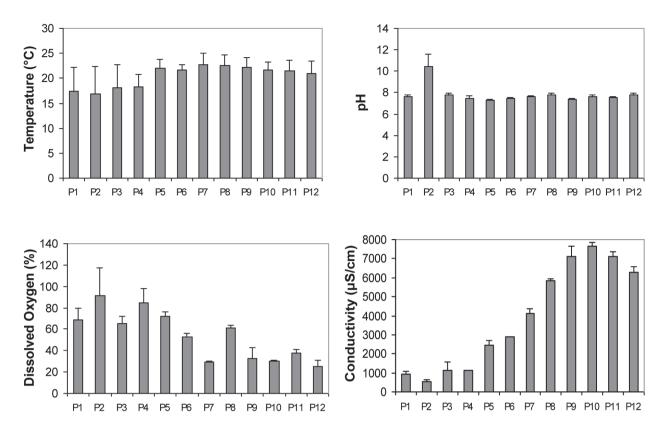


Fig. 2 - Mean value of the main groundwater physico-chemical variables in the Tafna aquifer. (Bars represent standard deviations).

It was scarcely influenced by climatic variations due to low rainfall during the study period. Overall, water levels were highest in March in relation to the high aquifer recharge during the rainy period (January-March) and lowest in August. Seasonal water variations in the wells had an amplitude of 3 to 4.5 m in the upper reach (p < 0.05) and became progressively less pronounced in the lower course of the river, where they did not exceed 2-3 m, in particular at wells P8, P11 and P12. Significant variations (p<0.05) recorded in other wells in the lower course were mainly related to their use for irrigation (Fig. 3).

The piezometric levels generally differed most between two types of wells: wells located near the riverbank (P1, P5, P6, P8), where the water levels reached their maximum, and wells far from the riverbank (P4, P9, P10, P12) (Fig. 3).

In the upper reach, the mean water level in the wells increased from P1 (-4.65 m below the ground surface) to P4 (-9.70 m). Downstream, it increased from P6 (-1.36 m) to P9 (-16.25 m) (p<0.05).

#### Faunal data

We collected 8309 individuals belonging to 37 taxa and 31 families (Table 2). Eight taxa were frequent (present in more than 50% of the samples) and 21 were rare (present in less than 20% of the samples); the remaining taxa were moderately frequent. Crustaceans represented 93% of the total fauna, 72% of which were Isopoda Cirolanidae. Insects represented only 5% of the total fauna and were dominated by larvae of Diptera Culicidae. Other taxa (Mollusca, Oligochaeta, Nematoda and Hydracarina) made up 2% of the total (Fig. 4).

The invertebrate fauna collected in the wells consisted of a combination of epigean and hypogean taxa, the latter represented mainly by crustaceans. Among the nine

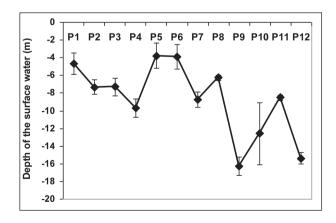


Fig. 3 - The piezometric level in the study area: main depth of the water surface of the aquifer. (Means and standard deviations).

		P1			P2			P3			P4			P5			P6	
Taxa		ind	ind/		ind/	ind/		ind/	ind/		ind/	ind/	ind.	ind/	ind/		ind/	ind/
Nematoda	ind. tot	/m <sup>3</sup>	trap	ind. tot	<b>m</b> <sup>3</sup>	trap	ind. tot 18	<u>m<sup>3</sup></u> 0.92	trap	ind. tot	<u>m<sup>3</sup></u> 0.16	trap	Tot	m <sup>3</sup>	trap	ind. tot	m <sup>3</sup>	trap
Oligochaeta							10	0.92		1	0.10							
Tubificidae																		
Limnodrilus sp.													23	0.6				
Lumbriculidae													23	0.6				
Trichodrilus sp.							1.1	0.52		1	0.00							
-							11	0.53		1	0.08							
Gastropoda Bythinellidae																1	0.02	
•								1.42		1	0.16		52	0.71	1	1	0.03	0.22
Physidae Planorbidae							9	1.43		1	0.16		53	2.71	1	3	0.08	0.33
										1	0.16					7	0.32	
Hydrobiidae																		
Copepoda																		
Cyclopidae	55	0.984	6.86	2		0.29	4	0.21		158	2.65	15.14	414	6	52.83	115	3.7	9.83
Isopoda																		
Cirolanidae																		
** <i>Typhlocirolana</i> sp.										6		0.86	6	1		2592	0.08	432
Asellidae																		
**Proasellus sp.							47	2.47		1	0.08					1		0.17
Microparasellidae **Microcharon sp.										2	0.16							
Amphipoda																		
Gammaridae																		
Gammarus sp.	274	3.012	35.86	1	0.08		2	0.16					17	0.89		72	4	4.17
**g. sp.		5.012	20.00		0.00		_	0.10					2	0.16		12	1.9	,
Salentinellidae **Salentinella cf.													2	0.10		12	1.9	
angelieri													6	0.47				
Ostracoda																		
Cyprididae																		
Heterocypris sp.	3	0.221					239	13.28		19	1.15							
Herpetocypris sp.		0.221					239	15.20		19	1.15		282	11.05	1.67			
Hydracarina				2	0.24		2	0.32		2	0.156		1	0.17	1.07			
Collembola							2	0.52					1	0.17				
				2	0.13					1	0.08							
Heteroptera Mesovellidae						0.00		0.00			0.00							
				2		0.28	1	0.08		1	0.08							
Corixidae													3		0.5	4		0.66
Naucoridae													6	0.08	0.83			
Pleidae										1	0.16		2		0.33			
Diptera																		
Culicidae				60	4.7		1	0.06		18	1.42		20	1.1		25	2.58	
Ptychopteridae													1	0.08				
Ceratopogonidae	5	0.793		1	0.13													
Chironomidae				3	0.24		2	0.29		14	1.1		2	1.08				
Dixidae																		
Coleoptera																		
Limnebiidae				8	0.42													
Elmidae	1	0.158		1	0.08		1	0.05										
Dytiscidae													7		1.17	36		6
Hydraenidae																1	0.03	
Haliplidae																7	0.08	1
Curculionidae																1	0.03	
Hydrophilidae																		
Trichoptera																		
Ecnomidae				1	0.05													
Glossosomatidae				1														
Total Taxonomic					0.05													
Richness		5			12			12			12			16			14	

	P7		P8			P9				P10			P11					
Taxa	ind. tot	ind/ m <sup>3</sup>	ind/ trap	ind. tot		ind/ trap	ind. tot	ind/ m <sup>3</sup>	ind/ trap	ind. tot		ind/ trap	ind. tot		ind/ trap	ind. tot	P12	ind/ trap
Nematoda	inu. tot	mu/ m	սոր		mu/m	uap	inu. tot	mu/ m	нар		IIIu/III	uap	inu. tot	mu/m	нар	inu. tot	inu/in	пар
Oligochaeta																		
Tubificidae																		
Limnodrilus sp.																		
Lumbriculidae																		
Trichodrilus sp.																		
Gastropoda																		
Bythinellidae		0.4																
Physidae	3	7		2	0.21		9	0.63	0.17	2	0.064					17	0.84	0.17
Planorbidae	3	/		-	0.21		9	0.05	0.17	2	0.004					1	0.84	0.17
Hydrobiidae				1	0.11											1	0.052	
Copepoda				1	0.11													
Cyclopidae		0.05	0.5								0.022	1.17		0.47			0.212	0.67
Isopoda	4	0.05	0.5							8	0.022	1.17	4	0.47		9	0.212	0.67
Cirolanidae																		
	150																	
** <i>Typhlocirolana</i> sp.	468		78	2598		433							332		55.33			
Asellidae																		
** <i>Proasellus</i> sp.																		
Microparasellidae																		
**Microcharon sp.																		
Amphipoda																		
Gammaridae																		
Gammarus sp.				3	0.29													
**g. sp.	3	0.16		4	0.08	0.5	1	0.05					4	0.42				
Salentinellidae **Salentinella cf.																		
angelieri				1	0.08													
Ostracoda																		
Cyprididae																		
Heterocypris sp.	5	0.55	0.17				4	0.31		2	0.064							
Herpetocypris sp.																3	0.105	0.17
Hydracarina																	0.100	0.17
Collembola																		
Heteroptera																		
Mesovellidae																		
Corixidae																		
Naucoridae																		
Pleidae																		
Diptera																		
Culicidae	1	0.16		17	1.79		7	0.47		10	0.25		41	4.24		15	0.86	0.17
Ptychopteridae	, i	0.10		1,	1.13		ĺ ĺ	U.T/		10	0.23		- <sup>-</sup>	7.27		1.5	0.00	0.1/
Ceratopogonidae							1	0.04					1	0.105		1	0.04	
Chironomidae							2	0.04	0.17	2	0.05		5	0.105		8	0.04	0.17
Dixidae							Ĺ	0.08	0.17	1	0.05		5	0.42		Ô	0.38	0.17
Coleoptera											0.012							
Limnebiidae																		
Elmidae																		
Dytiscidae																		
Hydraenidae																		
Haliplidae																		
Curculionidae																		
											0.012							
Hydrophilidae Trichontore										1	0.012							
<b>Trichoptera</b>																		
Ecnomidae Glossosomatidae																		
Total Taxonomic																		
Richness		6			7			6			7			6			7	

100

crustacean taxa, five were stygobiont. The most diverse groups were Isopoda (3 taxa) and Amphipoda (3 taxa), followed by Ostracoda (2 taxa) and Copepoda Cyclopidae. The last taxon might be a stygobitic species.

Larval stages of winged insects belonged to 18 families; the most diverse order was Coleoptera (7 families) followed by Diptera (5 families), Heteroptera (4 families) and Trichoptera (2 families).

The mean taxonomic richness recorded in all the wells was 10 taxa. Stygobionts were represented by 5 taxa: Isopoda Cirolanidae (*Typhlocirolana* sp.), Microparasellidae (*Microcharon* sp.) and Asellidae (*Proasellus* sp.), Amphipoda Salentinellidae (*Salentinella* cf. *angelieri*) and Gammaridae (stygobitic Gammaridae sp.). Specimens of all these Amphipoda were without eyes and pigment.

The mean density of invertebrates varied significantly among wells (p<0.05), being lowest in P9 (0.26± 0.56 ind/m<sup>3</sup>; 0.4 ± 0.51 ind/trap) and highest in the P6 (2.14±1.71 ind/m<sup>3</sup>; 452±247 ind/trap). These differences among wells were primarily due to crustaceans, among which only the Isopoda Cirolanidae showed strong variations in abundance. The difference in Cirolanidae abundance among wells was significant (ANOVA 1, p<0.05). Isopods were most abundant in P6 and P8, with a mean density of 452±232 ind/trap and 433±247 ind/trap respectively (Fig. 5). In contrast, the mean density of insects was low and varied among wells, albeit without significant differences.

The structure of the invertebrate fauna varied among wells. Although the mean taxonomic richness was similar among wells (Table 1), the total number of taxa showed significant differences (p<0.05) (Fig. 6). Well P5 was most taxa-rich (16 taxa) and P1 least taxa-rich (only 5 taxa). This difference in richness is explained mainly by the presence of insect families: 6 and 8 families were present in P4 and P5 respectively. The richness of wells P4 and P5 can also be explained by the presence of 3 and 5 stygobiont species respectively:

A strong dominance of a few taxa was generally observed, with one dominant taxon per well, often representing over 70% of the total fauna. This was particularly true for Cyclopidae in P4 (70%), for the Ostracoda Heterocypris sp. in P3 (71%), for the Gammaridae Gammarus sp. in P1 (81%) and for Diptera Culicidae in P2 (70%). In well P5, Cyclopidae represented 49% of the total fauna and the Ostracoda Herpetocypris sp. 33.37%. Similarly, stygobitic crustaceans were dominated by the single species Typhlocirolana sp. which represented 86-98% of the total abundance in wells P6, P7, P8 and P11. In P9 and P12, the invertebrate community was dominated by two epigean taxa: Physidae (Mollusca, Gastropoda) and Culicidae (Arthropoda, Insecta). The remaining taxa were only present in 1 to 3 wells with very low abundance (<3%), except for Chironomidae (Arthropoda, Insecta) which were present in most wells with low abundance.

In general, the taxonomic richness and total abundance did not vary significantly over time (p>0.05). For

Relative Abundance (%) **Total Fauna** 10 1 0.1 Crustacea Insecta Oligochaeta other Mollusca 100 Relative Abundance(%) Crustacea 10 1 0,1 SNOODIC Cannaidae 0,01 Cyclopidae Salentinella Gammatus Microcharon Citolanidae Ostracoda Relative Abundance (%) Insecta 0,1 Heteroptera Trichopters Coleopters Collembola Diptera

Fig. 4 - Relative abundance of invertebrates: total (epigean + stygobitic), epigean (insects) and stygobionts (crustaceans).

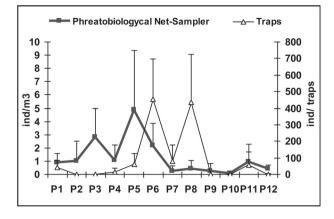


Fig. 5 - Mean density of total taxa (epigean + stygobitic) collected in the wells with the two sampling techniques.

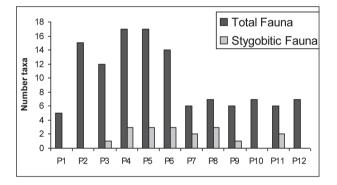


Fig. 6 - Total number of taxa (epigean + stygobitic) and of stygobitic taxa collected in the wells.

most wells, there was only a temporal change in composition. For example, among the stygobiont crustaceans, *Typhlocirolana* and the stygobitic undetermined Gammaridae species were present throughout the entire sampling period, while *Microcharon* sp., *Proasellus* sp. and *Salentinella* cf. *angelieri* were present only from March, in relation to the aquifer recharge.

#### Comparative use of two sampling methods

The phreatobiological net sampler allowed us to collect 35 taxa and 1393 individuals belonging to 4 higher stygofaunal taxonomic groups, primarily Crustacea Salentinellidae (*S.* cf. *angelieri*), Isopoda (*Microcharon*), Asellidae (*Proasellus*) and Ostracoda (Fig. 6). The nasse trap yielded only 17 taxa and 6916 individuals, consisting mainly of Cirolanidae (*Typhlocirolana*), Gammaridae (*Gammarus*) and Cyclopidae (Fig. 7). All taxa of other groups were collected with the net sampler, especially Culicidae, Ceratopogonidae, Trichoptera, Oligochaeta and nematodes which were absent in the trap (Fig. 7)

Comparison of the two techniques by ANOVA1 revealed a significant difference in taxonomic richness and abundance (p<0.05). The phreatobiological net had higher total richness but lower total abundance, whereas the trap had lower total richness but higher total abundance (Table 2).

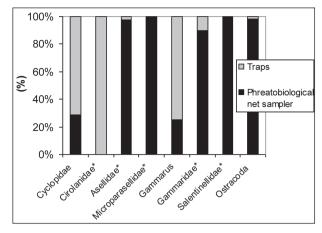


Fig. 7 - Relative abundance of crustaceans (epigean + stygobitic) collected with the two sampling techniques.

#### Spatial distribution of crustaceans

Copepoda, Ostracoda and the isopod *Typhlocirolana* sp. were widely distributed along the longitudinal transect, with a maximum number of 2500 individuals per well. The total abundance was significantly different among wells (p<0.01), being much higher in the wells near the riverbank (P1, P5, P6, P8). However, the correlation (Pearson) of crustacean total abundance and the distance to the river was not significant (P=0.11). Stygobitic Gammaridae coexisted with Cyclopidae, Ostracoda and *Typhlocirolana* sp., but Gammaridae were less abundant and frequent. Collections of *Microcharon*, Salentinella and Proasellus were rare (Fig. 8).

The results of the Correspondence Analysis (Fig. 9) show the spatial distribution of various crustacean species on the longitudinal transect of the Tafna alluvial aquifer for both sampling techniques:

#### Phreatobiological net sampling

The byplot explains 55% of the variability of the data matrix, with 30% of the variance explained by the first axis. The F1 axis is mainly related to four crustacean taxa (Proasellus, Cyclopidae, Salentinella and Ostracoda) located near the ends of the axis (Fig. 9 A). An inter-well discriminant analysis was performed to understand the contribution of the wells to the variability. Four wells were discriminated mainly by the F1 axis and are divided into two groups according to the distance that separates them from the bed of the wadi. The first group is negatively correlated to the axis and is associated with Proasellus and Microcharon. It is formed by wells P3 and P4, farthest from the stream. In contrast, the second group (wells P1, P6 and P8, all located near the stream bed) is associated mainly with Cyclopidae, S. cf. angelieri and stygobitic Gammaridae. The F2 axis isolates P5 and P12, characterized by the ostracod Herpetocypris.

#### Trap sampling

The byplot explains 70% of the variability of the data matrix, with 38% of the variance explained by the first

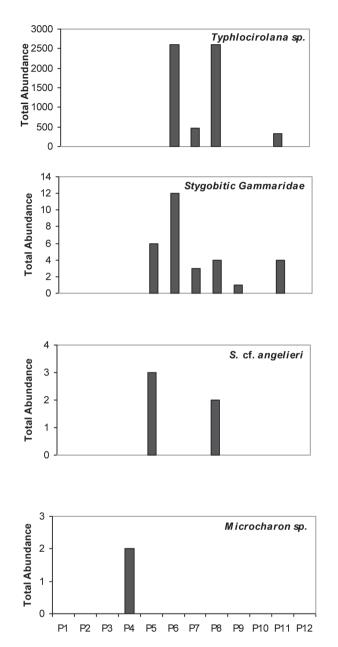


Fig. 8 - Spatial distributions of the five stygobiont taxa in the twelve wells.

axis. Three wells were discriminated mainly by the F1 axis. P7 is positively correlated to the axis and is associated with a low density of *Typhlocirolana*. In contrast, P6 and P8 are associated with a high abundance of this isopod. The F2 axis isolated P1, characterized by the dominance of *Gammarus*, and P12, associated with the ostracod *Herpetocypris* (Fig. 9 B).

#### DISCUSSION

The fauna of the wells in the Tafna alluvial aquifer is dominated by epigean taxa (86% of all taxa). The mean taxonomic richness in the 12 wells is lower than that observed

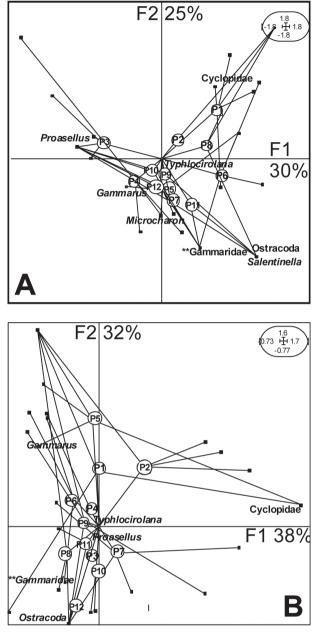


Fig. 9 - Between-group Correspondence Analysis of crustaceans sampled in the wells with the (A) phreatobiological net sampler and (B) trap sampler.

Position of the crustaceans and wells in the F1-F2 ordination graph. Position of taxa in the F1-F2 ordination graph indicated by small squares, position of wells by large circles.

in Morocco, e.g. by Boutin and Dias (1987) at Marrakech (a mean of 12 species in 11 wells), by Boulal (1988) in Tiznit region in the northern Anti-atlas (a mean of 14 species in 10 wells) and by Boutin and Idbennacer (1989) in the southern Anti-atlas (a mean of 10.8 species in 7 wells). Moreover, the subterranean aquatic fauna collected in this study is characterized by relatively low taxonomic richness: 11 and 18 stygobitic species were reported in previous studies of wells in Morocco (Boulal 1988; Boulanouar 1986).

Of the 34 species of stygobitic crustaceans identified in Algeria, 5 were already recorded and identified in the Tlemcen area: 4 Harpacticoida and 1 Asellidae (Decu et al 2001). Five taxa were recorded in the study wells. Microcharon sp. (Isopoda Microparasellidae) was recorded only in well P4. Two species of this genus have been cited for Algeria (Decu et al 2001): Microcharon zibani Pesce and Tetè 1978 and M. karamani Pesce and Tetè 1978, collected respectively in wells of Biskra and Dirah in eastern Algeria. Proasellus sp. (Isopoda Asellidae) was recorded in wells P3, P4 and P6. It should be noted that Proasellus notenboomi Henry and Magniez, 1981 was recorded in the study area in a karst spring between 900 and 1100 m altitude in Sebdou (Tlemcen) (Decu et al 2001; Henry and Magniez 2001). Eight species of this genus have been cited for Algeria, but only three of them are depigmented and anophthalmic, namely P. notenboomi in Tlemcen, P. delhezi in Kabily and P. bragadicus (Decu et al 2001; Henry and Magniez 2001). However, no Stenasellidae were recorded in the study wells, even though two species of this group have been cited for Algeria; Johannella purpurea Monod recorded in Msila and Metastenasellus leysi Magniez in Ain Sefra.

Typhlocirolana sp. (Isopoda Cirolanidae) was recorded in wells P4, P5, P6, P7, P8 and P11. Three species of Typhlocirolana were previously described for Algeria (Racovitza 1912), two in the north-west (T. buxtoni and T. gurney) and one in the south-east (T. fontis). Salentinella cf. angelieri (Amphipoda Salentinellidae) was recorded in wells P5 and P8 and stygobitic Gammaridae sp. (Amphipoda Gammaridae), depigmented and anophthalmic, was recorded in wells P5, P6, P7, P8, P9 and P11. One species of stygobitic Gammaridae had already been identified in Algeria, namely Echinogammarus tacapensis collected from a spring near Guelma in eastern Algeria (Stock 1986). However, no Melitidae and no Bogidiellidae were recorded in the study wells. Two stygobitic species of *Pseudoniphargus* were previously cited for Algeria, P. africanus Chevreux in Annaba and P. macrotelsonis Chevreux et Gauthier in Guelma, while two stygobitic species of Bogidiella were known for eastern Algeria, B. africana Karaman et Pesce in Biskra and B. chapuis Ruffo.

No specimens of the depigmented and anophthalmic Syncarida Parabathynellidae were collected in our samples, although this taxon was previously recorded in the hyporheic habitat of the Tafna wadi by Gagneur and Chaoui-Boudghane (1991) and Belaidi et al (2004); hence, it can be considered the sixth stygobitic taxon from the Tafna alluvial aquifer. It should be noted that *Cteniobathynella essameuri* Dumont Syncarida Parabathynellidae is a stygobitic species recorded in the central Sahara of Algeria (Decu et al 2001).

Copepoda Cyclopidae were collected in most of the study wells. This group is represented in Algeria by one stygobitic species, *Megacyclops donnaldsoni algericus* Kiefer recorded in Ain Sefra (Decu et al 2001). However, no Copepoda Harpacticoida were collected in the wells during the study period, despite the presence of five stygobitic species in Algeria identified by Rouch (1986). Four of them were recorded in Tlemcen: *Parapseudoleptomesochra minoricae* (Chapuis et Rouch), *Elaphoidella algeriensis*, *Parastenocaris numidiensis* Rouch and *Parapseudoleptomesochra* sp. The fifth species, *Nitocrellopsis petkovskii* Rouch, was recorded in Bechar. Moreover, Copepoda Harpacticoida were previously recorded in a hyporheic zone of the study area (Belaidi et al 2004).

The fauna collected in the study wells is mainly represented by the genus *Typhlocirolana* (Cirolanidae), which coexists in the wells with the undetermined stygobitic Gammaridae species collected with the same frequency but with lower abundance. The coexistence of these two species is related to the particle size of the sediment, which provides favourable conditions for the establishment of a large fauna; in fact, according to Boutin (1984), *Typhlocirolana* occurs where the sediment spaces are large enough to allow free space for the animal.

Among the rare taxa, *Salentinella* cf. *angelieri* (Amphipoda, Salentinellidae) was collected in only two wells, with low abundance and very low frequency. This confirms its preference for deep aquifers (Marmonier and Dole 1986). The Microparasellidae *Microcharon* sp. (Isopoda) is very abundant in the hyporheic zone of Tafna wadi (Gagneur and Chaoui-Boudghane 1991; Belaidi et al 2004) and was accidentally collected in one of the study wells. This is probably related to the sampling methods. A third taxon collected with low abundance and intermediate frequency in three of the wells is the isopod *Proasellus* sp. (Asellidae). The rarity of this taxon is related to water withdrawal from the wells for irrigation: in fact, its density increased during the entire study period when water was not pumped.

In general, the distribution of the stygofauna in the Tafna alluvial aquifer was very heterogeneous. The spatial distribution of species in the groundwater indicates that they are preferentially located in areas influenced by the river water. In fact, the species diversity and abundance were higher in wells located near the riverbed than those further away along the bank. This distribution may reflect environmental conditions in the groundwater which correspond to the species' requirements. Almost all of the oxygen and organic matter required by the stygofauna comes from surface water (Ward and Palmer 1994); hence, there is an attraction effect of surface water on stygobitic fauna (Maridet et al 1992). Indeed, mixing with the stream surface water was higher near the bed. It should be noted that during low water flow the river was fed by dam release water (Taleb et al 2004). These results confirm the crucial role of connectivity between groundwater and surface water identified by Dole-Olivier (1998), and particularly in the wadi system by Gagneur and Chaoui-Boudghane (1991), Belaidi et al (2004) and Taleb et al (2008) (the term "wadi" generally used in North Africa to name watercourses refers to the great variations in flow).

We did not detect a differential distribution of the stygobitic taxa along a longitudinal gradient, despite the north-south gradient of increasing conductivity recorded in the wells. Indeed, an extremely high gradient of water salinity is tolerated by cirolanids, which live in water with salinity ranging from perfectly fresh water to almost sea water (Botosaneanu 1986). This would explain the wide distribution of this species in the Tafna alluvial aquifer and, similarly, the wide distribution recorded for *Salentinella* sp., which belongs to a genus that may live in saline waters (Notenboom 1991).

#### CONCLUSION

At present, we are still far from knowing the actual groundwater biodiversity in the Tafna watershed. In Algeria, data on the stygofauna are still very fragmentary and rare due to the few sampling campaigns conducted in the past. Significant efforts are still required to increase our knowledge about the presence and distribution of stygobitic fauna, and it is likely that many taxa have yet to be discovered.

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## The first record of external abnormalities in the subterranean *Aegla marginata* Bond-Buckup & Buckup, 1994 (Crustacea: Decapoda: Aeglidae), from a karst area of Southeastern Brazil

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#### ABSTRACT

The occurrence of a morphologically altered individual identified as *Aegla marginata* is reported in this note. The specimen was found in the subterranean environment, as part of wider bioespeleological study. The observed abnormalities consist mainly of deformities in abdominal epimera, pleopods, telson and uropods, which could results in difficulties for reproduction and escape from predators. Genetic or nutritional factors related to the scarce food supply observed in the cave environment are admitted as being the most probable cause of the deformities reported here. We emphasize the need of new studies in the area as well as efforts to preserve subterranean environment.

Key words: Anomura, abdominal deformities, nutritional factors, genetic factors, cave environment

#### INTRODUCTION

External abnormalities or deformities are just one of the common problems affecting freshwater and marine crustaceans, and have been reported in many natural crustacean populations (see Lawler and Van Engel 1973; Lira et al 2006; Luppi and Spivak 2007; Beguer et al 2008; Follesa et al 2008; Gregati and Negreiros-Fransozo 2009). The most common problems are modifications on chelipeds (Morgan 1923; Shuster Jr. et al 1963; Zou and Fingerman 2000; Benneti and Negreiros-Fransozo 2003), carapace spines (Moncada and Gomes 1980; Gregati and Negreiros-Fransozo 2009), pereiopods (Lawler and Van Engel 1973) and abdomen shape (Mantellato et al 2000). These alterations could be attributed to genetic factors (Zou and Fingerman 2000), accidents or predation that occurred during the molting process (Moncada and Gomes 1980; Luppi and Spivak, 2007; Follesa et al 2008), besides stress and environmental contamination (Beguer et al 2008).

For crustaceans of the Infraorder Anomura Mac Leay 1838, these records are still scarce. We can cite Nickerson and Gray Jr. (1967) that describe abnormalities on pereiopods of *Paralithodes camtschatica* (Tilesius, 1815) (Lithodidae); Fantucci et al (2008) that report intersexual specimens of *Isocheles sawayai* Forest & Saint-Laurent, 1968 (Diogenidae); and Jara and Palacios (2001) that described the occurrence of conjoined twins in *Aegla abtao* Schmitt, 1942 (Aeglidae).

The Aeglidae Dana, 1852, constitutes a distinctive family of Anomura with characteristic morphology, ecology, and reproduction. They are the only freshwater anomurans. The family consists of two fossil genera and one extant genus, *Aegla* Leach, 1820, which is endemic to temperate South America. The genus contains approximately 70 species and subspecies spread out over Chile, Brazil, Argentina, Uruguay, Paraguay, and Bolivia (Bond-Buckup and Buckup 1994; Pérez Losada et al 2002) in habitats such as lakes, streams, swamps, and caves (Bond-Buckup and Buckup 1994).

The species *Aegla marginata*, is relatively little studied in comparison with other congeneric species. It is known that to occur in both epigean and subterranean environments in Parque Estadual Intervales (PEI), Iporanga city, São Paulo State, southeastern Brazil (Rocha and Bueno 2004). In this region, the *A. marginata* populations present some differences in pigmentation among each other (Morachiolli 1994). As the species is capable of completing its entire life cycle in both subterranean and epigean streams, it is considered as troglophiles (Barr and Holsinger 1985; Morachiolli and Trajano 2002).

The purpose of this note is to present information, for the first time about the occurrence of abdominal abnormalities on a subterranean population of *Aegla marginata*.

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#### METHODS

As part of a bioespeleological study in the Parque Estadual Intervales (PEI), located in the city of Iporanga in the São Paulo State (Fig. 1), Brazil, anomuran crabs of the genus Aegla were sampled inside the cave Gruta Colorida (24° 16'13" S; 48° 25'09" W, registration number SP 129) by means of covo traps, in May 2009. All of the collected individuals (n= 15) were kept in plastic bags and refrigerated until they were analyzed. The identification was performed according to Buckup and Bond-Buckup (1994). An individual bearing abdominal deformities was identified by similarity with other individuals collected in the same place and occasion, because the second abdominal epimera, an important taxonomic character, was modified (Fig. 2 c). The specimen was photographed and measured under a stereoscope microscope equipped with camera and distances measurement system, and is stored in the scientific collection of the Laboratório de Estudos Subterrâneos, under the following registration numbers: CC1036.

#### RESULTS

The female specimen with external abdominal deformities was identified as being *Aegla marginata*, measuring 15.5 mm of carapace width and 17.6 mm of carapace length (Fig. 2 a, c, e). Sex was identified by the presence of developed pleopods and the presence of the genital apertures in the coxa of third pair of pereiopods, characters which are present only in females. In ventral position, there are considerable changes in the insertion of pleopods in the abdominal epimera, resulting in a non functional morphology (Fig. 2 a). It is also observed the absence of the fourth and fifth pleopods on the right side of the abdomen.

In dorsal view, the second abdominal epimera is modified, and the third and fourth ones are in abnormal posi-

Fig. 1 - Location of Parque Estadual Intervales (PEI), Iporanga, São Paulo State, Brazil (modified from Google Earth (2010), Digital Globe satellite).

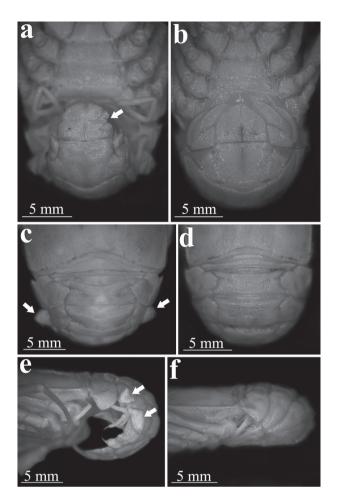


Fig. 2 - *Aegla marginata* on ventral, dorsal and lateral view. **a**, **c**, **e**) specimen found with carapace abnormalities; **b**, **d**, **f**) normal specimen found on same place. White arrows indicates the abnormalities on abdomen.

tion, folded backwards (Fig. 2 c, e). Between second and third abdominal somites is a flattening of the carapace, forming a scar. The telson is almost divided in two horizontal parts, forming an additional segment. The uropod is reduced to two buds, in both sides.

The other morphological characters are similar to the ones reported in the literature for the species *A. marginata* (Buckup and Bond-Buckup 1994) and for the morphological description of the genus *Aegla* (Martin and Abele 1988).

#### DISCUSSION

This malformation probably results in difficulties for egg incubation and aeration, as well as juvenile maintenance, since the genus shows evidence of parental care (López-Greco et al 2004). Moreover, as they present caridoid swim reflex (Martin and Abele 1988), probably the pleon deformities would result in greater difficulties to escape predators.

The regeneration of damaged appendages has been reported in the literature for several Decapoda, in case of injury or problems during molting (López-Greco et al. 2001; Luppi and Spivak 2007). This process is often flawed, resulting in scars and deformities (Luppi and Spivak 2007). In the case of the described specimen it is a hypothesis to be considered, although the apparent symmetry of the lesions in the uropods suggests an advanced process of regeneration that, given the extent of the lesions, would have resulted in the death of the individual.

High rates of incidence of anomalies in crustaceans have been associated with the presence of pollution by heavy metals and organophosphates (Betancourt-Lozano et al 2006; Beguer et al 2008; Sánchez et al 2005). Nonetheless, there is a low probability that these pollutants may cause the reported problem, because Moraes (2003), based on chemical analysis of water, sediment and fish tissues, established the levels of these substances as being below the risk levels for the area of Parque Estadual Intervales (PEI). However, new environmental analysis must be accomplished, mostly in benthonic invertebrates.

The subterranean environment depends on allochthonous food intake, which could mean food scarcity (Bichuette and Trajano 2003). As a result, the subterranean populations may have some differences when compared to the epigean ones, mainly in melanic pigmentation, metabolic rates, sex maturation and size (Poulson and White 1969; Mejía-Ortiz and López-Mejía 2005). Depending on the degree of nutritional deficiency, a high level of chronic distress develops, subjecting the individuals to attack by pathogens, which may cause of several deformities in crustaceans (Nunes and Martins 2002 *apud* Barroso 2005; Gregati and Negreiros-Fransozo 2009). Hence, this hypothesis must be considered in the reported case.

Studies with Decapoda species in cultivation environment has demonstrated that populations subjected to inbreeding can present some morphological effects as deformities (De Donato et al 2005), and asymmetries (Maia et al 2009a). In natural decreased populations without gene flow with other populations, the loss of genetic diversity is plausible. That is provided they are subjected to founder effect and genetic drift (Barr 1967; Poulson and White 1969; Trajano 2007), mainly if a reproductive connection with other populations does not occur to replace this loss. In that way, some populations could be subjected to endogamic depression effects and, consequently, to morphological effects in the form of deformities or malformations (Poulson and White op. cit.).

The cave system where the specimen was collected is developed in limestones that occur discontinuously and are interrupted by unsolvable rocks such as granites, phyllites and quartzites (Trajano 1991; Bichuette 1998). This fact suggests reduced gene flow between caves, since unsolvable rocks could limit the contact between populations by isolating the caves.

Evidence that *A. marginata* populations can be isolated was observed in the work of Morachiolli (1994), who found populations of the same species showing different levels of pigmentation in the same cave system. However, it is not known if that is due to genetic differences, pigments eaten by epigean individuals, food type, or absence of light in hypogean environment necessary to aggregate some pigments (Cullingford 1962; Morachiolli 1994).

The absence of light has also been reported in the literature as responsible for deformations in nontroglobitic (acidental) fishes. Rasqueen and Rosenbloom (1954) *apud* Poly and Boucher (1996), reported the occurrence of esqueletal deformations associated with darkness in epigean individuals of *Astyanax mexicanus* (De Filippi, 1853) maintained in absence of light in the laboratory, possibly due to hormonal imbalance. Other deformations that could be associated with subterranean environment were the lack of pelvic fins or deformed caudal fins in *Ameiurus natalis* (Lesueur, 1819) (Relya and Sutton (1973) *apud* Poly and Boucher (1996).

We speculate that absence of light is not responsible for deformations observed in *A. marginata*, because this species is generally considered to be a troglophile (Morachiolli and Trajano 2002). It is therefore well adapted and capable of completing its entire life cycle in a subterranean environment.

The distribution of the genus Aegla, which is restricted to temperate and subtropical South America (Bond-Buckup and Buckup 1994), contributes to its vulnerability (IUCN, 2001). In addition the situation is aggravated by the present drastic reduction of populations in the Ribeira Valley (Maia et al 2009 b) and, possibly, by the founder effect and increased homozygosity, both very common in subterranean populations (Barr 1967). According Brook et al (2002), Spielman et al (2004) and Buhay and Crandall (2005), loss of heterozigosity and inbreeding play an important role in the extinction of threatened species. Therefore, we could consider the need of studies related to effective population size and genetic diversity in this population (Buhay and Crandall 2005), as well as efforts to preserve subterranean environment.

In as much as only one specimen was registered with anomalies, it is not possible to establish if these problems occur by chance, or are influenced by the above mentioned factors. Genetic or nutritional factors are suggested as being the most probable cause of the deformities reported here.

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# A new species of the subterranean amphipod crustacean genus *Stygobromus* (Crangonyctidae) from a cave in Nevada, USA

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### ABSTRACT

*Stygobromus albapinus*, a new stygobiotic amphipod crustacean species in the family Crangonyctidae, is described from two pools in Model Cave in Great Basin National Park, White Pine County, Nevada, USA. The type specimens were collected on two different visits to the cave. The new species is assigned to the *hubbsi* group, bringing the number of described species in this group to 45, but many other provisionally recognized species assigned to this group remain undescribed. With exception of a single species from deep wells in southeastern Wisconsin, all other members of the *hubbsi* group are recorded from a wide variety of subterranean groundwater habitats (e.g., caves, springs, wells, etc.) in western North America, west of the Great Plains. Although the taxonomic affinities of *Stygobromus albapinus*, n. sp. need further study, the species does appear to share several important morphological characters with a species from a cave in western Utah located approximately 300 km east-northeast of Model Cave.

Key words: amphipod, Stygobromus, subterranean groundwater, stygomorphic, hubbsi group

## INRODUCTION

The genus Stygobromus is comprised exclusively of stygomorphic species (typically lacking eyes and pigment) that inhabit a variety of subterranean groundwater habitats, including cave pools and streams, phreatic water in wells, seeps (hypotelminorheic) and springs, the underflow (hyporheic) of surface streams, and rarely deep lakes (Ward and Holsinger 1981; Holsinger 2009, see also Culver and Pipan 2009). In North America, the genus is represented by 129 described species, although numerous new species have been discovered and are either undergoing description (22 in manuscript by JRH) or are provisionally recognized. In addition, four (and possibly five) species are recorded from Eurasia, bringing the total number of described species in the genus to 133 or 134 (Sidorov et al 2010). In North America more than 50 species are found in the western United States and southwestern Canada (Wang and Holsinger 2001), the majority of which lack sternal gills or processes and have been assigned to the hubbsi group (see Holsinger 1974; Wang & Holsinger 2001) primarily on the basis of this diagnostic character. The systematics of the genus in North America are treated in a series of major papers produced over several decades (e.g. Holsinger 1974, 1978, 2009; Wang and Holsinger 2001), along with several others more restricted in scope (Holsinger 1980; Bousfield and Holsinger 1981; Holsinger and Shaw 1986, 1987; Ward 1977). The most recent paper in this series by Sidorov et al (2010) includes descriptions of two new species from Siberia. These papers should be consulted

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for keys, taxonomic changes, and specific distributional information for the genus.

In the course of a survey of the cave fauna of Great Basin National Park (White Pine County, Nevada), an undescribed *Stygobromus* species of the *hubbsi* group was collected from two different pools in Model Cave in Great Basin National Park, and it is described herein. The description of S. *albapinus* n. sp. increases the number of species assigned to the *hubbsi* group to 45, all but one of which occur in the western Cordillera of North America.

#### METHODS AND MATERIALS

Specimens were collected directly from cave pools and preserved in 70% ethanol. In the laboratory permanent slide preparations were made by mounting dissected appendages in Faure's mounting medium (a modification of the conventional Hoyer's medium [Upton 1993]) on glass slides. Slide-mounted appendages were photographed using a Leica DM 2500 compound microscope, and stacked digital images were created using Image-Pro Express (version 5.1.1.14, ©2005, Media Cybernetics, Inc.). Images were imported into Adobe Illustrator CS2 (version 12.0.1, ©2005 Adobe Systems, Inc.), where final line drawings were produced. Drawings are of female specimens unless otherwise indicated.

In the description, usage of the descriptive terms "seta" and "spine" are in the original conventional sense and do not agree with terminology suggested by Watling (1989). We use the term "spine" for thick and stiff setae and the term "seta" for thin and flexible structures. No-

menclature for setal patterns on segment 3 of the mandibular palp follows the system introduced by Stock (1974). The term "defining angle" of the gnathopod propodi refers to the "angle" formed by the end of the palm and beginning of the posterior margin or the point at which the tip of the dactyl closes on the propodus.

## SYSTEMATICS

# Stygobromus albapinus Taylor & Holsinger, sp. nov. (Figs 1-5)

# Material Examined

USA, NEVADA. White Pine County, Great Basin National Park, Model Cave,  $\bigcirc$  holotype and 6 paratypes (1  $\bigcirc$ , 5  $\bigcirc$   $\bigcirc$ ) collected from pool of water by Gretchen M. Baker, 10 November 2008; 40 paratypes (1 immature  $\bigcirc$ , 39  $\bigcirc$   $\bigcirc$ ) collected by Gretchen and Ben M. Roberts from pool of water (7° C) and sump pool (4° C), 2 February 2009.

The holotype  $\bigcirc$  (4.5 mm) is deposited in the National Museum of Natural History (Smithsonian Institution), USNM 1135494; 5 paratypes are deposited in the Illinois Natural History Survey (INHS) Crustacean Collection (INHS 11447, INHS 11448); remaining paratypes are in the research collection of J. R. Holsinger at Old Dominion University (H-4564, H-4653).

#### Diagnosis

A relatively small stygobiotic species of the *hubbsi* group distinguished by structure of pereopods 6 and 7, which have relatively long, narrow bases lacking distinct distoposterior lobes; uropod 3, which has a very short ramus bearing 2 small, apical spines; and sub-rectangular-shaped telson with small V-shaped apical notch and 7 to 8 apical spines. This species appears to be morphologically more similar to *S. utahensis*, Wang and Holsinger (2001) from Pole Creek Cave in Duchesne Co., Utah and possibly *S. blinni*, Wang and Holsinger (2001) from Roaring Springs Cave in Coconino Co., Arizona than to any other species in the *hubbsi* group known to date. Largest  $\bigcirc$ , 5.0 mm; largest  $\bigcirc$ , 3.5 mm

#### Female (Fig. 1)

Antenna 1 (Fig. 2B) approximately 45% percent length of body, 25 % longer than antenna 2 (Fig. 2C) primary flagellum with 11 segments, most bearing aesthetascs (not shown); accessory flagellum approximately equal in length to 1<sup>st</sup> flagellar segment. Antenna 2



Fig. 1- *Stygobromus albapinus* new species, (ca. 5.0 mm) female paratype, collected 2 February 2009 in Model Cave, Great Basin National Park, White Pine County, Nevada.

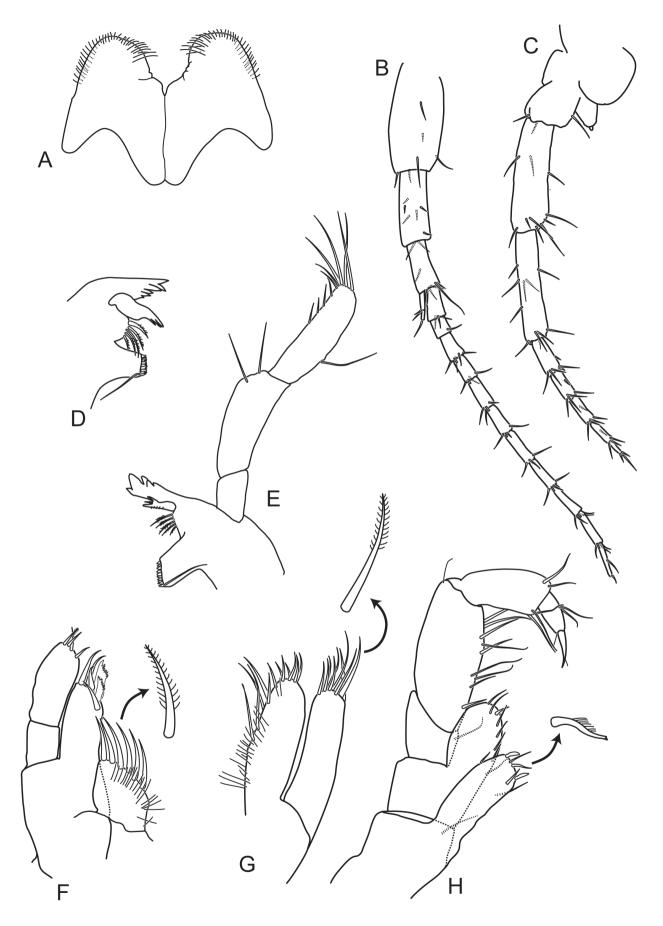


Fig. 2 - *Stygobromus albapinus*, new species, female paratype (5.0 mm), Model Cave, Nevada: A, lower lip; B, antenna 1; C, antenna 2; D, left mandible; E, right mandible; F, maxilla 1; G, maxilla 2; H, maxilliped.

(Fig 2C): peduncular segments approximately equal in length, flagellum with 5 segments. Mandibles (Fig. 2D,E) closely similar but differing in a few minor ways: spine row of both with 5 lightly plumose spines; lacinia mobilis of left with 2 or 3 tiny teeth, that of right apically forked with 3 tiny spines.; incisors normal for genus; mandibles cone shaped, protruding, each with single seta. Palp segments subequal; segment 2 bearing 2 or 3 setae on inner margin distally; segment 3 with 4 long E setae, 5 or 6 shorter D setae and 1 B seta; lacking both A and C setae. Lower Lip: inner lobes present. Maxilla 1 (Fig. 2F): inner plate with 6 apical plumose setae; outer plate with 6 apical spines (2 or 3 with serrated margins); palp with 4 short, apical setae. Maxilla 2 (Fig 2G): inner plate with 4 relatively long naked setae on distal half, followed by approximately 8 shorter setae apically; outer plate with 9 or 10 apical, plumose setae. Maxilliped (Fig. 2H): inner plate with 2 bladelike spines, 1 plumose spine, and 2 naked setae apically; outer plate with 7 or 8 short setae on upper, inner margin; palp segment 2 broader than and more than 2X longer than palp segment 1, inner margin with row of about 8 rather long setae; nail of dactyl (palp segment 4) relatively long and sharply pointed.

Gnathopod 1 (Fig. 3A): propodus smaller than that of gnathopod 2, palm straight and about 25 % longer than posterior margin, armed with double row of about 15 spine teeth in unequal double row; defining angle rounded, with 3 spine teeth on outside, 2 shorter ones on inside; posterior margin lacking setae; 1 superior medial setae, 3 inferior medial setae; dactyl nail short; coxa little broader than deep, ventral margin with 1 seta. Gnathopod 2 (Fig 3B): propodus longer than broad, palm nearly straight, margin irregular, armed with 11 or 12 spine teeth in double row; defining angle with 1 long spine tooth on outside, 2 shorter spine teeth on inside; posterior margin approximately 50 % length of palm, with 2 sets of doubly inserted setae; row of 4 superior medial setae, and 2 inferior medial setae. Coxa of gnathopod 2 slightly broader than deep, margin with 1 seta. Brood plates well developed in sexually mature females (Fig 3C).

Pereopods 3 and 4 (Fig. 4A, B) subequal; coxae subquadrate, slightly broader than deep, reaching about 25 percent of length of basis, ventral margins with 2 setae each. Pereopods 6 and 7 (Figs. 4D, E) subequal in length, 50% length of body, 1.25 X longer than pereopod 5 (Fig. 4C). Bases of pereopods 5-7 relatively narrow, slightly tapering distally but little broader proximally than distally, with few short marginal, distoposterior lobes obsolete; dactyls of pereopods 5-7 relatively long, 30-35 % length of corresponding propodi. Coxal gills (Figs. 4A-D) present on gnathopod 2 and 3-6 (absent from 7). Brood plates (Fig. 3C) relatively long, bearing long distal setae. Sternal gills (processes) absent.

Pleonal plates (Fig.5A): posterior margins weakly convex, each with 1 setule; distoposterior corners weakly rounded; ventral margins weakly convex to nearly straight, plate 3 with single setule. Pleopods (not shown) normal for genus. Urosomites free. Uropod 1 (Fig. 5B): Inner ramus slightly longer than outer ramus, about 78 % length of peduncle, with 9-10 spines; outer ramus with 7 spines; peduncle with 6 spines. Uropod 2 (Fig. 5C): inner ramus markedly longer and broader than outer ramus, nearly as long as peduncle, with 8 or 9 spines; outer ramus with 5 spines; peduncle with 2 spines. Uropod 3 (Fig. 5D): ramus tiny, much smaller than peduncle, bearing 2 tiny spines apically; peduncle at least 4X larger than ramus, with 1 tiny setule. Telson (Fig. 5E) nearly twice as long as broad, apex with distinct V-shaped notch and bearing 7 or 8 spines.

### Male

Closely similar to the female but apparently reaching sexual maturity at a slightly smaller size and differing in the morphology of uropod 1 (Fig. 5F) as follows: inner ramus equal in length to outer ramus, about 70 percent length of peduncle, armed with 7or 8 spines; outer ramus with 5 spines; peduncle with 4 small spines on dorsal margin; possession of distinct peduncular process approximately 20 % percent length of peduncle and bearing 5 tiny peg-like spines.

#### Etymology

The specific epithet *albapinus* is a contraction of *alba* (Latin for white) and *pinus* (Latin for pine), in reference to White Pine County, Nevada—the type-locality and only known population of this species.

#### Distribution & Ecology

The species is known only from two pools in Model Cave, Great Basin National Park, White Pine County, Nevada, where specimens were collected from both a mud/silt bottom pool with temperature of 7°C and a sump pool with temperature of 4°C. The majority of specimens were collected from the mud/silt bottom pool. Of the 47 specimens sampled, only one is a mature male, suggesting a tendency for parthenogenesis; a trend previously noted for several other species of *Stygobromus* (Culver and Holsinger 1969).

## DISCUSSION

The description of *S. albapinus* n. sp. brings the total number of species of *Stygobromus* described from western North America west of the Great Plains to 54, of which 45 are assigned to the *hubbsi* group. Descriptions of 17 new species of *Stygobromus*, and redescription of *S. hubbsi* Shoemaker (1942) from western North America, west of the Great Plains, were provided earlier by Holsinger (1974). This paper was followed more recently by descriptions of an additional 28 new species from the same region by Wang and Holsinger (2001). In addition, there are more than 10 provisionally recognized, undescribed new species of *Stygobromus* from California and

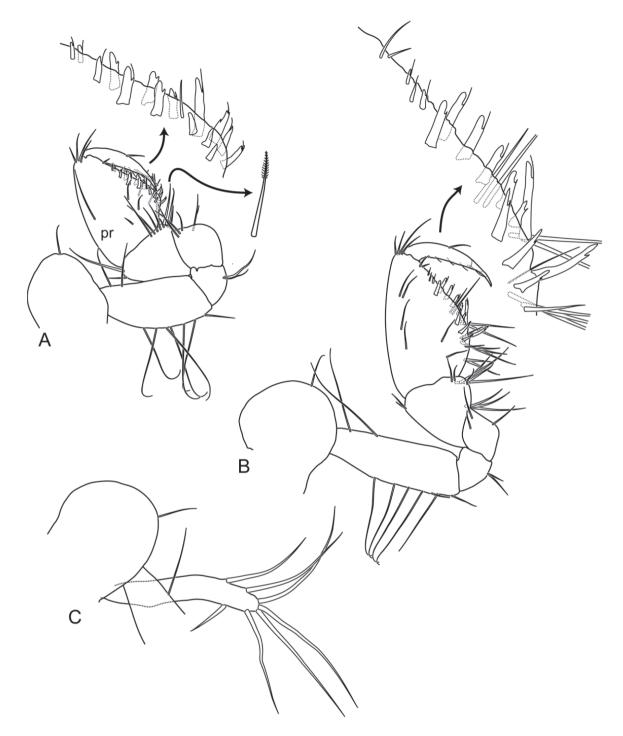


Fig. 3 - *Stygobromus albapinus*, new species, female paratype (5.0 mm), Model Cave, Nevada: A, gnathopod 1 (palm enlarged); B, gnathopod 2 (palm enlarged); C, coxal plate (in part) and setose brood plate of gnathopod 2.

Washington State and several more from southern Alberta, Canada and southeastern Alaska (J. R. Holsinger unpublished data). Most, if not all, of the undescribed species from California and Washington can be assigned to the *hubbsi* group. It is also of biogeographic interest that only one species (*Stygobromus putealis* Holmes) assigned to the *hubbsi* group has been found outside western North America in southeastern Wisconsin (Holsinger 1974; Wang and Holsinger 2001). Despite relatively close morphological similarities among most of the species, and a preliminary cladistic analysis combined with a list of character states (Wang and Holsinger 2001), the origin, interspecific relationships and biogeography of these species remain in need of further study, especially those assigned to the *hubbsi* group. In addition, given the cryptic habitats and seclusion of groundwater amphipods in western North America, many undescribed

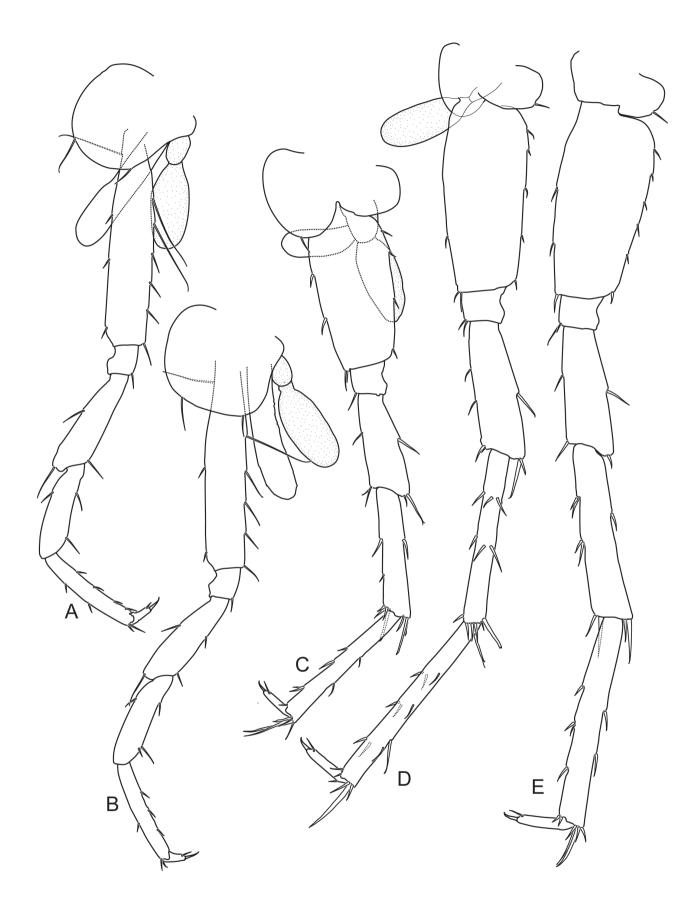


Fig. 4 - Stygobromus albapinus, new species, female paratype (5.0 mm), Model Cave, Nevada: A-E, pereopods 3-7, respectively.

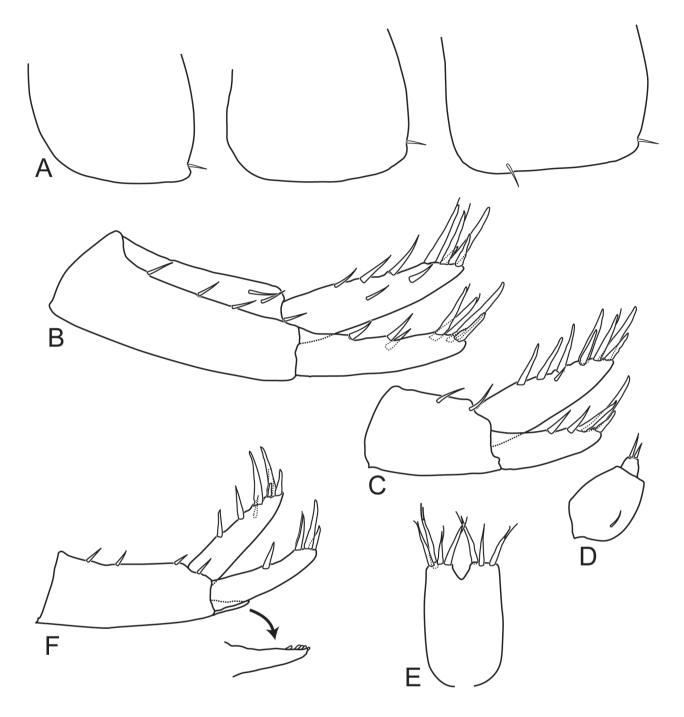


Fig. 5 - *Stygobromus albapinus*, new species, female paratype (5.0 mm), Model Cave, Nevada: A, pleonal plates 1-3; B, uropod 1; C, uropod 2; D, uropod 3; E, telson. Male paratype (3.5 mm), Model Cave: F, uropod 1 (peduncular process enlarged).

species probably remain undiscovered. Thus, future discovery and additional taxonomic study, including molecular analyses, should provide a better idea of the taxonomic and biogeographic relationships among western species.

Wang and Holsinger (2001) suggested that changes in the Pleistocene pluvial lake system of western North America could have affected the distribution of *hubbsi* group species in the Great Basin. Groundwater levels would have been elevated during glacial maxima, followed by disjunctions when groundwater levels were subsequently lowered, and this in turn may have provided the opportunity for speciation in this area. However, it was also noted that the majority of western species occur outside of the areas impacted by the pluvial lakes (Fig. 6), and that the *hubbsi* group is split roughly into a western subgroup that occurs in the Cascades-Sierra Nevada-Coastal Range and an eastern group that occurs in the Rocky Mountains and Colorado Plateau. Between these two groups, the *hubbsi*  Fig. 6 - Generalized distribution of described species of *Sty-gobromus* in western North America (closed circles) based in part on Wang & Holsinger (2001), with location of *Stygobromus albapinus* sp. nov. indicated by a star. Dashed line indicates extent of the Great Basin.

group is represented by only seven species that are recorded to date from the Great Basin and Columbia Plateau (Wang and Holsinger 2001). Finally, it is perhaps of interest that none of the non-*hubbsi* group western species occur in the Great Basin or further west (Wang and Holsinger 2001).

What does the occurrence of the new stygobiotic species from Great Basin National Park tell us about the Stygobromus fauna of the Great Basin as well as about other cave and groundwater adapted invertebrates? Recent inventories of cave-adapted invertebrate organisms in Great Basin National Park have resulted in the discovery of new species of millipeds and collembolans (Shear 2007, Shear et al 2009; Zeppelini et al 2009), as well as undescribed diplurans and pseudoscorpions. The description of S. albapinus n. sp. brings the total number of new invertebrate taxa described from caves in this small area as a result of recent bioinventory work to four. Studies of spring-inhabiting hydrobiid and pleurocerid snails in the Great Basin, many of which occur at one or few sites, have shown that high diversity of springsnail species in the Great Basin is a reflection of their strong association with points of groundwater resurgence and the regional history of aquatic connections (Brown et al 2008). Furthermore, a genetic analysis of amphipods in the epigean genus Hyalella (Talitridae) from numerous spring sites in the Great Basin identified a high level of genetic diversity

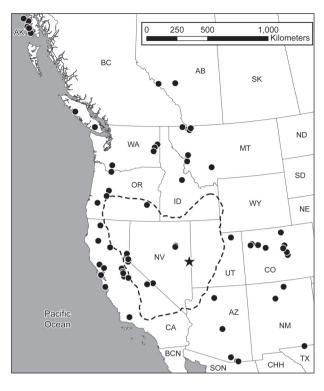
strongly indicating the presence of a multitude of cryptic, undescribed species of Hvalella in this region (Witt et al 2006). These findings suggest that the cave and spring faunas of the Great Basin region have not been thoroughly sampled to date. Moreover, sampling methods needed to obtain subterranean amphipods have not to our knowledge been widely implemented in surveys of Nevada's subterranean groundwater faunas. Despite extensive surveys for springsnails and epigean amphipods in the Great Basin, cryptic subterranean groundwater habitats commonly inhabited by Stygobromus are not as easily accessed and remain poorly sampled. Utilization of special techniques for sampling small groundwater crustaceans, such as the Bou-Rouch groundwater pump and similar tools (e.g. Leijs et al 2009) for probing relatively deep (~30 cm) into the substrate of spring outlets, may reveal additional undescribed species of Stygobromus. A number of factors have to date precluded rigorous sampling of potential groundwater habitats in the Great Basin, perhaps the most important being the marked physical isolation and relatively remote location of aquatic habitats common to Great Basin topography. The occurrence of numerous isolated springs associated with the bases of mountain ranges offers the potential of finding additional populations of Stygobromus species within this region. It is likely that the recent discovery of Stygobromus albapinus n. sp. will be followed by additional discoveries with further exploration of groundwater habitats in the Great Basin region.

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# The Mexican cavernicolous *Pseudosinella* (Collembola: Entomobryidae) with description of a new species

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### ABSTRACT

A compilation of the information on the genus *Pseudosinella* from Mexican caves was undertaken and one new species from Puebla State is described and illustrated: *P. rochezi*, sp. nov. It is similar to *P. bonita* Christiansen, 1973 and displays typical troglomorphic characters. An identification key for all Mexican cave species of the genus is presented.

Key words: Cave fauna, Pseudosinella, identification key, México, new taxa

## INRODUCTION

*Pseudosinella* is one of the most diverse genera of springtails with about 330 species (Bellinger et al 1996-2010). Members of the genus are found in all countries and in many diverse habitats, including soil, litter, on vegetation and in caves.

Eighteen species of the genus, including the new one described below, have been recorded from Mexican caves. However, some remain undescribed (sp. BB and sp QQ of Christiansen, 1982). The most important contributions to the taxonomy of the cavernicolous members of the genus in Mexico were made by Christiansen (1973, 1982) and Christiansen and Reddell (1986). One recent key for the identification of these species was prepared and is given here.

Those species with troglomorphisms are usually big in comparison with those living in soil and litter or even with those living in caves but without strong adaptations to cave life (Table 1). Here we describe a new taxon showing interesting morphologic characters, which are a clear indication of adaptation to cave life. (Table 1).

For description of the new species and construction of keys, we used the system of Gisin (1967) and the formula of Christiansen et al. (1990). In keeping the same system we use the same abbreviations as those in our recent contribution to the genus (Simón Benito and Palacios-Vargas 2007). Also, we use the keys posted on the web page of Christiansen (2007).

# SPECIES DESCRIPTION

# *Pseudosinella rochezi*, sp. nov. (Figs 1-6, 7-11, Table 2)

#### Material examined.

Mexico, State of Puebla, karst region of Tzontezecuiculi, in one small cave close to Tepepan Zaragoza, 19.II.2002. (GPS TZ 22 coordinates 14 Q 0720429 UTM 2038732, 1,450 m elevation. - 1 male (holotype) found in decomposing tree trunk.

# Etymology.

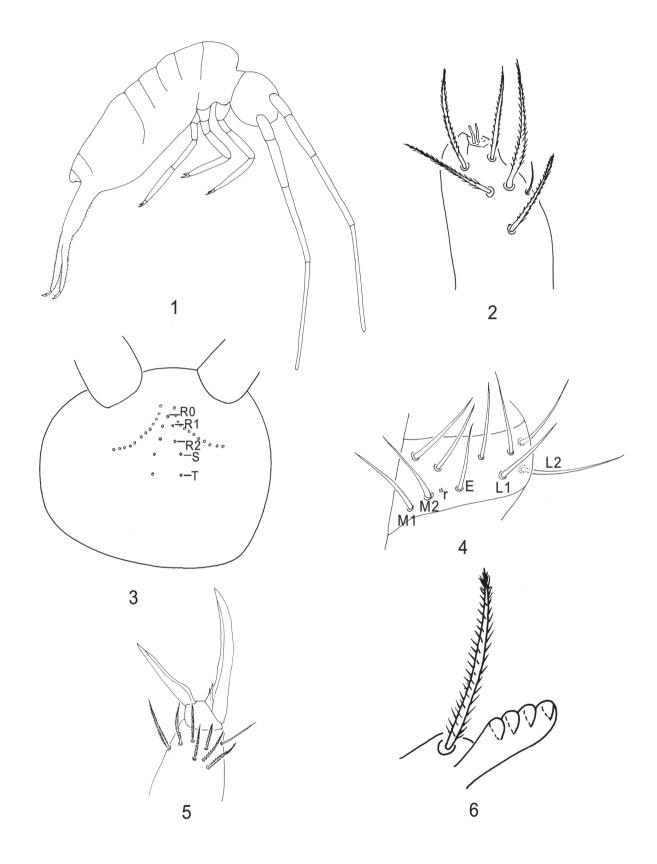
This species is named after Mr. Gaëtan Rochez, who collected the specimen.

*Description*. Length 2.45 mm. Body without pigment, 0+0 eyes.

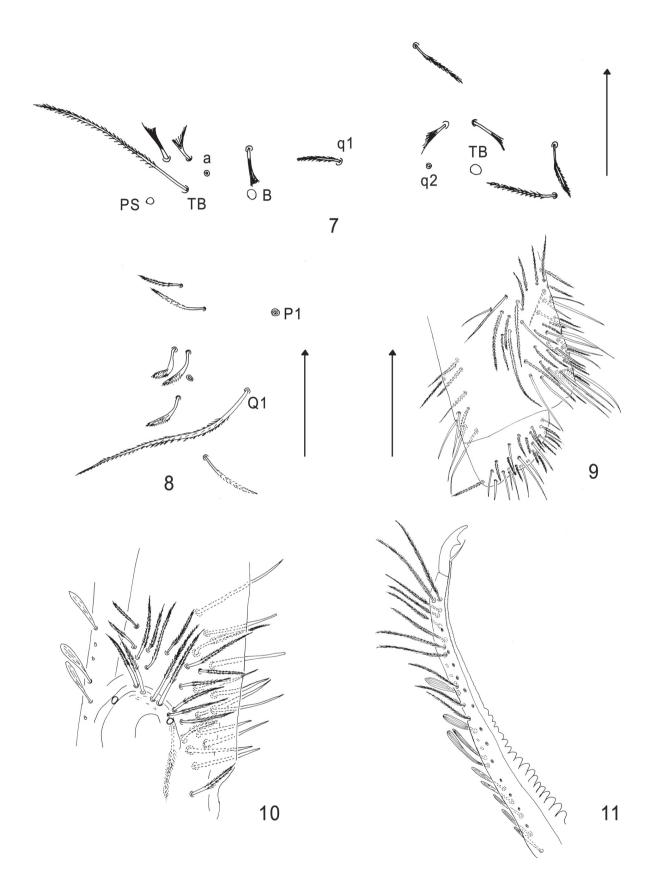
Antenna without pigment or scales, and 3.6 times longer than head. Ant. IV without a clear apical bulb. Ratio of antennal segments as 1: 1.6; 3.0; 4.0 (Fig. 1). Apical organ of third antennal segment with two straight short rods, difficult to observe (Fig. 2). Head with macrosetae R0, R1, R2, S and T; and Po macroseta absent (Fig. 3). Four triangular labral papillae. Differentiated setae of outer labial papilla (E) straight and its apex not reaching the apex of papilla, with 3 sublobular setae on the external maxillar lobule. Labial triangle formula:  $M_1$ ,  $M_2$ , r, E,  $L_1$ ,  $L_2$  (Fig. 4), all smooth. Five ciliated setae along the head ventral groove.

Mesothorax slightly protuded, pushing head into small hypognathus position. Legs without scales, tibiotarsus with ciliate and acuminate macrosetae, and also with some differentiated setae. Tenent hair acuminate and shorter than the internal length of ungues (ungues 3.6 times as long as tenent hair). Unguis long and slender, with two inner teeth; basal pair smaller, distal tooth located about 10 % of the internal side of unguis; distal unpaired tooth located about 17 % the internal side of unguis; lateral and outer tooth absent (Fig. 5). Unguiculus smooth, lanceolate, basally swollen and about the length of unguis (unguis is 1.3 times the length of unguiculus).

Dorsal macrosetae formula: R110/00/0101+2. Chaetotaxy of Abd. II:  $apBq_1q_2$ , setae "a" fan-like shaped ciliate (Fig. 7). Bothriotrical complex on Abd. IV without accessory seta "s" (Fig. 8).



Figs 1-6 - *Pseudosinella rochezi*, sp. nov.: 1, habitus; 2, sensorial organ of Ant. III; 3, head chaetotaxy; 4, labial triangle; 5, foot III complex; 6, half retinaculum lateral view.



Figs 7-11 - *Pseudosinella rochezi*, sp. nov.: 7, chaetotaxy of anterior bothriotrichal complex of Abd. II; 8, bothriotrichal complex of Abd. IV; 9, ventral tube in lateral view; 10, distal part of manubrium and basal part of dens; 11, distal part of dens and mucro.

	Locality	Biotope	Maximal size in mm
P. violenta (Folsom, 1924)	Durango, Chihuahua,	Soil and litter	2.1
	Coahuila		
P. sp. QQ	Mexico, Central America,	Cave	1.2
	West Indias		
P. petrustrinatii Christiansen, 1982	Guerrero, San Luis Potosi,	Cave	?
	Tamaulipas		
P. cava Christiansen & Reddell, 1986	Hidalgo, San Luis Potosí,	Cave	1.5
	Querétaro		
P. finca Christiansen, 1973	Puebla, Oaxaca, Morelos.	Cave	1.8
	Guatemala: Verapaz		
P. palaciosi Christiansen & Reddell, 1986	Guerrero	Cave	0.8
P. hautla Christiansen, 1982	Oaxaca	Cave	2.0
P. bonita Christiansen, 1973	Oaxaca	Cave	3.6
P. crypta Christiansen & Reddell, 1986	Sierra Madre Oriental	Potrero Redondo	3.0
P. yuca Christiansen, 1982	Yucatán	Cenote	0.8
P. reddelli Christiansen, 1973	Nuevo León, San Luis Potosí,	Cave	2.5 - 3.1
	Coahuila, Tamaulipas		
P. leoni Christiansen, 1982	Nuevo León, Oaxaca	Cave, Guano	2.5
P. sp. BB	México (country)	Litter	1.1
P. vera Christiansen, 1982	San Luis Potosí, Puebla,	Cave	1.7
	Veracruz		
P. voylesi Christiansen, 1982	Puebla	Cave	3.08
P. volca Christiansen, 1982	Veracruz	Cave	1.8
<i>P. rochezi</i> , sp. nov.	Puebla	Cave	2.4
P. strinatii Christiansen, 1973	San Luis Potosí, Tamaulipas	Cave	1.6

Table 1 - Comparison of size and habitat distribution among the different species of *Pseudosinella* found in Mexican caves.

Ventral tube with 12+12 barbulate distal setae and 10+10 smooth distal setae and 5+5 in the posterior medial region (Fig. 9). Retinaculum with 4+4 teeth and one barbulate seta (Fig. 6). Distal part of manubrium dorsally with 2-3 internal and 3-4 external setae separated by 2 pseudopores (Fig. 10). Dentes basally with long and smooth spine-like setae (Fig. 10). Mucro bidentate, mucronal spine smooth (Fig. 11).

# DISCUSSION

*Pseudosinella rochezi*, sp. nov. is similar to the Mexican cave species *P. vera*, *P. huautla*, *P. volca* and *P. bonita* in the habitus and the unusual elongation of antennal segment IV. Similarities and differences of the five species are shown in Table 2. The new species most closely resembles *P. bonita* (3.6 mm) but the two species differ in size (*P. rochezi* sp. nov. is 2.4 mm), and in addition the new species has dorsal head macroseta T and the smooth microseta r. For the number of inner and outer setae of manubrial setae of manubrial plate, it is more similar to *P. vera*. Dens in *P. bonita* has small spines, in *P. rochezi*, sp. nov. they are longer. The anntenal/cephalic diagonal in the new species is 3.7, more similar to *P. huautla*. The apical organ of third antennal segment is rod-like, differing from all other Mexican cave species. *P. rochezi*, sp. nov. was found in one cave in Puebla state and *P. bonita* was found in three caves in neighboring Oaxaca state (Table 1).

Key to Mexican species of Pseudosinella.

1	Unguiculus with a winged tooth P. violenta
-	Unguiculus without a winged tooth2
2	Base of dens with short spines
-	Base of dens without short spines
3	Unguis thin with three internal teethP. crypta
-	Unguis with less than three internal teeth
4	Tenent hair capitate
-	Tenent hair acuminate
5	Eyes present
-	Eyes absent
6	Five eyes per sideP. sp. QQ
-	Three eyes per side7

53

Table 2 - Comparison of characters of *P. rochezi*, sp. nov. with those of the closest species. Number in parenthesis is the character state proposed by Christiansen (2007).

	vera	hautla	volca	bonita	<i>rochezi</i> , sp. nov.
1. dorsal cephalic macrosetae S	absent (1)	absent (1)	absent (1)	present (2)	present (2)
2. dorsal cephalic macrosetae T	absent (1)	present (2)	present (2)	absent (1)	present (2)
3. Labial m1	smooth	smooth	smooth macroseta	smooth	smooth
	macroseta	macroseta	(3)	macroseta	macroseta
	(3)	(3)		(3)	(3)
4. Labial m2	smooth	smooth	smooth macroseta	smooth	smooth
	macroseta	macroseta (3)	(3)	macroseta (3)	macroseta
	(3)				(3)
5. r (ventral labial)	absent (5)	absent (5)	smooth microsetae	smooth	smooth
			(1)	microseta (1)	microsetae (1)
6. e (ventral labial)	smooth	smooth	smooth macrosetae	smooth	smooth
	macrosetae (3)	macrosetae (3)	(3)	macrosetae (3)	macrosetae (3)
7. L1 (ventral labial)	smooth	smooth	smooth macrosetae	smooth	smooth
	macrosetae (3)	macrosetae	(3)	macrosetae	macrosetae
		(3)		(3)	(3)
8. L2 (ventral labial)	smooth	smooth	smooth macrosetae	smooth	smooth
	macrosetae (3)	macrosetae (3)	(3)	macrosetae	macrosetae
				(3)	(3)
9.a (second abd seta)	smooth	smooth	smooth macrosetae	smooth	ciliate
	macrosaeta	macrosetae (3)	(3)	macrosaeta	microseta (2)
	(3)			(3)	
10.b (second abd seta)	smooth	smooth	smooth macrosetae	smooth	ciliated
	macrosaeta	macrosetae	(3)	macrosetae	macrosetae
	(3)	(3)		(3)	(4)
11. p (second abd seta)	absent (1)	absent (1)	absent (1)	absent (1)	absent (1)
12. q1 (second abd seta)	smooth	smooth	smooth macrosetae	smooth	smooth
	microsetae	microsetae	(1)	microsetae	microseta?
	(1)	(1)		(1)	
13. q2 (second abd seta)	smooth	smooth	smooth microsaeta	smooth	smooth
	microsaeta (1)	microsaeta	(1)	microsaeta (1)	microseta?
		(1)			
14. posterior thoracic seg 2	clavate (2)	truncate (3)	(?)	absent (4)	absent (4)
macrosetae				<b>^</b>	
15. posterior thoracic seg 2	1 (one)	2 (two)	1 (one)	0 (zero)	0 (zero)
macrosetae	1 (2)		1 (2)	1 (2)	1 (2)
16. thoracic seg 3 macrosetaee	absent (3)	(?)	absent (3)	absent (3)	absent (3)
18. anterior lateral(P) fourth	1(2)	1(2)	1(2)	1(2)	1(2)
abdominal dorsal macrosetaee	2(2)	2(2)	2(2)	2(2)	2(1) 0
19. median (M) 4th abd dorsal	2(2)	2(2)	2(2)	2(2)	2(1)?
macrosetaee					1 (1)
20. supplementary seta (4th	present (2)	present (2)	present (2)	present (2)	absent (1)
abd seg)	•	•	• • • • • • • • • • • • • • • • • • • •	•	• • • • • • •
21. tenent hair shape	acuminate $(1)$	acuminate $(1)$	acuminate (1)	acuminate (1)	acuminate $(1)$
22. number of teeth of inner	3(2)	3(2)	3(2)	1(2)	2(1)
unguis		1 (1)	1 (1)	1 (1)	1 (1)
23. ungual wing tooth	present (2)	absent (1)	absent (1)	absent (1)	absent (1)
24. unquiculus wing tooth	absent (1)	minute (2)	absent (1)	absent (1)	absent (1)
25. unguiculus shape	acuminate (1)	acuminate (1)	acuminate (1)	basally swollen	basally swollen
			basally swollen (3)	(3)	(3)
26. number of eyes per side	0-2(zero-three)	0 (zero)	0 (zero)	0 (zero)	0 (zero)

27. inner setae manubrial plate	2(2)	2-3(2-3)	3(3)	3(3)	2(2)
28. no. outer setae manubrial	3 (4-6)	range: 2 (2-6)	5(5)	6-10	4(four)
plate					
29. habitat	cave $(1)$	cave (1)	cave (1)	cave (1)	cave (1)
30. region located	mexico (3)	mexico (3)	mexico (3)	mexico (3)	mexico (3)
31. apical antennal bulb	absent (1)	absent (1)	absent (1)	absent (1)	absent (1)
32. apical organ of third	expanded (2)	expanded (2)	expanded (2)	paddle-shaped	peg or rod-like
antennal segment				(3)	(1)
33. maximum length	1.7 mm.	2 mm.	1.8 mm.	3.6 mm	2.4 mm
34. distance distal unpaired	55 %	?	?	17%	0.15 %
ungual tooth from base total					
unguis %					
35. antennal cephalic diagonal	1.3	3.5	2.5	4.5	3.7
36. differentiated inner seta on	not applicable	clear	clear? acuminate	?	clear acuminate
hind tibiotarsus		acuminate (1)	(1)		(2)
37. cephalic seta R0	(?)	present (2)	present (2)	present (2)	absent (1)
38. cephalic seta R1	present (2)	present (2)	present (2)	present (2)	present (2)
39. cephalic seta R2	present (2)	present (2)	present (2)	present (2)	present (2)
40. cephalic seta R3	absent (1)	absent (1)	absent (1)	absent (1)	absent (1)

7	Some basal setae of the labial triangle barbu-
	late
-	All labial setae smooth
8	Unguis with a basal tooth longer than the
	otherP. vera Ungual teeth subequalP. reddelli
9	
9	Abdominal segment IV with $0 + 2$ macrose-
	tae <i>P. petrustrinatii</i> Abdominal segment IV with 1 + 2 macrosetae 10
10	Head macrosetae S and/or T present
10	Head macrosetae S and/or 1 present
- 11	Both head macrosetae S and T present
11	Head macroseta S absent (=Only T
-	present?)P. sp. BB
12	Eyes present
-	Eyes absent
13	Unguis with a basal tooth longer than the other 14
-	Ungual teeth subequal
14	Head macrosetae S or T present
-	Head macrosetae S or T absent P. vera
15	With 2 + 2 eyes, separatedP. palaciosi
-	With $1 + 1$ or $2 + 2$ eyes, very close to each
	otherP. voylesi
16	Head macrosetae S and T absent P. vera
-	Head macrosetae S or T present 17
17	Abdominal segment IV with $0 + 2$ macrose-
	taeP. volca
-	Abdominal segment IV with 1 + 2 macrosetae 18
18	Antenna less than 2.4 times the cephalic diago-
	nalP. finca
-	Antenna more than 2.5 times the cephalic diago-
	nal
19	Adult 2.4 mm long, labial r seta present
	as smooth microseta, basal dental spines
	longP. rochezi, sp. nov.

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# A new subgenus and two new species of the troglobitic genus *Dongodytes* Deuve from Guangxi, China (Coleoptera, Carabidae)

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#### ABSTRACT

In the present paper, a new subgenus and two new species of the cave-dwelling genus *Dongodytes* Deuve, 1993 are described and illustrated: *Dongodytes (Dongodytodes) deharvengi*, subgen. and sp. nov. and *Dongodytes baxian*, sp. nov. from Du'an Xian, North Guangxi, China.

Key words: cave beetles, Trechinae, new subgenus, new species, Guangxi, China

### INTRODUCTION

Since *Sinaphaenops mirabilissimus* Uéno & Wang, the first blind trechine was described in mainland China (Uéno and Wang 1991), many new genera and species have been discovered in southern Provinces or Regions such as Guizhou, Guangxi, Yunnan, Hunan, Sichuan and Chongqing, making China one of the richest countries in cave-dwelling trechine beetles on both generic and specific aspects (Tian 2008). Furthermore, all Chinese genera and species of cave-dwelling beetles are endemic, most of them occurring in a very narrow area, generally known only from the type locality, i.e. one or a few caves (Latella and Chen 2008). Because China holds the largest karstic areas in the world it is supposed that more and more new taxa of cave-dwelling trechines will be discovered in near future even they are always so rare.

It is too early to discuss the phylogenetic relationships of cave-dwelling trechines in China because of poor knowledge on the cave fauna of the country. For instance, the peculiar genus *Dongodytes* Deuve, 1993 is one of the most cave adapted trechine genera in the world (Ueno 1998). It is composed of three species so far, *viz. D. fowleri* Deuve, 1993, *D. grandis* Uéno, 1998 and *D. giraffa* Ueno, 2005. But the relationship of this genus within Trechinae has not been well determined. It was regarded either close to *Sinaphaenops* Uéno & Wang (Deuve, 1993), or allied to European trechine *Aphaenops* series (Vigna Taglianti 1997; Uéno 1998); for the latter hypothesis, there would be a big challenge to explain how to link between both lineages from the zoogeographical point of view.

Our recent discovery might provide a very important evidence for this interesting topic. Sponsored by a World Bank project which managed by the Biodiversity Office, Guangxi Forestry Bureau, Nanning, Dr. Louis Deharveng (the Paris Museum of Natural History, MNHN), and the author, with assistance of Dr. Anne Bedos (MNHN) and Dr. Li Youbang (Guangxi Forestry Bureau, Nanning), made a twenty-day collecting exploration on cave fauna in several selected karstic areas of Guangxi Zhuang Minority Autonomic Region in April, 2010. Among the large collected material, we found two new species of *Dongodytes*, one of them represents a new subgenus which evidently different from the nominate subgenus.

## MATERIALS AND METHOD

All specimens used in the study were collected in Du'an Yao Minority Autonomic Xian, Hechi City, Guangxi. All specimens are deposited in the insect collection of South China Agricultural University (SCAU), except two paratypes of *Dongodytes* (*Dongodytodes*) *deharvengi* sp. nov. in MNHN, Paris. Abbreviations of measurement used in the text are the same as in Tian (2009).

### RESULTS

# Dongodytodes, subgen. nov. (Figs 1-5)

Type species: *Dongodytes* (*Dongodytodes*) *deharvengi*, sp. nov.

*Diagnosis. Dongodytodes*, subgen. nov. is similar to *Dongodytes* (s.str.) concerning the evidently swollen prothorax, the very elongate elytra without humeral angles, the unmodified protarsi of male, the bisetose labial palpomere 2 and the chaetotaxal pattern of elytra. However, the following peculiar features of *Dongodytodes* make it fall into another lineage rather than *Dongodytes* 



(s.str.): (1) head more stout and broadly convex posteriorly, widest at the level of the end of frontal furrows, and neck very short in Dongodytodes, versus long and narrowly gradually convergent posteriorly, widest at the level of anterior supraorbital pores, neck much longer in Dongodytes (s.str.); frontal furrows shorter, straight and subparallel-sided before divergence in Dongodytodes, versus longer, evidently sinuate and not parallel-sided before divergence in Dongodytes (s.str.); head with two pairs of supraorbital pores, posterior pores far from anterior ones and closer to neck in Dongodytodes, versus both anterior and posterior pores (the latter sometimes not well marked, and irregularly located, or wanted) closer to each other than the posterior one to the neck in Dongodytes (s.str.); (2) antennae stouter and much shorter, hardly extending to beyond apices of elytra in Dongodytodes, versus much longer, elongate, and evidently extending over elytral apices in *Dongodytes* (s.str.); (3) clypeus sexsetose in Dongodytodes, versus quadrisetose in Dongodytes (s.str.); (4) surface of body densely covered with long and erected hairs or pubescence, and more or less punctate on elytra in Dongodytodes, versus with only a few sparse setae on head or sometimes on pronotum, and glabrous and impunctate on elytra in Dongodytes (s.str.); (5) the elytral chaetotaxal pattern of Dongodvtodes is guite similar to that of Dongodvtes (s.str.), but stria 3 with only one setiferous pore at about 3/5 from base in Dongodytodes, versus two or three in Dongodytes (s.str.); (6) elytral striae more or less evident throughout in Dongodytodes, but rather faint in Dongodytes (s.str.); (7) abdominal ventrite VII of male with only one pair preapical setae in Dongodytodes, versus two pairs in Dongodytes (s.str.); ventrites IV-VI each with only one pair of paramedian setae in Dongodytodes, versus two pairs in Dongodytes (s.str.); (8) body of Dongodytodes smaller than that of the members of *Dongodytes* (s.str.); and (9) Dongodytodes with much slender, and longer male aedeagus than that of Dongodytes (s.str.) considering its smaller body size.

*Etymology.* The name of the new subgenus refers to its similarity to *Dongodytes* (s.str.).

Distribution. Guangxi (Du'an Xian).

# Dongodytes (Dongodytodes) deharvengi, sp. nov. (Figs 1-9)

Holotype: male, collected in an anonymous cave, about 4 km south of Xia'ao Xiang, Du'an Yao Minority Autonomic Xian, coordinate around 107.55 E /24.16 N, 26 April, 2010, Louis Deharveng leg., in SCAU; paratypes: 1 male and 6 females, same data as holotype, Anne Bedos, Louis Deharveng & Mingyi Tian leg., in SCAU and MNHN, respectively.

*Diagnosis.* Small sized, blind, dark reddish brown beetle, surface covered with dense and short bristly setae, head and pronotum smooth, elytra evidently rugose and

punctate; antennae not exceeding over the elytral apices; head evidently expanded posteriorly, posterior supraorbital pores close to neck; protarsomere 1 not modified in male; aedeagus small and slender, evenly arcuate throughout, parameres elongate, each with four long setae at apex.

## Description

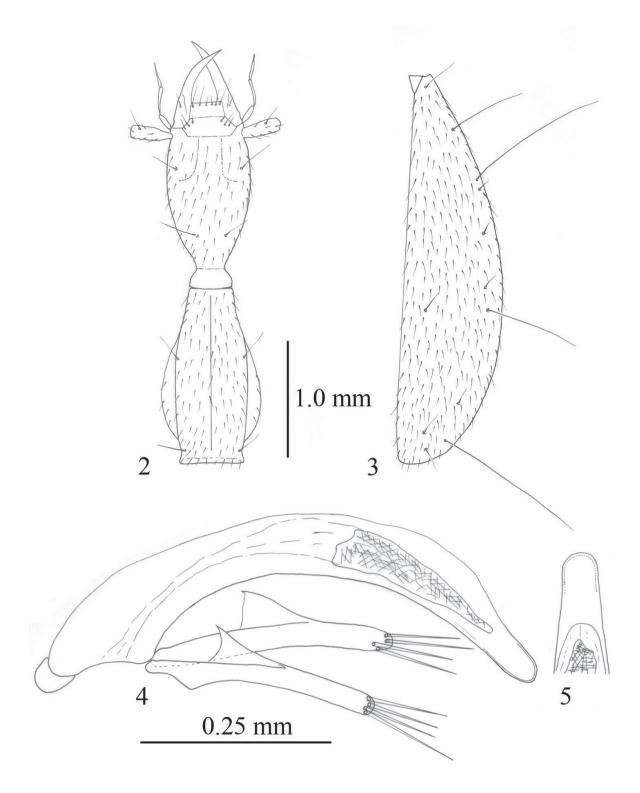
Length (including mandible): 6.2-6.4 mm.

Habitus as in Fig. 1. Whole body covered with dense and bristly setae, dark reddish brown. Surface moderately shiny, smooth on head and pronotum, more or less rugose and punctate on elytra. Microsculptural meshes fine transversal lines on head and pronotum, and strongly transversal polygons on elytra.

Head (Fig. 2) elongate and narrow, HL/HW=2.6-2.8; widest at about the level of end of frontal furrows, gently expanded from the level of posterior pores to frontal furrows, then suddenly and strongly constricted to neck; neck constriction evident, base of neck distinctly convex, ring-shaped; dorsal surface convex, frons and supraorbital area gently convex; frontal furrows comparatively short, well marked, subparallel-sided before the evidently divergence, ended a little behind the anterior supraorbital pores; the two pairs of supraorbital pores far from each other, posterior ones much closer to neck than to the anterior ones; mandibles moderately slender; labrum sexsetose, slightly bisinuate at frontal margin, with median margin nearly straight; clypeus sexsetose; mentum and submentum well separated by labial suture; mentum moderately concave, with the median tooth simple; ligula quite narrow, with two setae at apex; labial palpomere 2 bisetose at inner side; antennae rather long and slender, but not extending over elytral apices; antennomere 1 short and stout, slightly longer than antennomere 2; antennomeres 3, 4 and 5 subequal in length, each 2.3 times as long as antennomere 2.

Prothorax (Fig. 2) elongated subovate, shorter than head (including mandibles), HL/PL=1.3-1.5, but evidently wider, PW/HW=1.2; widest at about 3/7 from base, gradually narrowed towards apex but strongly towards base, suddenly sinuate before hind angles; much longer than wide, PL/PW=1.8; propleura markedly expanded, visible from above. Pronotum long, elongate, somewhat columnar; narrower than head, PNW/ HW=0.8-0.9; widest at a little behind middle, and more straightly narrowed towards apex than towards base; PNW/PP=1.1, PNW/PA=1.4-1.5; posterior margin evidently wider than anterior one, PP/PA=1.4; sides feebly bordered throughout, more or less parallel-sided behind middle, sinuate before hind angles; hind angles broadly rectangular, fore angles rectangular; base more or less bordered, almost straight; disc moderately convex, basal transverse impression well marked; middle line fine and clear. Scutellum small.

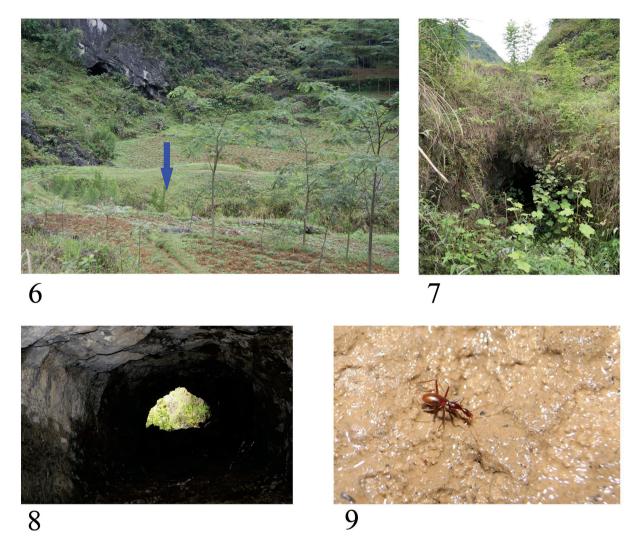
Elytra (Fig. 3) elongate oblong-ovate, very narrow at base, apex narrowly rounded; much wider than wide,



Figs 2-5 - *Dongodytes (Dongodytodes) deharvengi*, subgen. and sp. nov., holotype (2, head and prothorax; 3, right elytron; 4, aedea-gus, lateral view; 5, apical part of aedeagus, dorsal view).

EL/EW=1.9; widest at about 2/3 from base, much less, and evenly narrowed towards base than towards apices, without shoulders; much wider and longer than prothorax, EW/PW=1.7, EL/PL=2.5-2.6; disc strongly convex, but evidently depressed near base; sides finely

bordered throughout, smooth; scutellar area distinctly convex before the depression, scutellar striole absent; striation very faint but traceable; stria 3 with a single moderately setiferous pore at about 3/5 from base; preapical setae present, moderate in length; marginal



Figs 6-9 - *Dongodytes (Dongodytodes) deharvengi*, subgen. and sp. nov. (6, locality of the coal mine, indicated by the arrow-head; 7-8, entrance of the mine; 9. wandering beetle in the cave).

umbilicate pores similar in arrangement to those of *Dongodytes* (s.str.).

Ventral surface wholly pubescent, denser in median portion of ventrites; apical margin of apical abdominal ventrite complete, with a pair of anal setae in male but two in female; ventrites IV-VI each with one pair of paramedian setae in both sexes.

Legs quite slender, but more or less shorter than those of *Dongodytes* (s.str.); tarsomere 1 long, pro- and metatarsomere 1 as long as pro- and metatarsomeres 2-4 combined, respectively, mesotarsomere 1 longer than tarsomeres 2-4 combined; protarsi not modified in male, though both male and female with protarsomere 1 slightly stouter than others.

Male aedeagus (Figs. 4-5) with the median lobe small and slender, evenly arcuate throughout; inner sac armed with a long and rather thin sclerotized copulatory piece, which is about 1/3 as long as the median lobe, base orifice quite narrow; in dorsal aspect, apex broad and symmetric, nearly parallel-sided; parameres elongate, the right one as long as the left, each with four long apical setae.

*Sexual dimorphism.* No sexual dimorphism; male and female similar, the protarsi not modified in male, but abdominal ventrite VII with only one pair of setae in male, while two pairs in female.

*Remarks. Dongodytes (Dongodytodes) deharvengi,* sp. nov. is more or less similar to members of *Dongodytes* (s.str.). But it is very easy to separate this new species from all *Dongodytes* (s.str.) species by its smaller and pubescent body, stouter head, shorter antennae, sexsetose clypeus, punctuate elytra and slender aedeagus.

*Etymology*. This new species is named in honor of Dr. Louis Deharveng (MNHN, Paris), a well known specialist of Collembola and cave biodiversity.

*Distribution.* Guangxi (northern Du'an Xian). Known only from the type locality cave which is still anonymous. It is a ruined coal mine (Fig. 6-8). The entrance is

about 200 m from the main road (National Road G050) on the west side. First part of the passage is an horizontal mine, straight for about 30 m, then gradually going down for about 50 m, where it joins a natural cave passage. The natural passage continues towards the left (not explored); towards the right and upwards, it is a large and steep gallery for about 50 m; then, after a relatively narrower path, it goes down for about 50 m to a large room about 40 m wide. All the beetles were collected on the steep, wet and muddy slopes of this room and on formations. At the bottom of the room, the passage decreases in size and continues horizontally for at least 30 m, with some puddles. Most specimens were wandering when collected (Fig. 9) except two which were caught under stone.

# Dongodytes (Dongodytes) baxian, sp. nov. (Figs 10-13)

Holotype: male, Baxian Dong, a limestone cave in Baxian Park, at suburb of Du'an for about 8 km, 108.13'281"E / 23.91'404"N, 150 m, 26 April, 2010, Louis Deharveng leg., in SCAU.

*Diagnosis*. Small sized for *Dongodytes*, elongate and slender beetle; reddish brown, head, thorax covered with long and sparse setae, surface smooth, elytra glabrous; abdominal ventrites shortly pubescent; head a little more stout than other members of *Dongodytes* (s.str.), with only the anterior pair of supraorbital pores, the posterior ones wanted, with a few irregularly located setae; frontal furrows rather long; protarsomere 1 not modified in male; aedeagus very short and stout, with apical portion short and obtuse, and parameres narrow and slender, subequal in length, each with four long setae at apex.

#### Description

Length (including mandible): 7.3 mm.

Yellowish brown, palpi, antennomeres 5-11, and tarsi pale; surface moderately shiny, smooth on head and prothorax, faintly rugose on elytra; head and pronotum covered with long and sparse setae. Microsculptural meshes fine transverse or oblique lines on head and pronotum, irregularly polygons on elytra.

Head (Fig. 10) very long and narrow, HL/HW=3.1; widest at about the level of antennal articulation, and gradually narrowed to neck posteriorly; neck constriction quite elongate, base of neck evidently convex, ring-shaped; dorsal surface moderately convex; frontal furrows moderately long and well defined; with only anterior pairs of supraorbital pores, and a few irregularly located setae; labrum emarginated at frontal margin, cl-ypeus quadrisetose; mandibles slender, and well developed; eyes effaced; mental tooth evident, faintly bifid at tip; palpi slender, labial palpomere 2 with two setae at inner side. Antennae long and slender, evidently extending over elytral apices; antennomere 1 with a long preapical setae and a few sparse shorter ones, other antennomeres with denser pubescence; antennomere 1 short and stout,

3.4 times as long as wide; antenomeres 1 and 2, and 3 and 4 subequal in length, respectively.

Prothorax (Fig. 10) elongated subovate, PL/PW=1.7; much shorter than head, HL/PL=1.9, as wide as head; widest at about 2/5 from base, and much more gradually narrowed towards apex than towards base; propleura markedly expanded, visible from above. Pronotum elongate, PL/PNW=2.1, much narrower than head, PNW/ HW=0.3; widest at about 6/13 from base, almost straightly narrowed towards apex, and faintly sinuate towards base; PNW/PP=1.2, PNW/PA=1.8; posterior margin evidently wider than anterior one, PP/PA=1.6; sides well bordered throughout; both anterior and posterior angles rectangular, but the former more or less obtuse; with two pairs of marginal setae which is normally located. Scutellum comparatively large.

Elytra (Fig. 11) very slender, and narrow, widest at a little before 3/5 from base; EW/PW=2.0, EL/PL=2.8, EL/EW=2.0; sides well bordered throughout, smooth and not serrated; with very slight shoulders, feebly sinuate behind and almost straight before the faint shoulders; surface strongly convex exception of base area which is distinctly depressed; striae obsolete except for striae 1 and 3 which are more or less evident; stria 3 with two setiferous pores at 5/7 and 1/2 from base, respectively; marginal umbilicate pores similar to those in *D. fowleri* Deuve and *D. grandis* Ueno; without scutellar striole.

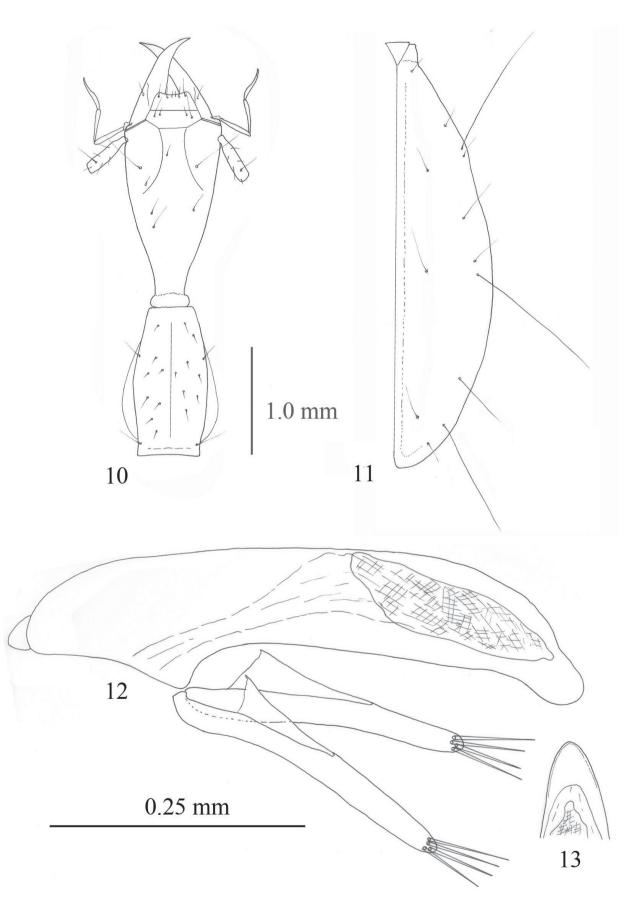
Underside of head and thoraxes smooth and glabrous, with sparse and long setae; abdominal ventrites shortly pubescent. Apical abdominal ventrite with two pairs of preapical setae; ventrites IV-VI each with a pair of setae at median portion.

Legs slender; procoxae glabrous, while meso- and metacoxae with long setae, denser on mesocoxae; tarsomere 1 long, protarsomere 1 shorter than protarsomeres 2 to 4 combined (0.8 times), while meso- and metatarsomere 1 are longer than meso- and metatarsomeres 2-4 combined (1.3 and 1.1 times), respectively; protarsomeres not modified in male.

Male aedeagus (Figs. 12-13) with the median lobe small, and short, but stout and well sclerotized, slightly membraneous on apical dorsal part, base orifice wide, apex shortly broad, inner sac armed with a very large, well-defined sclerotized copulatory piece, which is about 1/3 as long as the median lobe; in dorsal aspect, apex broad, gently protrude and rounded, slightly membraneous at tip; parameres long and slender, each with four long setae at apex.

*Remarks. D. baxian* sp. nov. is very similar to *D. fowleri* and *D. grandis* Uéno, but easily distinguished from them by the following characters: (1) aedeagus stouter and broader, parameres more elongate and slender; (2) pronotum covered with sparse and long setae; and (3) elytral sides slightly but obviously sinuate behind the very slight shoulders.

*Etymology.* This new species is named after its type locality: Baxian Dong. In Chinese, Ba Xian refers to the



Figs 10-13 - *Dongodytes baxian*, sp. nov., holotype (10, head and prothorax; 11, right elytron; 12, aedeagus, lateral view; 13, apical part of aedeagus, dorsal view).

eight immortals, viz. Zhang Guolao, Lu Dongbin, Cao Guojiu, Zhong Liguan, Li Tieguai, Han Xianzi, He Xiangu and Lan Caihe, who are symbols for good fortune. Distribution. Guangxi (Du'an Xian).

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# Scuttle flies (Diptera: Phoridae) from caves in Alabama and Georgia, USA

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#### ABSTRACT

Four species of scuttle fly are reported from caves in Alabama and Georgia.

Key words: Diptera, Phoridae, caves, new records, USA

### INRODUCTION

The most recent paper on scuttle flies (Diptera: Phoridae) recorded from caves in the USA (Disney et al 2010), reviewed previous identifications of Phoridae from caves in the USA and added new records from Arkansas and Missouri. Their review included revision of previous records for Alabama and Georgia (Reeves and Disney 1999) by showing that the three commonest cave dwelling species in the USA belong to a sibling species complex. In this paper we report on further collections from caves in the latter two states.

# METHODS

All of the flies were captured by JC with ramp-pitfall traps baited with liver and bananas. The traps were left in each cave for 14 days. The specimens were preserved in alcohol (70% ethanol) and were identified by RHLD, who slide mounted samples of each species in Berlese Fluid (Disney 2001).

Slide mounted samples are deposited in the University of Cambridge, Museum of Zoology (UCMZ). For these slide mounted specimens the labels include a code such as 31-98, which refers to RHLD's notebook 31 page 98. The distances in metres from the cave entrances are given thus 5 m, 75 m, etc.

# RESULTS

The following species were collected, all belonging to the enormous genus *Megaselia* Rondani 1856. This is one of the largest genera of Diptera known. Provisional keys to the Nearctic species were provided by Borgmeier (1964, 1966). Important revisions and additions are given by Robinson (1978, 1981), but these do not cover the section of the genus that includes the species *M. cavernicola* that has been repeatedly reported from American caves and mines. However, it is now realized that this species belongs to a sibling species complex (Disney et al 2010) and that some previous reports of *M. cavernicola* probably refer to other species or to mixed samples that may or may not include *M. cavernicola*. They also provide a key to the five Nearctic species of *Megaselia* known to be cavernicolous. Other species, such as the *M. scalaris* reported below, are only of casual occurrence in caves and are typically only found close to the cave entrance.

Megaselia breviterga (Lundbeck, 1920) Aphiochaeta breviterga Lundbeck, 1920: 33 (female) Apohiochaeta similata Lundbeck, 1921: 140 (male) Megaselia spelunciphila Disney in Reeves & Disney 1999: 211.

Material examined: ALABAMA, Jackson County, Sauta Cave, July 2008, 5 m, 43 M, 81 F. Morgan County, Anvil Cave, July 2009, 5m, 5 M, 9 F; 75 m, 1 M, 49 F; 150 m, 8 F; Cave Springs Cave, July 2008, 5 m, 6 F; 75 m, 1 M, 12 F; 75 and 125 m, July 2008, 2 M, 1 F. GEOR-GIA, Dade County, Byers Cave, June 2009, 75 m, 1 M; Howard's Waterfall Cave, June 2008, 5 m, 3 M, 27 F; 75 m, 1 M, 3 F. Walker County, Harisburg Cave, June 2008, 5 m, 12 M, 18 F; 75 m, 1 F; Pettyjohns Cave, May 2009, 5 m, 5 M, 13 F.

> *Megaselia cavernicola* (Brues, 1906) *Aphiochaeta cavernicola* Brues, 1906: 101.

Material examined: ALABAMA, Jackson County, Sauta Cave, July 2008, 5 m, July 2008, 1 M, 2 F; Morgan County, Anvil Cave, July 2009, 75 m, 5 M, 8 F; 150 m, 5 M, 5 F; Cave Springs Cave, July 2008, 5 m, 1 F: 75 m, 2 M, 2 F; and 125 m, 1 M, 2 F. GEORGIA, Dade County, Byers Cave, June 2009, 75 m, 5 M, 5 M; 225 m, 9 M, 11 F; Howard's Waterfall Cave, June 2008, 75 m, 16 M, 24 F; 150 m, 9 M, 23 F; 225 m, 11M, 18 F; Walker County, Harisburg Cave, June 2008, 5 m, 2 M, 6 F; 75 m, 10 M, 29 F; 150 m, 9 M, 31 F; 225 m, 19 M, 54 F; Howard's Waterfall Cave, June 2008, 75 m, 16 M, 24 F; 150 m, 9 M, 23 F; 225 m, 11M, 18 F; Pettyjohns Cave, May 2009, 5 m, 4 F; 75 m, 2 M, 23 F; 150 m, 6 M, 19 F; 225 m, 4 M, 6 F; 375 m, 2 M, 23 F; 600 m, 3 M, 12 F.

## *Megaselia scalaris* (Loew, 1866) *Phora scalaris* Loew, 1866: 53.

This species, whose larvae feed on an exceptionally wide range of decaying organic materials, has been carried around the world by man (Disney 2008). It has been reported from Santee Cave in South Carolina (Reeves, 2001) and from caves in Trinidad (Disney 1995).

Material examined: ALABAMA, Morgan County, Cave Springs Cave, July 2008, 5 m, 4 F.

Megaselia taylori Disney, 2010 Megaselia taylori Disney in Disney et al 2010: 86.

Material examined: ALABAMA, Jackson County, Sauta Cave, July 2008, 5 m, 24 M, 143 F. Morgan County, Anvil Cave, July 2009, 5m, 5 M, 31 F; 75 m, 34 F; 150 m, 8 F; Cave Springs Cave, July 2008, 5 m, 46 F; 75m, 1 M, 31 F. GEORGIA, Dade County, Byers Cave, June 2009, 5 m, 2 M, 18 F; Howard's Waterfall Cave, June 2008, 5 m, 1 M, 8 F; 75 m, 1 F. Walker County, Harisburg Cave, June 2008, 5 m, 6 F; Pettyjohns Cave, May 2009, 5 m, 4 F;

### DISCUSSION

The above records confirm that the regular cavernicolous species in caves in the southern half of the USA are *Megaselia breviterga*, *M. cavernicola* and *M. taylori*. 53% of the *M. breviterga* (n = 406) were caught at the 5 m point from the cave entrances, but only 9.7% of *M. cavernicola* (n = 426) were caught at this point in contrast to 75.4% of the *M. taylori* (n = 363). This suggests that *M. cavernicola* is the most cavernicolous of these three species and that *M. taylori* is the least so adapted. This confirms the conclusions of Disney et al (2010).

The sex ratios differ from those reported by Disney et al (2010) for flies procured with pitfall traps baited with Limburger cheese spread. Thus they reported 56.85% of the 489 *M. breviterga* they collected were females. By contrast, 81.2% of the 406 flies of this species reported above were females. Likewise of the 253 *M. cavernicola* they collected 84.58% were females but for our 530 flies of this species 70.38% were females; and for their 332 flies of *M. taylori* 82.83% were females compared with 90.9% for our 363 flies of this species. These differences are likely to reflect differences in the methods of collection. In their study their use of a different bait probably accounts for this difference. It is well established that every collecting method is selective with respect to which species are procured (e. g. Disney et al 1982). Our studies suggest that for some species there is also selectivity with regard to the ratios of the sexes caught.

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# **Sandro RUFFO**

# Remind of a master

# August 26, 1915 - May 7, 2010

On the 7th of May 2010, at age 94, Sandro Ruffo passed away.

He was born the 26<sup>th</sup> of August 1915 in Soave, near Verona, and he soon developed an interest for Nature and the passion for caves.

It would take too long to list the studies he conducted, and the prizes and the academic awards he was bestowed, which in his long life made him one of the preeminent Italian zoologists, the only Museum curator belonging to the "Accademia dei Lincei", and one of the most important amphipodologists in the world. He was professor of Zoology at Modena University, the director of the Natural History Museum of Verona, and was also presented with a Degree Honoris Causa in Conservation and Management of Natural Resources in 2007. However, we think this journal is the appropriate place to mention the Ruffo's important contribution to speleology and in particular to biospeleology.

Already during his high school years, Sandro started exploring the caves of Lessini Mountains, which border Verona to the Northeast. In 1929, at age 14, he visited for the first time the Natural History Museum of Verona to find out the name of an insect he collected along the Adige River: he probably could not have imagined then, that the museum would have become his second home. In those years two other young researchers were also frequent visitors of the Museum: Francesco Zorzi, attracted by prehistory, and Angelo Pasa, interested in geology and paleontology. Even if the three young researchers had different backgrounds and different scientific interest, the passion for the study of caves brought them together, and they explored numerous caves in the Veronese area. Their enthusiasm in research is shown by their first publication, co-authored by all three, where they described a series of research conducted in Grotta Damati (9 V/VR) near Verona, which lead Ruffo to notice the importance of biospeleology and of how "the zoological research should be integrated by records of temperature and water level, in order to characterize the physical environment of each cave". In the same year, the 19-year old Sandro Ruffo published a paper on the presence of a leech in the "Grotta A del Ponte di Veja" (117 V/VR) on Lessini Mountains and two years later he published two papers on subterranean amphipods of the genus Niphargus.



All data collected in those first years of research of Lessinian caves were gathered in a paper published in 1983 and titled: "Studio sulla fauna cavernicola della regione veronese (i.e., A study of cave fauna of Veronese area)". In the same year he graduated in Agricultural Sciences at the University of Bologna, and his paper was published in the "Bollettino dell'Istituto di Entomologia della Regia Università di Bologna".

In the war years Ruffo, who was drafted in 1939, was taken prisoner by the Germans in 1943 in France, and sent to prison camps first in Poland and than in Germany. In 1945 he returned to Verona and resumed his speleological and scientific activity, at the same time working for the reconstruction of the Museum, where he had officially assumed the position of zoological curator. In the first post-war years, his renewed enthusiasms for the scientific exploration of caves lead him to create the "Gruppo Grotte A. Massalongo" with headquarters at the Museum.

Between 1948 and 1959, he and Angelo Pasa organized five research campaigns in the Apulian caves and explored and mapped, among other caves, the famous Zinzulusa Cave. These research were summarized in Ruffo's publication "Le attuali conoscenze sulla fauna cavernicola della Regione Pugliese (i.e., Present knowledge on the cave fauna of Apulia)". This paper is still often cited not only because it is one of the first publications on the



Zorzi, Ruffo and Pasa at the entrance of the Damati Cave in 1933 (photo: archive of the Museo Civico di Storia Naturale di Verona)

Italian cave fauna, and the first one for Apulia, but also because it provided an innovative ecological classification of cave animals which is still largely used.

In June 1950 Ruffo is one of the organizers of the national meeting where, in the conference room of the Natural History Museum of Verona, the Italian Speleological Society (Società Speleologica Italiana) was born (or reborn, according to someone). The celebrations for the 25<sup>th</sup> and the 50<sup>th</sup> anniversary of the society were held in the same Museum; Ruffo was present to both, to give to the old and new speleologists his greetings and wishes that speleology would continue to arouse enthusiasm in new generations of explorers. He was supposed to do the same for the 60<sup>th</sup> anniversary, last June, but he left us before then.

In the '50s Sandro's scientific activity increased, and he also took part to cave explorations: in 1950 together with Pasa, Broglio, Allegranzi and Bartolomei, he explored the first 5 km of the "Buso della Rana". This cave was later on further surveyed for a total of 27 km length; it is the longest Italian cave with only one entrance.

In 1953 Ruffo and other Italian speleologist attended the First International Spelological Conference in Paris. There, Auguste Vandel, at the time director of the Subterranean Laboratory of Moulis (France) and co-organizer of the Conference, unexpectedly asked to Sandro to open the conference with a lecture on the distribution of cave amphipods, the small, unpigmented subterranean crustaceans that Sandro studied through all his life, passing his enthusiasm and passion to dozens of young researchers from all over the world.

In 1964 he became the director of the Natural History Museum of Verona, which turned into the meeting point for biospeleologists form different countries. In 1969 he founded an international working group for the study of cave amphipods and, in the same year, he organized the first meeting on the amphipods of the genus *Niphargus*. In 1978, Ruffo organizes the XXII National Conference of the Italian Biogeographical Society dedicated to the "Biogeografia delle Caverne Italiane (i.e., the biogeography of Italian caves)", which was, and still is, a milestone for the Italian biospeleology. In 1976 the Museum became the repository of the cadaster of the caves of Verona province, and started to host the Unione Speleologica Veronese (Veronese Speleological Union).

At the end of the '70s Ruffo contributed to the creation and development of the Société Internationale de Biospéologie (now International Society for Subterranean Biology), of which he was an Honor Member.

Sandro Ruffo left something else than his academic awards, the more than 300 scientific publications (listed in a subsequent and more complete paper), the description of 190 new species, 35 genera and 1 family: he left his teaching. He was the mentor of numerous Italian and foreign scientists; he was not only a teacher of science, but also a teacher of life. His kind-heartedness, warmth and willingness to help, and his joy of living, togehter with his great scientific knowledge, have taught to many



Sandro Ruffo drawings new species of amphipods

of us the love for life and the joy and importance of exploring caves and studying small creatures which are invisible to many.

Sandro Ruffo was fascinated by all aspects of Nature, and he loved caves and the small creatures living there. He taught us the importance of the astonishment that generate the curiosity in front of all the aspect of nature, and, as he said: "curiosity is the mother of the knowledge".

Ciao Sandro, thank you.

Leonardo Latella and Beatrice Sambugar Museo Civico di Storia Naturale di Verona

# New taxa described in this volume

# CRUSTACEA

Amphipoda, Crangonyctidae

*Stygobromus albapinus*, sp. nov. Taylor and Holsinger: USA, Nevada, White Pine County, Great Basin National Park, Model Cave.

# COLLEMBOLA Entomobryidae

*Pseudosinella rochezi*, sp. nov. Palacios-Vargas and Mejia Recamier: Mexico, State of Puebla, karst region of Tzontezecuiculi, in one small cave close to Tepepan Zaragoza.

# INSECTA

Coleoptera, Carabidae, Trechinae

*Dongodytodes*, subgen. nov Tian: China, Du'an Yao Minority Autonomic Xian, anonymous cave, about 4 km south of Xia'ao Xiang.

*Dongodytes (Dongodytodes) deharvengi*, sp. nov. Tian: China, Du'an Yao Minority Autonomic Xian, anonymous cave, about 4 km south of Xia'ao Xiang.

Dongodytes (Dongodytes) baxian, sp. nov Tian: China, Baxian Dong, a limestone cave in Baxian Park, at suburb of Du'an for about 8 km.

# World karst science reviews

Cave and Karst Science of British Cave Research Association Volume 37, (1), 2010

CONTENTS

Monitoring the population of the linyphid spider *Porrhomma rosenhaueri* (L. Koch, 1872) (Araneae: Linyphiidae) in Lesser Garth Cave, Cardiff, UK. *Julian Carter, Adrian Fowles and Catalena Angele* 

Derbyshire pipe veins – deep-seated speleogenesis. Trevor D. Ford

21st British Cave Research Association Cave Science Symposium Abstracts: University of Bristol, 6 March 2010

# **International Journal of Speleology** Of the International Union of Speleology **Volume 39(2), July 2010**

CONTENTS

Results of palaeomagnetic research of karst sediments in Slovenia. *Nadja Zupanhajna, Andrej Mihevc, Petr Pruner and Pavel Bosak* 

Spatial distribution of soda straws growth rates of the Coufin Cave (Vercors, France). *Yves Perrette and Stéphane Jaillet* 

Karst geology and cave fauna of Austria: a concise review. *Erhard Christian and Christoph Spotl* 

Modern C, O and H isotope composition of speleothem and dripwater from Modrič Cave, Eastern Adriatic (Croatia). *Maa Suri, Zvjezdana Roller-Lutz, Magda Mandi, Ines Krajcar Broni, Mladen Jura* 

Uranium-series age data and revised stable isotope curves for New Zealand speleothems: frequency distribution, sample depth and palaeoclimatic implications. *Paul W. Williams, J-X Zhang, H. Neil* 

A re-appraisal of the stratigraphy, palaeontology and dating of Cow Cave, Chudleigh, Devon, England. *Jim W. Simons* 

Do Karst Rivers "deserve" their own biotic index? A ten years study on macrozoobenthos in Croatia. *Biljana Raa and Sanja Puljas* 

BOOK REVIEW — Angel Ginés, Martin Knez, Tadej Slabe & Wolfgang Dreybrodt 2009 Karst rock feature Karren sculpturings, Carsologica 9, Postojna/Ljubljana. *Jo De Waele*  Journal of Cave and Karst Studies of the National Speleological Society Volume 72(1), April 2010

CONTENTS

Stochastic modelling of surface stream flow at different time scales: Sangsoorakh Karst Basin, Iran. *M. Reza Ghanbarpour, Karim C. Abbaspour, Goudarz Jalavand, and Ghodsieh Ashtiani Moghaddam* 

Distribution of cyanobacteria at the Gelada Cave (Spain) by physical paramters. *Antonia Martinez and Antonia Dolores Asencio* 

Direct measurement of present-day tectonic movement and associated radon flux in Postojna Cave, Slovenia. Stanka Šebela, Janja. Vaupotič, Blahoslav Košťák, and Josef Stemberk

Developing a GIS-based inventory for the implementation of cave management protocols in Withlacoochee State Forest, Florida. *Grant L. Harvey, Philip P. Reeder, Jason S. Polk, and Philip E. van Beynen* 

The sulfidic thermal caves of Acquasanta Terme (Central Italy). Sandro Galdenzi, Filippo Cocchioni, Giampaolo Filipponi, Luciana Morichetti, Stefania Scuri, Rosario Selvaggio, and Mario Cocchioni

BOOK REVIEW — Subterranean Twin Cities *Joseph C. Douglas* 

Journal of Cave and Karst Studies of the National Speleological Society Volume 72(2), August 2010

CONTENTS

Coastal cave in Bahamain eolian calcarenites: Differentiating between sea caves and flank margin caves using quantitative morphology. *Willapa J. Waterstrat, John E. Mylroie, Athena M. Owen, and Joan R. Mylroie* 

Secondary minerals in volcanic caves: Data from Hawai`i. *William B. White* 

Description of a new *Macrobrachium* species (Crustacean: Caridea: Palaemondidae) from a cave in Guangxi, with a synopsis of the stygobiotic Decapoda in China. *Yitao Pan, Zhonge Hou, and Shuqiang Li* 

Ground-penetrating radar investigation of a rapidly developed small island in a lake in southern Georgia, USA. *Can Denizman, Eric C. Brevik, and Jim Doolittle* 

The subterranean asellids of Maryland: Description of *Caecidotea nordeni*, new species, and new records of *C. holsingeri and C. franzi* (Crustacea: Malacostraca: Isopoda). *Julian J. Lewis and Thomas E. Bowman* 

*Heperonemastoma smilax*, n. sp., a remarkable new harvestman from a cave in West Virginia, with comments on other reported cave-dwelling *Hesperonemastoma* species (Opiliones, Ischyropsalidoidea, Sabaconidae). *William A. Shear* 

Stable isotopes of subfossil bat guano as a long-term environmental archive: Insights from a Grand Canyon cave deposit. *Christopher M. Wurster, Donald A. McFarlane, Michael I. Bird, Philippa Ascough, and Nancy Beavan Athfield* 

Upper Pleistocene *Gulo gulo* (Linné, 1758) remains from the Srbsko Chlum-Komin hyena den cave in the Bohemian Karst, Czech Republic, with comparisons to contemporary wolverines. *Cajus G. Diedrich, and Jeffrey P. Copeland* 

BOOK REVIEW — Geological Monitoring *Arthur N. Palmer* 

# Karstologia

Of the Fedration Française de Spéléologie and Assocaiation Française de Karstologie **Volume 53, 2009** 

## CONTENTS

Spatial and morphological analysis by high-resolution 3D modeling of a stalagmites forest in Orgnac's karstic network (Ardèche, France). Souhail Hajri, Benjamin Sadier, Stéphane Jaillet, Estelle Ployon, Elisa Boche, Aiman Chakroun, Georges-Marie Saulnier, Jean-Jacques Delannoy-

Use of natural canyoning sites in France : a socio-cultural approach.. *André Suchet et Dominique Jorand* 

Pseudokarst in sandstone of the coastal eocene flysch, Guipuzcoa. Carlos Galan, José Rivas et Marian Nieto

Carte des karsts autour de la Méditerranée et en Europe moyenne. *Jean Nicod* 

An example of karst water resources exploitation: snow production in Villard-de-Lans - Corrençon-en-Vercors (Isère, France) Pierre Paccard

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- Bergstrom, D.E. 1997. The phylogeny and historical biogeography of Missouri's *Amblyopsis rosae* (Ozark cavefish) and *Typhlichthys subterraneus* (southern cavefish). Unpublished M.Sc. thesis, University of Missouri-Columbia.

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