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Zoological Research

Scanning Electron Microscopy of Antennae of *Aphidoletes aphidimyza* (Diptera: Cecidomyiidae)

ZHANG Jie, YANG Mao-fa*

（*Key Laboratory for Plant Pest Management of Mountainous Region of Guizhou Province, Institute of Entomology, Guizhou University, Guiyang* 550025, *China*）

Abstract: The morphology of antennae of *Aphidoletes aphidimyza* was observed with a scanning electron microscope. The results showed that both male and female were fourteen segmented, the male was approximately 2000μm and the female was 1050μm. Six types of sensillae on the antenna were observed, viz. chaetica (Ch), trichoidea (Tr), basiconica (Ba), cavity (Ca), styloid (St) and circumfila (Ci) on the antennae of *A. aphidimyza*. Sensillae Ch had a long external-process, with a base surrounded by membranous sockets and a length of about 67.5μm. Sensillae Tr were distally curved and inserted into a depression, 61.0μm long. Sensillae Ba were peg-like and 4.7μm long on the antennae. Sensillae Ca were pit-like in appearance and the diameter of the pit was 1.2μm. Sensilla St was found on the second sub-segment flagellum of the male antennae. The length of the sensilla was about 21μm and the diameter was 1.5μm. The circumfila, which are a unique type of sensilla found only on cecidomyiid antennae, formed loops around each of the antennal sub-segments, and were attached to the surface by a series of stalks. Sensilla St was only present on male antenna. The number of Ba and Tr was almost the same in both sexes. There were more Sensilla Ca on the male antenna than on the female, while there was more Ch on the female.

Key words: *Aphidoletes aphidimyza*; Scanning electron microscopy；Antenna; Sensillae; Ultrastructure

食蚜瘿蚊触角的扫描电镜观察

张 洁，杨茂发*

（贵州山地农业病虫害重点实验室，贵州大学 昆虫研究所，贵州 贵阳 550025）

摘要：用扫描电子显微镜对食蚜瘿蚊的触角进行了观察。结果显示，雌雄触角都为14节，其中雄性约为2 000 μm，雌性约为1 050μm。电镜下可观察到食蚜瘿蚊触角有6种类型的感受器，即：刺形感受器、毛形感受器、锥形感受器、腔形感受器、柱形感受器和环丝。刺形感受器较长，约67.5μm，基部有膜状的窝。毛形感受器长约61μm，末端弯曲。锥形感受器呈钉状着生在表皮上，长约4.7μm。腔形感受器呈凹陷状，腔的直径约为1.2μm。柱形感受器着生在雄虫鞭节的第二亚节，长约21μm，直径约为1.5μm。环丝，是瘿蚊类昆虫触角中特殊的结构，它通过着生在一系列腔中的的短梗，连结成环状附着在触角各亚节的表面。刺形和锥形感受器在数量上，雌雄之间差别不大；柱形感受器只在雄虫中发现；雄虫触角上的腔形感受器在数量上要比雌虫多。

关键词：食蚜瘿蚊；扫描电镜；触角；感受器；超微结构
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Antennae in insects are organs of taste, smell and stimulation (Wigglesworth, 1972). They serve as sensory structures, as claspers in grasping prey and taxonomic placement in certain instances, and also help in sex differentiation, as in mosquitoes. The antennae also play kinetic roles and normally keep the nervous system in a state of tone in which it responds to stimuli of all kinds. Antennae of insects vary greatly in length, overall size, size of the individual segments, segmentation, setation and other aspects with the structures being closely related to their function.

Extensive scanning electron microscope (SEM) studies were made on the antennae of lepidopterans (Yponomeutidae) and some coleopterans (Cerambycidae, Curculionidae, Carabidae, Elateridae, Anobiidae) (Dyer & Sea brook, 1975; Isidoro & Solinas, 1992; Kim &

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*通讯作者（Corresponding author），杨茂发，E-mail :yangmaofa68@hotmail.com

Yamasaki, 1996; Merivee, 1992; Okada et al, 1992). However, few studies on sensillae of dipteran have been conducted. The antennae of Cecidomyiidae have been of particular interest to entomologists since they were first studied at the end of the last century (Felt, 1897; Keiffer, 1900). They have an unusual arrangement of various sensory hairs and the possession of the looped sense organs known as circumfila. Despite several cecidomyiid species being important crop pests, to date there have been few detailed studies of the antennal sensilla. Slifer & Sekhon (1971) used scanning and transmission electron microscopy to study the antennal sensilla of sorghum midge, *Contarinia sorghicola*, an important pest of sorghum and other grasses in many parts of the world. Solinas & Nuzzaci (1987) studied in detail the antennal sensilla of another cecidomyiid midge, *Mycodiplosis erysiphes*.

Aphidoletes aphidimyza (Diptera) is one of the most popular, attractive, and economically important biocontrol agents for many insect pests (such as predator), and has a wide geographical distribution (Xie et al, 2000). But, there have been very little morphological or functional studies conducted on its sense organs. SEM may reveal the desired information in this area. Antennae being one of the primary sensory organs, such studies may reveal differences in the antennal structure and types of sensillae, which may be indicative of their different functions. Hence, SEM investigations of the antennae of both sexes of *A. aphidimyza* was to look at sex specific differences in the antennal sensillae and to understand probable roles of the various sensillae of male and female *A. aphidimyza*.

1 Materials and Methods

1.1 Sources of tested insects

Newly-emerged male and female adult *Aphidoletes aphidimyza*, from cocoons collected from Key Laboratory for Plant Pest Management of Mountainous Region of Guizhou Province.

1.2 Methods

Isolated midge heads were mounted on specimen stubs with either carbon glue or double-sided tape and coated with carbon diaphragm in HUS-5GB sputter coater. Specimens were coated for 15 minutes, being tilted and rotated between each run to maximize coating and reduce subsequent charging. Specimens were coated without using freeze drying or critical point drying techniques as these methods damage the antennal sensilla. Ten midges of each sex were examined with KYKY-1000B microscope at an accelerating voltage of 25 KV.

The sensory structures found were named following the terminology of Faucheus (1985) and Jourdon et al (1995).

2 Results

2.1 General structure of antennae

The female antennae were shorter than that of the males, with a mean antennal length of 1 050 μm compared to 2 000 μm. Unless stated otherwise, the following descriptions of the antennal morphology applies to both sexes.

The antenna of *Aphidoletes aphidimyza* consisted of scape, pedicel and flagellum. The antennal flagellum, consisted of twelve sub-segments, was attached to the head via a cup-shaped pedicel and scape. The pedicel of the female measured 45 μm in diameter, the male's was slightly larger at 50 μm. The antennal sub-segments of the male were separated by a stalk measuring 60 μm in length, thus male antennas seem to be longer than female's where the sub-segments were fused directly. Microtrichia (minute, hair-like, non-innervated cuticular structures) were present on all antennal sub-segments but absent on the stalks between the male sub-segments.

2.2 Ultra-structure of antennal sensillae in two sexes of *A. aphidimyza*

Six sensillae types occurred on the antennae of *A. aphidimyza*: sensilla chaetica(Ch), sensilla trichodea (Tr), sensilla basiconica (Ba), sensilla cavity (Ca), sensilla styloid (St) and circumfila (Ci).

2.2.1 Sensilla chaetica. Only one type of sensilla chaetica occurred on *A. aphidimyza* antennae (Figs.1, 3-5). These mechanoreceptors were present on each flagellum sub-segment of the male and female and occurred in whorls at the distal region of each sub-segment. Numbers of sensilla per sub-segment ranged from 8–15 and 12–17 for female and male antennae respectively. Significantly more sensilla chaetica were found on male antennae than on female. These blunt tipped hairs were between 1.5–2 μm in width and 65–70 μm in length. Each hair possessed a bulbous body at the base where it met the socket opening (Fig.3). The socket of the tactile hair had a circular outline and measured 5 μm in diameter, its opening had a ridged structure on the distal rim.

2.2.2 Sensilla trichodea. Sensilla trichodea are chemoreceptors which occurred on all sub-segments of both male and female antennae except the scape of females. There were slightly more sensilla trichodea on the male antennae compared to the female. Numbers of sensilla per sub-segment ranged from 18–24 and 15–20 for the male and female antennae respectively. The sensilla trichodea consisted of a hair shaft which was slightly longer in the female than the male becoming thinner towards the tip. A series of longitudinal ridges ran parallel along the length of the hair shaft from the smooth horseshoe shaped socket (Figs.1-3, 5-6).

2.2.3 Sensilla basiconica. The sensilla basiconica were peg shaped structures between 4–5 μm in length. There was no significant difference in the number of sensilla basiconica found on male and female antennae. Up to three may be found on any sub-segment although the usual number was one or two. Although these sensilla were very similar to the surrounding microtrichia they could be distinguished by the fact that they possessed a socket (Figs.3, 6).

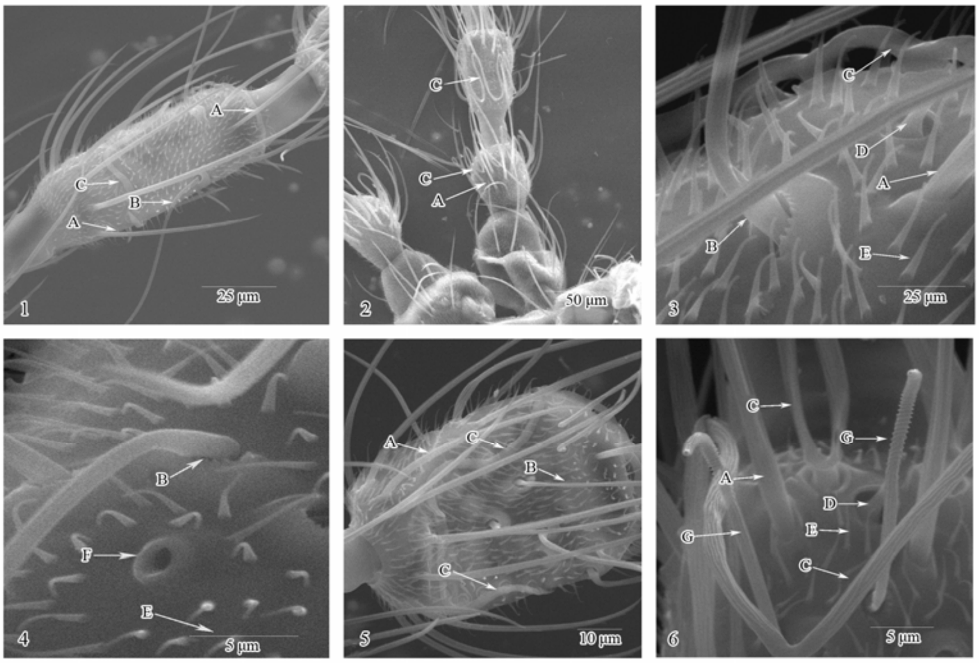
2.2.4 Sensilla cavity. These Sensilla were present on antennae of male and females. There were significantly more sensilla chaetica on male antennae than on female. Numbers of sensilla from one to ten sub-segment flagellum ranged from 10–20 for the male and 0–3 female antennae respectively. The diameter of it was approximately 1.2 μm (Fig.4).

2.2.5 Sensilla styloid. The sensilla styloid was found on the second sub-segment flagellum of the male antenna. This was recorded for the first time on the antenna of males of Diptera. The length of the sensilla was about 21 μm and the diameter was 1.5 μm. This was not found on female antenna. Its surface was not smooth; it was pore-less with transverse lines and only a pore on the terminal (Fig. 6).

2.2.6 Circumfila. Each sub-segment of the flagellum was encircled by looped structures. The loops of the circumfila were attached to the antennal surface by a series of stalks situated within inverted sockets (Figs.1, 2, 3, 5, 6). Each stalk was split into two approximately 2–3 μm above the antennal surface where it joined its two neighboring stalks to form a continuous looped structure. The male circumfila were more complex than the female. Each female sub-segment had 1–2 loops of circumfila running around its circumference. Each male circumfilum consisted of one loop with a band running around the proximal circumference of each sub-segment and each sub-segment of the flagellum had three circumfila (Fig.2). The female had more stalks per sub-segment than the male antenna.

3 Discussion

The present observations confirm that *Aphidoletes aphidimyza* has a similar array of sense organs to two other cecidomyiid species, *Contarinia sorghicola* (Slifer & Sekhon, 1971) and *Mycodiplosis erysiphes* (Solinas &



Figs. 1-6 Scanning electron micrographs about the antenna sensilla of *Aphidoletes aphidimyza*

(1, 3-5: female; 2, 6: male) A: Sensilla trichoidea; B: Sensilla chaetica; C: Circumfila; D: Sensilla basiconica; E: Microtrichia; F: Sensilla cavity; G: Sensilla styloid.

Nuzzaci, 1987). The general function or functions of the sensilla are inferred from their ultra-structure, although verification will require behavioral and electrophysiolog ical studies.

On both male and female antennae the sensilla chaetica were the most prominent. They had the typical appearance of mechanoreceptors. The morphology of these sensilla closely resembled sensilla chaetica found on other Cecidomyiid species (Slifer & Sekhon, 1971; Solinas & Nuzzaci, 1987). The bulbous body at the hair shaft base, along with the ridged socket opening may act to limit the hair's range of movement. These sensilla might protect the more delicate underlying structures found on the antennae or may act to determine whether the antennae is in contact with a substrate, such as a leaf. The sensilla chaetica of the female were more numerous than on the antennae of the male. This suggests that the male may also utilize these sensilla to orientate to sound as was common in male mosquitoes which are tuned to the flight sound of the female (Slifer & Sekhon, 1971; McIver & Hudson, 1972).

Sensilla trichodea has been identified as the main olfactory sensilla for several insect species (Schneider, 1964; Zacharuk, 1980). Sensilla trichodea of *A. aphidimyza* made up approximately 25% and 23% of the total number of sensilla found on male and female antennae respectively. These sensilla closely resembled the sharp-tipped sensory hairs on both *C. sorghicola* (Slifer & Sekhon, 1971) and *M. erysiphes* (Solinas & Nuzzaci, 1987). Solinas & Nuzzaci (1987) recorded that there were slightly more sensilla trichodea on the female antennae than on the male, so they postulated that these olfactory sensilla played an important role in finding oviposition sites on plants and/or fungi. However, on ten of the thirteen antennal sub-segments on *A. aphidimyza* there were significantly more of these sensilla on male than on female antennae suggesting that they may be more important in the detection of a female releasing sex pheromone.

The sensilla basiconica of *A. aphidimyza* closely resembled the 'grooved pegs' of *M. erysiphes* (Solinas & Nuzzaci, 1987) in their morphology, both had grooved hair-shafts. Insects which possess 'grooved pegs' with a single pore at the tip are extremely rare (Toh, 1977). Pore channels had a low electron density similar to that of the cuticle of the peg, very narrow dimensions, and an often convoluted structure within the cuticle. This made them very difficult to see and they can therefore be easily overlooked (Slifer, 1970; Cribb & Jones, 1995).

Sensilla cavity present on both male and female antennae. Significantly more sensilla cavities were found on male than on female antennae. Maybe it is a receiver to receive sound waves.

Two sensilla styloids were found on the flagellum of male antennae. This is the first report of this characteristic on the antenna of male of Diptera. Though it was not found on female antenna, it revealed sexual dimorphism in the antennae of male and female *A. aphidimyza*. Its function is as yet not known; but possibly facilitates the selection of mate or as a sex pheromone receptor.

The circumfila of *A. aphidimyza* shared many features with those of other cecidomyiid species (Slifer & Sekhon, 1971; Solinas & Nuzzaci, 1987). In the two previous studies of cecidomyiid circumfila, the threadlike sensilla were seen to encircle the flagellomers quite differently between males and females. For example, in *C. sorghicola* (Slifer & Sekhon, 1971) the male circumfila consisted of a series of elongated loops, measuring up to 36 μm in height compared to the female circumfila which consisted of two circular loops surrounding each sub-segment, which were at a height of 2–3 μm above the antennal surface. Due to the fact that the circumfila had features commonly found in olfactory receptors and that the circumfila of the male had a much more elaborate structure than the female, Solinas & Nuzzaci (1987) suggested that the circumfila of *M. erysiphes* may be important as a sex pheromone receptor. The structure of *A. aphidimyza* circumfila upholds this premise as the male sensilla appeared to be more elaborate than the female. The circumfila of *A. aphidimyza* however, no doubt played an important olfactory role.

In conclusion, detailed descriptions of the sensillae of *A. aphidimyza* provided the basis for future electrophysiological studies of the sensilla involved in chemical ecology.

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杨增明　厦门大学生命学院
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