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ORIGINAL ARTICLE

# Characterization of Antennal Sensilla of Male Adult Mulberry Silkmoth, *Bombyx mori* L.

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## ABSTRACT

The antennal sensilla plays a critical role in chemosensation, tactile perception, and environmental interaction, facilitating the modulation of essential behaviours during its life. The present study provides a comprehensive characterization of the antennal sensilla of the male adult mulberry silkmoth, *Bombyx mori* L. using scanning electron microscopy (SEM). The sensilla present on antenna of male adults were identified and classified as Böhm bristles and sensilla trichoidea are found on the scape and pedicel. The flagellum consists of sensilla chaetica (SCH), sensilla trichoidea (ST), sensilla trichoidea curvata (STC), sensilla basiconica (SB), sensilla squamiformia (SQU) and sensilla styloconica (SS). Sensilla trichoidea is further divided into sensilla trichoidea-I (ST-I) and sensilla trichoidea-II (ST-II). The flagellum contains SCH, ST-I, ST-II, STC, SB-I, SB-II, SQU, SS, SC and microtrichia in males. Morphological features, including size, shape, and distribution patterns have also been described.

**KEY WORDS:** Mulberry silkmoth,Antenna,*Bombyx mori*, Characterization, SEM, Sensilla

## INTRODUCTION

The *Bombyx mori* L., commonly known as the mulberry silkworm, is a model organism for studying insect physiology, especially in reproduction, behaviour, and sensory biology. The antennae of adult *B. mori* serve as the primary sensory organs, detecting chemical, mechanical, and environmental stimuli. These sensory inputs are crucial for behaviours like mate finding, oviposition site selection, and navigation toward host plants. The antennal sensilla are specialized sensory structures on the antennae, responsible for detecting these stimuli.

There are many commercial silkworm species, but *B. mori* is the most widely and intensively studied silkworm. The silk moth, *B. mori* is dioecious i.e., sexes are separate. The female moth is larger than the male moth. The adult moth is quite robust and in creamy white colour. Its body is divisible into three regions that are head, thorax, and abdomen. The head bears a pair of compound eyes, a pair of branched or feathery antenna and the mouth parts (Jolly *et al*., 1979).

Antennae are paired, highly mobile and segmented organs; located between or behind the compound eyes and mostly well-developed in adult stages. The antenna is fixed in a socket of the insect brain's cranium and its base is connected by an articulatory membrane which allows free movement of the antennae. The basal segment that is conspicuously larger than succeeding segments is called a scape, followed by another antennal segment called pedicel. The remaining annuli or flagellomeres are known as flagellum.

Antenna are highly evolved chemical sensing devices for detecting stimuli that have adaptive value so that most of their receptors are sensitive to a specific range of available stimuli and this sense has become further differentiated into two categories *i.e.,* chemo and mechano-receptors (Tousson & Gaaboub, 2004).

Typical modality-specific features of the cuticular apparatus and of the receptor cell endings in many cases allow for correct prediction of the functional modality of a sensillum type before electrophysiological recording can yield final confirmation (Hunger & Steinbrecht, 1998).

The adult antennal flagellum is divided into many small annuli and annulations are an important step in the

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metamorphosis of adult antenna. It is followed by the formation of adult organs (histogenesis) during pupal stage when a pair of large compound eyes, a pair of large antennae, fore and hind wings, legs, etc. become prominent (Snodgrass, 1935).

Scanning electron microscopic (SEM) studies on the antenna of male mulberry silk moths reveal the presence of various types of sensilla showing ultrastructural variation. The antennae are major sense organ that detects chemicals including food and pheromones (chemicals secreted into air by the opposite sex). They receive environmental information like smell, humidity changes, and variation in temperature, vibration, wind velocity and directionthrough a variety of sensillum receptors. The sensilla are cuticular special appendages for chemoreceptive, olfactory, hygrothermal-receptive and mechanoreceptive functions. Several types of sensilla including Böhm bristles, sensilla chaetica (SCH), sensilla trichoidea (ST), sensilla styloconica (SSC), sensilla basiconica (SB), and sensilla coeloconica (SSC) are known to have specialized functions.

This study aims to identify and elucidate the surface micro-morphology of antennal sensilla, which can provide insights into the sensory ecology of *B. mori* and may have applications in pest control strategies using pheromones and other sensory cues.

## MATERIALS AND METHODS

### Study Area and Collection

The disease-free layings (DFLs) were brought from NSSO, Bengaluru. The larvae were reared till the emergence of adults from silk cocoons in the laboratory at, Centre of Sericulture and Bioresource Management Research (CSBR), RTM Nagpur University, Nagpur. Male pupae were selected;and incubated and adult moths emerged from pupae used in subsequent experiments.After adult emergence from the pupae, male moths were sacrificed,and antenna were separated with anemphasis on understanding the sensillar morphology and their probable functions.

Table 1: Types of sensilla present on antenna of male adult moth, *B. mori*.

| | Male |
|---|---|
| Scape | Bohm's bristles<br>Sensilla trichoidea-I (ST-I)<br>Sensilla trichoidea-II(ST-II)<br>Sensilla basiconica (SB) |
| Pedicel | Bohm's bristles<br>Sensilla trichoidea |
| Flagellum | Microtrichia<br>Sensilla chaetica (SCH)<br>Sensilla trichoidea (ST)<br>Sensilla basiconica-I(SB-I)<br>Sensilla basiconica-II (SB-II)<br>Sensilla squamiformia (SQU)<br>Sensilla styloconica (SSC)<br>Sensilla coeloconica (SC) |

### Scanning Electron Microscopy (SEM)

Antennae of adult male silk moths, *B. mori* were examined using Scanning Electron Microscopy (SEM). The imputed antennae from male moths were washed thoroughly with distilled water, fixed in 10% formalin for of 12 h, dehydrated in various grades of alcohol, cleared in acetone, dried, and mounted on the metallic stub at different angles, processed for platinum coating for scanningusing Scanning Electron Microscope (JEOL (JSM 6380A) at Metallurgical & Materials Engineering Department, Visvesvaraya National Institute of Technology (VNIT), Nagpur.

## RESULTS

### Morphological Characterization of Antennal Sensilla

The male adult silkmoths show bipectinate (double comb like) antennal segments with long slender lateral processes on both sides (Fig. 1a). SEM analysis revealed several types of sensilla on the antennae of male *B. mori* adults:

1. **Böhm bristles (BB):** These are thread-like structures found on the scape and pedicel of antenna. They are long sharply tapered and had sclerotise ring at the basein male adults (Fig. 1b).
2. **Sensilla Trichoidea (ST):** Long, hair-like structures predominantly located on the dorsal surface of the antennae, ranging from 50 to 100 µm in length, these are the longest on the antennae and most likely involved in detecting volatile compounds. The sensilla trichoidea had been characterized by their length as longand short (ST-Iand ST-II) trichoid sensilla (Fig. 1c, 2f, 3b, 3d, and 3e).
3. **Sensilla TrichoideaCurvata (STC):** These are characterised as long, pointed shafts that are slightly curved towards the apex (Fig. 1d).
4. **Sensilla Chaetica (SCH):** These are the bluntspikes found at the middle region of each segment, and distributed on terminal end of the barbs. Based on length SCH was differentiated into long sensilla chaetica (SCH-I) and short sensilla chaetica (SCH-II). The surface of SCH covered with radial ridges (Fig. 3c).

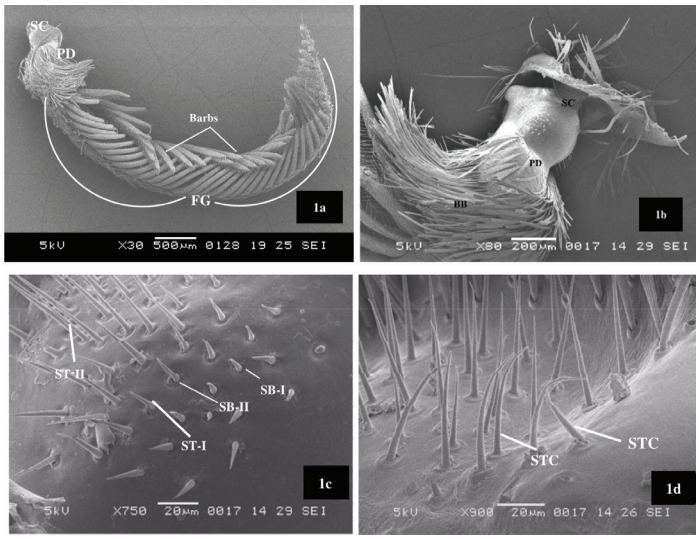


Fig. 1(a-d): Scanning electron microscopic(SEM) photomicrograph of antennae in adult male of *B. mori*; 1a: SEM of antenna showing scape (SC), pedicel (PD), flagellum (FG) and barbs; 1b: Posterior portion of antenna showing scape (SC), pedicel (PD) and Bohm's bristles (BB). 1c: Magnified photograph of scape showing senisilla trichoidea-I&II (ST-I&II) and sensilla basiconica-I&II (SB-I&II); Fig. 1d: Photomicrograph of scape showing sensilla trichoidea curvata (STC).

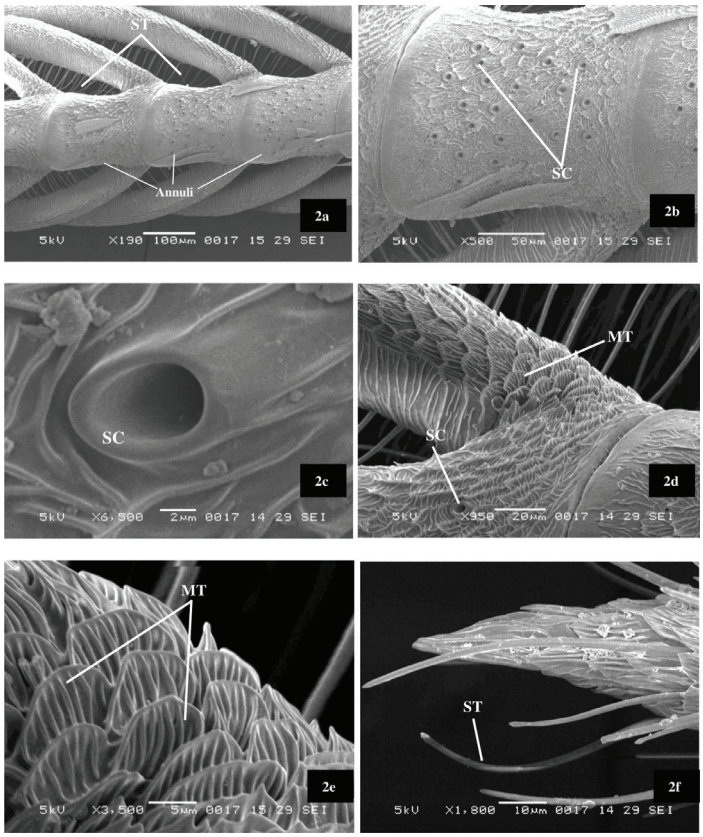


Fig. 2(a-f): Scanninge lectronmicroscopic(SEM)photomicrograph of antennae in adult male of *B.mori;* 2a: Mid annuli of flagellum showing various types of sensillae trichoidea (ST); 2b: Magnified view of annuli showing sensilla coeloconica (SC); 2c: Magnified view of one of the sensilla coeloconica (SC) on the annuli; 2d: Annuli of flagellum showing sensilla coeloconica (SC) and a barb with microtrichia (MT); 2e: Photomicrograph of a barb with microtrichia (MT); 2f: Tip of a barb showing sensilla trichoidea (ST).

5. **Sensilla Basiconica (SB):** Cone-shaped sensilla, observed in clusters on both the ventral and dorsal surfaces, approximately 10-15 µm in diameter at the base and 30-40 µm in length, implicated in detecting pheromones. The sensilla basiconica had been characterized based on their length as long and short (SB-I and SB-II) (Figure 1c).
6. **Sensilla coeloconica (SC):** Sensilla with a conical base and large central pore, found primarily on the distal segments of the antennae, typically 10-20 ìm in diameter, likely involved in detecting airborne chemicals. The most proximal segments of flagellum are devoid of them. The coeloconic sensilla of *B. mori* is double-walled, located in pits, and longitudinally grooved in their distal (Fig. 2b, 2c, 2d).
7. **Sensilla squamiformia (SQU):** Sensilla squamiformia are short, scale like. They were broad at the base and pointed towards the tipand abundantly distributed on the flagellum of the male moth (Fig. 3a).
8. **Sensilla styloconica:** The sensilla styloconica of *B. mori* are slender located in narrow grooves at the very tip of the antennal branches. They may be one, two or three on a single branch tip and located on a prominent stylus amidst the sensillum field of the sensilla trichodea and sensilla basiconica (Fig. 3f).

The distribution of these sensilla may differ between males and females, with males exhibiting a higher density of sensilla basiconica.

## DISCUSSION

The study highlights the sophisticated sensory system of *B. mori*. The identification of distinct sensilla types, each specialized for different sensory functions, supports the hypothesis that these structures mediate

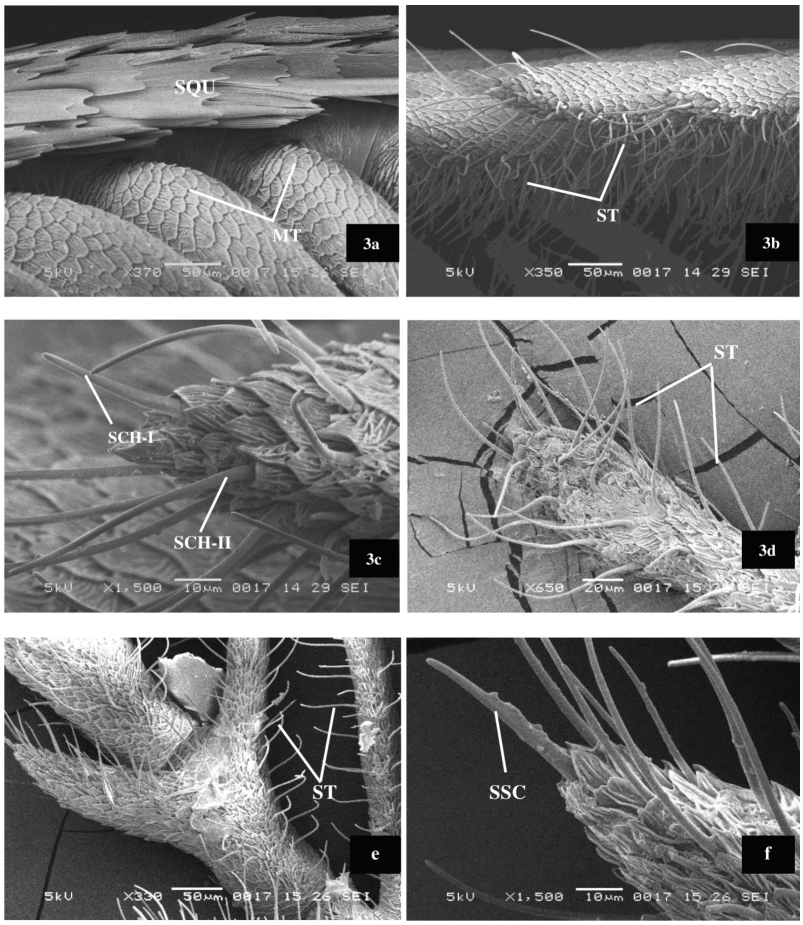


Fig. 3(a-f): Scanning electron microscopic(SEM) photomicrograph of antennae in adult male of *B.mori;* 3a: Flagellomere of flagellum showing sensilla squamiformia (SQU) and barb showing microtrichia (MT); 3b: Barbs of flagellum showing sensilla trichoidea (ST); 3c: Tip of a barb showing various types of sensillae chaetica-I&II; 3d: Photomicrograph of tip of flagellum showing sensilla trichoidea (ST); 3e: Early annuli of flagellum showing barbs having sensilla trichoidea (ST); 3f: Terminal end of flagellum showing sensillas tyloconica (SSC).

specific ecological and reproductive behaviours in the silkworm.

The Böhm bristles are spine-like sensilla and are present, on the antenna of *Trichoplusia ni, Heliothis zea, Prodenia ornithogalli* and *Spodoptera exigna.* Callahan *et al*. (1968) observed that, the Böhm bristles are dielectric wave guide aerials in the visible region and may also have a mechanoreceptive function in saturniid species. Schneider (1964) confirmed the mechanoreceptive properties of the Böhm bristles through electro-physiological studies in *Apis mellifera*.

The present SEM observations also confirmed the presence of leaf-like Böhm bristles on the scape and pedicel of male antenna of *B. mori* might be performing the functions of mechanoreceptors and chemoreceptors. The sensilla chaetica (SCH) are blunt striking spines found on the middle annuli as well as barbs end. A similar arrangement of the sensilla chaetica has been confirmed earlier by Albert & Seabrook (1973), the sensilla chaetica usually functions as mechanoreceptors and contact chemoreceptors., thus present study supported by the earlier observations by Barsagade *et al.* (2014), who observed that the antennae of both sexes of erisilk moth *Philosami aricini* are bipectinate type segmented consisting of scape, pedicel, and flagellum. Different types of sensilla are present on the scape, pedicel and flagellum functioning as a sensory perception. Böhm bristle and sensilla trichoidea are found on scape and pedicel.

Structurally, the body of ST-I is found to possess spiral ridges as observed by earlier workers on the male antenna of saturnid moths (Jefferson *et al*., 1970; Mayer *et al*., 1981; Eid *et al.,* 1990). Bland (1991) observed the trichoid sensilla present on the antenna of adult *Hyperapostica* has blunt tip. Similar observations were made by Barsagade *et al.* (2014) in eri silkworm *Philosamia ricini,* the flagellum consists of sensilla chaetica (SCH), sensilla trichoidea (ST), sensilla basiconica (SB) and sensilla squamiformia (SQU). Sensilla trichoidea is further divided into sensilla trichoidea-I (ST-I) and sensilla trichoidea-II (ST-II).

In the present study, similar types of sensilla have been observed inmale adult *B. mori.* It was also noted that trichoid sensilla II (ST-II) are shorter and more curved compared to the longer ST-I. The ST-I sensilla present on male adultsare sensitive to the sex pheromone released by female adults of *B. mori*, confirming their role in sex specificity, likely being involved in detecting the female sex pheromone during mate finding. Additionally, some sensory scales were also observed on the flagellum, as reported earlier by Loundon and Koehi (2000).

Schneider & Kaissling (1957) identified sensory scales on the antennae of *Bombyx mori*, which they termed sensilla squamiformia, and suggested that these structures function as receptors. Gaaboub *et al*. (2016) reported six different types of sensilla in the silk moth, *Bombyx mori* antennae: Olfactory sensilla trichodea, sensilla basiconica, sensilla. coeloconica, putative gustatory sensilla chaetica are multimodal receptors, sensilla styloconica, are thought to be thermo- and hygroreceptors and the mechano-sensory sensilla (squamiformia).

In the present investigation, similar types of sensillae have been observed on the antennae of the male silk moth of *B. mori*. These scale-like sensilla squamiformia are abundant on the antennae of male and are likely to be involved in sensory perception. Additionally, the sensilla basiconica on the antennae, which are more prominent in males, are believed to act as chemoreceptors, responding to chemical cues originating from mature female adults for opposite-sexfinding which leads to coupling and successful mating of both the sexes.

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## CONCLUSION

The antennal sensilla of adult *B. mori* are integral to the moth's ability to interact with its environment, from mate selection to host plant finding. In the present study, the antennae of male mulberry silk mothswere examined using scanning electron microscopy. It was observed that the antennae are bipectinate type segmented consisting of scape, pedicel, and flagellum, consisting of 38-40 annuli. Various types chemoreceptors and mechano-receptors of sensilla are present on the scape, pedicel, and flagellum. It may be sensitive to sex pheromone bombykol emitted by the female moth which helps in mate finding for mating.

## REFERENCES

Albert, P.J.& Seabrook, W.D. (1973). Morphology and histology of the antenna of the male eastern spruce budworm, *Choristoneura fumiferana* (Clem.) (Lepidoptera:Tortricidae) *Can. J. Zool.,* 51: 443-448.

Barsagade, D.D., Khurad, A.,Chamat, M., Meshram, H., Thakre, M., Gharade, S., Gathalkar & Thakre, R. (2014). Sex

Specificity of Antennal Sensilla in Eri Silkworm *Philosamia ricini. J. Entomol. Zool. Stud.*, 2(4): 206-211

Bland, R.G. (1991). Antennal and mouthpart sensilla of Tetrigidae (Orthoptera). *Ann. Entomol. Soc. Amr.,* 84: 195-200.

Callahan, P.S.,Taschenberg, E.F. & Carlysle T. (1968)The scape and pedicel dome sensora dielectric aerial waveguide on the antennae of night-flying moths. *Ann. Entomol. Soc. Amr*., 61(4): 934-937.

Eid, M., Salem, M.S. & El-Maasaraswy, S. (1990) Morphology and histology of antennae of eri silkworm moths, *Philosamia ricini* (Boisd.) (Lepidoptera: Saturniidae). *Ind. J. Seric*., 29(I): 13-23.

Faucheux, M.J. (1990). External structure of sensilla on the male and female flagellum of *Noctuapronuba* L. (Lepidoptera; Noctuidea). *Annaledela Societe Entomologiquede France*, 26: 173-184.

Gaaboub, I., Amira M. El-Shewy & Magda, A. & Salem (2016) Effect of feeding Silkworm (*B. mori*) on mulberry treated with bio and inorganic fertilizer on antennal sensilla. *J. Biosci. Appl. Res.,* 2(7): 440-451.

Hunger, T. & Steinbrecht, R. A. (1998) Functional morphology of a double-walled multiporous olfactory sensillum: the sensillum coeloconicum of *Bombyx mori* (Insecta, Lepidoptera). *Tissue & Cell,* 30(1): 14-29.

Jefferson, R.N., Rubin, R.E., Mofarland, S.U. & Shorey, H.S. (1970). Sex pheromones of noctuid moths. XXII – The external morphology of the antennae of *Trichoplusia ni, Heliothis zea, Prodenia ornithogalli* and *Spodoptera exigua. Ann. Entomol. Soc. Amr.,* 63(5): 1227-1238.

Jolly, M.S., Sen, S.K., Sonwalakar, T.N. & Prasad, G.K. (1979). Nonmulberry silks in Agricultural service. *Bull., F.A.O.United Nation*, 29: 1-178.

Loundon, C. & Koehi, M.A. (2000). Sniffing by a silkworm moth: wing fanning enhances air penetration through and pheromones interception by the antennae. *J. Experim. Biol.,* 203: 2977-2990.

Mayer, M.S., Mankin, R.W. & Carlysle, T.C. (1981) External antennal Morphometry of *Trichoplusia ni* (Hubner) (Lepidoptera: Noctuidae). *J. Insect Morphol and Embryol*., 10(3): 185-201.

Schneider, D. (1964). Insect antennae. *Ann. Rev. Entomol*., 9: 103-122.

Schneider, D. & Kaissling, K.E. (1957) Der Bau der Antennedes Seidenspinners *Bombyx mori* L. II-Sensillen, Cuticulare Bildungen und innerer Bau. *Ibid.,* 76: 223-250.

Snodgrass, R.E. (1935). The head appendages. In "Principles of Insect Morphology." Mcgraw-Hill Bookcomp, New York, 130-156.

Tousson, E. & Gaaboub, I., (2004). Neuroanatomical Relationships between Sensory Afferent Arborizations in the Locust Paraproctal Sensory Systems. *The Intern. Conf. Biol. Sci.,* 3(1): 594-612.