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ULTRASTRUCTURES OF AEDEAGUS AND SPERMATHECA OF TWO SPECIES OF THE SUBGENUS *CHAETOCNEMA* (*HORTENSOIDES*) ÖZDİKMEN, 2021 (CHRYSOMELIDAE: GALERUCINAE: ALTICINI) BY SCANNING ELECTRON MICROSCOPE

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ABSTRACT: The paper presents unknown ultrastructures of aedeagus and spermatheca of *Chaetocnema arenacea* (Allard, 1860) and *Chaetocnema aridula* (Gyllenhal, 1827) which is two of the species of the subgenus *Chaetocnema* (*Hortensoides*) Özdikmen, 2021 are studied and figured under both stereo microscope and Scanning Electron Microscope from Turkey for the first time. The studied specimens were collected from Çankırı and Kayseri provinces in 2013, 2014, 2015, 2016 and 2018. Aedeagi and spermathecae so far were studied on the basis of only stereo microscopic studies within the genus *Chaetocnema* Stephens. Until now, there is only one ultrastructural study with SEM on male and female genitalia of *Chaetocnema arida* in the subgenus *Chaetocnema* (*Hortensoides*) Özdikmen, 2021, therefore, the present study is the first for these two species mentioned above. In accordance with this, to reveal and to obtain new diagnostic characters with ultrastructural and detailed investigations of aedeagus and spermatheca of the species *Chaetocnema arenacea* (Allard, 1860) and *Chaetocnema aridula* (Gyllenhal, 1827) are basically aimed with this study. It is revealed new diagnostic characters for both the aedeagus and spermatheca by investigations on ultrastructural morphologies in SEM.

KEY WORDS: Ultrastructure, aedeagus, spermatheca, SEM, *Chaetocnema arenacea, Chaetocnema aridula*, Turkey

Chaetocnema Stephens, 1831, a member of the tribe Alticini Newman, 1835, is a cosmopolitan genus and includes a large number of species (about 500) that spread all over the world (Palaearctic, Nearctic, Neotropical, Oriental, Afrotropical, Madagascar and Australian regions). The genus is represented with a total of 27 species in Turkey that is in the Palaearctic region (Ekiz et al., 2013; Özdikmen, 2014, 2021; Çilbiroğlu Aslan & Bayram Partal, 2023; Bezdek & Sekerka, 2024). Species of the genus *Chaetocnema* Stephens are often divided into the subgenera *Chaetocnema* s. str. and *Tlanoma* Motschulsky, 1845, but according to White (1996), the nasal keel which is supposed to distinguish the

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latter subgenus from the former is highly variable and therefore a poor character for grouping species. In addition, White (1996) argued that *Tlanoma* cannot be treated as a subgenus of *Chaetocnema* because both have the same type species (*C. concinna*). Consequently, subgeneric classification of the genus *Chaetocnema* Stephens was recently reviewed and arranged by Özdikmen (2021). He proposed a total of 13 subgenera for the genus *Chaetocnema* Stephens.

According to Konstantinov et al. (2011), *Chaetocnema* species are habitually very similar and cannot be separated only with external characters. Genitalic characters have been used traditionally and are diagnostically important. However, works on aedeagi and spermathecae of *Chaetocnema* species so far were studied on the basis of only stereo microscopic studies such as revisional studies of White (1996), Konstantinov et al. (2011), Ruan et al. (2019) and the others, for example, Ruan et al. (2014a,b), while ultrastructures of aedeagi and spermathecae of *Chaetocnema* species with SEM are currently very few.

As known, aedeagal morphology in the genus *Chaetocnema* Stephens, 1831, like in most genera of the tribe Alticini, seems to be diagnostic at the species level because of there are many new species described based on aedeagal structure. On the contrary of aedeagal morphology, spermathecal morphology seems to be not diagnostic at the species level because of there is no new species described only based on spermathecal structure. Even, it must be because of this, spermathecae of Nearctic species in revision of White (1996) did not studied.

In Chrysomeloidea, the spermatheca has been useful to define subfamilies, genera and species (Reid, 1989; Hernández, 1993; Biondi, 2001; Borowiec & Świętojańska, 2001, Borowiec & Skuza, 2004; Borowiec & Opalinska, 2007; Borowiec & Pomorska, 2009; Rodríguez-Mirón & Zaragoza-Caballero, 2017; Rodríguez-Mirón et al., 2017). According to Rodríguez-Mirón et al. (2017), the spermatheca provides characters to diagnose genera and subgenera, and also they stated that the spermatheca has a high taxonomic value for diagnosing taxa at various ranks.

The species *Chaetocnema arenacea* (Allard, 1860) and *Chaetocnema aridula* (Gyllenhal, 1827) have Palaearctic (Europeo-Mediterranean) and Palaearctic (Sibero-Europeo-Mediterranean) + introduced to Oriental chorotypes respectively. There is not much information available about the female and male genital structures of *Chaetocnema arenacea* (Allard, 1860) and *Chaetocnema aridula* (Gyllenhal, 1827), and with this study, the male and female genital structures of these species have been studied and presented in detail.

MATERIALS AND METHODS

The available specimens for the present work were collected in Çankırı and Kayseri provinces from Turkey in 2013, 2014, 2015, 2016 and 2018. The specimens were deposited at Gazi University (Turkey, Ankara) and Nazife Tuatay Plant Protection Museum (Turkey, Ankara).

The aedeagi and spermathecae were dissected from the abdomen and the remaining tissue was removed with fine tweezers. After cleaning, the samples were placed in 70% ethanol and examined with Olympus SZX7 stereo microscope and Leica Z-16 APO stereo microscope for stereo microscopic examination. For scanning electron microscopy (SEM), the cleaned samples were dehydrated using an increasing series of ethanol (70%, 80%, 90% and 100%) and then air-dried.

The samples were then mounted on SEM stubs using double-sided adhesive tape, coated with gold using Polaron SC 502 Sputter Coat and examined with JEOL JSM 6060 Scanning Electron Microscope (SEM) at 10 kV in Prof. Dr. Zekiye Suludere Electrone Microscopy Center at Gazi University. The main spermathecal and aedeagal terminologies used in the present study are given below.

RESULTS AND DISCUSSION

Genus *Chaetocnema* Stephens, 1831
Type sp.: *Chrysomela concinna* Marsham, 1802

Subgenus *Hortensoides* Özdikmen, 2021
Type sp.: *Altica hortensis* Geoffroy, 1785

The cosmopolitan genus is represented with a total of 27 species of 6 *Chaetocnema* subgenera in Turkey that is in Palaearctic region (Ekiz et al., 2013; Özdikmen, 2014, 2021; Bezdek & Sekerka, 2024). According to the number of species, the subgenus *Chaetocnema* (*Hortensoides*) Özdikmen, 2021 includes fourteen species in Turkey (about 52% of the total species number in Turkey). These are; *Chaetocnema aerosa* (Letzner, 1847), *Chaetocnema arenacea* (Allard, 1860), *Chaetocnema arida* Foudras, 1860, *Chaetocnema aridula* (Gyllenhal, 1827), *Chaetocnema confusa* (Boheman, 1851), *Chaetocnema hortensis* (Geoffroy, 1785), *Chaetocnema igori* Konstantinov et al., 2011, *Chaetocnema imitatrix* Gruev, 1990, *Chaetocnema mannerheimii* (Gyllenhal, 1827), *Chaetocnema montenegrina* Heikertinger, 1912, *Chaetocnema obesa* (Boieldieu, 1859), *Chaetocnema sahlbergii* (Gyllenhal, 1827), *Chaetocnema subcoerulea* (Kutschera, 1864) and *Chaetocnema wollastoni* Baly, 1877 (Özdikmen, 2021; Çilbiroğlu Aslan & Bayram Partal, 2023).

Some external characters for the subgenus. Frontal ridge between antennal sockets wide and flat. Frons evenly covered with relatively short, white setae. Vertex flat, situated about on same level as orbit. Surface of vertex with evenly and densely punctures. Punctures about as large as pronotal punctures. Pronotum with evenly and densely punctures. Diameter of pronotal punctures subequal to distance between them. Pronotal punctures usually about as large as elytral punctures. Basal stria of punctures on pronotum absent. Base of pronotum without longitudinal impressions. Elytra with convex sided. Elytral punctures confused including periscutellar rows at least in basally or entirely regular including periscutellar rows. Moreover, periscutellar row of punctures on elytron regular and single, confused or more than one (geminate) and second through sixth rows of punctures at base of elytron regular (Özdikmen, 2021).

***Chaetocnema arenacea* (Allard, 1860)** (Figs. 1A-C, 2A-B)
arenacea Allard, 1860: 569 (*Plectroscelis*)

Materyal examined: Çankırı prov.: Eldivan, Maruf village, 40˚38'26"N, 33˚25'59"E, 1196 m, 23.IV.2013, 1 specimen; Eldivan, Akçalı Çukuroz return, 40˚36'31"N, 33˚28'21"E, 1068 m, 23.IV.2013, 1 specimen; Şabanözü, between Şabanözü-Eldivan road 3rd km, 40˚28'48"N, 33˚18'37"E, 1141 m, 24.IV.2013, 1 specimen; Korgun, Çankırı-Korgun road, Aşağıçavuş village, 40˚41'08"N,

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33˚36'41"E, 837 m, 21.IV.2013, 1 specimen; Kurşunlu, 5 km to Eskiahır village, 40˚50’39”N, 33˚15’30”E, 1152 m, 25.VII.2013, 1 specimen; Korgun, between Karatekin and Yolkaya village, 40˚41'30"N, 33˚29'20"E, 957 m, 22.IV.2013, 4 specimens; Central, Hasakça plain, 40˚40’01,6”N, 33˚47’14”E, 873 m, 12.VII.2014, 2 specimens; Central, entry of Dereçatı village, 40˚45’6,7”N, 33˚40’15,7”E, 1060 m, 15.VII.2014, 1 specimen; Yapraklı, Buluca-İgdir return, 40˚45’25,4”N, 33˚47’7,24”E, 1195 m, 15.VII.2014, 1 specimen; Yapraklı, exit of Büyük Yayla, 40˚48’27”N, 33˚46’36,9”E, 1505 m, 15.VII.2014, 1 specimen; Korgun, Karatepe (Şıhlar) village, 40˚46’58,8”N, 33˚32’4”E, 1019 m, 16.VII.2014, 1 specimen; Ilgaz, Mesutören village, 40˚49’43,3”N, 33˚34’26,3”E, 1226 m, 16.VII.2014, 1 specimen; Yapraklı, Büyük Yayla forest, 40˚49’38,7”N, 33˚43’37,4”E, 1517 m, 16.VII.2014, 1 specimen; Yapraklı, Kaymaz village, 40˚43’45”N, 33˚54’21,3”E, 1137 m, 16.VII.2014, 1 specimen; Kızılırmak, between Yukarıalagöz and Alaca villages, 40˚22’28”N, 33˚54’54”E, 619 m, 25.IV.2014, 1 specimen; Kurşunlu, entry of Hacımuslu, 40˚51’3”N, 33˚16’20”E, 1146 m, 20.VII.2014, 4 specimens; Korgun, Ayan return, 40˚41’6,4”N, 33˚36’14,4”E, 821 m, 09.VIII.2014, 1 specimen; Kızılırmak, between Cacıklar and Karamürsel villages, 40˚23’36,7”N, 34˚02’22”E, 527 m, 11.VIII.2014, 1 specimen; Centra, Tuz Cave env., 40˚31’38”N, 33˚45’55”E, 699 m, 26.IV.2014, 1 specimen; Kurşunlu, Köprülü return, 40˚50’29”N, 33˚15’38”E, 1130 m, 20.V.2014, 1 specimen; Korgun, Sanı plateau, 40˚37’00”N, 33˚24’10”E, 1363 m, 20.V.2014, 1 specimen; Orta, entry of İncecik village, 40˚35’33”N, 32˚55’59”E, 1600 m, 22.V.2014, 1 specimen; Şabanözü, entry of Kamış village, 40˚33’45”N, 33˚20’13”E, 1221 m, 23.V.2014, 1 specimen; Korgun, exit of Bugay village, 40˚42’50,8”N, 33˚29’40,1”E, 857 m, 08.VII.2014, 2 specimens; Şabanözü, between Maruf and Kavuş villages, 40˚37’22”N, 33˚23’36”E, 1361 m, 08.VII.2014, 1 specimen; Eldivan, entry of Çukuröz village, 40˚36’31,8”N, 33˚26’09,9”E, 1102 m, 09.VII.2014, 1 specimen; Kurşunlu, entry of Köpürlü, 40˚47’12,5”N, 33˚16’49,7”E, 1006 m, 10.VII.2014, 15 specimens; **Kayseri prov.:** Pınarbaşı, Toklar, 38°35‟58”N, 36°7‟55”E, 10.VI.2015, 1425 m, 1 specimen; Yahyalar, 38°1‟24”N, 35°29‟47”E, 11.VI.2015, 1295 m, 1 specimen; Develi, Güneyaçağı, 38°21‟53”N, 35°28‟16”E, 21.VI.2016, 1164 m, 3 specimens; Yahyalı, Kirazlı, 38°3‟1”N, 35°24‟0”E, 22.VI.2016, 1408 m, 1 specimen; Yahyalı, Kirazlı, 38°3‟1”N, 35°24‟0”E, 22.VI.2016, 1408 m, 1 specimen; Yahyalı, Dikme, 38°1‟10”N, 35°32‟3”E, 22.VI.2016, 1404 m, 1 specimen; Kocasinan, Kalkancık, 38°53‟22”N, 35°8‟38”E, 23.VI.2016, 1111 m, 2 specimens; Kocasinan, Emmiler, 38°52‟9”N, 35°23‟59”E, 23.VI.2016, 1234 m, 1 specimen; Kocasinan, Karakimse, 38°55‟8”N, 35°9‟42”E, 23.VI.2016, 1154 m, 1 specimen; Melikgazi, Sarımsaklı, 38°51‟23”N, 35°47‟8”E, 23.VI.2016, 1222 m, 1 specimen; Kocasinan, Yuvalı, 38°50‟0”N, 35°5‟0”E, 24.VI.2016, 1115 m, 2 specimens; Tomarza, Köprübaşı, 38°25‟45”N, 35°58‟29”E, 12.VII.2016, 1350 m, 4 specimens; Bünyan, Sümer, 38°49‟3”N, 35°55‟17”E, 21.IX.2016, 1407 m, 2 specimens; Bünyan, Sümer, 38°49‟3”N, 35°55‟17”E, 21.IX.2016, 1407 m, 2 specimens; Kocasinan, Himmetdede, 38°53‟45”N, 35°6‟42”E, 05.V.2018, 1160 m, 1 specimen; Kocasinan, Beydeğirmeni, 38°49‟43”N, 35°10‟4”E, 28.V.2018, 977 m, 4 specimens; Pınarbaşı, Karahacılı, 38°38‟20”N, 36°18‟8”E, 05.VI.2018, 1563 m, 1 specimen; Pınarbaşı, Aşağıbeyçayırı, 38°39‟25”N, 36°27‟1”E, 05.VI.2018, 1622 m, 1 specimen; Felahiye, Kayapınar, 39°6‟48”N, 35°37‟13”E, 06.VII.2018, 1554 m, 4 specimens; Felahiye, Büyüktoraman, 39°9‟28”N, 35°40‟24”E, 06.VII.2018,

1473 m, 1 specimen; Felahiye, Darılı, 39°7‟1”N, 35°32‟2”E, 30.VII.2018, 1331 m, 1 specimen; Felahiye, Kepiç, 39°6‟16”N, 35°29‟45”E, 30.VII.2018, 1212 m, 5 specimens; Felahiye, Cumhuriyet, 39°5‟11”N, 35°33‟28”E, 30.VII.2018, 1268 m, 4 specimens; Pınarbaşı, Aşağıkızılçevlik, 38°50‟31”N, 36°32‟24”E, 31.VII.2018, 1622 m, 3 specimens; Pınarbaşı, Aşağıkızılçevlik, 38°50‟31”N, 36°32‟24”E, 01.VIII.2018, 1622 m, 1 specimen; Yahyalı, Dikme, 38°1‟10”N, 35°32‟3”E, 22.VI.2016, 1404 m, 1 specimen; Kocasinan, Emmiler, 38°52‟9”N, 35°23‟59”E, 23.VI.2016, 1234 m, 1 specimen; Kocasinan, Kalkancık, 38°53‟22”N, 35°8‟38”E, 23.VI.2016, 1111 m, 2 specimens; Kocasinan, Beydeğirmeni, 38°49‟43”N, 35°10‟4”E, 28.V.2018, 977 m, 4 specimens; Pınarbaşı, Karahacılı, 38°38‟20”N, 36°18‟8”E, 05.VI.2018, 1563 m, 1 specimen; Felahiye, Kepiç, 39°6‟16”N, 35°29‟45”E, 30.VII.2018, 1212 m, 3 specimens; Pınarbaşı, Aşağıkızılçevlik, 38°50‟31”N, 36°32‟24”E, 31.VII.2018, 1622 m, 3 specimens.

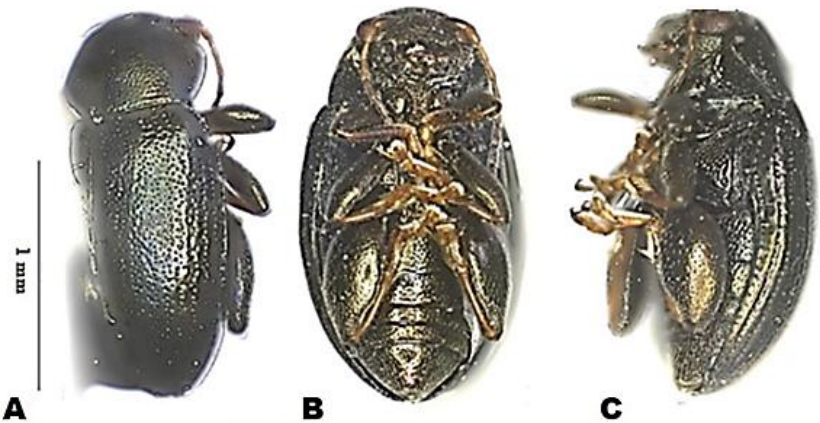

Figure 1. *Chaetocnema arenacea* (Allard, 1860): A. Dorsal, B. Ventral, C. Lateral view (Stereo microscope).

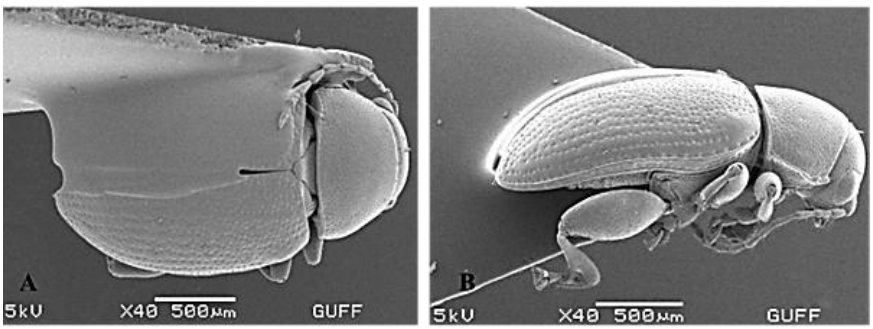

Figure 2. *Chaetocnema arenacea* (Allard, 1860): A. Dorsal, B. Lateral view (SEM).

The Palaearctic (Europeo-Mediterranean) species is distributed in Europe (Albania, Austria, Bulgaria, Crimea, Croatia, Czeck Republic, France, Greece, Hungary, Italy, Portugal, Romania, Russia (South European Territory), Serbia, Slovakia, Spain, Switzerland, Turkey, Ukraine), Asia (Armenia, Azerbaijan, Iran, Turkey), North Africa (Algeria, Morocco) (Bezdek & Sekerka, 2024).

The flea beetle is a rather widely distributed species for Turkey and it has been recorded from 17 of 81 provinces and six of seven regions in Turkey until now as Adana, Aksaray, Ankara, Çankırı, Denizli, Eskişehir, Erzurum, Gümüşhane, Hakkâri, Kayseri, Konya, Mersin, Nevşehir, Niğde, Sivas, Yozgat provinces in Anatolia (Asian Turkey) and Edirne province in European Turkey (Thrace) (Ekiz et al., 2013; Özdikmen, 2014; the present study) (Fig. 3).

Figure 3. Provincial and regional distribution of *Chaetocnema arenacea* (Allard, 1860) in Turkey.

Classification systems have mainly based on the external morphological characters, however, most of these classifications change constantly because of homoplasy and symplesiomorphy within these character sets. To provide a more stable classification, therefore, taxonomists have studied internal and genital morphological features in combination with the external morphological characters (e.g. Kasap & Crowson, 1979; Mann & Crowson, 1983; Suzuki, 1988).

As known, the structures of aedeagus and spermatheca are taxonomically important. For this reason, to reveal more diagnostic characters than hitherto known, ultrastructural and detailed investigations of aedeagus and spermatheca of *Chaetocnema arenacea* (Allard, 1860) from Turkey were firstly studied with scanning electron microscope (SEM) and stereo microscope. Obtaining observations are presented below.

Aedeagus (Stereo microscope) (Figs. 4A-C)**:**

Under the stereo microscope, the aedeagus is completely light brown. Only in the lateral view is the ventral part slightly darker. When the median lobe is viewed from the dorsal and ventral angles, except for the narrowing at the base and tip, it is almost parallel-sided. The distal width of the aedeagus relative to the basal aperture is equal to the width just before the apical slope. The tooth at the apex of

the median lobe is bluntly terminated in the form of a valve when viewed from the dorsal and ventral angles, but when viewed from the lateral angles, it becomes increasingly narrower towards the tip and ends in a pointed shape at the very tip. The aedeagus is in the form of an arch bent ventrally when viewed from the lateral angle. The apex of the median lobe ends with a tooth structure that extends from the orifice.

Figure 4. Aedeagus of *Chaetocnema arenacea* (Allard, 1860): A. Dorsal view in stereo microscope, B. Ventral view in stereo microscope, C. Lateral view in stereo microscope.

Aedeagus (SEM)

Ventral view (Figs. 5B)**:**

The median lobe is almost completely parallel-sided except for the slight expansion laterally in the middle part. The apical part of the aedegus is gradually narrowed at the apex. The apex of the median lobe is rectangular in shape and ends in a narrow elongated tongue at the wide end at the base. The width of the aedeagus is almost equal to the width just before the apical slope from the distal to the basal aperture. When the median lobe is viewed from the dorsal and ventral sides, except for the constriction at the base and the tip, the other parts are almost parallel-sided. The surface of the median lobe is convex apically, medially and basally from the lateral part of the median groove. The median groove is gradually narrowed from the base to the apical part and the depth of the groove decreases towards the apical part. The median groove has a deeper groove compared to other species. The width of the longitudinal groove in the middle of the median lobe is narrower than the distance between the groove and the lateral margin. There are quite distinct transverse wrinkles in the median groove from the base to the middle of the median lobe. There are distinct, scattered and sparse sensilla on the lateral edges of the median groove and in the parts close to the apex. While the sensilla are pit-shaped in other species, there are seta-like sensilla in this

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species. The median foramen is not completely circular. Its upper and lateral edges are curved (indented and wavy towards the median lobe).

Lateral view (Figs. 5C, 6, 7A,B)**:**
The median lobe is almost equal and quite distinctly curved towards the ventral side. The most curved part of the median lobe is the apical 1/3. The apex of the median lobe is quite narrow and pointed. A transverse depression is seen between the basal aperture of the median lobe and the middle part (approximately 1/4 of the part close to the base). All parts of the median lobe are seen to have ultrastructural sensilla (sensory receptors) in the form of scattered, sparse and distinct setae, increasing from the base to the apical part. The integrated form of the aedeagus (when we place the lateral image oppositely and symmetrically, the image obtained is a quadrata (square) structure, united at the top and expanded at the bottom. Although the end parts of the aedeagus come together in the integrated form, there is a quite wide gap between the base parts.

Dorsal view (Figs. 5A, 7C)**:**
The median lobe is almost completely parallel except for being slightly widened laterally in the apical 2/3 part. The median lobe is quite clearly convexly curved ventrally from the base to the apex, especially from the middle part. The apical part of the aedegus is gradually narrowed at the apex. Although there is a tooth structure at the apex of the median lobe (seen in stereo), this structure is not seen in the SEM photograph because of the tissue remains. The dorsal plate has a 3-part structure. While the parts on the sides extend from the apical 1/3 part of the median lobe to the apex by narrowing, the single part in the middle, the dorsal plate, is almost equally wide at the base starting from almost ½ part of the median lobe and widening towards the apical part, elongating in the form of a tongue, and its tip narrows again and curves inward. The dorsal plate fuses with the dorsal surface of the middle lobe at the base and divides the orifice into four. The orifice is distinct but narrowed. It is basically U-shaped or obtuse-angled V-shaped.

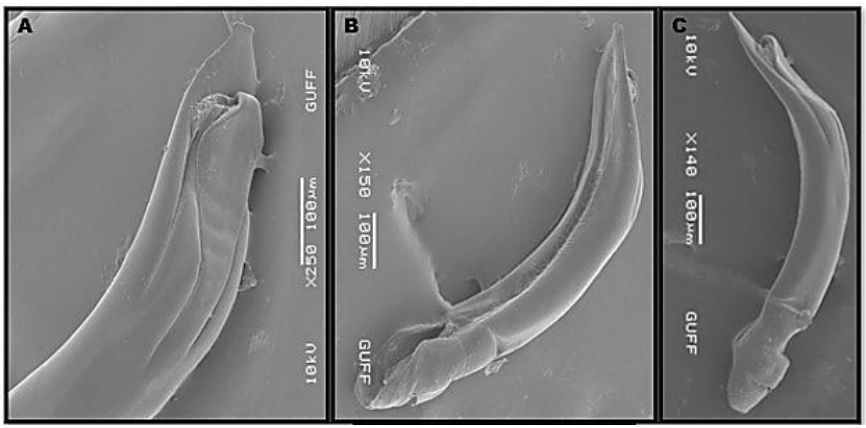

Figure 5. Aedeagus of *Chaetocnema arenacea* (Allard, 1860): A. Dorso-lateral view in SEM, B. Ventro-lateral view in SEM, C. Lateral view in SEM.

Figure 6. Integrated form of aedeagus.

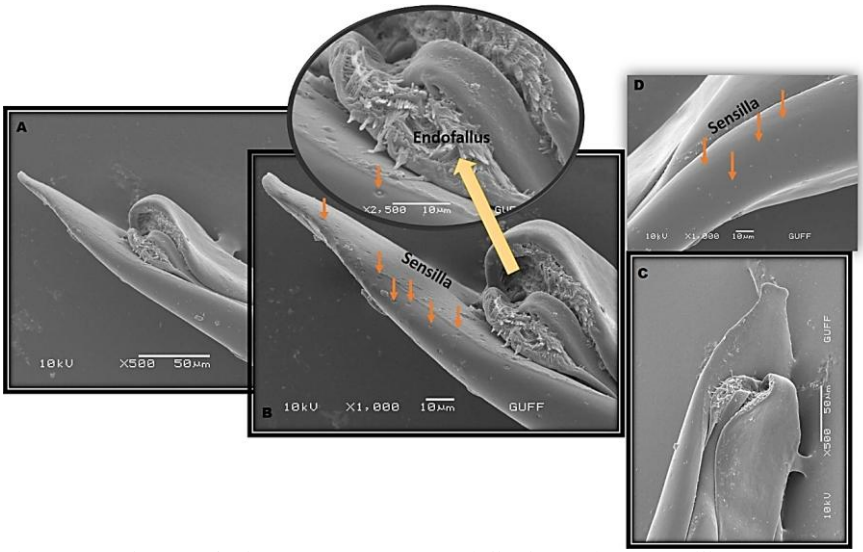

Figure 7. Aedeagus of *Chaetocnema arenacea* (Allard, 1860): A. Sensillar structure and density on the apical part of the lateral surface of the median lobe, B. Sensillar structure and density on the apical part of the dorsal surface of the median lobe and endophallus structure, C. Dorsal and dorsal wall structure of the median lobe, D) Sensillar structure and density on the medio-lateral walls of the median lobe (SEM).

Spermatheca (Stereo microscope) (Fig. 8B)**:**

The spermatheca is generally light brown. The protrusion at the tip of the spermathecal pump is dark brown, while the apical part of the receptacle is darker brown. The spermathecal receptacle is generally cylindrical, slightly curved in the shape of the letter S near the middle of the receptacle, and ends in an oval shape

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at the base. The spermathecal receptacle is almost equally wide in all areas. The spermathecal receptacle is almost 4 times longer than the pump. The spermathecal pump has a skin protrusion that is curved ventrally, duck-head-shaped, and has an elongated beak-like tip. The spermathecal canal curves from the base of the receptacle and extends apically to 1/2 of the receptacle.

Spermatheca (SEM) (Figs. 8A, 9A-D)**:**

The spermathecal receptacle is generally cylindrical, slightly curved in the shape of the letter S near the tip, and ends in an oval shape at the base. The spermathecal pump is cylindrical and its apical part is curved ventrally. There is a very distinct elongated skin protrusion at the apex of the spermathecal pump. Therefore, the structure at the apex of the pump appears dark brown in the stereo microscope image. This additional integument gives a duck beak appearance at the apex.

The spermathecal pump is connected to the middle of the apex of the spermathecal receptacle. The surface of the spermathecal receptacle is almost uniformly rugulose (with fine wrinkles). The spermathecal receptacle is almost equally wide in every area. The spermathecal canal originates from the middle part of the spermathecal receptacle base. The spermathecal canal curves ventrally from the base of the receptacle and extends to ½ of the receptacle. The exit point of the spermathecal gland duct is almost at the level of the middle part of the spermathecal receptacle. The spermathecal canal is straight at the base where it meets the receptacle, but it curves towards the middle and extends straight towards the distal.

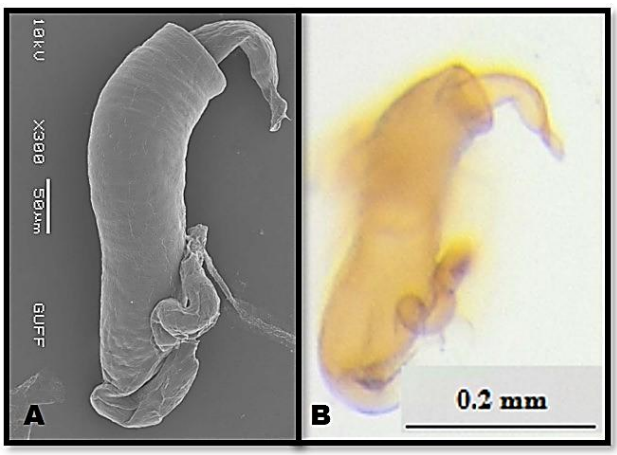

Figure 8. *Chaetocnema arenacea* (Allard, 1860): A. Spermatheca in SEM, B. Spermatheca in stereo microscope.

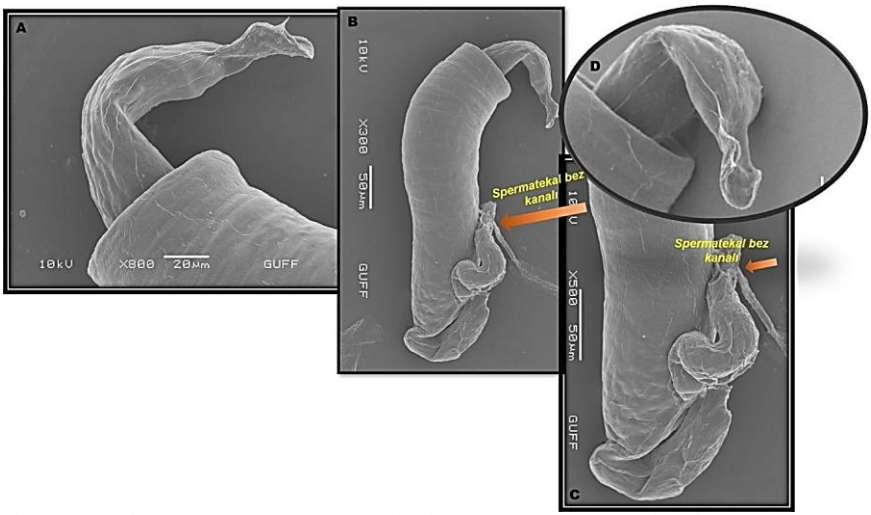

Figure 9. *Chaetocnema arenacea* (Allard, 1860): A. Spermathecal pump part, B. Spermathecal structure in lateral view, C. Spermathecal receptacle structure and spermathecal canal structure, D. Apical part of spermathecal pump (SEM).

***Chaetocnema aridula* (Gyllenhal, 1827)** (Figs. 9A-C, 10A-B)

aridula Gyllenhal, 1827: 663 (*Haltica*)
weisei Gerhardt, 1906: 238 (var.)
medvedevi Paliy, 1968: 15

Materyal examined: Kayseri prov.: Tomarza, Güzelce, 38°26‟16”N, 36°3‟4”E, 12.VII.2016, 1458 m, 3 specimens.

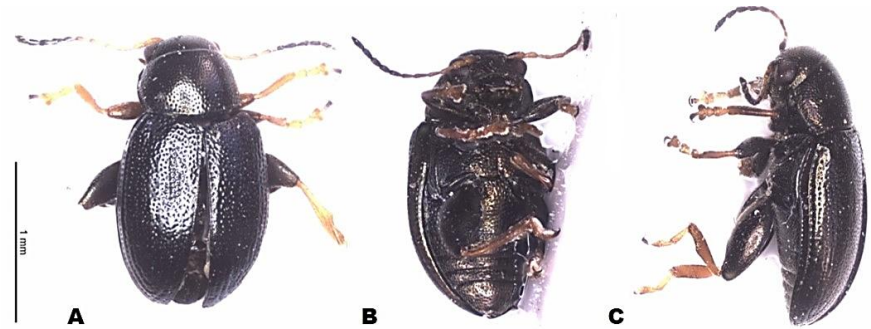

Figure 9. *Chaetocnema aridula* (Gyllenhal, 1827): A. Dorsal, B. Ventral, C. Lateral view (Stereo microscope).

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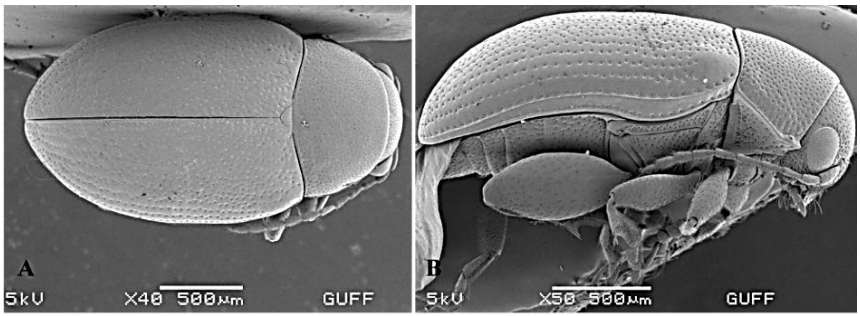

Figure 10. *Chaetocnema aridula* (Gyllenhal, 1827): A. Dorsal, B. Lateral view (SEM).

The Palaearctic (Sibero-Europeo-Mediterranean) + introduced to Oriental species is distributed in Europe (Albania, Austria, Belarus, Belgium, Bosnia & Herzegovina, Bulgaria, Crimea, Croatia, Czeck Republic, Denmark, England, Finland, France, Germany, Greece, Hungary, Italy, Kazakhstan, Latvia, Liechtenstein, Lithuania, Luxemburg, Malta, Moldavia, Montenegro, Netherland, North Macedonia, Norway, Poland, Portugal, Romania, Russia (Central, North and South European Territories), Serbia, Slovakia, Slovenia, Spain, Sweden, Switzerland, Turkey, Ukraine), Asia (Azerbaijan, Cyprus, Iran, Kazakhstan, Kirgizia, Russia (E, S and W Siberia), Turkey), North Africa (Algeria), and introduced to Oriental region (Bezdek & Sekerka, 2024).

The flea beetle is a rather little-known species for Turkey and it has been recorded from 9 of 81 provinces and four of seven regions in Turkey until now as Ankara, Erzurum, İstanbul, Kayseri, Manisa, Sivas, Yozgat provinces in Anatolia (Asian Turkey) and Edirne, Kırklareli provinces in European Turkey (Thrace) (Ekiz et al., 2013; Özdikmen, 2014; the present study) (Fig. 11).

Figure 11. Provincial and regional distribution of *Chaetocnema aridula* (Gyllenhal, 1827) in Turkey.

As known, the structures of aedeagus and spermatheca are taxonomically important. For this reason, to reveal more diagnostic characters than hitherto known, ultrastructural and detailed investigations of aedeagus and spermatheca

of *Chaetocnema aridula* (Gyllenhal, 1827) from Turkey were firstly studied with scanning electron microscope (SEM) and stereo microscope. Obtaining observations are presented below.

Aedeagus (Stereo microscope) (Figs. 12A-C)**:**

Under the stereo microscope, the aedeagus is completely light brown. When viewed from the dorsally and ventrally, the median lobe is narrow at the base and tip and is widest in the middle. The distal width of the aedeagus relative to the basal aperture is equal to the width just before the apical slope. The tooth at the apex of the median lobe is bluntly terminated in the form of a valve when viewed from the dorsally and ventrally, but when viewed from the lateral side, it becomes narrower towards the tip and ends sharply at the tip. When viewed from the lateral side, the aedeagus is in the form of an arch bent ventrally. The apex of the median lobe ends with a tooth structure that extends from the orifice.

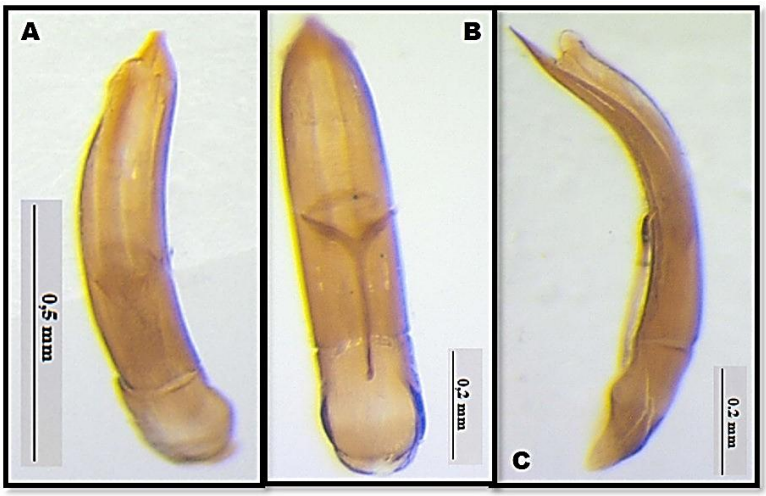

Figure 12. Aedeagus of *Chaetocnema aridula* (Gyllenhal, 1827): A. Dorsal view in stereo microscope, B. Ventral view in stereo microscope, C. Lateral view in stereo microscope.

Aedeagus (SEM)

Ventral view (Figs. 13B)**:**

The median lobe almost completely parallel except for slightly widening laterally in the middle part. Apical part of aedegus gradually narrows towards apex. Apex of median lobe ends with well differentiated, prominent, broad, flat-topped, square-shaped and elongated tooth which appears truncated. Width of aedeagus from distal to basal aperture almost equal to width just before apical slope. Median lobe narrows at base and apex when viewed dorsally and ventrally, widest in middle. Surface of median lobe convex apically, medially and basally from lateral to median groove. Median groove gradually narrows from base to apical part and depth of groove decreases towards apical part. Width of median

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longitudinal groove narrows narrower than distance between groove and lateral margin. Transverse wrinkles are present in median groove from base to middle part of median lobe.

Lateral view (Figs. 13C, 14, 15D)**:**

The median lobe is almost equal and quite distinctly curved towards the ventral side. The apex of the median lobe is quite narrow and pointed. The most curved part of the median lobe is the apical 2/3. A transverse depression is seen between the basal aperture of the median lobe and the middle part (approximately 1/4 of the part close to the base). All parts of the median lobe are seen to have increasingly scattered and sparse ultrastructural sensilla from the base to the apical part. The integrated form of the aedeagus (when we place the lateral image oppositely and symmetrically, the image obtained is suboval in a wide upper and narrowed lower form. Although the aedeagus end parts come together in the integrated form, there is a gap between the base parts.

Dorsal view (Figs. 13A, 15A,B)**:**

The median lobe is almost completely parallel except for being slightly widened laterally in the middle part. The median lobe is convexly curved ventrally from the base to the apex, especially from the middle part. The apical part of the aedegus is gradually narrowed to the apex. The apex of the median lobe ends with a well-differentiated, distinct, wide, flat-topped, square-shaped and elongated tooth that appears to be cut. The dorsal plate has a 3-part structure. While the parts on the sides extend from the apical 1/3 of the median lobe to the apex by narrowing, the single part in the middle, the dorsal plate, starts from almost 1/2 of the median lobe and is almost equally wide at the base, widens towards the apical part and is extended in the form of a tongue and its tip is curved inward. The dorsal plate fuses with the dorsal surface of the middle lobe at the base and divides the orifice into four. The orifice is distinct but narrowed. It is basically U-shaped or wide-angled V-shaped.

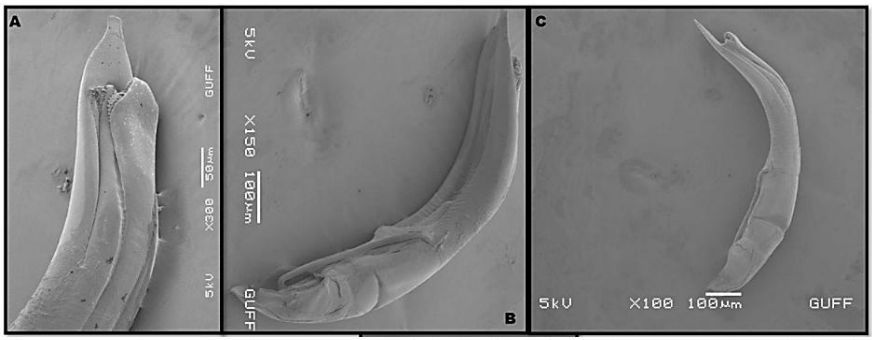

Figure 13. Aedeagus of *Chaetocnema aridula* (Gyllenhal, 1827): A. Dorso-lateral view in SEM, B. Ventro-lateral view in SEM, C. Lateral view in SEM.

Figure 14. Integrated form of aedeagus.

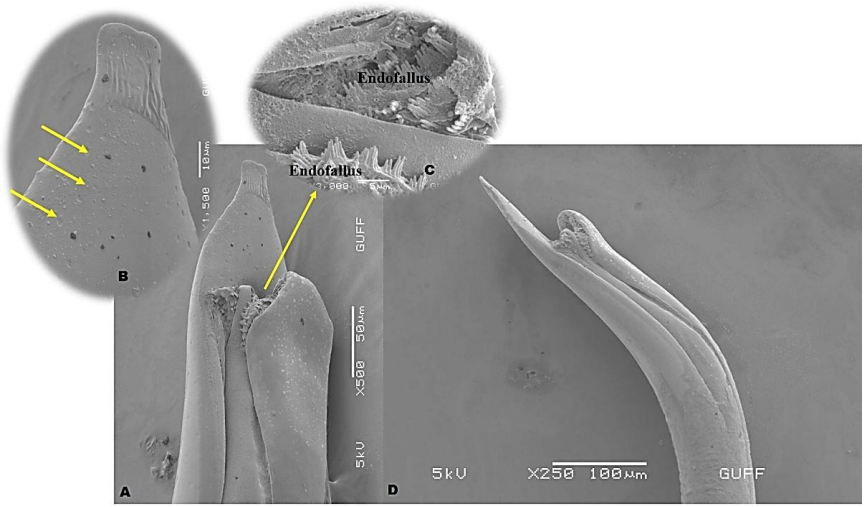

Figure 15. Aedeagus of *Chaetocnema aridula* (Gyllenhal, 1827): A. Apical part and dorsal wall structure of the median lobe, B. Sensillar structure and density on the apical part of the dorsal surface of the median lobe, C. Endophallus structure, D. Apical part of the lateral surface of the median lobe (SEM).

Spermatheca (Stereo microscope) (Fig. 16B)**:**

The spermatheca is generally light brown. The spermathecal receptacle is generally cylindrical, thin and long. The spermathecal receptacle narrows in the middle and widens at the base and apex. The spermathecal receptacle is almost 3 times longer than the pump. The spermathecal pump is curved ventrally, has a duck-head-shaped skin protrusion with a beak-like tip. The spermathecal canal curves and extends to 1/2 of the receptacle.

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Spermatheca (SEM) (Figs. 16A, 17A-C)**:**

The spermathecal receptacle is generally cylindrical, thin and long. The spermathecal pump is curved ventrally. The apex of the spermathecal pump is more or less cylindrical or round. There is a very distinct elongated skin protrusion at the apex of the spermathecal pump. Therefore, the structure at the apex of the pump appears dark brown in the stereo microscope image. This additional integument gives a duckbill appearance at the apex. The spermathecal pump is connected to the middle of the apex of the spermathecal receptacle.

The surface of the spermathecal receptacle is almost uniformly rugulose. The receptacle narrows in the middle and its maximum width is at the base and apex. The spermathecal receptacle is almost the same width from the base to the tip except for the middle constriction. The spermathecal canal is shorter than the receptacle. The spermathecal canal extends to approximately 1/2 of the receptacle. The base part of the spermathecal canal is curved and the apical part is more or less straight. It is connected to the lateral part of the base of the spermathecal receptacle.

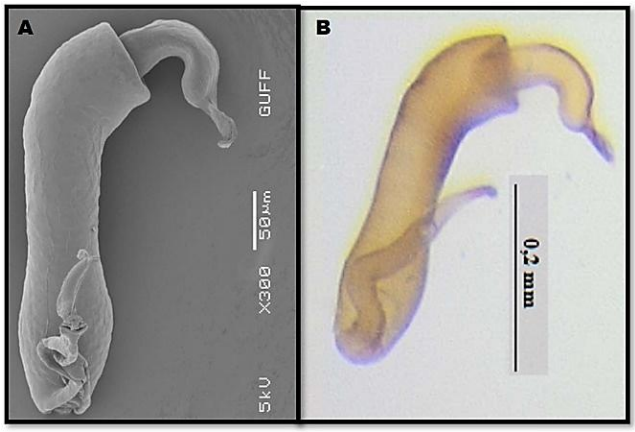

Figure 16. *Chaetocnema aridula* (Gyllenhal, 1827): A. Spermatheca in SEM, B. Spermatheca in stereo microscope.

Figure 17. *Chaetocnema aridula* (Gyllenhal, 1827): A. Spermathecal pump part, B. Spermathecal structure in lateral view, C. Spermathecal receptacle structure and spermathecal canal structure (SEM).

SEM images of aedeagi and spermathecae of *Chaetocnema arenacea* (Allard, 1860) and *Chaetocnema aridula* (Gyllenhal, 1827) are presented to the scientific world with the present study for the first time. When diagnosing specimens belonging to the genus *Chaetocnema* Stephens, 1831, the useful characters in the structure of the aedeagus are general form of aedeagus and apical part of the median lobe chiefly.

As a result of the SEM images obtained with the present study, some new diagnostic characters have been added to these characters: for aedeagus: Dorsal plate in apical part of the median lobe, the structure and density of the sensillae on the median lobe and chitinization rate of aedeagus and the integrated form of the aedeagus (when we place the lateral image mutually and symmetrically), for spermatheca: General form of spermatheca, attaching points to spermathecal receptacle of spermathecal pump and spermathecal duct, and chitinization rate of spermatheca mainly. As a result of all these, the findings of the present study and Ekmekci et al. (2025) fully confirm and support the subgeneric arrangement of the genus *Chaetocnema* by Özdikmen (2021) and the proposed subgenus *Chaetocnema* (*Hortensoides*) Özdikmen, 2021 (Figs. 18, 19).

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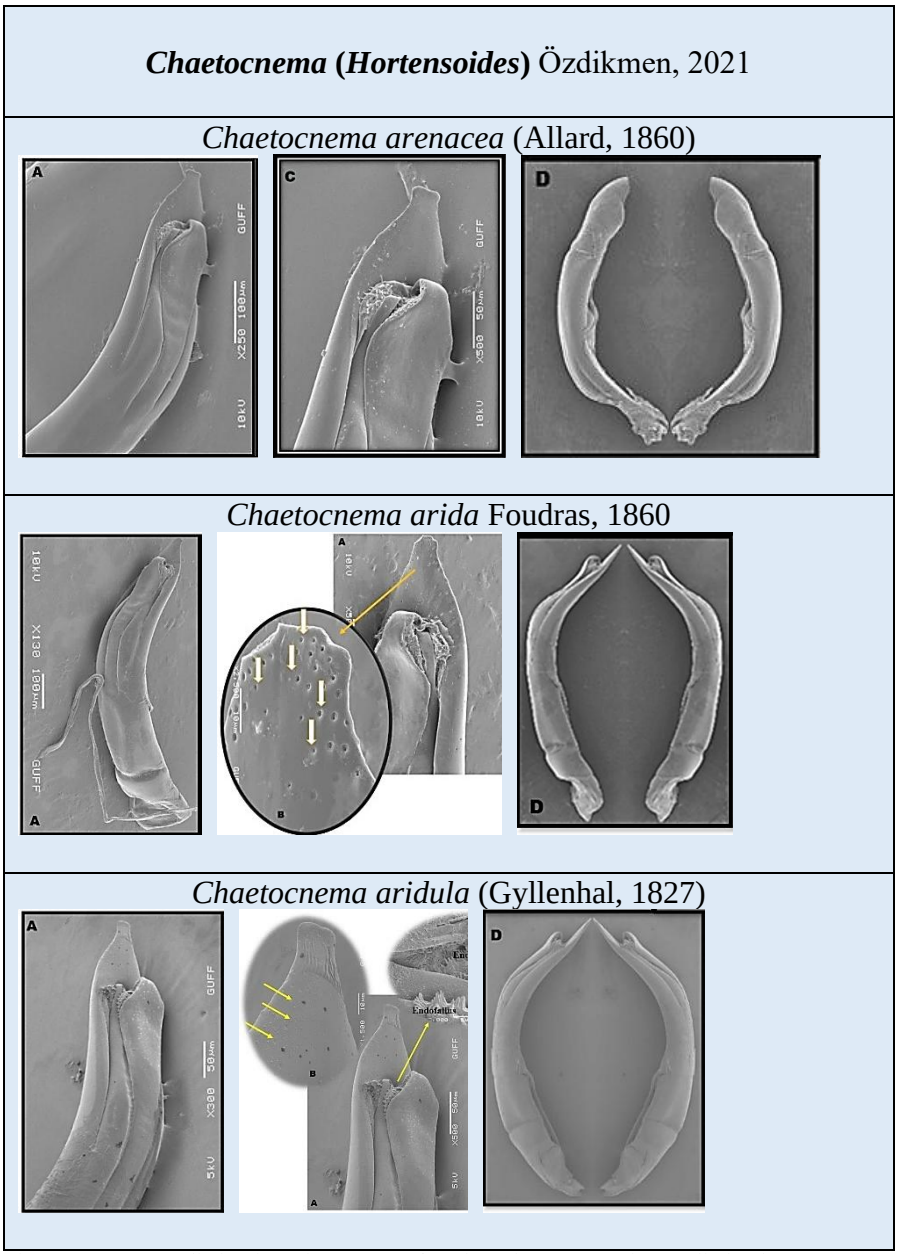

Figure 18. Some distinctive characters of aedeagal structure in studied *Chaetocnema* (*Hortensoides*) Özdikmen, 2021 species.

***Chaetocnema* (*Hortensoides*)** Özdikmen, 2021	
Chaetocnema arenacea (Allard, 1860)	
Chaetocnema arida Foudras, 1860	
Chaetocnema aridula (Gyllenhal, 1827)	

Figure 19. Some distinctive characters of spermathecal structure in studied *Chaetocnema* (*Hortensoides*) Özdikmen, 2021 species.

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