

**Notes on the behavior of the advanced troglobite *Eukoenenia maquinensis* Souza & Ferreira 2010 (Palpigradi: Eukoeneriidae) and its conservation status**

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**Cave Palpigrads**

Most of the Palpigradi species currently described are found in Africa and Europe, although these arachnids are also distributed in the Americas, Asia and Oceania. Many northern species are found exclusively in caves, showing morphological adaptations in response to evolution in these environments<sup>1</sup>.

Within the order Palpigradi, the most expressive troglomorphisms are found in the genus *Eukoenenia* Börner 1901. The morphological adaptations to the cave environment in this group consist mainly of the increase of corporal size, elongation of the appendages (that also become thinner) and the increase in number of elements that constitute the lateral organs. Furthermore, the setae of the propeltidium, represented by ten pairs in edaphomorphic species, have a tendency to become tiny and sometimes less numerous in the troglotic species<sup>2</sup>.

According to Condé (1998), the length of the basitarsus related to the length of the tibia provides an index of adaptation to the cave environment. In edaphomorphic species, the tibia is longer than the basitarsus, which indicates a ratio less than one; while in the troglobite species, the ratio will be close, equal to, or higher than one. The ratio between the lengths of the propeltidium and the basitarsus IV is also used to indicate troglomorphism; in the edaphomorphic species, that value is approximately between 3 and 4 and in the troglomorphic species it is less than 2.

The knowledge about troglomorphisms in neotropical palpigradi is still very restricted. Up to date, *Eukoenenia orghidani* Condé & Juberthie 1981 from Cuba and the Brazilian species *E. maquinensis* Souza & Ferreira 2010 and *E. spelunca* Souza & Ferreira 2011 represent the troglotic neotropical palpigrades<sup>3, 4, 5</sup>. Among these species, *E. maquinensis*, from Gruta de Maquiné cave (Cordisburgo, Minas Gerais state) deserves attention, since it represents the most advanced troglotic palpigrade known from Americas.

### General traits of *E. maquinensis*

*Eukoenenia maquinensis* is the first troglobite species described from South America, corresponding to one of the most troglomorphic Palpigradi species described until now (Figure 1). This species presents a large body size (around 2 mm), has a flagellum formed by elongated segments (length equal to 3,865  $\mu$ m), has six blades on the prosomal lateral organs, the propeltidium has seven pairs of short setae and an extremely elongated basitarsus IV, 8.8 times longer than wide. Furthermore, this species has a Basitarsus VI/Tibia ratio equal to 1.07 and a Propeltidium/ Basitarsus IV ratio equal to 1.58 – 1.7. These values are close to those presented in the highly troglomorphic Greek species *E. naxos* Condé 1989<sup>6</sup>.



**Figure 1.** Habitus of a living specimen of *Eukoenenia maquinensis*.

### Species habitat

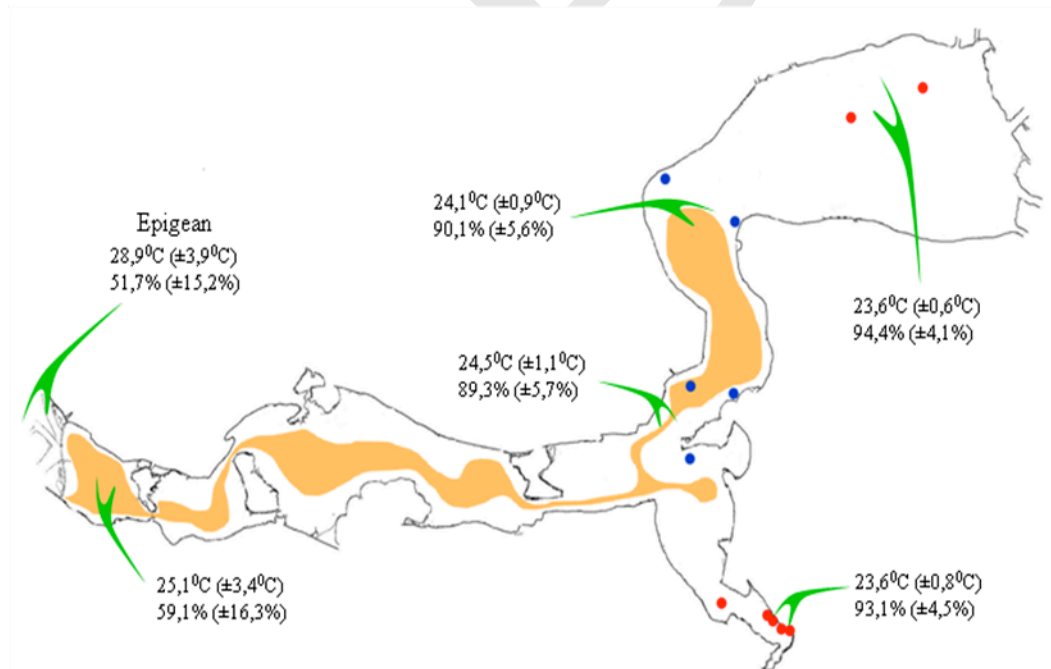
Maquiné cave is the only known habitat for *E. maquinensis*. This cave is located in the municipality of Cordisburgo, Minas Gerais state, Brazil. It comprises the oldest show cave in Brazil, and has been exploited for this purpose since 1908. A gate with screen installed near the cave entrance has prevented use by bats for decades. Guano, which could be a resource of great importance in this permanently dry system, was basically absent until recent years. In 2009, the gate was removed and bats have recolonized the cave. Organic resources in the cave consist of material left during the installation of the touristic infrastructure, such as wood scraps and food left by tourists during their visitation of the cave. Small plants grow near the cave mouth. We encountered the fauna of the cave mainly through the touristic parts of the system, where organic resources are present. In the areas closed to visitation, we found few inhabitants.

### Distribution of *E. maquinensis* within the cave

The first individual of *E. maquinensis* was found in 1999 within the tourist area. Between 1999 and 2001, three other individuals were also observed, all in the same chamber. Climatic traits of this region were relatively stable when compared to the epigean surrounding environment (Figure 2). In 2009, one specimen was observed in a chamber located out of the tourist route. In 2010, during studies concerning the management plan for the cave, six specimens were observed, five of them in non-tourist areas. Finally, in 2011, a specimen was also observed in a non-touristic area. This specimen was observed for minutes and its behavior was registered through photographs.

The shift of specimens in recent years from tourist to non-tourist areas apparently resulted from the removal of a gate in the cave entrance. After that, bats settled in non-visited areas of the cave, producing guano. Guano resources may attract potential prey items of *E. maquinensis* (such as springtails and mites).

All individuals were found in areas with moisture reaching from 89% up to 95% and temperatures ranging from 23.6°C to 24.5°C (Figure 2).



**Figure 2.** Schematic map of Maquiné cave. The red spots represent the individuals found in non-visited areas while the blue spots indicates the individuals observed in the touristic areas of the cave.

## Behavioral traits

Observations were sporadic; most of the behaviors that we describe here were registered from one live specimen of *E. maquinensis* in the study system on 25/04/2011 at 5:00 p.m.

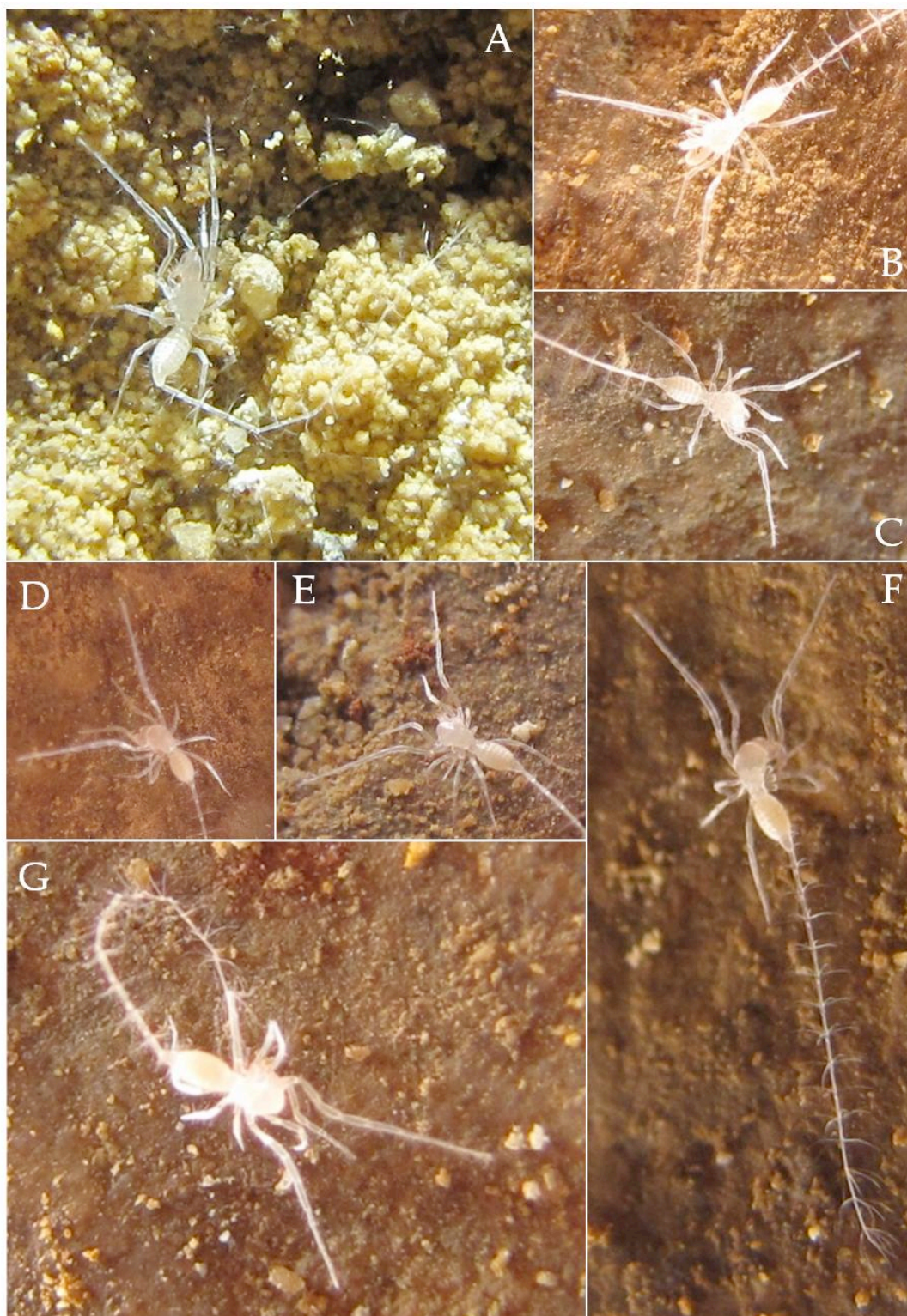
The focal individual kept moving almost all the time in which it was observed. It repeatedly swerved and did not demonstrate any directional movement. While moving, the individual kept the flagellum upward, moving it laterally. We hypothesize that the uplifted flagellum was associated with perception of the environment.

The focal specimen entered a depression with an empty spider web. As soon as it made contact with the web, the individual stopped moving. It kept immobilized for several seconds before starting to move again (Figure 3A). However, instead of struggling like most invertebrates caught in spider webs, the individual began to move slowly, removing the threads of the web. This behavior lasted roughly 2 minutes. Using the chelicerae, the specimen appeared to have cut the web, but due to its small size, it was impossible to see exactly how the web was removed. The slow movements of the individual to get rid of the web were remarkable. Although speculative, it is plausible to assume that this behavior may prevent a predator to quickly attack the prey.

The observed specimen spent considerable time cleaning itself (Figure 3 A-G). We hypothesize that this organism spends time cleaning because of its need to keep sensory structures devoid of waste. However, the frequency of this behavior could be caused by the contact with the spider web. The cleaning was always made with the chelicerae, the organism curved them in a ventral position and performed the appendage cleaning. It occurred quickly (generally, less than 10 seconds) and always one appendage at a time. The pedipalps (Figure 3B), legs (Figures 3C, D, E, F) and flagellum (Figura 3G) were cleaned.

The species *E. florenciae* (endogenous, also found in caves from Brazil) clean the flagellum through the full flexion of it under the opisthosoma horizontally (Ferreira, unpublished data). The shorter flagellum present on endogenous species allows this movement. Otherwise, in *E. maquinensis* the long flagellum prevents it to be flexed under the opisthosoma. It is bent laterally, allowing the individual to reach the last flagellar segments (Figure 3G).





**Figure 3.** Different behavior exhibited by *E. maquinensis*: A) Specimen trapped in a spider web; B) Specimen cleaning the right pedipalp; C) Specimen cleaning a leg II (right); D) Specimen cleaning a leg IV (left); E) Specimen cleaning a leg III (right); F) Specimen cleaning a leg IV (right); G) Specimen cleaning the flagellum.

## Threats

The history of commercial use with this cave indicates a sequence of events that we hypothesize has impacted the hypogean biological community across the last 200 years. The floor excavation (to remove fossils or saltpeter) has changed the topography, modifying pristine microhabitats. The obstruction of the cave entrance certainly has caused the depletion of resources in the cavity, since the supply of an important trophic resource (bat guano) has been interrupted for decades. In addition, the implementation of tourism has also led to several changes in habitats. For example, the last decades have seen the cave formations washed with a certain frequency to remove the moss/algae that grew due to light from the spotlights. In addition, other severe impacts have also occurred, such as the application of poison inside the cave (in 1990) to prevent the proliferation of fleas.

Perhaps the main threat to the species consists of commercial use of the cave. Since *E. maquinensis* are very small, they can easily be trampled by visitors. Unfortunately, it is impossible to determine the frequency of such occurrences, as well as the percentage of population mortality caused by tourists.

However, with the removal of the entrance gate, the bats began to recolonize the cave in the last three years. Bat colonies have established mainly in areas not visited by tourists, which may have contributed to the attractiveness of invertebrates for those regions (due to the continuous deposition of fresh guano). The specimens observed in the last two years were only found in non-touristic areas. The persistence of *E. maquinensis* in the areas of non-commercial visitation provides hope for the future of this endemic species.

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