

## EXTERNAL MORPHOLOGY OF THE ANTENNAE OF *Rhinocricus padbergi* Verhoeff, 1938 (DIPLOPODA, SPIROBOLIDA)

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### ABSTRACT

The antennae of millipedes (Diplopoda) have an important role in detecting various types of environmental stimuli. The structural organization of the antennae is closely related to the degree of sensorial perception. The antennae generally consist of eight segments, the most distal (apical) of which characteristically contains four cone-shaped sensilla. In this paper, we describe the external morphology of the antennae of the diplopod *Rhinocricus padbergi*. Antennae from anesthetized male and female millipedes were fixed in Karnovsky solution and processed for scanning electron microscopy. Intraspecific variation was observed in the number of apical cones (13-18) in segment VIII. This number of cones differed from that usually found (4) in other millipede species. Segments I, II and III lacked sensilla while the remaining segments had different types of sensilla, including trichoidea, chaetica and basiconica. The presence of the latter types of sensilla suggests that *R. padbergi* antennae have chemoreceptor, mechanoreceptor and thermal/hygrosensitive functions, respectively.

**Key words:** Diplopoda, millipedes, scanning electron microscopy (SEM), sensilla external morphology

### INTRODUCTION

Diplopod antennae have an important sensorial function in providing the animals with information about the environment. This importance was first reported by Cloudsley-Thompson [2], who demonstrated that removal of the antennae kept the animal in a general state of depression that persisted until death. Carey and Bull [1] observed that males without antennae were unsuccessful in copulating with females.

The antennae occur in pairs and consist of eight segments each. The apical segment is usually small and contains characteristic cone-like sensilla. The antennae are covered with a variety of sensorial structures, including mechanoreceptors and contact chemoreceptors that have taste and olfactory functions. The mechanoreceptors and chemoreceptors have internal structures very similar to those found in insects [3]. Thermal and hygroreceptors are probably also present [5-16].

The antennae of the Iulidae contain five types of sensorial structures, the most prominent of which are

the apical cones; these structures are contact chemoreceptors and probably also have taste, tactile and olfactory functions. The other four types of sensorial structures and their functions include: a) sensilla basiconica spiniformis – gustatives, b) sensilla chaetica – mechanosensitive, c) sensilla trichoidea - contact chemoreceptors and, d) sensilla basiconica - thermal and hygrosensitive [3,9,13,16,17]. According to Nguyen Duy-Jacquemin [13], these sensilla of the Diplopoda are classified based on characteristics such as the absence or presence of pores, the location of the pores and the absence or presence of an articulated base.

In this study we examined the external morphology of the antennae of the Brazilian diplopod–*Rhinocricus padbergi*. This species is widely distributed in the State of São Paulo, where it occurs in a variety of habitats, including grasslands, forests and urban residences. This species apparently adapts easily to different environments.

### MATERIAL AND METHODS

Male and female specimens of *Rhinocricus padbergi* Verhoeff, 1938 were collected on the UNESP campus in Rio Claro, SP. The animals were anesthetized with ether and the antennae were removed and fixed in Karnovsky fixative [4]. After critical point drying, the antennae were mounted on aluminum stubs with adhesive tape, sputter coated with gold and examined with a Jeol scanning electronic microscope (SEM).

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## RESULTS

The antennae of *R. padbergi* (Order Spirobolida) are small when compared to those of other Diplopoda species belonging to different orders and hardly reach the ground during locomotion of the animal. Segments I-VI are nearly equal in size (around 300  $\mu\text{m}$  long) whereas the last two (VII and VIII) are much smaller than the others (Fig. 1A). The VIIth segment is about 40  $\mu\text{m}$  long and the last segment (VIII) is essentially hidden by segment VII (Fig. 1B) and can only be seen in frontal view.

The cuticle of each segment is formed by juxtaposed plates (Fig. 1C,D) that make up the base of the segment (arrows in Fig. 1B). These plates, which have denticles on their apical border (arrow in Fig. 1C), gradually disappear and are absent from the middle and apical areas of the segment (Fig. 1D).

Openings among the plates (solid arrows in Figs. 1D and 2C,D) and rows of small depressions, probably associated with glands (Figs. 1D and 2A,C,D), were seen in all of the segments except the last one. A small amount of material was seen deposited over some of these depressions (hollow arrows in Fig. 1D), suggesting a probable secretory function for these structures.

The four types of sensilla observed in *R. padbergi* antennae included trichoidea, chaetica, basiconica and apical cones. The sensillum trichoidea consisted of an articulated base inserted into a cupule and an apical pore (Figs. 2A,C,D and 4A). The sensillum chaetica showed a progressive decrease in its diameter and, as in the trichoidea sensillum, was inserted into a cupule (Figs. 1B and 2B); there was no apical pore. The sensillum basiconica had no articulation at the bases and no pores (Fig. 4B). The apical cones showed several longitudinal grooves on their surface (arrow in Fig. 3C) and an apical pore (Fig. 3A-C).

Segments I, II, III lacked sensilla (Fig. 1A) whereas segment IV had 3-5 sensilla of the trichoidea type.

Segment V had a larger number of sensilla than segment IV, and were located mainly on the lateral surface (Fig. 1B). The sensilla in this segment were of two types: the sensillum chaetica and the sensillum trichoidea (Figs. 1B and 2B-D). The trichoidea sensillum seen in this segment had the same external morphology as those in segment IV (Figs. 2C,D).

Segment VI had large number of sensilla trichoidea on its surface (Figs. 1B and 2B), although the density was greatest laterally (Fig. 4A).

Segment VII was covered by numerous sensilla (Figs. 1B and 3A) of two types: sensilla trichoidea (similar to those found in the other segments) and sensilla basiconica (Figs. 3B,C and 4B). The latter were located amongst the sensilla trichoidea, mostly on the apical face of the segment.

Segment VIII was covered by groups of small cuticular expansions (Fig. 3B,C) throughout its surface and also had apical cones (Figs. 1B and 3A). The number of apical cones in 10 specimens varied from 13 to 18.

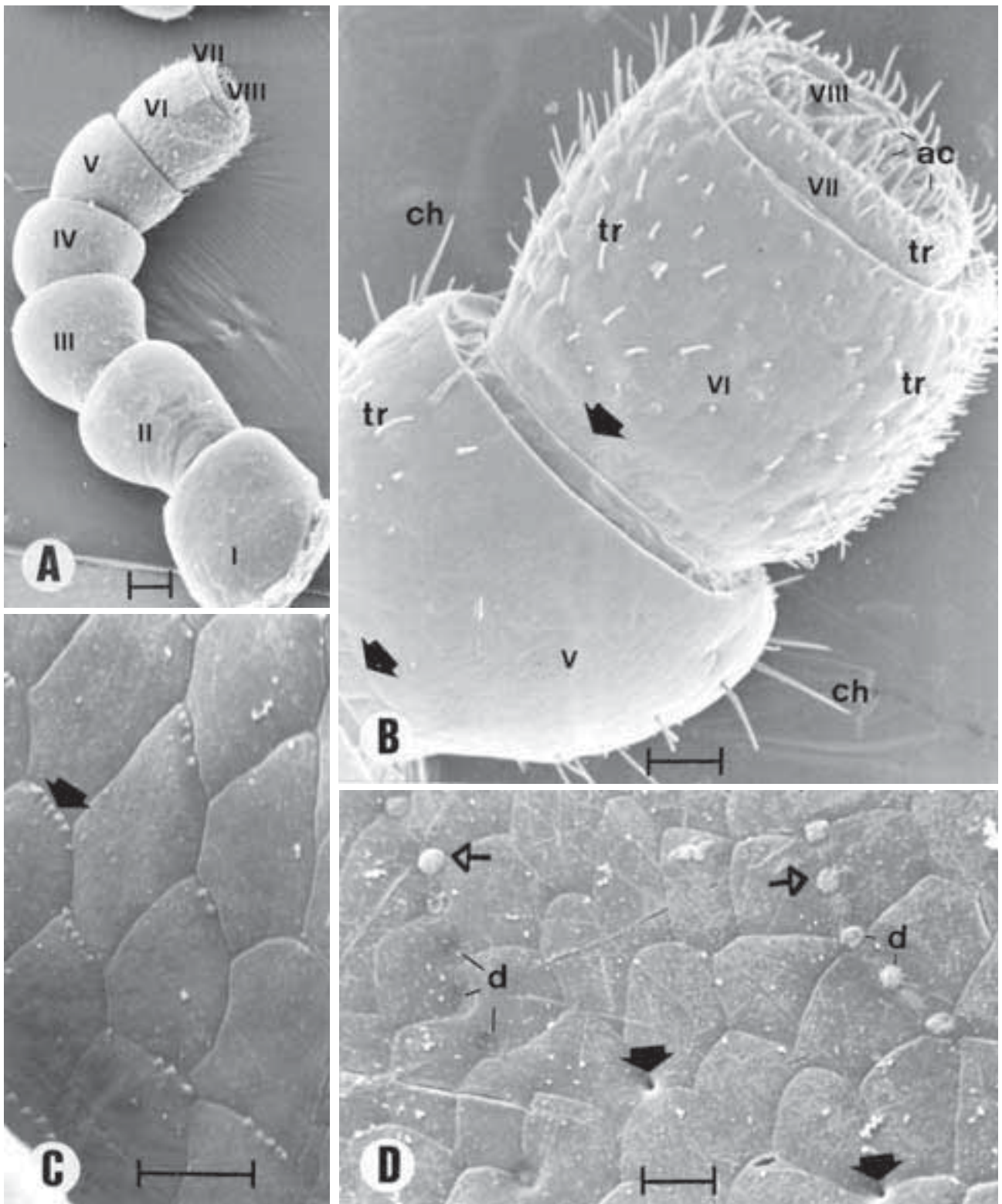
## DISCUSSION

Diplopods constantly tap the substrate with their antennae while moving. The importance of the antennal sensilla of Diplopoda for orientation, location of food and reproduction has been demonstrated [1, 2].

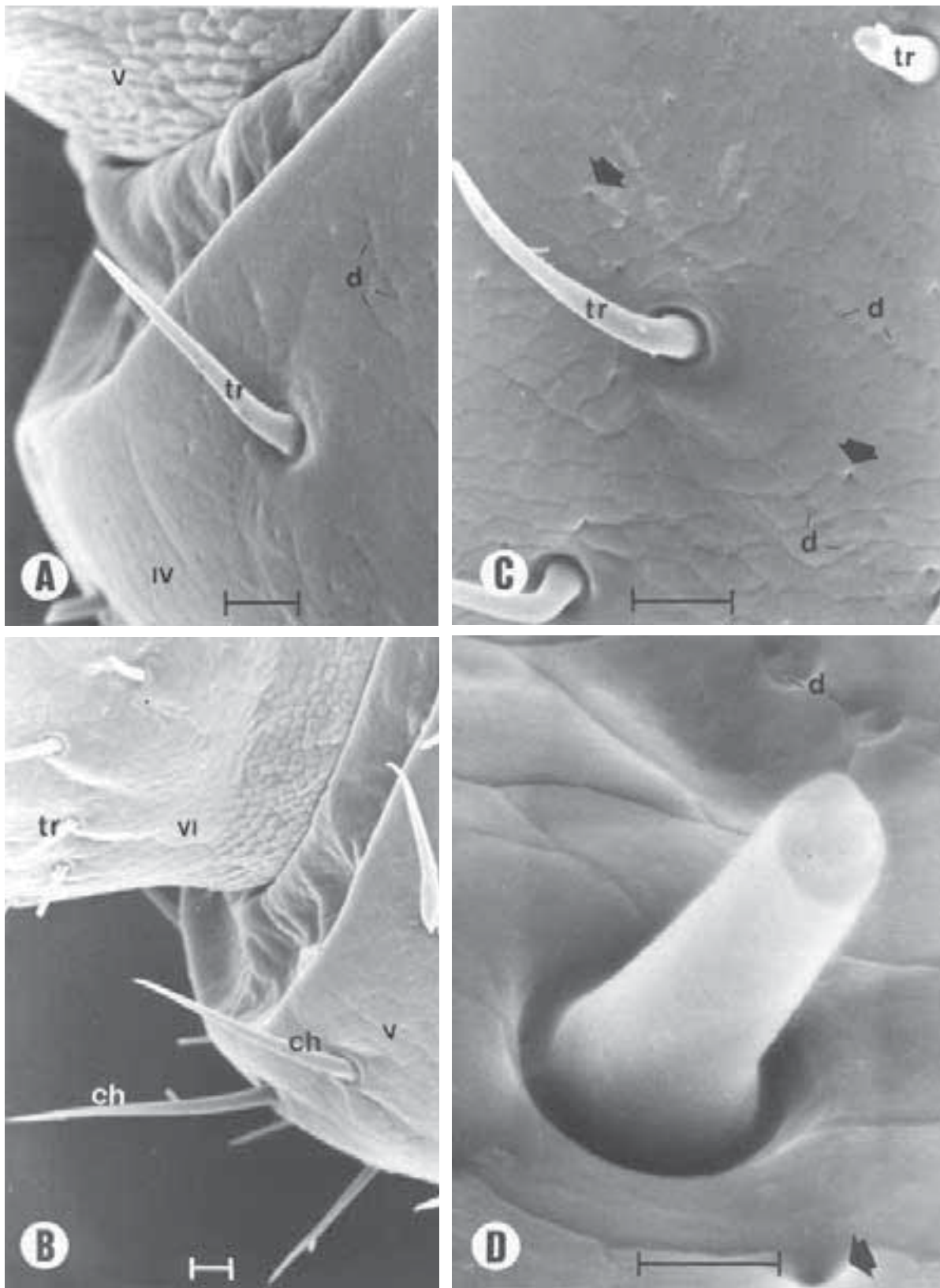
The structure and function of the different antennal sensilla in diplopods were well described by Nguyen Duy-Jacquemin for European species. The function of the sensillum chaetica in Chilognatha is probably that of mechanoreception while the sensillum trichoidea has a contact chemoreceptor function [13]. According to Nguyen Duy-Jacquemin [13], the function of sensilla basiconica without an apical pore as seen in segment VII is unknown, although a thermal-hygrosensitive role has been postulated.

The apical cones of *Cylindroiulus punctatus* (Iulidae) have similar cuticular expansions to those seen in segment VIII of *R. padbergi* [9]. According to Nguyen Duy-Jacquemin [9], these structures represent a small evagination of the exocuticle.

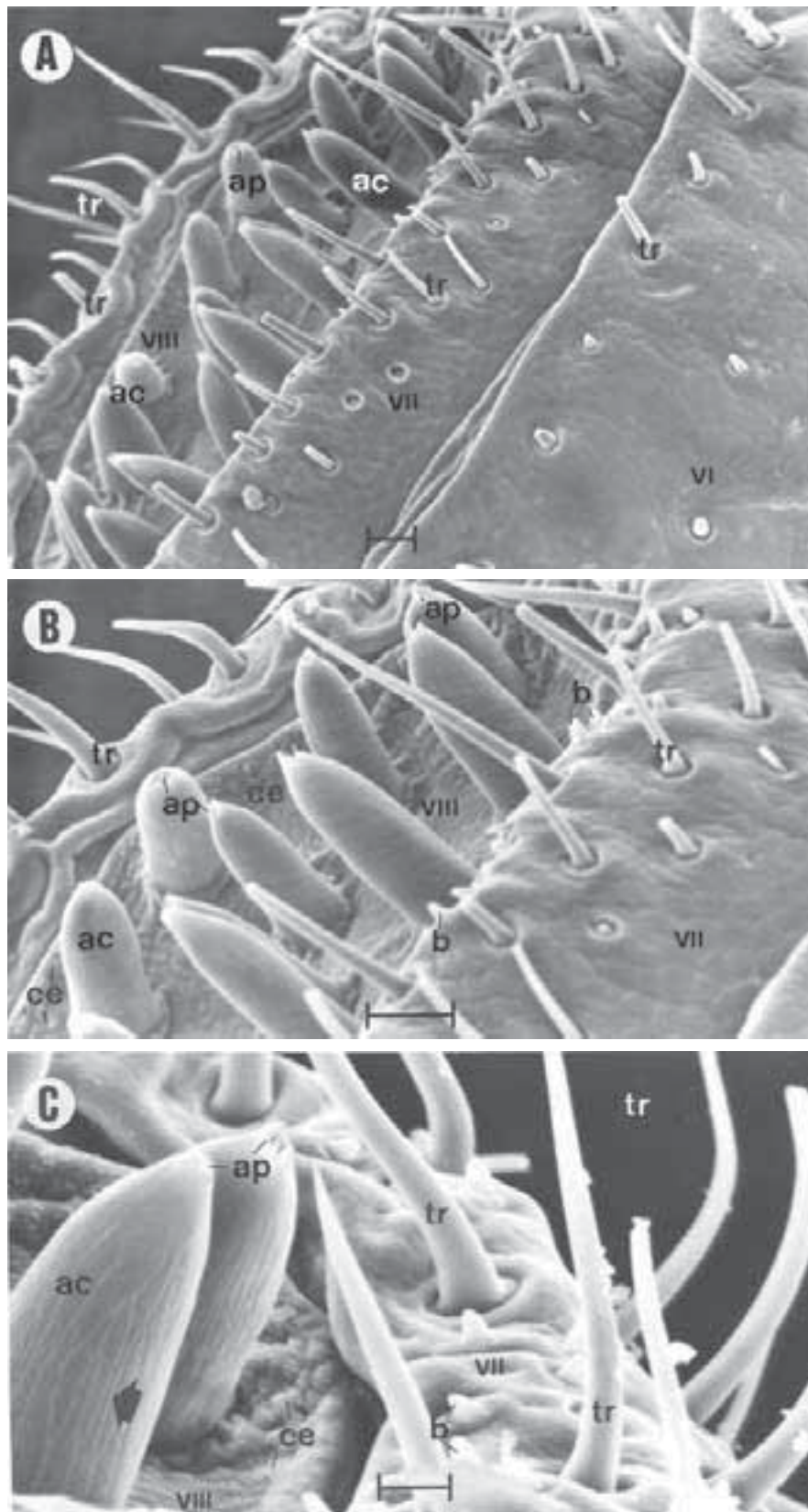
The apical cones are important sensorial organs specific to Diplopoda (16). These cones have mechanosensitive, gustative and olfactory functions and all are articulated at the base. The cones are stimulated by direct contact with the substrate [15,16]. In *Blaniulus* spp (Blaniulidae), each cone consists of a varying number of sensory units and is provided with glandular cells with a duct [15]. Apical cones occur in all diplopod, and are typically four in number. As shown here, the number of cones in *R. padbergi* (13-17) was much greater than for other species (4) [3,9,13,16]. Intraspecific variation in the number of apical cones has not yet been reported. Other species of this genus will need to be studied in order to determine whether this characteristic is typical of the genus or of this species. Such information would contribute to our understanding of the phylogenetic relationships of this group.



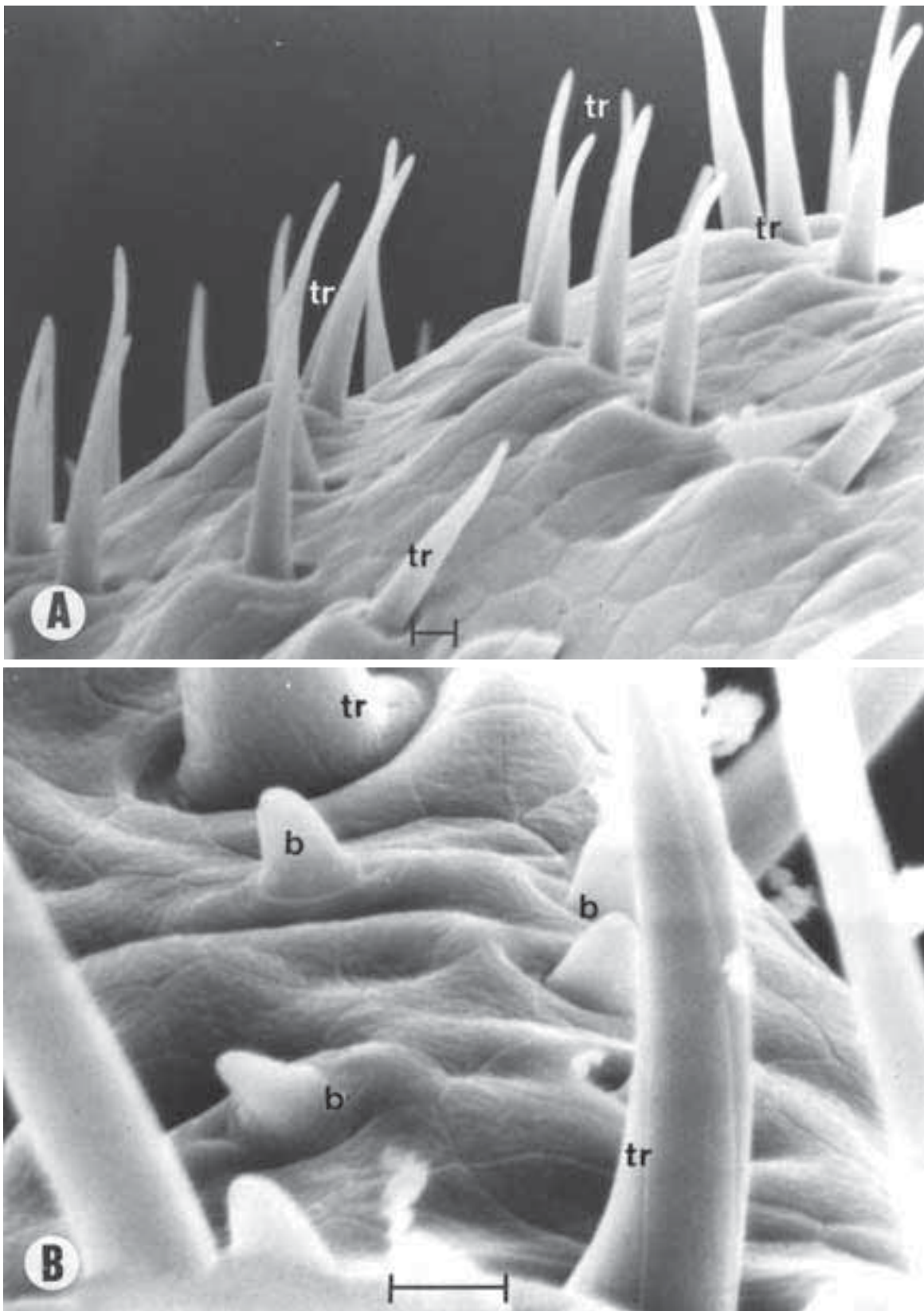
**Figure 1.** SEM of an antenna of *Rhinocricus padbergi*. **A.** General view – segments numbered from I to VIII. **B.** View of the end region of the antenna (**arrows** indicate the basal portion of the segments where the cuticular plates have denticles). **C.** Details of the cuticular plates with denticles (**arrow**) on their apical border. **D.** Details of the cuticular plates in the most apical portion of the segments (**solid arrows** indicate glandular openings among the plates, **hollow arrows** indicate material deposited in the putative glandular depressions). **ac** - apical cones, **ch** - sensillum chaetica, **d** - depressions, **tr** - sensillum trichoidea. Bars: A = 100  $\mu$ m, B = 50  $\mu$ m, C, D = 2.5  $\mu$ m.



**Figure 2.** A. Details of segments IV and V. B. Details of segments V and VI. C. Trichoidea sensilla of segment V. D. Details of a trichoidea sensillum of segment V. **ch** - sensillum chaetica, **d** - depressions, **tr** - sensillum trichoidea. **Arrows** indicate pore openings amongst the cuticular plates. Bars = A-C = 10  $\mu$ m, D = 2.5  $\mu$ m.



**Figure 3.** SEM of the apical region of an antenna of *Rhinocricus padbergi*. **A.** Details of segments VI, VII and VIII. Note the number of apical cones. **B** and **C.** Details of segments VII and VIII. **ac** - apical cone, **ap** - apical pore, **b** - sensillum basiconica, **ce** - cuticular expansions, **tr** - sensillum trichoidea. The **arrow** in **C** indicates a longitudinal sulcus in the apical cone. Bars = A, B = 10  $\mu$ m, C = 5  $\mu$ m.



**Figure 4.** A. Details of the lateral portion of segment VII. B. Details of the apical region of segment VII. **b** - sensillum basiconica, **tr** - sensillum trichoidea. Bars = 2.5  $\mu$ m.

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## REFERENCES

- Carey CJ, Bull CMM (1986) Recognition of mates in the Portuguese millipede *Ommatoiulus moreleti*. *Austr. J. Zool.* **34**, 837-842.
- Cloudsley-Thompson SL (1951) On the responses to environmental stimuli and the sensory physiology of millipedes (Diplopoda). *Proc. Zool. Soc. London* **121**, 253-277.
- Hopkin SP, Read HJ (1992) *The Biology of Millipedes*. Oxford University Press: New York.
- Karnovsky MJ (1965) A formaldehyde-glutaraldehyde fixative at high osmolarity for use in electron microscopy. *J. Cell Biol.* **11**, 137-140.
- Nguyen Duy-Jacquemin M (1972) Description d'un organe sensoriel antennaire cupuliforme chez *Polyxenus lagurus* (Diplopode, Penicillates). *C.R. Acad. Sci. Hebd. Séanc. l'Acad. Sci.* **275D**, 251-253.
- Nguyen Duy-Jacquemin M (1981) Ultrastructure des organes sensoriels de l'antenne de *Polyxenus lagurus* (Diplopode, Penicillate). I. Les cônes sensoriels apicaux du 8e. article antennaire. *Ann. Sci. Nat. Zool.* **13**, 95-114.
- Nguyen Duy-Jacquemin M (1982) Ultrastructure des organes sensoriels de l'antenne de *Polyxenus lagurus* (Diplopode, Penicillate). II. Les sensilles basiconiques des 6e. et 7e. articles antennaires. *Ann. Sci. Nat. Zool.* **13**, 211-229.
- Nguyen Duy-Jacquemin M (1983) Ultrastructure des organes sensoriels de l'antenne de *Polyxenus lagurus* (Diplopode, Penicillate). III. Les sensilles coeloconiques des 6e. et 7e. articles antennaires. *Ann. Sci. Nat. Zool.* **13**, 207-220.
- Nguyen Duy-Jacquemin M (1985a) Ultrastructure des cônes sensoriels apicaux et des sensilles basiconiques spiniformes du 7e. article des antennes de *Typhloblaniulus lorifer* et *Cylindroiulus punctatus* (Diplopodes, Iulides). *Ann. Sci. Nat. Zool.* **13**, 67-88.
- Nguyen Duy-Jacquemin M (1985b) Structures dendritiques des cônes antennaires apicaux de diplopodes (Myriapoda). *Bijdragen tot de Dierkunde* **55**, 159-170.
- Nguyen Duy-Jacquemin M (1988) Ultrastructure des organes sensoriels de l'antenne de *Polyxenus lagurus* (Diplopoda, Penicillate). IV. Les sensilles setiformes à base renflée. *Ann. Sci. Nat. Zool.* **13** 161-175.
- Nguyen Duy-Jacquemin M (1989) Ultrastructure des sensilles basiconiques baciliformes des antennes du diplopode cavernicole *Typhloblaniulus lorifer* Brolemann (Myriapoda, Diplopoda). *Mem. Biospeol.* **16**, 251-256.
- Nguyen Duy-Jacquemin M (1990) Connaissances actuelles déduites de l'étude ultrastructurale des sensilles, sur le rôle de l'antenne dans la perception des stimuli chez les myriapodes. In: *Proceedings of the 7th International Congress of Myriapoda*. (Minelli A, ed), pp. 97-108, E.J. Brill: Leiden.
- Nguyen Duy-Jacquemin M (1996a) Nombre et répartition des sensilles basiconiques antennaires dans le genre *Polyxenus* (Myriapode, Diplopode, Penicillate). *Bull. Soc. Zool. Fr.* **121**, 125-127.
- Nguyen Duy-Jacquemin M (1996b) Comparative ultrastructure of the apical antennal cones of two blaniulids millipedes: the cave dwelling *Blaniulus lorifer* (Brolemann, 1921) and the edaphic *Blaniulus guttulatus* (Bosc, 1792). *Mem. Biospeol.* **23**, 237-240.
- Nguyen Duy-Jacquemin M (1997) Fine structure and possible functions of antennal sensilla in *Polyxenus lagurus* (Diplopoda, Penicillata, Polyxenidae). *Ent. Scand. Suppl.* **51**, 167-178.
- Nguyen Duy-Jacquemin M, Arnold G (1991) Spatial organization of the antennal lobe in *Cylindroiulus punctatus* (Leach) (Myriapoda: Diplopoda). *Int. J. Insect Morphol. Embryol.* **20**, 204-214.

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