THE ANTENNAL SENSILLA OF THE NYMPH OF *EPHEMERA DANICA*

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**Abstract**

In the present study, a first ultrastructural investigation under Scanning and Transmission Electron Microscopy (SEM, TEM) on the sensilla located on the flagellum of the nymphal antennae of *E. danica* is presented. Each antenna consists of scape, pedicel and a flagellum of 26–27 segments. The flagellar sensilla are mainly located on the dorsal side of the flagellum and they are represented by long trichoid sensilla and shorter basiconic sensilla. The internal structure of these sensilla reveals that the long trichoid sensilla are mechanoreceptors while the shorter basiconic sensilla are uniporous sensilla with a chemo-mechanical function. These chemo-mechanosensory sensilla seem to be very common in mayflies. The presence of these sensilla is discussed in relation with the behavioral ecology of this burrowing insect.

Key words: Sensilla; aquatic insects; mechanoreceptors; chemo-mechano-receptors.

**Introduction**

*Ephemera danica* is a burrowing mayfly commonly present in freshwater streams and characterized by a semivoltine life-cycle. At present, the knowledge on the sensory structures of the nymph of this genus and, in particular, on its sensilla, is limited to an ultrastructural investigation by Schmidt (1974), who described the mononematic scolopidia located on the pedicel of the nymph of *Ephemera* sp. The author considered this structure as homologous of the Johnston’s organ of other insects, thereby performing a mechanosensory function allowing insects to sense the movement of the flagellum over the pedicel.

The present study represents the first ultrastructural investigation under scanning and transmission electron microscopy (SEM, TEM) on the sensilla located on the flagellum of the nymphal antennae of *E. danica*.

**Methods**

Mature nymphs of *Ephemera danica* Müller, 1764 were collected in the Nera River (Perugia, Umbria Region, Italy) in spring 2003. The antennae of the nymphs were dissected under a stereomicroscope. The antennal flagella were fixed in 2.5% glutaraldehyde buffered in cacodylate, pH 7.2, for 12 h, repeatedly rinsed in the same buffer and postfixed in 1% osmium tetroxide for 1 h. For scanning electron
microscopy (SEM) observations, the specimens were dehydrated by using ethanol gradients, followed by critical-point drying in a CO2 Pabisch CPD apparatus. Specimens were mounted on stubs with silver conducting paint, sputter-coated with gold-palladium in a Balzers Union Evaporator, observed and photographed in a Philips EM XL30 of the Electron Microscopy Centre of the University of Perugia.

For transmission electron microscopy (TEM) analysis, selected material was dehydrated in the graded ethanol series and embedded in Epon-Araldite mixture resin. Thin sections, cut on a Reichert ultramicrotome, were collected on formvar-coated copper grids and stained with uranyl acetate and lead citrate. The thin sections were examined with Philips EM 400 transmission electron microscope of the Electron Microscopy Centre of the University of Perugia.

Results

The antenna are held horizontally over the mandibles (Fig. 1), pointing anteriorly to the head. Each antenna consists of scape, pedicel and a flagellum of 26–27 segments. The flagellar sensilla are mainly located on the dorsal side of the antenna (Fig. 2), whose cuticle is irregularly spiny (inset of Fig. 2). The ventral side of the antenna is almost hairless (Fig. 3) and shows a smooth cuticle (inset of Fig. 3). The dorsal side shows in its proximal portion numerous setae located in the middle of each segment (Fig. 2). These setae are represented by two lateral groups of long (500–800 µm long, 7–8 µm wide) trichoid sensilla and one central group of shorter (150–200 µm long, 6–7 µm wide) basiconic sensilla (Fig. 4). Each lateral group of trichoid sensilla shows 6 hairs while the central group of basiconic sensilla is composed of 4–6 hairs. The sensilla tend to decrease in number towards the antennal distal region where only few groups of 2–4 basiconic sensilla are located (Fig. 5).

The very long and thin trichoid sensilla are mechanoreceptors: indeed they emerge from a well developed socket (Fig. 6), showing in section an elastic joint membrane connecting the hair to the socket and a socket septum supporting the tubular body (Fig. 7). The basiconic sensilla emerge from a socket (Fig. 6) and show an apical pore (Fig. 8). In section, the socket shows an elastic joint membrane connecting the hair to the socket and a socket septum supporting the tubular body (Fig. 9). The shaft has two longitudinally separated lumina as shown in longitudinal (Fig. 10) and in cross sections (Fig. 11): the inner lumen containing two dendrites extending along the shaft up to the apical pore and an outer lumen (Figs. 10 and 11). The three dendrites innervating the sensillum (one ending at the base of the hair with a tubular body and two extending along the hair up to the terminal pore) are surrounded by a dendritic sheath (Fig. 12). This morphology is consistent with a chemo-mechanosensory (gustatory) function.
Figures 1–12. Flagellar sensilla of the nymph of *E. danica* under SEM (Figs. 1–6, 8) and TEM (insets of Fig. 2 and 3, Figs. 7, 9–12): Fig. 1. Ventral view of the mandibles (M) and antenna with scape (S), pedicel (P) and flagellum (F); Fig. 2.
Dorsal side of the flagellum in its proximal portion. Note the numerous sensilla (long trichoid sensilla, T and short basiconic sensilla, B) located in the middle of each flagellar segment. Inset shows the irregularly spiny appearance of the cuticle; Fig. 3. Ventral side of the flagellum in its proximal portion. Inset shows the smooth surface of the cuticle; Fig. 4. Detail of Fig. 2 showing two lateral groups of long trichoid sensilla (T) and one central group of shorter basiconic sensilla (B); Fig. 5. Distal portion of the flagellum. Note the low number of basiconic sensilla (B) in this area; Fig. 6. Long trichoid sensilla (T) and basiconic sensilla (B) emerging from a well developed socket. Note the spiny appearance of the cuticle; Fig. 7. Transversal section of a mechanoreceptor at the level of the socket. Note the elastic joint membrane (JM) connecting the hair (H) to the socket and the socket septum (SS) supporting the tubular body (TB); Fig. 8. Apical portion of a basiconic sensillum showing the apical pore (arrow); Fig. 9. Transversal section of a basiconic sensillum at the level of the socket. Note the elastic joint membrane (JM) connecting the hair (H) to the socket and the socket septum (SS) supporting the tubular body (TB); Fig. 10. Longitudinal section of a basiconic sensillum at the level of the shaft. Note the two longitudinally separated lumina. The inner lumen contains two dendrites (D) extending along the shaft; asterisks point out the outer lumen; Fig. 11. Transversal section of a basiconic sensillum at the level of the shaft. Note the two longitudinally separated lumina. The inner lumen contains two dendrites (D) extending along the shaft; asterisk points out the outer lumen; Fig. 12. Three dendrites (D) innervating the basiconic sensillum wrapped by the dendritic sheath (DS).

**Discussion**

This ultrastructural investigation showed that all the hairs located on the dorsal antennal surface of the nymph of *E. danica* are sensilla. Most of them are groups of very long mechanoreceptors among which groups of shorter uniporous basiconic sensilla with chemo-mechanosensory (gustatory) function are located. The different distribution of sensilla on the two sides of the antenna (the gathering of sensilla on the dorsal side and their absence on the ventral side of the antenna) is in agreement with the behavioural ecology of this species. Indeed, *E. danica* nymphs live in very depressed “U” shaped burrows in sandy sediment (Ladle and Radke, 1990). The burrows are generally closed by coarse sediment anteriorly and the nymph, with upward thrusting movements of the head, creates and maintains a sediment-free space beneath the head and body. This sediment-free space is important for the water current around the insect. Ladle and Radke (1990) stress the importance of the antennae “thickly clothed in long, stiff hairs and held horizontally” in preventing coarse particle of sediment from falling into the space anterior to and ventral to the head. From our investigation, it emerges that the longest hairs are mechanoreceptors probably involved in sensing the presence and dimensions of the sediment over the
head. The relevance of tactile stimuli on the dorsal surface of the body in the burrowing of *Ephemera* sp. nymphs was already reported by Grandi (1960) on the base of some behavioural observations.

Also, the spiny pattern of the antennal dorsal cuticle may be correlated with the distribution of the sediment around the nympha1 body: the spiny pattern could protect the cuticle from the abrasive effect of the sand grains during the burrowing of the insect. This assumption is confirmed by the smooth surface of the antennal ventral side, which is not in contact with the sediment.

The uniporous chemo-mechanosensory basiconic sensilla show an internal structure very similar to that of the so-called “flat-tipped sensillum”, first described in the larvae of *Baetis rhodani* (Gaino and Rebora 1996, 1997, 1998, 1999a) and then observed on their antennae (Gaino and Rebora 1998), legs, tergites and cerci (Gaino and Rebora 1999b), maxillae and labial palps (Gaino and Rebora 2003). A sensillum similar to the flat-tipped sensillum has been also observed on the nymphal antennae of the heptageniid *Rhithrogena* and *Ecdyonurus* (Gaino and Rebora 1996). The bulk of data indicate that this sensillum, having both a mechanical and a chemical sensory function, is fairly common in Ephemeroptera. Behavioral studies on Ephemeroptera proved *Baetis tricaudatus* nymphs use chemical and hydrodynamic stimuli to detect Plecopteran predators (Peckarsky 1980, Peckarsky and Penton 1989). These two types of stimuli seem to interact together, as hypothesized by Ode and Wissinger (1993) in a leptophlebiid. On the basis of previous investigations, it seems acceptable that this sensillum, scattered on the whole insect body, can support a “general gustatory function”, useful not only to detect predators but also to locate feeding resources. The position of these gustatory sensilla on the antennae of *E. danica* is particularly relevant in consideration of the peculiar feeding of this mayfly. Indeed, suspended particles that the nymph is going to ingest are drawn by the water current through the “plug” of sediment just above the antennae (Ladle and Radke 1990).

**Literature Cited**


