

**External Morphology of Antennae Sensilla on Silverleaf Whitefly, *Bemisia Tabaci*,
(Hemiptera: Aleyrodidae)**

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Abstract

The silverleaf whitefly, *Bemisia tabaci*, (Gennadius), is important pests to pest to a wide range of crops around the world. The external morphology and distribution of sensilla on the antennal in adult male and female *B. tabaci*, was studied using scanning electron microscopy (SEM). Five types of sensory organs in both sexes on the flagellum were identified, including sensilla microtrichia, sensilla basiconic, sensilla trichodea, sensilla chaetae, sensilla coeloconic and finger-like sensilla. Four coeloconic sensilla, were observed on the antenna sub-segments, two in the first sub-segment, one in the third sub-segment, and one in the fifth sub-segment. basiconic sensilla (BS) were present on the antenna sub-segments of both sexes as single BS in the first, fourth, and fifth sub-segments with deep lengthwise grooves. In contrast, chaetae sensilla were found exclusively on the pedicel in both sexes. Significant differences were found in the length of the antennae, whereas females have greater longer of the antennae than males. These results would be helpful for further studies on detailed in the olfactory response of *B. tabaci*.

Keywords: Whitefly; Antennae; Sensilla; Morphology .

الشكل الخارجي للاعضاء الحسية على قرون الاستشعار في الذبابة الفضية البيضاء ، بميسيا توباكو (نصفية الاجنحة : الدقيقيات)

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الملخص :

الذبابة الفضية البيضاء ، *Bemisia tabaci* ، (Gennadius)، هي آفات مهمة لمجموعة واسعة من المحاصيل في جميع أنحاء العالم. تمت دراسة الشكل الخارجي وتوزيع الاعضاء الحسية على قرون الاستشعار في كلا من الذكور والإناث البالغين من *B. tabaci* باستخدام الفحص المجهر الإلكتروني (SEM). تم التعرف على خمسة أنواع من الأعضاء الحسية في كلا الجنسين على قرن الاستشعار وهي الحسية الدقيقة ، والحسية الأساسية ، والتريكوديا ، والحسية الالهلاب ، والحسية المتجاورة والحسية الشبيهة بالإصبع. لوحظ وجود أربعة أحاسيس متجاورة على الأجزاء الفرعية لسوط ، اثنان في الجزء الفرعي الأول ، وواحد في الجزء الفرعي الثالث ، وواحد في الجزء الفرعي الخامس. كانت الحسية الأساسية موجودة في الأجزاء الفرعية لسوط لكلا الجنسين وواحدة في الأجزاء الفرعية الأولى والرابعة والخامسة ذات الأخاديد الطولية العميقة. في المقابل ، تم العثور على اهلابا

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حسية حصريًا على السوقة في كلا الجنسين. تم العثور على اختلافات كبيرة في طول الاسواط ، حيث أن الإناث لديها قرون الاستشعار أطول من الذكور. ستكون هذه النتائج مفيدة لمزيد من الدراسات حول الاستجابة الشمية لذبابة البيضاء

الكلمات الرئيسية: الذبابة البيضاء؛ قرون الاستشعار؛ الحسية. الشكل المورفولوجي



Introduction

The silverleaf, whitefly, *Bemisia tabaci*, (Gennadius), was investigated as important pest of many vegetable and ornamental plants around the world [1]. Both nymphs and adults feed on the sap of the phloem of hundreds of plant species, causing direct damages as staining, yellowing and leaf drop, reducing plant vigor, and indirect damage, due to the excretion of 'honeydew' favor development of the fungi, and the transmission of virus. However, Studies on how insects perceive chemical signals from host plant volatiles has revealed the importance of the insect's olfactory system in such as host-seeking behaviour, oviposition site selection, and predator avoidance. Furthermore, insect antennae have sensory receptors called sensilla [2], these sensilla types have been reported in a variety of insect [3,4], and described previously as olfactory receptor systems with different functions important to an insect's ability to detect and respond to its environmental by facilitating the detection of sensory stimuli [5].

The importance of olfactory cues involved in the behavior of *B. tabaci*, has increased in the recent years, and shown that whitefly preference is affected by plant volatile organic compounds (VOCs) emitted among host plant varieties [6,7]. Therefore, better understanding the insect antennae morphology, would contribute to the elucidation of a comprehensive model of olfactory systems [8], for orientation behaviour of the individuals, e.g., toward food sources or mates [9]. Moreover, several published studies describe the external morphology antennal sensilla of whitefly species *Bemisia tabaci* using electron microscopy techniques [10-12]. However, despite the crucial functions of antennae, little is known about their morphology, and distribution of antennal sensilla of *B. tabaci*. The current study aimed at investigating and describing the morphology, type, and distribution of sensilla on the antenna of male and female *B. tabaci*, using scanning electron microscopy (SEM). To provide a morphological basis for future behavioural and electrophysiological studies.

Materials and Methods

Preparation of Antenna for (SEM)

Newly emerged whiteflies adults were collected, and were sexed under a stereomicroscope (50X). Insects were placed in small vials before the start the experiment. For scanning

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electron microscopy (SEM), ten adult *B. tabaci* females and males of insects for each were first anaesthetized for 2 min in a freezer (4°C). The entire antenna was carefully cut off under a stereomicroscope, and the samples were fixed immediately for 24 h in 5% glutaraldehyde and then dehydrated in a graded alcohol series as follows: 30, 50, 70, 90 and 100%, in each case for 10 min each. Each individual antenna was immersed in 1–2 ml of hexametyldisilazane (HMDS) for 10 min. Then, HMDS was decanted from the specimen vials and was allowed to dry at room temperature. Dehydrated samples specimens were mounted on aluminium SEM stubs with double-sided sticky tapes to stabilize the antennae in one place for viewing in the SEM chamber and were coated with gold in a high-resolution sputter coater (Hitachi E- 1010), to be observed under SEM (JEOL – JSM 636, Japan). Micrographs were taken of the antennae; flagellar segments and distribution of the sensilla on the antennae for both sex's males and females, at electron microscope unit / National University of Malaysia.

Data Analysis

All antennal length, sensilla number, distribution for each antennae segment, were measured directly from the printed SEM images. Data on antenna measurements (segments) were compared between both sexes male and female and tested for significance ($P < 0.05$) using Student's *t*-test. The Minitab Statistical Package (v. 16), has been used to analyze all of the data.

Results

General description

Antennae in both sexes, were similar and consisted three segments, which were attached to the head via a cup-shaped basal scape (Sc), pedicel (Pd), with flagellum (Fl) with five flagellum sub-segments (S5), from the base to the tip respectively in both sexes (Fig.1). No significant differences were found in the scape length between sexes ($P < 0.05$). While, the pedicel in female shown to be significantly ($P < 0.05$), longer than male. Moreover, among the five flagellum sub-segments, (S5), there was no significant difference between males and females in the lengths, except (S1, S4), were significantly ($P < 0.05$) longer in female than in male. The length average of the whole antennae of female was significantly ($P < 0.05$) greater than male antenna (Table 1).

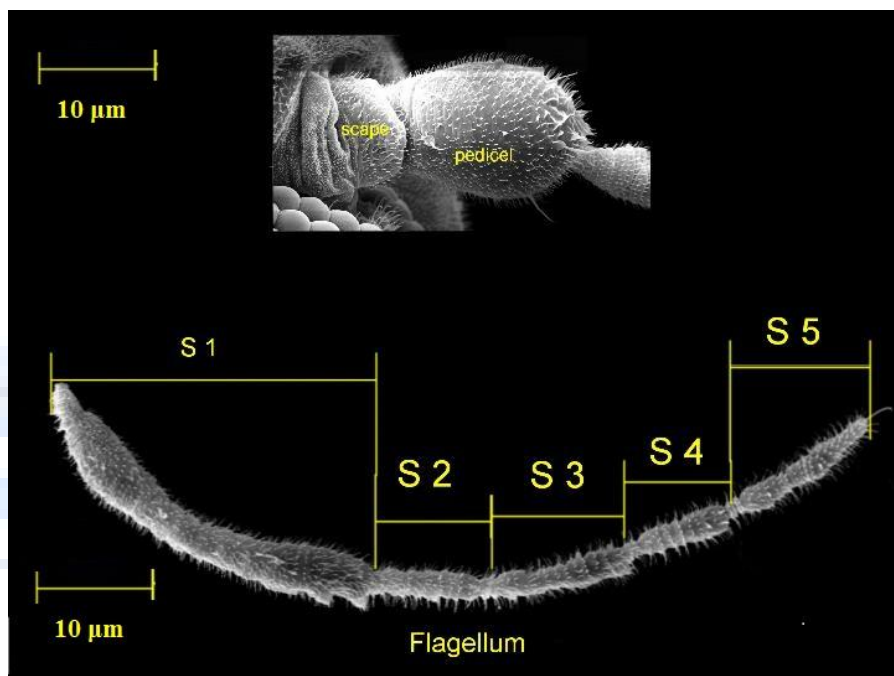


Fig. 1. SEM, General overview of antenna of *B. tabaci* female, showing the scape (Sc), pedicel, and flagellum formed by five sub-segments. The general shape of the antenna male and female are being similar.

Table.1 The length, in the antennae segments of females and males *B. tabaci* (Mean \pm SE).

| Section of Antenna | Female ($\mu\text{m} \pm \text{SE}$) | Male ($\mu\text{m} \pm \text{SE}$) |
|--------------------|--|--------------------------------------|
| Scape | 16.44 \pm 2.2a | 11.91 \pm 0.18a |
| Pedicel | 41.57 \pm 1.2a | 33.80 \pm 1.9b |
| S.1 | 96.50 \pm 1.6a | 90.09 \pm 3.0b |
| S.2 | 19.95 \pm 1.4a | 17.95 \pm 1.7a |
| S.3 | 30.93 \pm 0.98a | 31.31 \pm 1.7a |
| S.4 | 29.90 \pm 1.4a | 24.20 \pm 1.4b |
| S.5 | 37.42 \pm 3.0a | 34.92 \pm 2.2a |
| Total | 272.71 \pm 5.89a | 244.18 \pm 6.04b |

Means followed by the same lower-case letter on the lines are not statistically significant different

Distribution and Abundance of Antennal Sensilla

On both sexes antennae, there were six major types of sensilla recognized; microtrichia sensilla (MT), basiconic sensilla (BA), grooved surface trichodea sensilla (GT), chaetae sensilla (CH), coeloconic sensilla (CO), and finger-like sensilla (FS) (Fig 2,3). The dorsal and ventral surfaces of the scape, pedicel, and flagellum of the antennae of both sexes were found to be uniformly covered with the minute, hair-like microtrichia sensilla (MT). and seem to be most numerous type sensilla on the antennae. has small hair-like structure with smooth surface, with no socket at the basal part of each MT (Fig. 2A). On the ventral region of the flagellum sub-segments, three types of (BA) were observed as single BS in both sexes in the first, fourth, and fifth sub-segments. The peg was in the centre of the pit, but there were no spines on this form of sensilla. Furthermore, the sensilla's surface was flat, but there were small cuticular ridges near the base (Fig. 2A). In addition to basiconic sensilla (BA), there were only one grooved surface trichodea sensilla (GT), with a smooth cuticle, sat upright from sockets on the antenna surface, whereas were observed only on the dorsal region of the scape in both sexes of male and female (Fig. 2). There were differences in the abundance of the chaetae sensilla (CH) on the ventral and dorsal sides of the pedicel region antennal between the sexes, with 5 CH in the females and 8 in the males (Fig. 2B). Coeloconic sensilla (CO), were identified only on the ventral surface of the flagellum sub-segments 2 in the first, 1 in the third, and 1 in the fifth sub-segments flagellum in both sexes, had a flower-like shape with a peg in the centre, which was encircled by inward-facing spines that looked like petals (Table 2 and Fig. 2A). The surface of CO was with deep lengthwise grooves (Fig. 3) and finger-like sensilla (FS), this sensilla was found on the terminal of the fourth flagellum sub-segments as a small peg, with needle-like in both sexes (Fig. 4).

Table 2. The number of antennal sensilla in both sexes of *B. tabaci*.

| Type of sensilla | Microtrichia sensilla | Basiconic sensilla | Trichodea sensilla | Chaetae sensilla | Coeloconic sensilla | Finger-like sensilla |
|------------------|-----------------------|--------------------|--------------------|------------------|---------------------|----------------------|
| Male | More | 3 | 1 | 8 | 4 | 1 |
| Female | More | 3 | 1 | 5 | 4 | 1 |

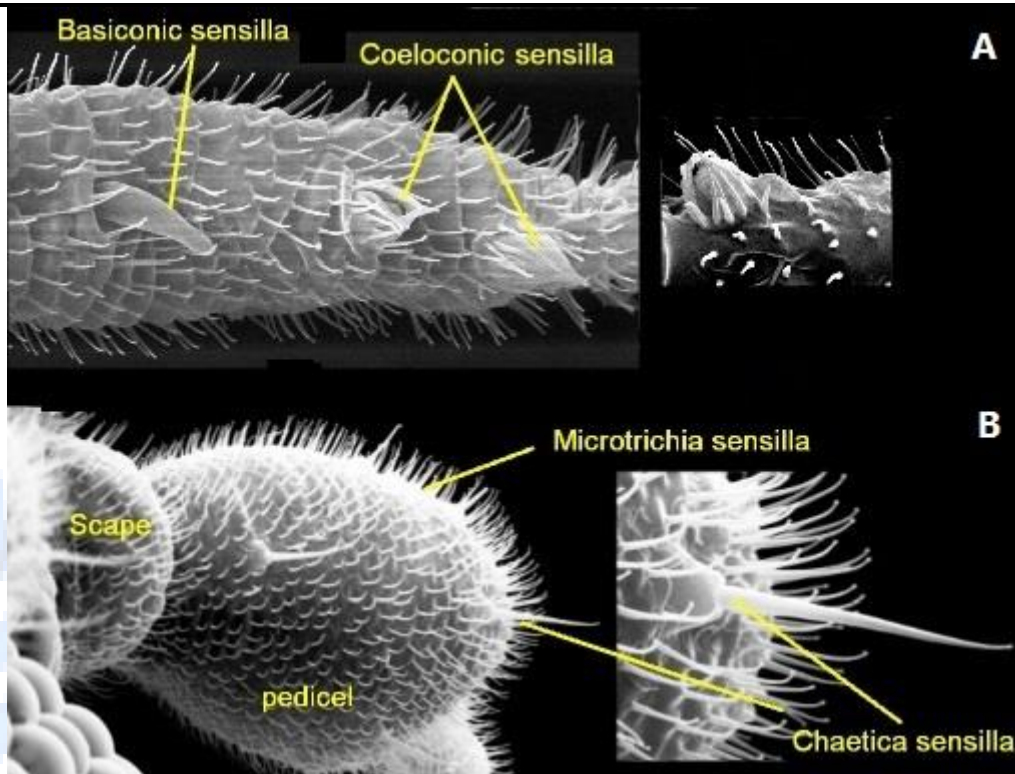


Fig. 2. Scanning Electron Microscopy, (A) basiconic sensilla and coeloconic sensilla. (B) Scape and pedicel with chaetica sensilla and covered with microtrichia sensilla. Scale bars: 1 μm in (a): 1 μm in (b)

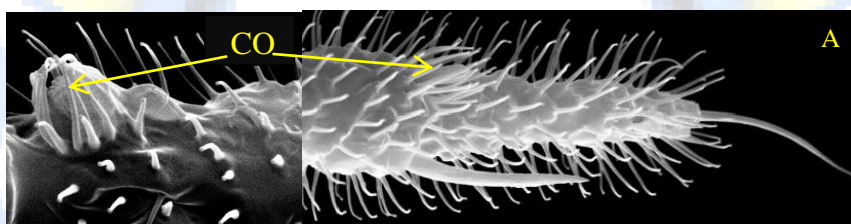


Fig. 3. SEM, (A) First and fourth sub-segment of the flagellum, showing Coeloconic sensilla (CO), with deep lengthwise grooves. Scale bars: 1 μm in (a): 1 μm in (b).

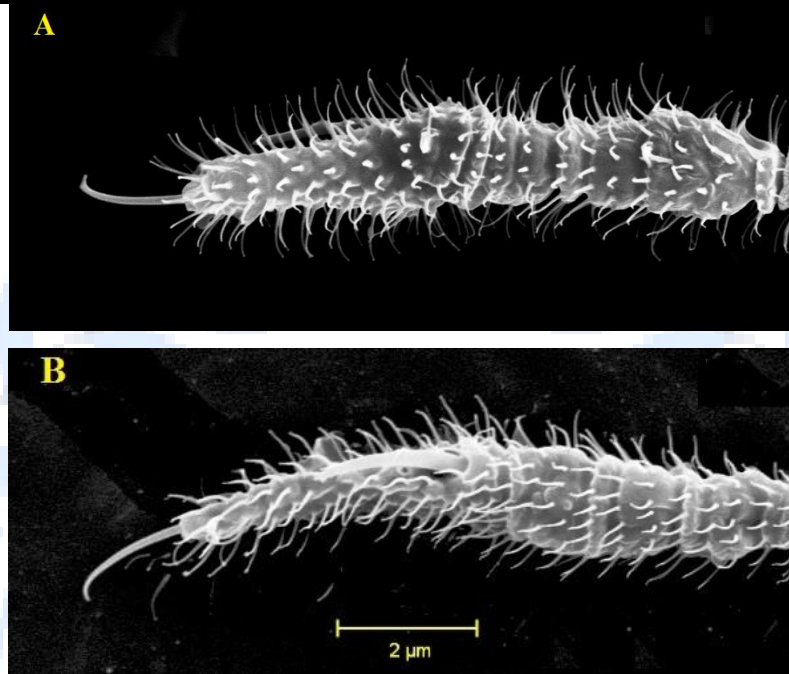


Fig. 4. SEM, Fifth sub-segment of the flagellum of adult male (A) and female (B), with terminal hair

Discussion

To our knowledge, the insect antenna is important sensory organs, they are involved in important role in the recognizing numerous stimuli for e.g., finding acceptable habitats and food as well as locating mates [13]. Sensory organs, basiconic, coeloconic, and chaetae sensilla on the insect's antenna are described as olfactory receptor systems for locating mates, hosts, habitats, and oviposition sites [14,15]. In the recent findings, despite the morphological similarities, of the general structure, and distribution patterns of sensilla were found in both sexes. and similar to other species of whitefly i.e., *Trialeurodes vaporariorum* and *Aleyrodes proletella* [16]. Our study revealed some degree of variability in the length of the antennae segment in females, which are significantly longer than in males. This difference was mainly due to subtle differences in the sizes of a few of the antennal segments. For instance, the size of the first and fourth flagellum (sub-segment) as well as the pedicle in female are significantly different from other in male, which results in a great increase in the length of their antennae compared to male antennae. The longer female antennae can be correlated, to their function by providing greater surface area to help translate into an increased sensitive

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attraction to plant volatiles, which detect many kinds of chemical stimuli, than male's antenna. Furthermore, females of *B. tabaci* have been shown to have a stronger reaction to plant volatile chemicals in olfactometer bioassays than males. [6, 17]. More work is necessary to support this hypothesis. In the current study morphological types of sensilla were identified into six types (Table 2). The types, abundance, and distribution of these sensilla are similar between male and female, except for the number of CH, which was classified into olfactory sensilla on the ventral surface of the pedicel region antennal 8 in the males and 5 females were present. In contrast, to our study by Zhang et al. (2015), only found 7 sensilla chaetica in males and 5 females on pedicel region in *B. tabaci* biotype (B and Q). Also, study by [18] et al. (1994), showed that only 5 sensilla chaetica were observed in on the pedicel, in both sexes. Some studies suggested that, the types, distribution, numbers, and function of antennal sensilla, might vary significantly among species even within the same genus. [18-20]. In addition, in both sexes, (CH) was most abundant on the ventral and dorsal sides of the pedicel area. The results differed from those of a previous study by [11] (1995), who discovered that (CH) are more frequent exclusively on the ventral side of the antennae in three whitefly species. several studies strongly indicate that these sensilla are mechanoreceptors [21-22]. However, microtrichia sensilla (MT), found in this study resembles those found in *Trialeurodes vaporariorum* and *Aleyrodes proletella* [11]. densely distributed on all regions of the scape, pedicel, and flagellum segments, surface of antennae in both sexes male and female.

The present results indicate that, four Coeloconic sensilla (CO), carried in a cup-like depression of the antennal wall, they are usually referred to as pitted pegs. 2 in first sub-segments, 1 in the third and 1 in the fifth sub-segments flagellum were are observed on both sexes, antennae. This result is consistent with that for three species of whitefly, whose antennae are covered with same number of CO [16]. For first report, our results show that the morphology of CO surface was deep lengthwise grooves on both sexes male and female. In contrast to our study, deep lengthwise grooves on Coeloconic sensilla (CO) are not reported in Mellor and Anderson, (1995). Based on their structure and the previous reports, (CO) may function as olfactory receptors for detecting vapour, CO₂ and plant odors. [23-26].

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In gender-specific olfactory receptor and responses to volatile compounds during host selection are relatively poorly studied. As a result, more research on the function of the sensilla is required, with transmission electron microscopy (TEM) in both male and female *B. tabaci*. This may aid in providing background information for our ongoing investigations on host localization processes in *B. tabaci*, such as olfaction behavioral studies.

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