ULTRASTRUCTURAL STUDIES ON THE ANTENNA OF DIPTERA OF FORENSIC IMPORTANCE

Mustafa Cemal Darılmaz*, Dilek Karataş* and Hafize Taşatan*

* Department of Biology, Faculty of Science and Letters, Aksaray University, Aksaray, TURKEY. E-mail: mdarilmaz@yahoo.com

[Darılmaz, M. C., Karataş, D. & Taşatan, H. 2019. Ultrastructural studies on the antenna of Diptera of forensic importance. Munis Entomology & Zoology, 14 (1): 197-205**]**

ABSTRACT: Insects play an important role in the decomposition of corpses, and this role of insects helps to elucidate criminal events. Forensic entomology requires that the species identification should be executed without error. The most important insect group used in estimate the postmortem interval is Diptera. By using scanning electron microscope, the external morphology of insects can be examined better and identification can be made faster and more accurately. The purpose of this study is to investigate some anatomical structures of the antennas by using scanning electron microscopy of five species of adult Diptera with forensic importance and to contribute to the insect database with forensic importance. The insect species studied in this study are*: Calliphora vicina* Robineau-Desvoidy, 1830, *Chrysomia albiceps* (Wiedemann, 1819), *Lucilia cuprina* (Wiedemann, 1830), *Lucilia sericata* (Meigen, 1826) and *Muscina stabulans* (Fallen, 1817). SEM images of five species of Diptera with forensic importance were examined by photographing. The surface of the antennal segments are covered with various number chaetic sensilla and trichoidea sensilla. The position and number of chaetic sensillans on scape and pedicel can be used as a taxonomic character.

KEY WORDS: Forensic science, forensic entomology, ultrastructure, antenna, Diptera

Forensic entomology is described by entomologist Jason Byrd as "the use of insects living in decomposed corpse and their other arthropod relatives for the purpose of helping forensic investigations" or as evidence for the use of insects in the litigation of certain events (Byrd & Castner, 2010).

A corpse (it does not matter that is a human or an animal) is a source of nutrients for many living things and contributes to the rapidly changing ecosystem by breaking up. After an actual death event, the body follows the sequence of certain decays. During this decomposition, physical, chemical and biological degradation take place. An analysis of the colonization sequence of insects on the body can determine the time after death. This method can provide precise and usable methods for determining the time since death. It is also used in many murder investigations around the world (Byrd & Castner, 2010).

The Diptera order is the most interesting group in forensic studies. In forensic studies and PMI estimation, Calliphoridae (Blow flies), Sarcophagidae (Flesh flies) and Muscidae (Muscid flies) families and their larvae are especially used.

Identification of some Diptera by using a normal stereo microscope is not possible. Especially in the larval stage, the species identification is not easy, and the larvae of some species are still undefined. Ultrastructural examination involves a detailed examination of the specimen structure of the specimen being examined. Scanning Electron Microscopy (SEM) provides us very important data for examining the ultrastructural investigation of structures of the samples (Ubero-Pascal et al., 2010).

The ultrastructure in adult Diptera have been the subject of many researchers; Sukontason et al. (2004, 2006, 2007, 2008) who studied, by using SEM, the analyzed the ultrastructure of the following families: Calliphoridae, Muscidae and

Sarcophagidae; Carriço et al. (2015) analyzed some characteristics of the head morphology of the Muscidae; Pezzi et al. (2016) studied of the antenna of *Sarcophaga tibialis*.

Terrestrial studies on forensic entomology have been conducted in our country. Some of those are: Ozdemir & Sert (2009), the determination of forensically significant Coleoptera fauna in Ankara; Sabanoglu & Sert (2010), Calliphoridae species on carcass and seasonal distribution in Ankara; Sert et al. (2012), the identification of forensically significant Coleoptera and Calliphoridae (Diptera) species on a decaying dog carcass *Canis lupus familiaris* in Ankara. There are no studies on the investigation of the ultrastructure of insects with forensic importance in Turkey. In this study, the external morphology of the antenna was examined in detail using five Diptera species scanning electron microscopy with forensic importance.

MATERIAL AND METHODS

The species *Calliphora vicina, Chrysomya albiceps, Lucilia cuprina, L. sericata and Muscina stabulans*, were obtained from the Forensic Entomology Project regularly performed by our group.

Adult fly specimens obtained in these studies were kept in 70% alcohol and specimens were made into standard museum material. For electron microscopy studies, the head of adult samples was dissected.

Specimens were placed on stub, and examined under FEI Quanta FEG 250 scanning electron microscope (SEM). Using low vacuum, it was scanned with the electron beam at 5 kV from different angles. It has been scanned with the electron beam at 5 kV from different angles by using low vacuum and it has nothing to do with any kind of coating, fixing and drying, because of the low vacuum and the natural chitin coverage of the specimens. Smith (1986), Dobson (2013) and Spzila (2012) sources were used to identify the specimens.

RESULTS

Calliphora vicina Robineau-Desvoidy, 1830

The antennae of *C. vicina* are composed of three segments: a proximal scape (S), pedicel (P), and distal funiculus (Fn) that bears a slender arista laterally (Figs. 1-2). Scape is the shortest antennal segment of *C. vicina*, and the only sensilla on antennal scape are the socketed chaetic sensilla (Ch) (Figs. 2-3), which are arranged in a single row. The entire surface of this segment is covered with trichoidea sensilla (Fig. 3).

The surface of the pedicel segment is covered with various chaetic sensilla and trichoidea sensilla. Chaetic sensilla (ChI) is one and quite long, shorter ones are ChII. For example, some of the ChII on the subject was broken. The side of the pedicel is covered with short chaetic sensilla (ChIII) (Fig. 3).

The entire surface of antennal funiculus is densely covered with trichoidea sensilla. The antennal arista is comprised of three segments, two basal short ones and a distal long one. There are long sensillae on arista (Fig. 2).

Chrysomya albiceps (Wiedemann, 1819)

The antennae of *C. albiceps* are composed of three segments: a proximal scape (S), pedicel (P), and distal funiculus (Fn) that bears a slender arista laterally (Figs. 4-5). Scape is the shortest antennal segment of *C. albicaps*, and the only sensilla

on antennal scape are the socketed chaetic sensilla (Ch) (Fig. 5). The entire surface of this segment is covered with trichoidea sensilla (Fig. 5).

The surface of the pedicel segment is covered with various chaetic sensilla and trichoidea sensilla. There are three types of chaetic sensilla: ChI, ChII and ChIII (Fig. 6).

The entire surface of antennal funiculus is densely covered with trichoidea sensilla. The antennal arista is comprised of three segments, two basal short ones and a distal long one. There are long sensilla on Arista (Figs. 5-6).

***Lucilia cuprina* (Wiedemann, 1830)**

The antennae of *L. cuprina* are composed of three segments: a proximal scape (S), pedicel (P), and distal funiculus (Fn) that bears a slender arista laterally (Figs. 7-8). Arista was broken and we could not review it. Scape is the shortest antennal segment of *L. cuprina*, and the only sensilla on antennal scape are the socketed chaetic sensilla (Ch) (Figs. 8-9). The entire surface of this segment is covered with trichoidea sensilla (Fig. 9).

The surface of the pedicel segment is covered with various chaetic sensilla and trichoidea sensilla. There is a single type of chaetic sensilla (ChI) (Fig. 9). ChI is broken.

The entire surface of antennal funiculus is densely covered with trichoidea sensilla. The antennal arista is broken (Figs. 8-9).

***Lucilia sericata* (Meigen, 1826)**

The antennae of *L. sericata* are composed of three segments: a proximal scape (S), pedicel (P), and distal funiculus (Fn) that bears a slender arista laterally (Figs. 10-11). Scape is the shortest antennal segment of *L. sericata*, and the only sensilla on antennal scape are the socketed chaetic sensilla (Ch) (Fig. 11). The entire surface of this segment is covered with trichoidea sensilla (Fig. 12).

The surface of the pedicel segment is covered with various chaetic sensilla and trichoidea sensilla. There are two types of chaetic sensilla: ChI and ChII (Figs. 11-12).

The entire surface of antennal funiculus is densely covered with trichoidea sensilla. The antennal arista is comprised of three segments, two basal short ones and a distal long one. There are long sensillae on Arista (Fig. 11).

***Muscina stabulans* (Fallen, 1817)**

The antennae of *M. stabulans* are composed of three segments: a proximal scape (S), pedicel (P), and distal funiculus (Fn) that bears a slender arista laterally (Figs. 13-14). Scape is the shortest antennal segment of *M. stabulans*, and the only sensilla on antennal scape are the socketed chaetic sensilla (Ch) (Fig. 14). The entire surface of this segment is covered with trichoidea sensilla (Fig. 14).

The surface of the pedicel segment is covered with various chaetic sensilla and trichoidea sensilla. There are two types of chaetic sensilla: ChI and ChII (Figs. 14-15).

The entire surface of antennal funiculus is densely covered with trichoidea sensilla. The antennal arista is comprised of three segments, two basal short ones and a distal long one. There are long sensillae on Arista (Fig. 14).

DISCUSSION

In the forensic entomology studies, identification of the species is one of the most important issues. In order to the PMI estimate to be accurate, the correct

identification of the insect collected over the corpse should be done first. As it is known, Diptera are the most used group of insects in forensic entomology studies and PMI prediction. In this study, it is aimed to investigate the scanning electron microscopy and antenna morphology of fly samples obtained in forensic entomology studies conducted in Aksaray. SEM images of five species of Diptera with forensic importance were examined by photographing. The five species examined in SEM images are found in chaetic sensilla on scape and pedicel segments. All antenna segments are covered with trichoidea sensilla. The position and number of chaetic sensillans on scape and pedicel can be used as a taxonomic character. We think that the results obtained will contribute to the fly database with forensic design. This study, which is the first in terms of our working group, should be supported with further studies.

ACKNOWLEDGEMENTS

This work was supported by the Aksaray University scientific research project [BAP 2015-054, 2016]. The authors are grateful to the Aksaray University for financial support and Aksaray University Scientific and Technological Research and Application Center (ASUBTAM) for the provision of laboratory facilities. The authors declare that they have no conflict of interest.

LITERATURE CITED

Byrd, J. H. & Castner, J. L. 2010. Forensic Entomology, The Utility of Arthropods in Legal Investigations. Boca Raton, Florida: CRC Press.

Carriço, C., Mendonça, P. M., Cortinhas, L. B., Mallet, J. R. & Queiroz, M. M. 2015. Ultrastructural studies of some character of Diptera (Muscidae) of forensically importance. Acta Tropica, 142: 96-102.

Dobson, M. 2013. Family-level keys to freshwater fly (Diptera) larvae: A Brief review and a key to European families avoiding use of mouthpart characters. Freshwater Reviews, 6: 1-32.

Özdemir, S. & Sert, O. 2009. Determination of Coleoptera fauna on carcasses in Ankara province, Turkey. Forensic Science International. 183 (1-3): 24-32.

Pezzi, M., Whitmore, D., Chicca, M., Semeraro, B., Brighi, F. & Leis, M. 2016. Ultrastructural Morphology of the Antenna and Maxillary Palp of *Sarcophaga tibialis* (Diptera: Sarcophagidae). Journal of Medical Entomology, 53: 807-814.

Sert, O., Kabalak, M. & Şabanoglu, B. 2012. Determination of forensically important Coleoptera and Calliphoridae (Diptera) species on decomposing dog (*Canis lupus familiaris* L.) carcass at Ankara province. Hacettepe Journal of Biology and Chemistry, 40 (1): 99-103.

Smith, K. G. V. 1986. A Manual of Forensic Entomology. London: British Museum.

Sukontason, K., Methanitikorn, R., Chaiwong, T., Kurahashi, H., Vogtsberger, R. C. & Sukontason, K. L. 2007. Sensilla of the antenna and palp of *Hydrotaea chalcogaster* (Diptera: Muscidae). Micron, 38: 218-223.

Sukontason, K., Methanitikorn, R., Kurahashi, H., Vogtsberger, R. C. & Sukontason, K. L. 2008. External morphology of *Chrysomya pinguis* (Walker) (Diptera: Calliphoridae) revealed by scanning electron microscopy. Micron, 39: 190-197.

Sukontason, K., Sukontason, K. L., Piangjai, S., Boonchu, N., Chaiwong, T., Ngern-klun, R., Sripakdee, D., Vogtsberger, R. C. & Olson, J. K. 2004. Antennal sensilla of some forensically importante flies in families Calliphoridae, Sacorphagidae and Muscidae. Micron, 35: 671-679.

Sukontason, K. L., Bunchu, N., Methanitikorn, R., Chaiwong, T., Kuntalue, B. & Sukontason K. 2006. Ultrastructure of adhesive devise in fly in families Calliphoridae, Muscidae and Sarcophagidae, and their implication as mechanical carries of pathogens. Parasitology Research, 98: 477-481.

Szpila, K. 2012. Key for identification of European and Mediterranean blowflies (Diptera, Calliphoridae) of medical and veterinary importance – adult flies. In: Gennard D, editor. Forensic Entomology, an Introduction. 2st ed. Willey-Blackwell, pp. 77-81.

Şabanoglu, B. & Sert, O. 2010. Determination of Calliphoridae (Diptera) fauna and seasonal distribution on carrion in Ankara province. Journal of Forensic Sciences, 55 (4): 1003-1007.

Ubero-Pascal, N., Arnaldos, I., López-Esclapez, R. & García, M. D. 2010. Microscopy and Forensic Entomology. In: Méndez-Vilas A, Díaz J, editors. Microscopy: Science, Technology, Applications and Education. Formatex Research Center, pp. 1548-1556.

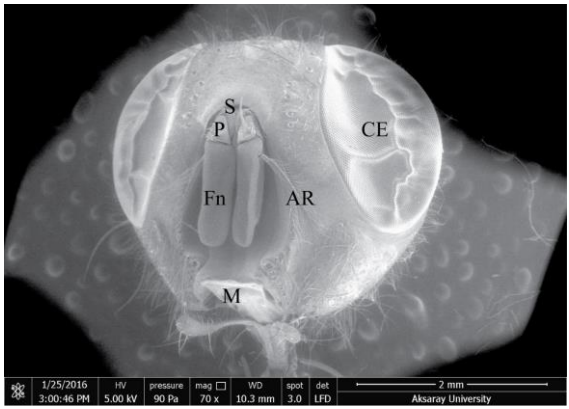

Figure 1. The head of adult female *C. vicina* SEM micrograph is displaying the antennae. These antennae have been seated centrally between large compound eyes (CE) by attaching the arista (AR) to the flagellum (F) dorso-laterally. The scape (S), pedicel (P), funiculus (Fn), and a section of the mouthparts (M) have been shown.

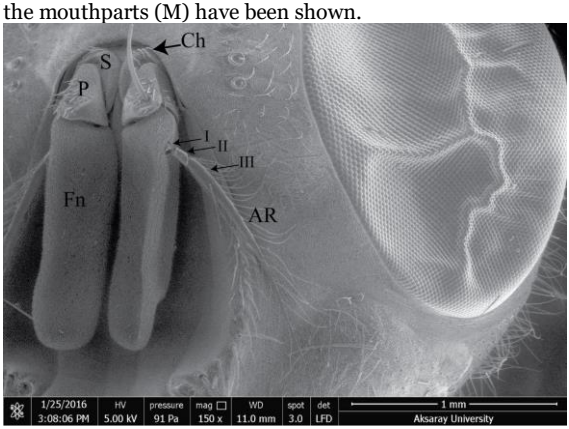

Figure 2. SEM micrograph of the antenna of adult female *C. vicina*. scape (S), pedicel (P), funiculus (Fn), and the chaetic sensilla on scape (Ch) arista (AR), arista segments (I, II, III).

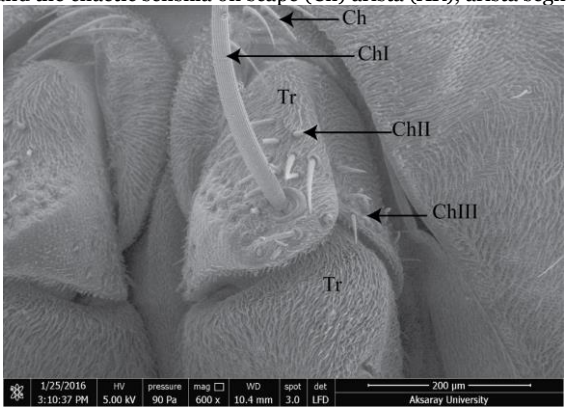

Figure 3. SEM micrograph of the scape of adult female *C. vicina*. chaetic sensilla on scape (Ch), chaetic sensilla (ChI), chaetic sensilla (ChII), chaetic sensilla (ChIII), trichoidea sensilla (Tr).

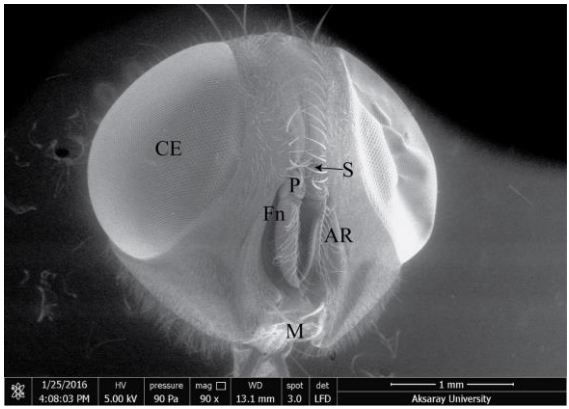

Figure 4. The head of adult female *Chrysomya albiceps* SEM micrograph is displaying the antennae. These antennae have been seated centrally between large compound eyes (CE) by attaching the arista (AR) to the flagellum (F) dorso-laterally. The scape (S), pedicel (P), funiculus (Fn), and a section of the mouthparts (M) have been shown.

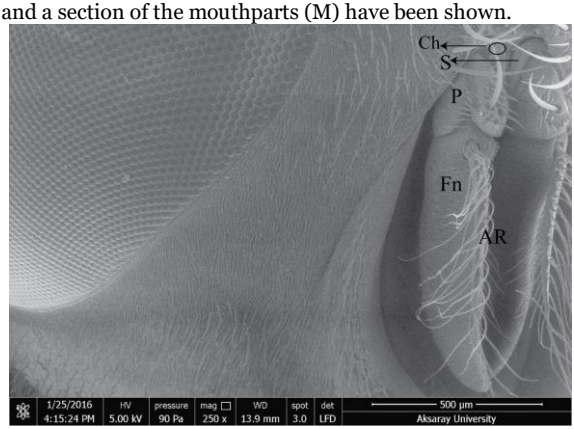

Figure 5. SEM micrograph of the antenna of adult female *Chrysomya albiceps*. scape (S), pedicel (P), funiculus (Fn), chaetic sensilla on scape (Ch) and the arista (AR).

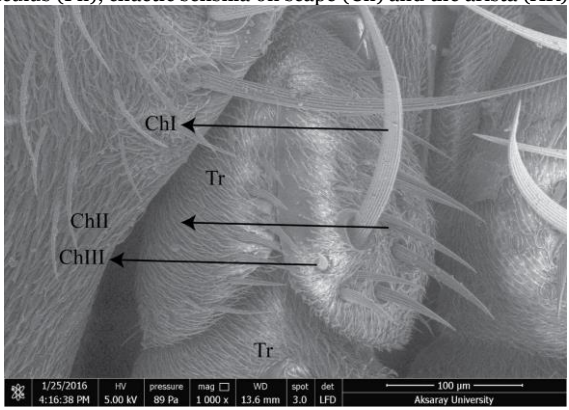

Figure 6. SEM micrograph of the scape of adult female *Chrysomya albiceps*. chaetic sensilla (ChI) chaetic sensilla (ChII) chaetic sensilla (ChIII) trichoidea sensilla (Tr).

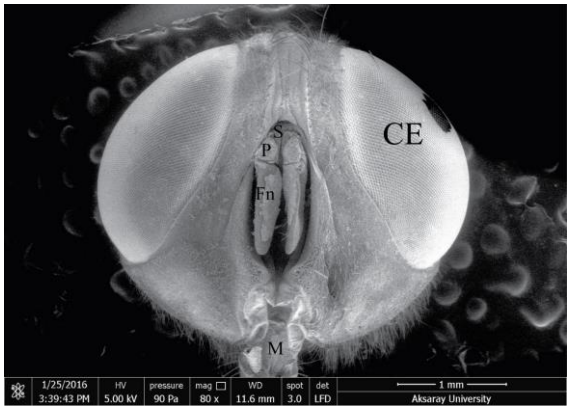

Figure 7. The head of adult female *Lucila cuprina* SEM micrograph is displaying the antennae. These antennae have been seated centrally between large compound eyes (CE) by attaching the arista (AR) to the flagellum (F) dorso-laterally. The scape (S), pedicel (P), funiculus (Fn); flagellum (F), and a section of the mouthparts (M) have been shown.

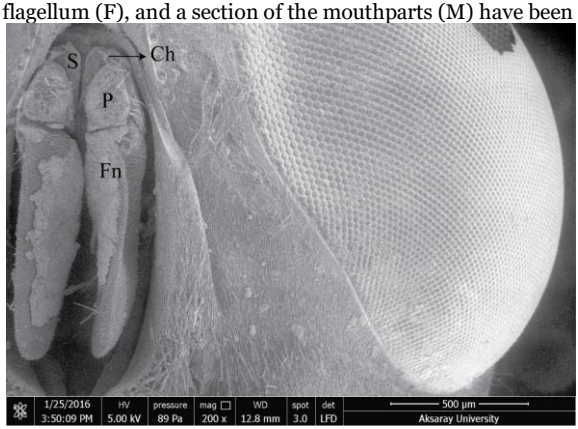

Figure 8. SEM micrograph of the antenna of adult female *Lucila cuprina*. scape (S), pedicel (P), flagellum (F), chaetic sensilla on scape (Ch) and the arista (AR).

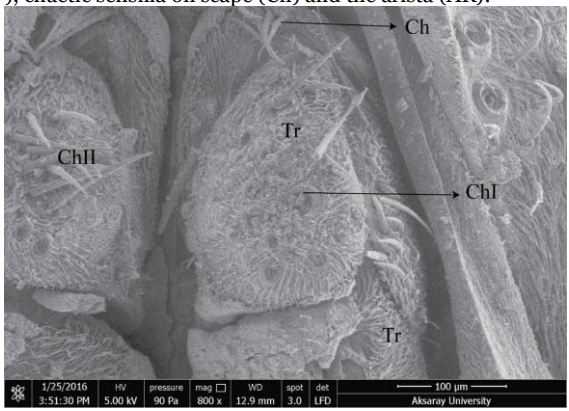

Figure 9. SEM micrograph of the scape of adult female *Lucila cuprina*. chaetic sensilla (Ch) chaetic sensilla (ChI) trichoidea sensilla (Tr).

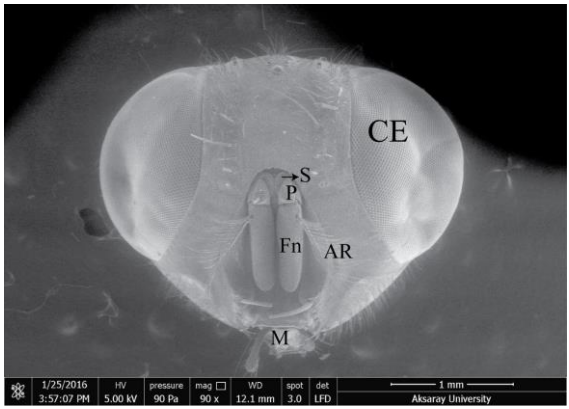

Figure 10. The head of adult female *Lucila sericata* SEM micrograph is displaying the antennae. These antennae have been seated centrally between large compound eyes (CE) by attaching the arista (AR) to the flagellum (F) dorso-laterally. The scape (S), pedicel (P), funiculus (Fn); flagellum (F), and a section of the mouthparts (M) have been shown.

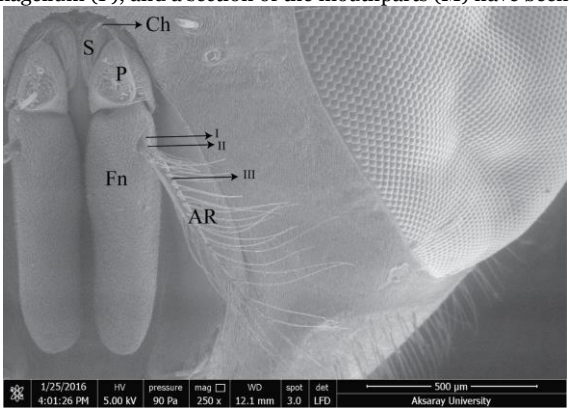

Figure 11. SEM micrograph of the antenna of adult female *Lucila sericata*. scape (S), pedicel (P), funiculus (F), and the chaetic sensilla on scape (Ch) arista (AR), arista segments (I, II, III).

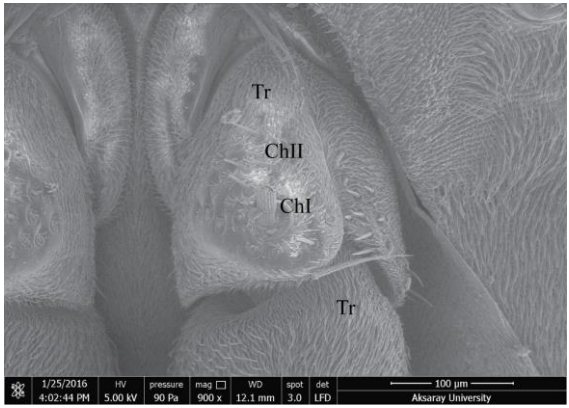

Figure 12. SEM micrograph of the scape of adult female *Lucila sericata*. chaetic sensilla (ChI) chaetic sensilla (ChII) trichoidea sensilla (Tr).

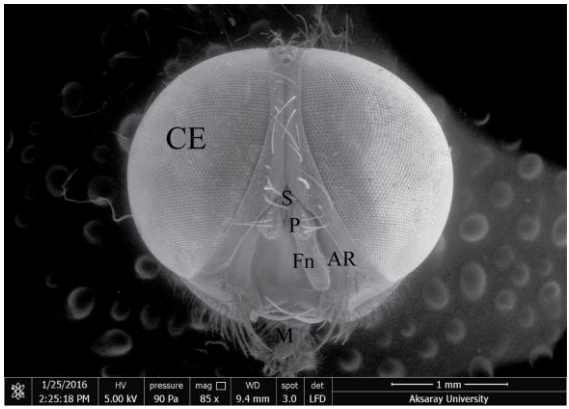

Figure 13. The head of adult male *Muscina stabulans* SEM micrograph is displaying the antennae. These antennae have been seated centrally between large compound eyes (CE) by attaching the arista (AR) to the flagellum (F) dorso-laterally. The scape (S), pedicel (P), funiculus (Fn); flagellum (F), and a section of the mouthparts (M) have been shown.

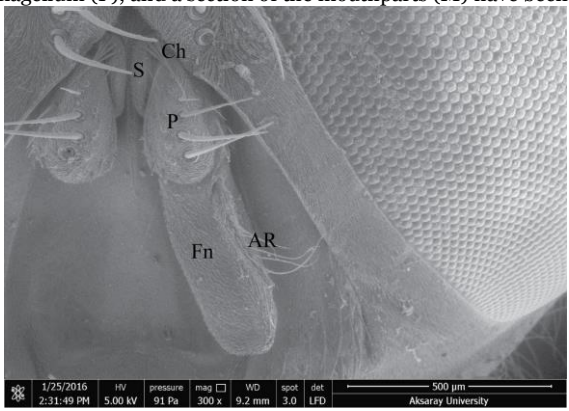

Figure 14. SEM micrograph of the antenna of adult male *Muscina stabulans*. scape (S), pedicel (P), funiculus (Fn), chaetic sensilla on scape (Ch) and the arista (AR).