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Labral gland in soldiers of the neotropical termite *Cornitermes cumulans* (Isoptera: Termitidae: Syntermitinae)

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ABSTRACT

Cornitermes cumulans is a termite species of the subfamily Syntermitinae with a nasute mandibulate soldier caste, members of which are morphologically equipped with structures and organs specialized for the defence of the colony. We investigated the labrum of soldiers of *C. cumulans* and described the labral gland, an exocrine structure present in this appendage. The labrum of *C. cumulans* soldiers presented two distinct regions, the hyaline tip and the proximal region connected to the head. The hyaline tip exhibited a thick cuticle composed of a loose endocuticle involving an epithelium of class 1 cells, which synthesize the glandular product into a subcuticular space that function as a reservoir prior to release the final secretion. The proximal region of the labrum had an epithelium composed of class 1 and class 3 cells, which released the secretion onto both ventral and dorsal surfaces. The ultrastructure showed abundant smooth endoplasmic reticulum and glycogen in the class 1 cells, whereas the class 3 cells had many electron-lucent vesicles that varied in size. We associated the labral gland with the production of toxic substances that may act inside enemy wounds made by the soldiers' mandibles. Other possible functions for the labral secretion are also discussed.

1. Introduction

The soldiers of Isoptera are highly specialized for defensive functions, and thus they display unique physiology and anatomy. Members of this caste are generally sterile and are not able to feed themselves (Noirot, 1969). Although soldiers are a terminal stage (i.e., they are not able to develop further into other castes), they are not considered adults because their prothoracic glands remain developed (Noirot and Bordereau, 1991).

The termite buccal apparatus is adapted for biting and is composed of the labrum, epipharynx, labium, mandibles, maxillae and hypopharynx (Chapman, 1998). Some termite soldiers exhibit important modifications of these buccal parts, like powerful mandibles and differentiation of the labrum. The labrum may completely cover the mandibles, as in *Glossotermes oculatus* (Šobotník et al., 2010), or be modified as a brush used to spread frontal secretions onto enemy wounds (Quennedey, 1975).

According to Deligne et al. (1981), a labrum with a transparent, thick, pointed apex, known as a hyaline tip, is present in soldiers of *Macrotermes*, *Acanthotermes*, *Pseudocanthotermes*, *Synacanthotermes*, and *Allodontotermes* (Macrotermitinae) and in some soldiers of basal Syntermitinae. These authors suggested the occurrence of a glandular apparatus in the hyaline tip, named the labral gland, which was responsible for the secretion of an abundant and flocculent liquid. This was later confirmed by Quennedey (1984) in soldiers of *Macrotermes bellicosus*, and this author named this organ as cibarial gland because of the similarity between the secretory epithelia of the labrum and the hypopharynx. In spite of the high degree of labrum development in the soldiers of *Cornitermes*, *Syntermes*, *Labiotermes*, *Apicotermes*, and *Foraminitermes*, no morphological information was provided for this structure in these taxa (Quennedey, 1984).

The function of the labral gland is hypothesized to be related to defence, and its secretion is thought to either coat the soldier mandibles prior to an attack on an enemy or be released directly onto the enemy wound made by the mandibles (Quennedey, 1984; Šobotník et al., 2010). The chemical composition of the secretion remains unknown.

The labral gland seems to vary among termite species, with different morphologies influencing the defensive mechanisms in soldiers. The minor soldier of *Schedorhinotermes* presents only class 3 glandular cells, and the soldier brushes the labrum impregnated with the secretion on the enemy's body during defensive behaviour (Quennedey, 1975). The labrum of *M. bellicosus* soldiers presents a

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hyaline tip and the labral gland is composed of only class 1 cells (Quennedey, 1984).

The morphology of neotropical termites of the subfamily Syntermitinae is poorly studied, and new information about this topic will contribute to the knowledge and understanding of their phylogeny, mainly because this subfamily is quite heterogeneous (Rocha et al., 2012). In the present study, the labrum of *Cornitermes cumulans* soldiers, which possess a hyaline tip, is investigated and the morphology of its labral gland is described. We also discuss possible functions of the labral gland in these soldiers.

2. Material and methods

2.1. Insects

Soldiers of *C. cumulans* Kollar 1832 were collected from nests located on the campus of São Paulo State University (UNESP) in the city of Rio Claro, SP, Brazil.

2.2. Scanning electron microscopy

Heads of *C. cumulans* soldiers were fixed in Karnovsky fixative, dehydrated in graded acetone series and dried to critical point in a CPD 030 Balzers desiccator. The heads were mounted on stubs with double-stick tape, sputter-coated with gold in a vacuum metalizer and examined in Jeol JSM-300 scanning electron microscope.

2.3. Transmission electron microscopy

The labrum of *C. cumulans* soldiers were isolated and fixed for 24 h in 3% glutaraldehyde buffered with 0.1 M sodium cacodylate (pH 7.3). After two washings in the same buffer solution, they were postfixed in 1% OsO_4 buffered with 0.1 M sodium cacodylate. After dehydration in ethanol and acetone series, the material was included in Epon-araldite resin, sectioned with a Porter Blum ultramicrotome and examined using EM9S2 and Philips electron microscopes. Semi-thin sections were stained with methylene blue and azur II and examined under a Leica photomicroscope.

3. Results

The labrum of *C. cumulans* soldiers is short, does not exceed the mandibles, and has a translucent tip with a crystal-like appearance (Fig. 1).

Fig. 1. Soldier of *Cornitermes cumulans*. The white arrow points to the hyaline tip of the labrum.

A dorsal view of the hyaline tip under scanning electron microscopy showed a round and swollen region that lacked long hairs but had characteristic peg-like sensilla (Fig. 2A–C). Additionally, the proximal region below the tip displayed two rows of hairs of different lengths (Fig. 2A). Among these hairs, in the latero-dorsal region, we observed many campaniform sensilla, some peg-like sensilla, and numerous glandular pores (Fig. 2D and E).

The ventral view of the hyaline tip was similar to the dorsal view, with the same wrinkled appearance and lack of hairs (Fig. 2F). The latero-ventral region had numerous glandular pores, which were absent in the median dilated part (Fig. 2G).

Successive transversal sections of the hyaline tip showed a glandular epithelium composed exclusively of class 1 secretory cells, as classified by Noirot and Quennedey (1974, 1991), about 15 μm long (Fig. 3A–D). This glandular epithelium was associated with a thick cuticle that was separated from the glandular epithelium by a subcuticular space filled with secretion (Fig. 3B). This cuticle was composed of a thin epicuticle and a thick and loose endocuticle. The epicuticular layer presented many pores and the endocuticular layer showed numerous pore canals. Class 1 secretory cells had elliptic nuclei and they were provided with extensive and deep plasma membrane invaginations in their basal regions (Fig. 3C and E). Neighboring cells displayed occlusive apical junctions and basal septate desmosomes (Fig. 3F). Many compacted microvilli were present in the apical portions of these cells (Fig. 3C and G). The apical cytoplasm presented enormous electrondense structures wrapped by concentric membrane layers (Fig. 3F and G). The glandular cells contained few profiles of rough endoplasmic reticulum, glycogen granules, numerous mitochondria, vesicles of different densities, abundant vesicular smooth endoplasmic reticulum and myelin figures (Fig. 4A and B). Glycogen was found throughout the secretory cells.

Transverse sections of the labrum, proximally from the hyaline tip, showed groups of class 1 and class 3 secretory cells (Fig. 5A and B). Each class 3 secretory cell was provided with a cuticular duct, composed of a receiving canal that collected the secretion and a conducting canal that crossed the thick cuticle to discharge the secretion. The receiving canal was composed of an interrupted inner epicuticle with few and sparse microvilli (Fig. 5C–E). The conducting canal was surrounded by a canal cell with an oval nucleus and spots of chromatin aggregates (Fig. 5F). This canal presented a continuous cuticular layer (Fig. 5G) that extends through the labral cuticle and open as a pore on the cuticular surface (Fig. 2G). The cytoplasm of class 3 cells contained many clear vesicles of different sizes with fine electron-dense particles (Fig. 5D).

4. Discussion

Termites possess many exocrine glands associated with chemical communication and the production of defensive substances (Costa-Leonardo and Haifig, 2010). In general, the exocrine glands associated with the buccal apparatus have the same name as the appendage to which they are connected. Noirot (1969) mentioned the presence of structures containing sensory elements in the termite labrum, which might correspond to the glandular cells previously named labral glands by Holmgren (1909).

Quennedey (1975) studied the ultrastructure of the labral gland in minor soldiers of *Schedorhinotermes* and showed that this gland was composed only of class 3 glandular cells, according to the classification of Noirot and Quennedey (1974, 1991). The author also observed many sensorial structures, two types of trichoid, and one type of campaniform sensilla associated with the labrum. Sensorial structures were also present on the labrum of *C. cumulans*, extending from the base to the hyaline tip on the dorsal surface. The presence of these structures on the surface of the labrum reinforces