Morphological Studies on the Antennal Sensilla of the Cigarette Beetle, *Lasioderma serricorne* (F.) (Coleoptera: Anobiidae)

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In order to identify the pheromone sensitive antennal sensilla of the male cigarette beetle, distribution and frequency of sensilla on the antennal flagellum of both sexes were investigated by scanning electron microscopy. On the flagellar segments, *Sensillum basiconicum*, *S. styloconicum*, *S. capitulum*, and three types of bristles were found. Among these types of sensilla, *S. basiconicum* and *S. styloconicum* have structural features indicating them to be olfactory sensilla. *S. basiconicum* is abundant on the male antenna, three times more so than that on the female antenna. In contrast, no sexual dimorphism was observed in *S. styloconicum*. Furthermore the number of *S. styloconicum* is much fewer than *S. basiconicum*. These findings suggest that *S. basiconicum* on the male flagella is responsible for detecting the sex pheromone produced by the female.

Key words: *Lasioderma serricorne*, cigarette beetle, antennal sensilla, sex pheromone, sexual dimorphism

INTRODUCTION

The cigarette beetle, *Lasioderma serricorne* (F), prevails throughout the world and harms a broad variety of dry foodstuffs as well as cured tobacco leaves. This species is able to breed on dried tobacco leaves and is a major pest at cigarette factories. The existence of the volatile sex pheromone produced by female cigarette beetles has been reported by BURKHOLDER (1970). CHUMAN et al. (1985) have carried out a chemical study of this pheromone, and have identified the major pheromone component as (4*S*, 6*S*, 7*S*)-4,6-dimethyl-7-hydroxynonan-3-one (serricornin). Recently, MORI et al. (1986) have pointed out the inhibitory action of the (4*S*, 6*S*, 7*R*)-stereoisomer against the pheromonal activity of the serricornin. This inhibitor has essentially no sex pheromonal activity, but evoked a strong EAG response (MOCHIZUKI et al., 1984).

LEVINSON and LEVINSON (1987) have dealt with morphology of the male antennal sensilla in their biological study of the cigarette beetle's pheromone. However, there has been no comparative observation between male and female, nor investigation on the frequency and distribution of the flagellar sensilla. The purpose of this paper is to clarify the types and distribution of sensilla on the antennae of the cigarette beetle, and to identify the sensillum responsible for pheromone detection.

MATERIALS AND METHODS

Beetles at an age of 3–10 d after adult ecdysis were obtained from cultures that had been maintained for at least 100 successive generations at our institute.

In order to remove the cuticular wax on the surface of the sensilla, whole insects were sonicated in *n*-hexane for 30 s, and air-dried. Heads with antennae were excised from the body, mounted on specimen stubs with two-sided adhesive tape, and coated with gold in a JEOL FC-1100 ion sputter. Five antennae of both sexes were examined using a JEOL JSM-35CF scanning electron microscope at 15 kV. Flagellar segments were observed from all sides.

RESULTS

The configuration of antenna is almost identical for both sexes (Fig. 1). The antenna consists of a bean-shaped scape (segment #1), a small pedicel (segment #2) and nine flagellar segments (segment #3–#11). Entire antennal length is about 750 μm . As the scientific name of this species "*serricorne*" indicates, the outline of a whole antenna appears serrated due to the triangular-shaped segments (#4–#10). Segment #3, with an average axial length of 72 μm and a maximal width (below the apex) of 50 μm , is smaller than the remaining segments. Segment #4 is larger than segment #5–#10 and has an average axial length of 90 μm and a maximal width of about 73 μm . Segments #5–#10 are distinctly triangular. The axial lengths are from 65 to 75 μm and maximal widths are from 59 to 70 μm . The terminal segment (#11) is ellipsoid, with an axial length of about 100 μm , and has a maximal width about 45 μm . All segments of the flagellum have a narrow base ranging from 19 to 23 μm dia.

Only bristle-type sensilla of various lengths with sockets at their base were found

Fig. 1. Left antenna of the male (1a) and female (1b) cigarette beetle, lateral inner side. The antenna consists of scape (segment #1), pedicel (segment #2) and 9 flagellar segments (segment #3–#11). No sexual dimorphism is observed in the configuration of the antenna. Scale: 100 μm .

Fig. 2. a. *S. basiconicum*, Arrows indicate olfactory pores. Scale: 1 μ m. b. *S. styloconicum*. Scale: 1 μ m. c. *S. capitulum*. Scale: 1 μ m. d. Bristle type I. Scale: 10 μ m. e. Bristle type II. Scale: 1 μ m. f. Bristle type III. Scale: 1 μ m. g. A pit located close to bristle type I. Scale: 1 μ m. h. Sensilla fractured at their bases by over-sonication, *S. basiconicum* (above) and Bristle type II (below). Scale: 1 μ m.

on the scape and pedicel. These are considered to be mechanoreceptive sensilla.

Sensilla come out from the overlapping region of the scale-like cuticular structure on the surface of flagellar segments. Six distinctly-shaped sensilla were observed on the flagellar segments.

Sensillum basiconicum

S. basiconicum length varies from 4.5 to 6.8 μm , and from 1.5 to 2.1 μm in base diameter. Fine pores (about 20 per μm^2 density) occur on the surface of the shaft (Fig. 2 a). The shaft of this sensillum seems to be made of single-walled cuticle (Fig. 2 h). *S. basiconicum* is abundant on the male antenna and 50–65 of the sensilla are distributed over the distal and lateral regions of a segment (Fig. 3 a; Fig. 4). On the other hand, only 10–18 *S. basiconica* have been observed on a female segment and the distribution is restricted to the distal region of segments (Fig. 3 b; Fig. 4).

The angle between the distal axis and *S. basiconicum* is less than 30° , and the high concentration on the distal region seems to enhance the antennal directivity toward the distal axis.

Sensillum styloconicum

This sensillum varies in length from 4.1 to 4.3 μm , and from 1.0 to 1.1 μm in base diameter (Fig. 2 b). It is characterized by deep longitudinal grooves from the central part to the tip of the shaft. The number of *S. styloconicum* per segment is 3–5 from segment #4 to #10, and 7–8 on the terminal segment. The distribution is restricted to the distal region of segments and no quantitative difference is visible between the male and female *S. styloconica*.

Sensillum capitulum

This type of sensillum is surrounded by a ring of raised cuticle (4.0–4.5 μm dia.), and has a non-perforated smooth surface shaft 2.3–2.6 μm axial length and 1.0–1.1 μm basal width (Fig. 2 c). A small pore is often observed near the base of the shaft. *S. capitulum* is rare; only one sensillum is observed at the distal portion of a segment, except for the terminal segment where 2–4 *S. capitula* are found.

Bristle type I

Bristle type I is prominent because of a long shaft with a slightly grooved surface. The length is up to 52 μm , and the diameter is 1.5–1.8 μm at the base (Fig. 2 d). The shaft without a full pore system is inserted in a flexible cuticular socket. There is a terminal pore opening at the tip of the sensillum.

Bristle type II

This type of sensillum is the most numerous on the flagellar segments. The shaft is set in a socket and tapers to a point (Fig. 2 e). The range of axial lengths and basal widths are from 15 to 27 μm and from 0.9 to 1.2 μm , respectively. No pores can be observed on its surface or tip. *Bristle type II* is distributed throughout segments but is rare in their distal regions.

Bristle type III

This type of sensillum is abundant on the lateral area of the female segments. The length vary from 5.5 to 6.1 μm , and the diameter is 1.0–1.3 μm at the base. Basal sockets are absent (Fig. 2 f). No pore system can be observed on the smooth surface of the shaft or tip.

In addition to the above sensilla, some pits are observed (Fig. 2 g). Pits are 1.4–1.8 μm in diameter, positioned on the marginal area of segments close to *Bristle type I*.

Fig. 3. Lateral surface (inner side) of segment #7; the male (3a) and the female (3b). Scales: 10 μ m.

Fig. 4. Typical distributions of the sensilla on segment #6; the male (left) and the female (right).

Most of them have two or three smaller pits inside, but sensilla are absent. In this paper, we do not venture to classify the pits as sensilla.

DISCUSSION

Which type of sensilla is responsible for sex pheromone detection?

Possible functions of each sensillum can be discussed according to external shape and distribution on a segment. The structural features and possible functions of the flagellar sensilla are summarized in Table 1. According to LEVINSON and LEVINSON (1987), flagellar sensilla on the male antenna are classified into six types based on external criteria. In their classification, *S. basiconica* on the male antenna were sub-classified into long and short types. But in our measurements, when the elevation angle of the sensilla was compensated for by length, *S. basiconicum* could not be divided into two separate types.

Sensilla that are supposed to have olfactory function are *S. basiconicum* and *S. styloconicum*. A male antenna generated 3–4 mV of maximal EAG responses to the sex pheromone, while it responded only weakly to general odors, such as dried tobacco leaf extracts and some kinds of essential oils derived from plants (CHUMAN et al., 1982; OKADA, unpublished data). These findings suggest that the antenna of the male

Table 1. Structural features and possible functions of the sensilla on cigarette beetle antennae

	Length (μm)	Width at base ^a (μm)	Pore system	Basal socket	Possible function
<i>S. basiconicum</i>	4.5–6.8	1.5–2.1	Entire surface		Olfactory
<i>S. styloconicum</i>	4.1–4.3	1.0–1.1	?		Olfactory, Auditory?
<i>S. capitulum</i>	2.3–2.6	1.0–1.1	No pore		Hygro-, and thermo- reception
Bristle type I	30–52	1.5–1.8	Tip of the shaft	+	Contact chemo- reception
Bristle type II	15–27	0.9–1.2	No pore	+	Mechanoreception
Bristle type III	5.5–6.1	1.0–1.3	No pore		?

^a Not including the cuticular socket.

Fig. 5. Frequency of sensilla(s) on single flagellar segments of *L. serricorne*.

cigarette beetle is highly specialized for detecting the sex pheromone produced by the female, and most of its olfactory sensilla are tuned for the sex pheromone. Frequency of each type of sensillum is shown in Fig. 5. The number of *S. styloconica* on a single male antenna is only 30–40, and seems to be insufficient to evoke a strong EAG response to the sex pheromone. On the other hand, the number of *S. basiconica* per male antenna is about 700. In the sexual communication system of this species, as well as that of most other insects, the females produce a volatile sex pheromone and attract conspecific males. EAG response of the female was very weak to serricornin (OKADA, unpublished data) and the number of *S. basiconica* on the female antenna is much lower than on the male. In this context, it seems reasonable to consider that *S. basiconica* on the male antenna are responsible for the sex pheromone detection.

The possible functions of other sensilla and adequate stimuli

Bristle type I, with a long shaft and a single pore at the tip, has typical features of the contact chemoreceptive sensillum. The long shaft makes it easy for its tip to come in contact with stimulants. Usually eight *Bristle type I*s occur from the marginal area of a segment, and each bristle seems to cover stimuli from a different direction (Fig. 4). The females of the cigarette beetle produce and apply oviposition deterrents, i.e. α - and β -serricornone, so as to avoid overlapping oviposition (IMAI et al., 1990). This type of sensillum on the female antenna may be responsible for detecting these oviposition deterrents in addition to the ordinary gustatory stimuli such as sugar, salt, water etc. The flexible socket at the base of this sensillum suggests *Bristle type I* also has mechano-sensitive function.

Bristle type II is a proprioceptive or an exteroceptive mechanoreceptor judging from the fact that its shaft, lacking a pore system, is inserted into the cuticular socket. This sensillum is the most abundant on the antenna and has a relatively long shaft. *Bristle type II*s may protect *S. basiconica* and *S. styloconica* scattered between them from physical damage when the antenna contacts an object.

Bristle type III has no basal cuticular socket, and is distinctive from *Bristle type I* and *type II*, and rather resembles *S. basiconicum* in this regard. *Bristle type III* is abundant on the female antenna where *S. basiconicum* is infrequent (Fig. 4). The distribution pattern of *Bristle type III* on the lateral area of the female segments corresponds to that of *S. basiconicum* on the male. Therefore, *Bristle type III*s on the female segments seem to be homologous to *S. basiconica* on the lateral area of the male segments. The function of this sensillum could not be inferred from only its external structure.

Structural features and the small number of *S. capitulum* are similar to the American cockroach's *S. capitulum* that has been identified to be hygro- and thermoreceptive sensillum by electrophysiological studies (YOKOHARI and TATEDA, 1976). *S. capitula* of the cigarette beetle are located on the distal area of a segment, and surrounded by other types of sensilla that seem to prevent them from becoming wet. *S. capitulum* of this species also seem to be a hygro- and thermoreceptive sensillum.

S. styloconicum is generally assumed to be mechano- or chemosensitive (ZACHARUK, 1985). *S. styloconica* of this species are distributed on the distal area of a segment and are surrounded by *Bristle type II* and *type I* that prevent its direct contact with chemical substances. This sensillum, therefore, may be responsible for olfactory or mechanical (vibratory and auditory) reception, rather than contact chemoreception.

The possible functions of the sensilla discussed in this paper should be confirmed

by electrophysiological studies, and the types of sensilla may be subdivided according to the spectrum of response. Electrophysiological studies are now in progress.

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