

Morphological Investigations of the Antennal Sensilla of the Banana Corm Borer (Coleoptera: Curculionidae)¹

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J. Entomol. Sci. 52(4): 379–386 (October 2017)

Abstract The antennal morphology and sensilla of the banana corm borer, *Cosmopolites sordidus* Germar (Coleoptera: Curculionidae), were examined. The antennae of *C. sordidus* were composed of eight antennomeres, a scape, a pedicel, and a flagellum composed of six flagellomeres. The use of scanning electron microscopy facilitated the recognition of five different sensilla types on the antennae of *C. sordidus*, including sensilla trichodea, sensilla chaetica type 1 and 2, sensilla basiconica, and sensilla furcata; cuticular plates were also found on the antennae of both males and females. These results provide necessary background information for our ongoing study on electrophysiology and chemical ecology of *C. sordidus*.

Key Words *Cosmopolites sordidus*, antennal club, scanning electron microscopy, sensilla, ultrastructure

The banana weevils (Coleoptera: Curculionidae) are recognized worldwide as the major insect pests of banana and plantain, and *Cosmopolites sordidus* Germar, the banana corm borer, mainly feeds on the stem base and corm in the field and seriously affects banana growth, which can cause yield losses up to 100% through sucker death, toppling, and snapping (Koppenhöfer et al. 1994, McIntyre et al. 2001, Rukazambuga et al. 1998). *Cosmopolites sordidus* is one of the most destructive pests of bananas in China (Yin et al. 2015). A male-produced aggregation pheromone attracts both males and females (Budenberg et al. 1993), and olfactory attractants seem to be promising for their management (Lopes et al. 2014).

A number of morphological studies have been conducted on the antennae of various Coleoptera, including curculionids *Hylobius abietis* L. (Mustaparta 1973), *Hypera meles* F. (Smith et al. 1976), *Odoiporus longicollis* Olivier (Gao et al. 2011), and *Polytus mellerborgi* Boheman (Yin et al. 2016). However, until now, no detailed reports on the antennal structure of *C. sordidus* have been published. The aim of this study was to describe morphological types, distributions, and sexual dimorphism of antennal sensilla in *C. sordidus* by scanning electron microscopy

¹Received 23 January 2017; accepted for publication 14 May 2017.

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(SEM). The results could provide basic information for developing control methods of the species based on its olfactory and gustatory systems.

Materials and Methods

Adult males and females of *C. sordidus* were obtained in Changjiang (N19°32', E108°95'), Hainan Province, China. For SEM observation, the antennae were rinsed in 70% ethanol in an ultrasonic cleaning instrument (Jieli Co. Ltd., Shanghai City, China) for 1 min and then dehydrated in a graded ethanol series (80, 90, 95, and 100%). After air drying, the specimens were mounted on a holder using electric adhesive tape and then coated with gold and observed using a Hitachi S-3000N SEM (Hitachi Corp., Tokyo, Japan). A total of 10 antennae per sex were studied under SEM.

The terminology and classification of sensilla used here followed those of Schneider (1964) and Keil (1997). The length and width of different types of sensilla were measured using Nano Measurer 1.2 software (Department of Chemistry, Fudan University, Shanghai, China). The data were analyzed using SPSS 19.0 for Windows (SPSS Inc., Chicago, USA). Differences in the length and width between the same sensilla of the males and females were determined using a *t* test. Statistical results were expressed as means $\pm$ SD.

Results

Cosmopolites sordidus antennae are geniculate, including a long scape, a pedicel, and a flagellum composed of six flagellomeres. The last flagellomere was termed the antennal club. General structure of the antennae of both sexes was similar. Most sensilla were located on the flagellum, especially on the antennal club. Five types of sensilla—sensilla trichodea (St), sensilla chaetica (Sch) type 1 and 2, sensilla basiconica (Sb), and sensilla furcata (Sf)—and cuticular plates were found on the antennae of both males and females.

St were the most abundant type of sensilla observed and were located exclusively on the antennal club in both males and females. St were identified based on their length and shape, with a conical base and slightly curved tip. St of both sexes were similar in shape (Figs. 1, 2). These sensilla varied from 25.8 to 33.0 μm in length, with a diameter of the hair base ranging from 0.9 to 1.5 μm . The length of St of males ($31.3 \pm 1.7 \mu\text{m}$) was greater than that of females ($28.4 \pm 2.6 \mu\text{m}$) (Table 1).

Sch were the longest on the antennae and were divided into two subtypes. Sch1 were long, robust, bristle-like sensilla with a sharp tip and sit almost perpendicular with respect to the antennal surface (Figs. 1, 2). Significant differences ($t = 5.595$, $P = 0.001$) in lengths between males and females were observed (Table 1). Sch2 were also long, robust, bristle-like sensilla, but extended from the antenna at roughly a 20° angle and had shallow longitudinal veins on the bristle shaft and closely apposed cuticular finger-like projections at their tips (Figs. 1, 2). Sch2 were distributed regularly along the lateral side of the flagellomeres.

Sb were common and distributed randomly on the antennal club (Figs. 1, 2). In adult males and females, Sb were blunt-tipped and shorter than St, and the length of Sb in the females was similar to that found in the males (Table 1).

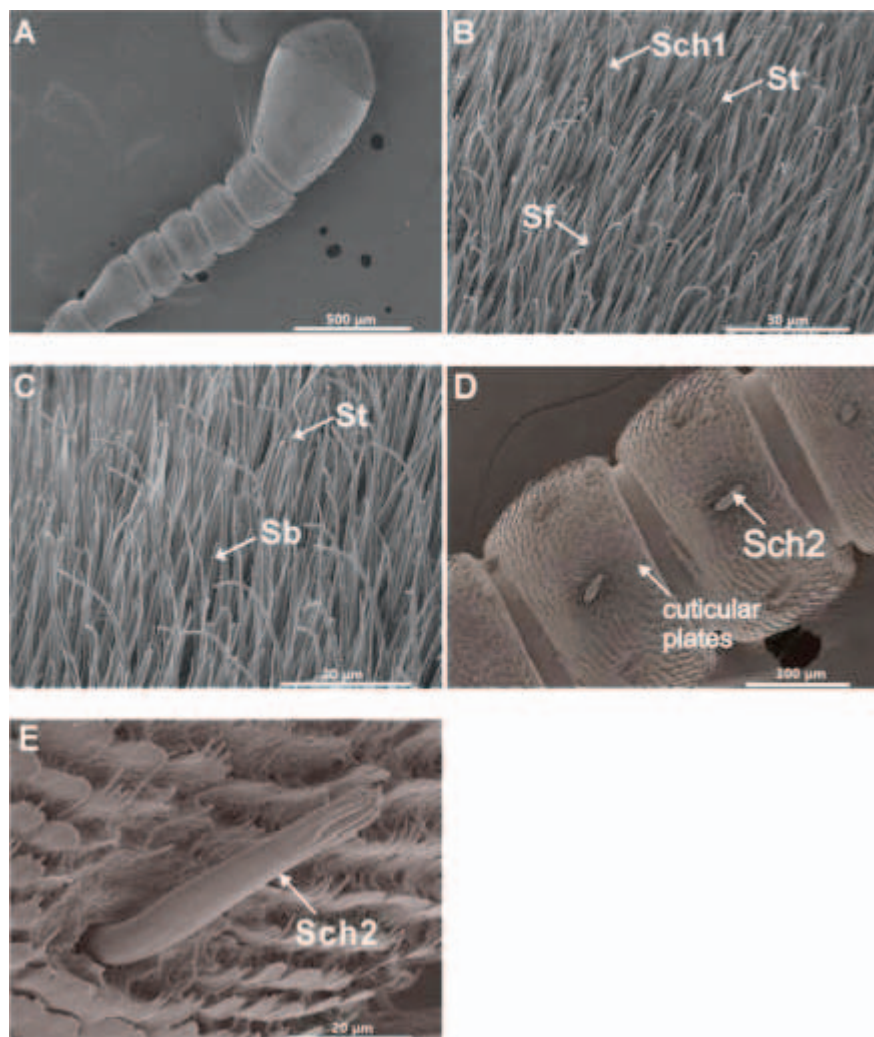


Fig. 1. Scanning electron microscopy micrographs of features on the antennal flagellum of male *Cosmopolites sordidus*. (A) antennal flagellum; (B) sensilla chaetica type 1 (Sch1), sensilla trichodea (St), and sensilla furcata (Sf) on the antennal club; (C) sensilla trichodea (St) and sensilla basiconica (Sb); (D) sensilla chaetica type 2 (Sch2) and cuticular plates covered whole flagellum; (E) sensilla chaetica type 2 (Sch2) on other flagellomeres.

Sf had a strong base and furcation at the tip, and appeared similar in profile to Sb. Sf ranged from 24.5 to 34.9 μm in length with a diameter at the base of 1.9 to 2.7 μm . Female Sf measured $27.7 \pm 1.8 \mu\text{m}$ in length while male Sf measured $29.7 \pm 5.2 \mu\text{m}$ (Table 1).

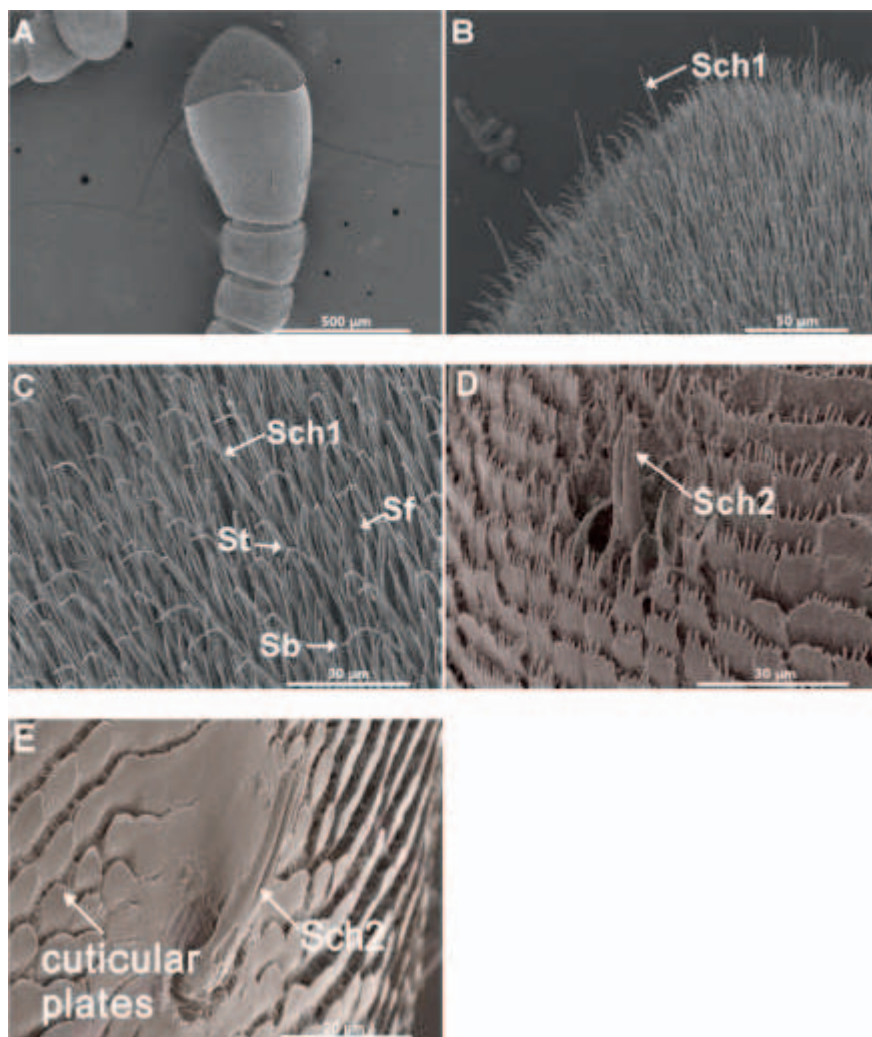


Fig. 2. Scanning electron microscopy micrographs of features on the antennal flagellum of female *Cosmopolites sordidus*. (A) A part of antennal flagellum; (B) sensilla chaetica type 1 (Sch1) on the antennal club; (C) sensilla chaetica type 1 (Sch1), sensilla trichodea (St), sensilla basiconica (Sb), and sensilla furcata (Sf) on the antennal club; (D) sensilla chaetica type 2 (Sch2) on other flagellomeres; (E) sensilla chaetica type 2 (Sch2) and cuticular plates covered whole flagellum.

Discussion

This study demonstrated that both sexes of *C. sordidus* contained five types of sensilla on the antennal flagellum; however, no significant sexual differences were

Table 1. Measurements (mean $\pm$ SD) of sensilla on the antennal flagellum of *Cosmopolites sordidus*.

Sensilla Type	Sex	Length (μm)	Width (μm)
Sensilla trichodea	♂	31.3 $\pm$ 1.7	1.3 $\pm$ 0.2
	♀	28.4 $\pm$ 2.6	1.0 $\pm$ 0.1
Sensilla chaetica 1	♂	42.8 $\pm$ 5.9a*	1.9 $\pm$ 0.1
	♀	59.2 $\pm$ 2.7b	2.4 $\pm$ 0.2
Sensilla chaetica 2	♂	29.3 $\pm$ 3.3	6.5 $\pm$ 0.7
	♀	37.3 $\pm$ 8.0	7.4 $\pm$ 1.2
Sensilla basiconica	♂	26.3 $\pm$ 2.9	1.7 $\pm$ 0.0
	♀	26.4 $\pm$ 3.6	1.4 $\pm$ 0.1
Sensilla furcata	♂	29.7 $\pm$ 5.2	2.6 $\pm$ 0.1
	♀	27.7 $\pm$ 1.8	2.1 $\pm$ 0.2

* Different lowercase letters indicate significant differences between the lengths of the same types of sensilla in males and females ($P < 0.01$).

observed in their morphological features except for the length of Sch1. These findings are in agreement with the observation that both males and females are attracted to male-produced aggregation pheromone (Budenberg et al. 1993). The antennal club of *C. sordidus*, composed of only one flagellomere, differs from that in other Curculionidae. For example, in *H. meles*, the antennal club is composed of four flagellomeres separated by three constriction bands (Smith et al. 1976). In *Eucryptorrhynchus chinensis* (Olivier) and *Eucryptorrhynchus brandti* (Harold), the antennal club is formed by three flagellomeres (Yu et al. 2013).

St are usually the most numerous sensory structures on coleopteran antennae and are presumed to function as olfactory receptors in *Agriotes obscurus* L. (Merivee et al. 1997). Sch and Sb are the most numerous sensory organs in Elateridae (Merivee et al. 1998, 1999) and Carabidae (Merivee et al. 2000, 2001, 2002). Most of them function as mechanoreceptors and olfactory receptors, respectively. External shape, large measurements, specific location at the antennal apex, small number, and almost perpendicular position on the antennal surface indicate that the Sch1 are most probably taste sensilla, which occur on the antennae of all beetles studied. They have been found on the antennae of Carabidae (Merivee et al. 2000, 2001, 2002), Elateridae (Merivee et al. 1997, 1998, 1999), and many other beetles. By electrophysiological recordings, it has been demonstrated that these sensilla are innervated by five neurons: four chemoreceptor neurons and one mechanoreceptor neuron. Similar sets of neurons are typical for insect taste sensilla. In Carabidae, one of the neurons is sensitive to water-soluble salts (Merivee et al. 2004), the second neuron responds to pH (Merivee et al. 2005, Milius et al. 2006), the third neuron responds to plant sugars and amino acids (Merivee et al. 2007, 2008, 2012), and the fourth neuron is

sensitive to plant secondary compounds such as alkaloids and glucosides (Milius et al. 2011). The fifth mechanoreceptor neuron responds to touch (Merivee et al. 2008). In Elateridae, three neurons have been electrophysiologically identified in these taste sensilla: salt, sugar, and touch-sensitive neurons (Tooming et al. 2012). Sb have been reported on the antennae of many coleopteran species (Alm and Hall 1986; Bartlet et al. 1999; Bland 1981; Dai and Honda 1990; Daly and Ryan 1979; Dyer and Seabrook 1975; Jourdan et al. 1995; Merivee et al. 1998, 1999, 2000, 2002; Okada et al. 1993; Ritcey and Mciver 1990) and have been shown to function as olfactory receptors responding to plant odors in *H. abietis* (Mustaparta 1975) and *Phoracantha semipunctata* F. (Lopes et al. 2002). Sf are rare on the antennae of Coleoptera. Wang et al. (2012) assumed that the sensillum furcatea is a morphological subtype of sensillum trichodea and may have the same olfactory function. In addition, sensilla coeloconica, sensilla styloconica, and sensilla placodea, commonly found on insect antennae (Okada et al. 1992, Yan et al. 2011, Zacharuk 1980), are absent on the antennae of *C. sordidus*.

In conclusion, our study demonstrates importance of olfaction and gustation in the behavioral ecology of *C. sordidus*. The study serves as a prerequisite for further electrophysiological studies on the antennal chemoreceptors of the species including chaetoid gustatory sensilla, which are crucial in final stages of host plant detection and selection.

Acknowledgments

This study was financially supported by National Key Technology Research and Development Program of the Ministry of Science and Technology of China (2007BAD48B05-01) and National Nonprofit Institute Research Grant of CATAS-EPPI (2008hzs1J008).

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