TÜBİTAK

Research Article

Turk J Zool
34 (2010) 1-21
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doi:10.3906/zoo-0806-26

Morphological analyses of some Palaearctic *Stenopogon* Loew, 1847 (Diptera: Asilidae: Stenopogoninae) based on the spermatheca structure

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Received: 26.06.2008

Abstract: Females of some *Stenopogon* species were compared both systematically and phylogenetically utilizing spermatheca morphology. The spermathecae of 8 species and 2 subspecies of *Stenopogon* (*S. coracinus, S. elongatus S. flavibarbis, S. junceus, S. laevigatus, S. nigriventris, S. schisticolor, S. strateagus, S. sabaudus harpax*, and *S. xanthotrichus xanthomelas*) were examined using a scanning electron microscope and subsequently analyzed with cluster analysis. Taxa of evaluated *Stenopogon* were found to be separated into 2 primary groups, A and B, based on the number of spirals and the diameter of the reservoir. Group A further separated into the sub-groups A1, A2, and A3, while group B separated into 2 sub-groups, B1 and B2. Species of both group A and group B were found to be monophyletic.

Key words: Asilidae, cluster analysis, Diptera, morphological structure, SEM, spermatheca, *Stenopogon*

Bazı Palaearktik *Stenopogon* Loew, 1847 (Diptera: Asilidae: Stenopogoninae: Stenopogonini) türlerinin spermateka yapısına dayalı morfolojik analizi

Özet: Bu çalışmada bazı *Stenepogon* türlerinin dişileri sistematik ve filogenetik olarak spermateka morfolojisi yardımı ile değerlendirilmiştir. *Stenepogon*'un sekiz türü ve iki alttürünün (*S. coracinus, S. elongatus S. flavibarbis, S. junceus, S. laevigatus, S. nigriventris, S. schisticolor, S. strateagus, S. sabaudus harpax, S. xanthotrichus xanthomelas*) spermatekaları Taramalı Elektron Mikroskobu kullanarak incelenmiş ve cluster analizi ile değerlendirilmiştir. Değerlendirilen *Stenepogon* türleri spiral sayısı ve rezervuarın çapına göre, A ve B olmak üzere iki ana gruba ayrılmaktadır. A grubu daha sonra A1, A2 ve A3 olmak üzere üç, B grubu ise B1 ve B2 olmak üzere iki alt gruba ayrılmaktadır. Hem A hem de B grubu monofiletiktir.

Anahtar sözcükler: Asilidae, cluster analizi, Diptera, morfolojik yapı, SEM, spermateka, *Stenopogon*

Introduction

There are 3 spermathecae in specimens representing all genera of Asilidae except in some American genera (*Proctacanthus* group), in which there are only 2 (Theodor, 1980). Each spermatheca (Figure 1) has a reservoir at the apex in which sperm

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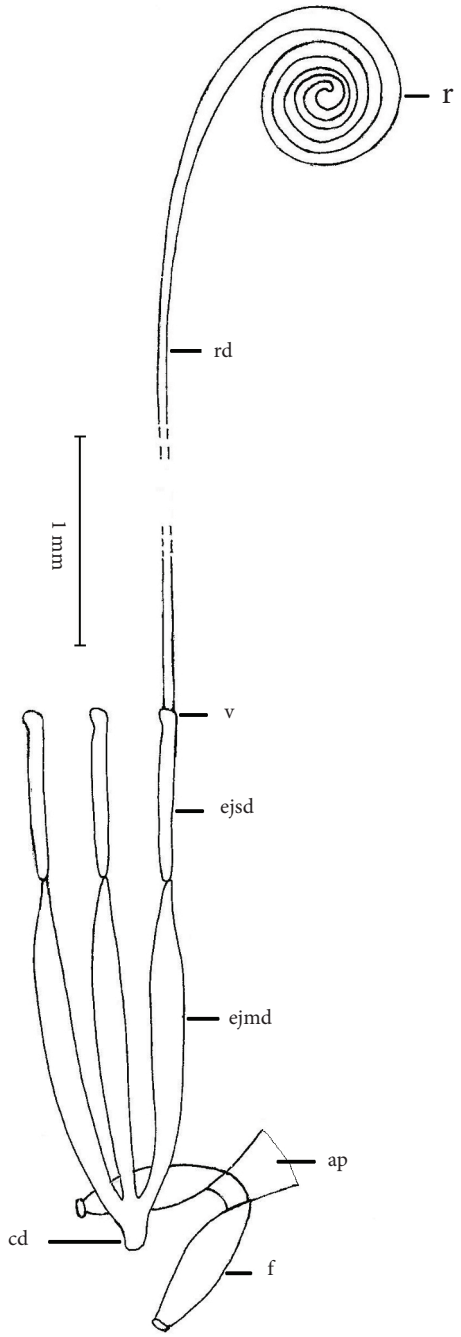

Figure 1. General view of spermatheca, *Stenopogon schisticolor*. (f: furca, cd: common duct, ap: apodeme, ejmd: muscular duct of ejection apparatus, ejsd: sclerotized duct of ejection apparatus, v: valve, rd: reservoir duct, r: reservoir).

is stored. Reservoirs may be more or less sclerotized and have various shapes, such as tube, spiral, oblong, pear, elliptical, and sausage. Reservoirs may have entrances with short or long ducts with various parts showing differentiations. Additionally the valves that control sperm ejection may be of different shapes, such as conical or ring shaped. Common ducts that open to the vagina in the middle of the furca may be long or short. The furca, the modified sternite 9, may be U-V-Y or bar shaped (Theodor, 1980).

Species of *Stenopogon* are large and generally 20-30 mm in length. They can be easily recognized by their long, tapering abdomens, metapleuron with micropubescence, 3rd antennal segment with style, fore tibia without curved spine, and the male genitalia not rotated. Males can be fairly easily separated based on characters of the male genitalia (Efflatoun, 1937; Hull, 1962; Lehr, 1963; Richter, 1963, 1968, 1988; Theodor, 1976, 1980). However, it is difficult to separate females, based on external morphological characteristics.

There are 152 described species of *Stenopogon* worldwide, with the great majority of them occurring in the Holoarctic region, while 63 are Palaearctic and 54 are Nearctic. The remaining species occur in the Oriental (20), Neotropical (12), Afrotropical (2), and Australasian (1) regions (Geller-Grimm, 2008).

The female abdomen of *Stenopogo*n species consists of 8 segments. The 9th sternite forms the furca, while the 9th tergite appears as a dorsal plate. The 10th tergite, which is triangular, is divided into 2 parts, with the acanthophorite bearing 6-8 strong spines (Theodor, 1976, 1980; McAlpine, 1981).

Theodor (1976, 1980) described the spermathecae of 6 *Stenopogon* species found in Israel but only illustrated the furca and spermatheca of *S. elongatus* (Meigen, 1804), using a light microscope. He reported that in general the furca of *Stenopogon* species is U or V shaped and possesses a short apodeme at its apex. The common duct is short and the reservoirs are spiral and narrow.

Artigas and Papavero (1993) have provided illustrations of the spermathecae of some American species in the tribe Stenopogonini. They showed that the reservoir of *Stenopogon inguinatus* Loew extends to the middle of the 6th abdominal segment, the furca is V shaped, the common duct is short, and the reservoir is curled into a spiral shape.

Investigations into the phylogeny of Asilidae have been carried out by various researchers. These studies are generally based on external morphological characters (Karl, 1959; Hull, 1962; Lehr, 1969, 1996; Papavero, 1973; Oldroyd, 1974; Cannings, 2002). In only one study was the phylogeny of Asilidae based on molecular evidence and that was only to the subfamily level (Bybee et al., 2004). Phylogenetic studies at the generic level have been done only for species in other subfamilies, such as those for Dasypogoninae: *Blepharepium* (Papavero and Bernardi, 1973), and Apocleinae: *Mallophora* (Artigas and Angulo, 1980), and Ommatiniae: *Ommatius* (*pumilus* group) (Scarbrough, 1990).

The aim of the present study was to characterize some species of *Stenopogon* found in Turkey on the basis of the morphology of the female spermatheca and analyzed using cluster analysis based on spermathecae morphology.

Materials and methods

The species of *Stenopogon* (*S. coracinus*, *S. elongatus S. flavibarbis*, *S. junceus*, *S. laevigatus, S. nigriventris, S. schisticolor*, *S. strateagus, S. sabaudus harpax*, and *S. xanthotrichus xanthomelas*) used in this study were collected from different regions of Turkey and are preserved in the Zoological Museum of Gazi University (ZMGU), Ankara, Turkey. Females were relaxed in the softening jar. After that their abdomens were removed and macerated in 5% KOH for 24 h at 25 °C. The abdomens were opened and the spermathecae were removed. The spermathecae were washed with distillated water and the remaining tissues were removed by forceps.

Cleaned spermathecae were stained with methylene blue and photographed with a digital camera mounted on a stereomicroscope. To provide standardization only impregnated females were studied. Spermathecae were mounted using double-sided tape on SEM stubs, coated with gold in a Polaron SC 502 sputter coater, and examined with a JEOL JSM 5600 scanning electron microscope at 15 kW.

Cluster analysis

Selected morphological features of spermathecae were designated as operational taxonomic units (OTUs). These included 9 quantitative and 13 qualitative characters plus 75 character states that were chosen for the cluster analysis (Table). Data were entered into a spreadsheet program, Microsoft Excel, version 7. The spreadsheet was subsequently transformed into a file format suitable for cluster analysis. Cluster analysis was carried out using Multivariate Statistical Package (MVSP) version 3.13 m, with a matrix of standardized data. The data were standardized to eliminate the distorting effects of different character states on the output results. For cluster analyses, an unweighted pair group method using arithmetic averages (UPGMA) was used. This clustering algorithm was performed on a Gower general similarity coefficient, resulting in a phenogram depicting similarity between the OTUs.

Results

The general morphology and the fine structure of the spermathecae of 8 species and 2 subspecies of *Stenopogon* were examined and analyzed using cluster analysis based on spermatheca morphology.

Spermathecae may extend from between the 5th and 6th abdominal segments to the middle of the 3rd abdominal segment. Furca are sclerotized plates in a U (*S. nigriventris, S. xanthotrichus xanthomelas, S. junceus,* and *S. elongatus*) or V (*S. coracinus, S. schisticolor, S. strataegus, S. sabaudus harpax, S. flavibarbis*, and *S. laevigatus*) form. Common ducts of all examined species were short and widely open to the genital chamber. The portion surrounded by a layer of muscles of the ejection apparatus, which ejects sperm, may be of different sizes. The distal part of the ejection apparatus is strongly sclerotized and its surface corrugated, gnarled or striate. Moreover, the surface of the sclerotized portion of the ejection apparatus is covered with canaliculi of different density and size. Valves, which control the passing of sperm, are conical or ring shaped. Reservoir ducts may be very long and strongly sclerotized at the beginning; this sclerotization is reduced close to the reservoir in *S. coracinus, S. schisticolor, S. flavibarbis, S. laevigatus, S. junceus*, and *S. elongatus*. The surface of the strongly sclerotized parts of the ducts may be corrugate or striate. In some species sclerotization may increase on the close to the reservoir and in some

Table. List of characters and character states used in the cluster analysis.

Characters	State
Number of spines on acanthophorites	0 = 8
	1 = 7
Shape of projection of dorsal plate	0 = Narrow
	1 = Wide
Position of spermatheca in the abdomen	0 = middle of 3rd segment
	1 = between 3rd and 4th segments
	2 = middle of 4th segment
	3 = middle of 5th segment
	4 = between 5th and 6th segments
	5 = middle of 6th segment
General shape of furca	0 = forceps
	1 = V shaped
	2 = U shaped
Proportion of Apodeme base to the furca base	0 = same
	1 = narrow
Proportion of Apodeme length to furca length	0 = 1/2
	1 = 1/3
	2 = 1/4
	3= 1/8
Shape of Apodeme	0 = rectangular
	1 = wine glass
Shape of groove between furca arms	0 = Narrow U
	1 = Wide U
	2 = V
Proportion of portion surrounded by a layer of muscles of ejection apparatus to sclerotized portion of ejection apparatus	0 = 1 times
	1 = 0.5 times
	2 = 2 times
	3 = 1.5 times
Diameter of sclerotized portion of ejection apparatus	0 = 40-60 μm
	1 = 61-80 μm
	2 = 80-100 μm
Surface of sclerotized portion of ejection apparatus	0 = transverse gnarled
	1 = irregular corrugated
	2 = longitudinally undulate
	3 = transverse corrugated
Shape of valves	0 = conical
	1 = thick ring
	2 = narrow ring
Diameter of valves	0= 70-80 μm
	1 = 81-90 μm
	2 = 91-100 μm
	3 = 101-110 μm
	4 = 111-120 μm
Sclerotization of reservoir ducts	0 = weakly sclerotized

Table 1. (Continued).

Characters	State
	1 = strongly sclerotized
Diameter of reservoir ducts	0 = 35-40 µm
	1 = 41-45 µm
	2 = 46-50 µm
	3 = 51-55 µm
	4 = 56-60 µm
	5 = 61-65 µm
Surface of reservoir ducts	0 = transverse undulate
	1 = longitudinally undulate
Density of canaliculi on reservoir ducts	0 = sparse
	1= dense
Shape of reservoirs	0 = elliptical
	1 = circular
Number of reservoir spirals	0 = spirals more than 3
	1 = spiral number 3
Diameter of beginning of reservoirs	0 = 15-20 µm
	1 = 21-25 µm
	2 = 26-30 µm
	3 = 31-35 µm
	4 = 61-65 µm
	5 = 76-80 µm
Surface of reservoirs	0 = porous
	1 = smooth
	2 = reticulate
	3 = punctate
	4 = regular spiral
	5 = irregular spiral
Sperm openings	0 = not visible
	1 = visible

species sclerotization may increase suddenly. Reservoirs may be spiral, formed by coiled tubes. These spirals are elliptically or circularly coiled. The numbers of spirals vary from 3 to 5. In some species, because spirals are intermingled, it is hard to separate one reservoir from another. In all examined species reservoirs are strongly sclerotized, their surfaces may be with regular (*S. junceus* and *S. elongatus*) or irregular (*S. laevigatus, S. nigriventris, S. sabaudus harpax,* and *S. xanthotrichus xanthomelas*) sclerotized cords, or may be more or less porous (*S. coracinus*), reticulate (*S. schisticolor*), smooth (*S. flavibarbis*), or punctuate (*S. strataegus*). Sperm were detected in the reservoirs of all species and found in the pores or openings formed by broadening of cords of sclerotized spirals.

***Stenopogon coracinus* (Loew, 1847)**

Character States: Ovipositor bears 8 spines on the acanthophorites. Projection of dorsal plate wide at the apex, ends globular (Figure 2A). Spermatheca extends to the middle of 4th abdominal segment. Furca V shaped, its arms swollen at the base, tapering to apex. Apodeme rectangular, its base as wide as furca base. Proportion of apodeme length to furca length is 1/4. Groove between furca arms narrow, rectangular (Figure 2B). Common duct is short. Portion surrounded by a layer of muscles of ejection apparatus approximately as long as sclerotized portion of ejection apparatus. Diameter of sclerotized portion of ejection apparatus is 43 µm, its surface transverse gnarled and covered with small, sparse canaliculi (Figure 2C). Valve short, conical, approximately 70

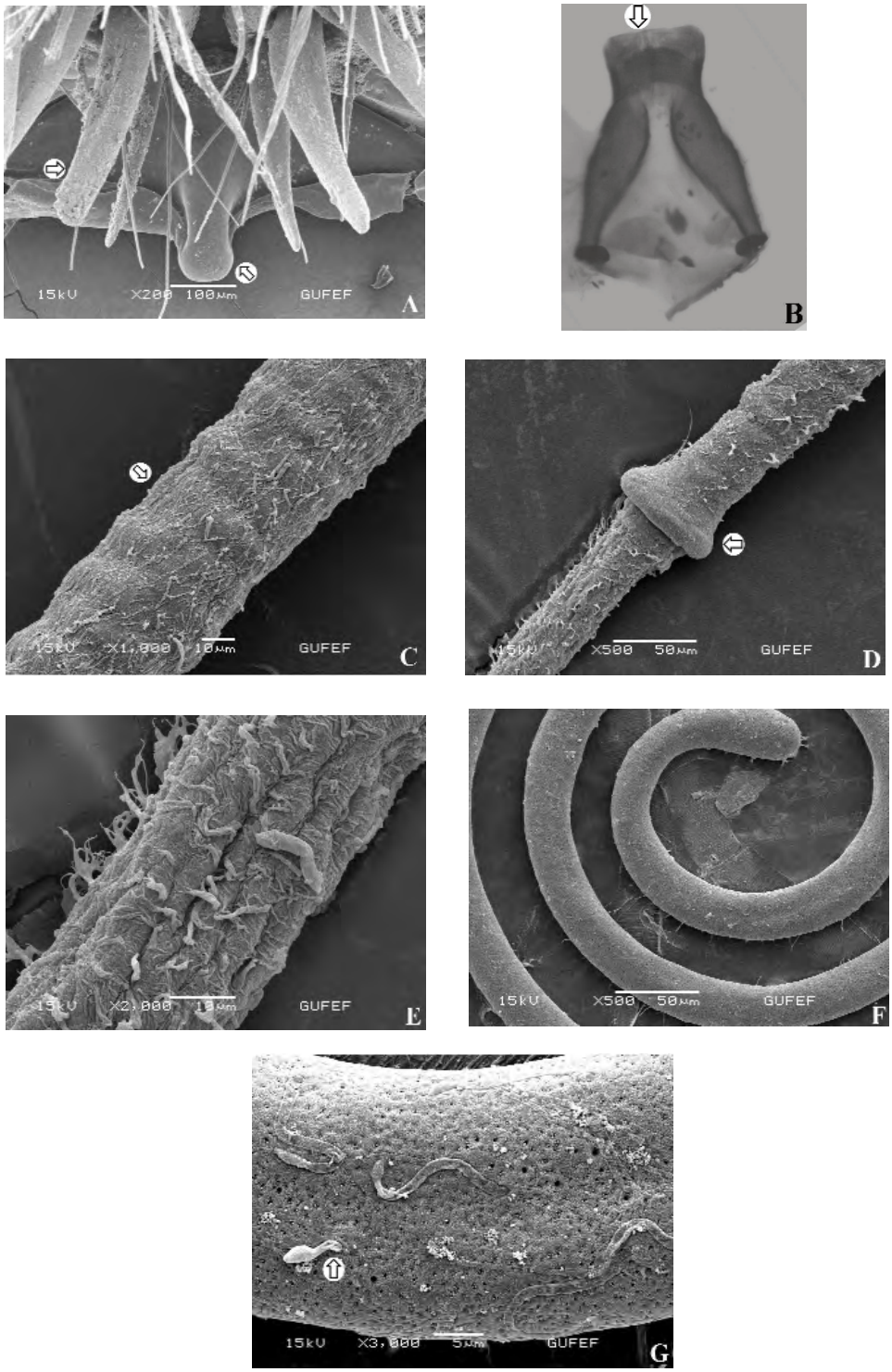

Figure 2. Spermatheca of *S. coracinus*. A. SEM micrograph view of dorsal plate, B. Furca (light microscope view), C. SEM micrograph of sclerotized duct of ejection apparatus, D. SEM micrograph of valve, E. SEM micrograph of reservoir duct, F. SEM micrograph of reservoir, G. SEM micrograph of reservoir. (Symbols: spine of acanthophores ⇨, projection of dorsal plate ⇖, apodeme ⇩, valve ⇦, sperm and opening ⇧).

μm in diameter (Figure 2D). Reservoir ducts weakly sclerotized, becoming less so as they approach the reservoir, 36 μm in diameter. Surface of sclerotized parts of the ducts longitudinally striate and covered with dense, wide canaliculi (Figure 2E). Individual reservoir is circular spiral with 3 thick coiled tubes and almost 530 μm in diameter. Each reservoir tube approximately 30 μm in diameter, strongly sclerotized (Figure 2F) and surface with densely porous with sparse, irregular dispersed sperm openings (Figure 2G).

***Stenopogon flavibarbis* Enderlein, 1934**

Character States: Ovipositor bears 7 spines on the acanthophorites. Projection of dorsal plate short, tapering towards the apex slightly, apex extends backwards (Figure 3A). Spermatheca extends to the middle of 6th abdominal segment. Furca forcep arms are tapering to apex slightly, apex with blunt tip sloping inwards. Apodeme wine glass shaped, its base as wide as furca base. Proportion of apodeme length to furca length is 1/3. Groove between furcal arms V shaped (Figure 3B). Common duct short and wide. Portion surrounded by layer of muscles of ejection apparatus half as long as sclerotized portion of ejection apparatus. Diameter of sclerotized portion of ejection apparatus is 45 μm, its surface irregularly corrugated and covered with small, dense canaliculi (Figure 3C). Valve short, conical, approximately 75 μm in diameter. Reservoir ducts quite long, weakly sclerotized, this sclerotization reduced close to the reservoir, 43 μm in diameter (Figure 3D). Surface of the sclerotized parts of the ducts longitudinally striate and covered with dense, long canaliculi (Figure 3E). Individual reservoir is circular spiral with 3 thick coiled tubes and these spirals almost 580 μm in diameter. Approximately 35 μm diameter reservoir tubes strongly sclerotized (Figure 3F) with smooth surface and sperm openings sparse and clearly visible (Figure 3G).

***Stenopogon schisticolor* Gerstaecker, 1861**

Character States: Ovipositor bears 4 spines on each acanthophorite. Projection of dorsal plate short, tapering towards the apex, extends backwards (Figure 4A). Spermatheca extends to the middle of 5th abdominal segment. Furca V shaped, its arms narrow elliptical, tapering to apex slightly, apex blunt. Apodeme quite long, its base as wide as furcal base, widening like a wine glass towards to the apex. Proportion of apodeme length to furca length is 1/2. Groove between furca arms narrow rectangular (Figure 4B). Common duct short and wide. Portion surrounded by a layer of muscles of ejection apparatus 2 times longer than sclerotized portion. Diameter of sclerotized portion of ejection apparatus is 60 μm, its surface corrugated and covered with dense canaliculi (Figure 4C). Valve narrow, ring shaped, approximately 80 μm in diameter (Figure 4D). Reservoir ducts quite long, weakly sclerotized, this sclerotization decreases close to the reservoir and 45 μm in diameter. Surface of sclerotized portion of the ducts longitudinally striate and covered with dense, fine, long canaliculi (Figure 4E). Individual reservoir circular, almost 600 μm in diameter, spiral with 3 thick coiled tubes. Each reservoir tube, approximately 40 μm diameter, strongly sclerotized (Figure 4F) with reticulate surface and numerous sperm openings clearly visible (Figure 4G).

***Stenopogon strataegus* Gerstaecker, 1861**

Character States: Ovipositor bears 4 spines on each acanthophorite. Projection of dorsal plate extends at the apex, ends truncated tip (Figure 5A). Spermatheca extends to the middle of 5th abdominal segment. Furca broadly V shaped, its arms narrow, elliptical, tapering to apex slightly, apex blunt. Apodeme rectangular, quite long, its base as wide as furca base, widening at the apex. Proportion of apodeme length to furca length is 1/3. Groove between furca arms wide, rectangular (Figure 5B). Common duct is short and wide. Portion surrounded by a layer of muscles of ejection apparatus 2 times longer than sclerotized portion of ejection apparatus. Diameter of sclerotized portion of ejection apparatus is 50 μm, its surface densely pilose and covered with dense papilla like canaliculi (Figure 5C). Valve short, conical, approximately 75 μm in diameter (Figure 5D). Reservoir ducts quite long, strongly sclerotized, 45 μm in diameter. Surface of the sclerotized parts of the ducts longitudinally undulate and covered with dense, canaliculi (Figure 5E). Individual reservoir circular, almost 500 μm in diameter, spiral with 3 thick coiled tubes; each reservoir tube 65 μm in diameter at its base, its diameter decreasing to 30 μm towards apex (Figure 5F) with punctuate surface but the numerous

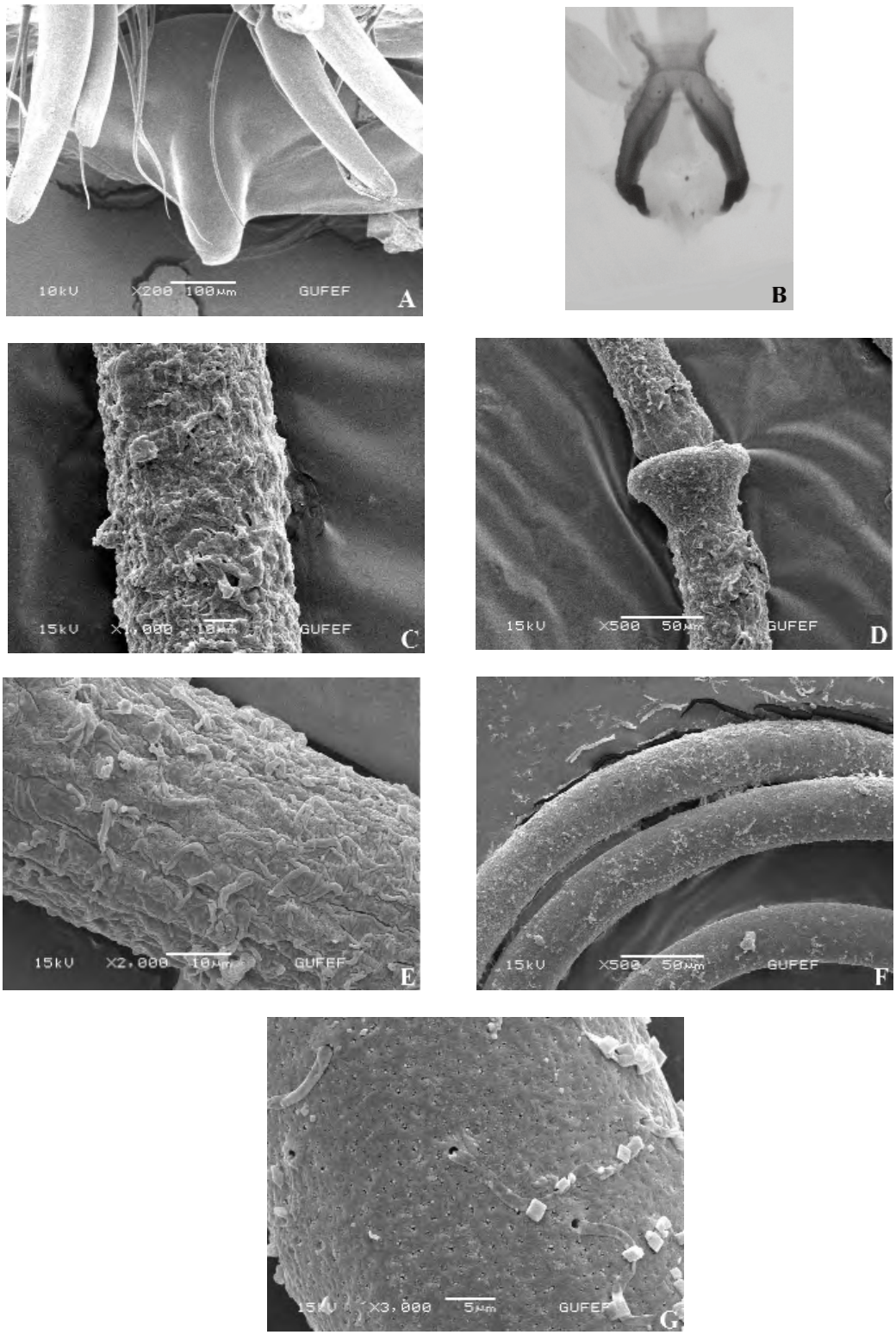

Figure 3. Spermatheca of *S. flavibarbis*. A. SEM micrograph view of dorsal plate, B. Furca (light microscope view), C. SEM micrograph of sclerotized duct of ejection apparatus, D. SEM micrograph of valve, E. SEM micrograph of reservoir duct, F. SEM micrograph of reservoir, G. SEM micrograph of reservoir.

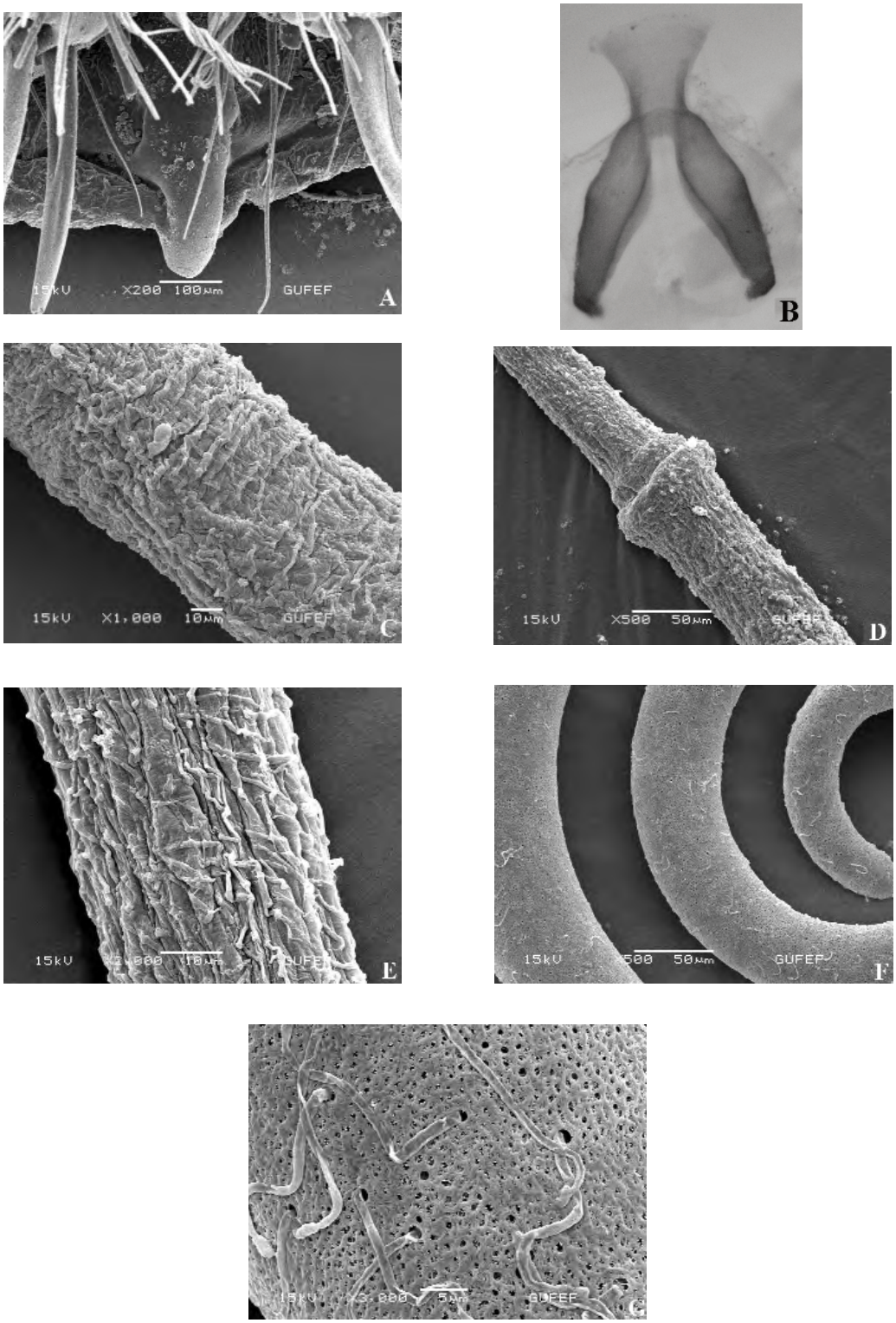

Figure 4. Spermatheca of *S. schisticolor*. A. SEM micrograph view of dorsal plate, B. Furca (light microscope view), C. SEM micrograph of sclerotized duct of ejection apparatus, D. SEM micrograph of valve, E. SEM micrograph of reservoir duct, F. SEM micrograph of reservoir, G. SEM micrograph of reservoir.

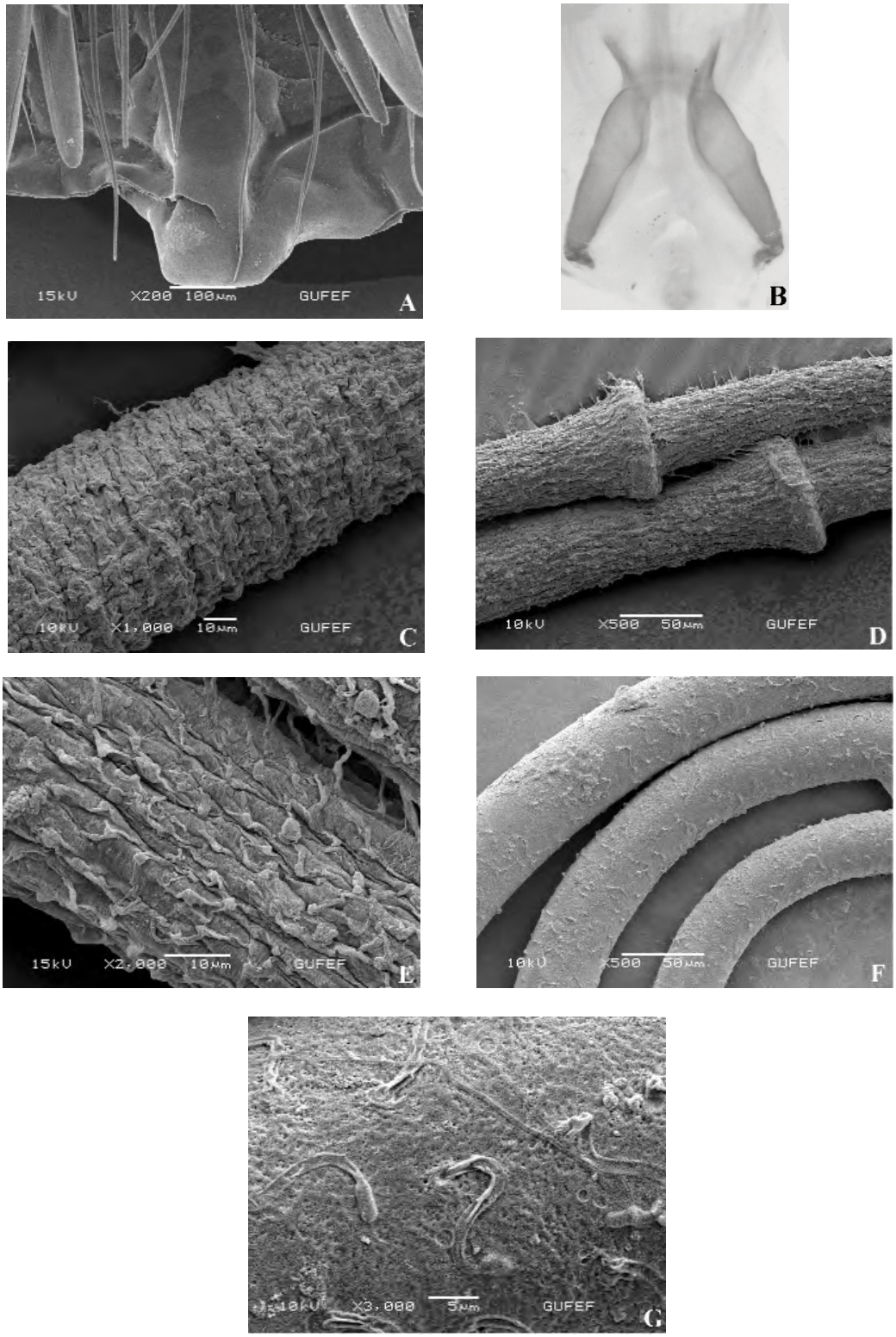

Figure 5. Spermatheca of *S. strataegus*. A. SEM micrograph view of dorsal plate, B. Furca (light microscope view), C. SEM micrograph of sclerotized duct of ejection apparatus, D. SEM micrograph of valve, E. SEM micrograph of reservoir duct, F. SEM micrograph of reservoir, G. SEM micrograph of reservoir.

sperm openings are less clearly visible than those in *Stenopogon schisticolor* (Figure 5G).

***Stenopogon laevigatus* (Loew, 1851)**

Character States: Ovipositor bears 4 spines on each acanthophorite. Projection of dorsal plate extends like a tongue at the apex (Figure 6A). Spermatheca extends to area between the 5th and 6th abdominal segments. Furca forceps aperture shaped, its arms are nearly equally wide from basal to distal, and tapered suddenly at the apex. Apodeme short, rectangular, its base as wide as furca base. Proportion of apodeme length to furca length is 1/4. Groove between furca arms wide, rectangular (Figure 6B). Common duct is short. Portion surrounded by a layer of muscles of ejection apparatus 1.5 times longer than sclerotized portion of ejection apparatus. Diameter of sclerotized portion of ejection apparatus is 60 µm, its surface transversely corrugated up to the valves and covered with a few canaliculi (Figure 6C). Valve thick, ring shaped, approximately 75 µm in diameter (Figure 6D). Reservoir ducts quite long, weakly sclerotized, 46 µm in diameter, and sclerotization reduced upon their approach to the reservoir. Surface of the sclerotized parts of the ducts longitudinally undulate and covered with sparse and short canaliculi (Figure 6E). Individual reservoir elliptical with many coiled tubes in the abdomen. Width of the reservoir is 0.7 mm and length is 1.3 mm. Approximately 20 µm diameter strongly sclerotized reservoir tubes (Figure 6F), with irregular sclerotized cords on surface, sperm openings not clearly visible, sperms found in the openings formed by broadening of cords (Figure 6G).

***Stenopogon nigriventris* Loew, 1868**

Character States: Ovipositor bears 7 spines on the acanthophorites. Projection of dorsal plate tapering from basal to apex, ends obtuse (Figure 7A). Spermatheca extends to the middle of the 5th abdominal segment. Furca U shaped, its arms swollen at the base, and tapered straight to the apex. Apodeme very short, rectangular, its base narrower than furca base. Proportion of apodeme length to furca length is 1/8. Groove between furca arms deep, wide, rectangular (Figure 7B). Common duct is short. Portion surrounded by a layer of muscles of ejection apparatus 1.5 times longer than sclerotized portion of ejection apparatus. The diameter of sclerotized portion of the ejection apparatus is 60 µm, its surface longitudinally striate and covered with sparse, short canaliculi (Figure 7C). Valve thick, ring shaped, approximately 85 µm in diameter (Figure 7D). Reservoir ducts are quite long and strongly sclerotized, 40 µm in diameter. The surface of the sclerotized portion of the ducts is longitudinally striate and covered with sparse, papilla shaped canaliculi (Figure 7E). The individual reservoir is elliptical containing many coiled tubes. The width of the reservoir is 0.8 mm and its length is 1.8 mm. Each reservoir tube is strongly sclerotized (Figure 7F), approximately 20 µm in diameter, surface with irregular sclerotized cords, sperm openings not clearly visible, sperms found in the openings formed by broadening of cords (Figure 7G).

***Stenopogon sabaudus harpax* Loew, 1868**

Character States: Ovipositor bears 7 spines on the acanthophorites. Projection of the dorsal plate long, base and apex are equal in width, apex of projection blunt (Figure 8A). Spermatheca extends to the middle of the 5th abdominal segment. Furca V shaped, its arms swollen at the base, and tapered towards apex, ends blunt. Apodeme long, wine glass shaped, its base as wide as furca base. Proportion of apodeme length to furca length is 1/4. Groove between furca arms deep, wide, rectangular (Figure 8B). Common duct is short and narrow. Portion surrounded by a layer of muscles of ejection apparatus approximately 1.5 times longer than sclerotized portion of ejection apparatus. Diameter of transverse, thick, gnarled sclerotized portion of ejection apparatus is 70 µm, which is covered with sparse, fine, long canaliculi (Figure 8C). Valve thick, ring shaped, approximately 85 µm in diameter (Figure 8D). Reservoir ducts quite long, weakly sclerotized, 35 µm in diameter. Surface of the sclerotized parts of the ducts longitudinally striate and covered with sparse, thick and long canaliculi (Figure 8E). Individual reservoir is elliptical with many coiled tubes. Width of the reservoir is 0.65 mm and length is 2.3 mm (Figure 8F). Approximately 25 µm diameter reservoir tubes strongly sclerotized, surface with irregular sclerotized cords, sperm openings not clearly visible (Figure 8G).

***Stenopogon xanthotrichus xanthomelas* Loew, 1868**

Character States: Ovipositor bears 7 blunt spines on the acanthophorites. Projection of dorsal plate

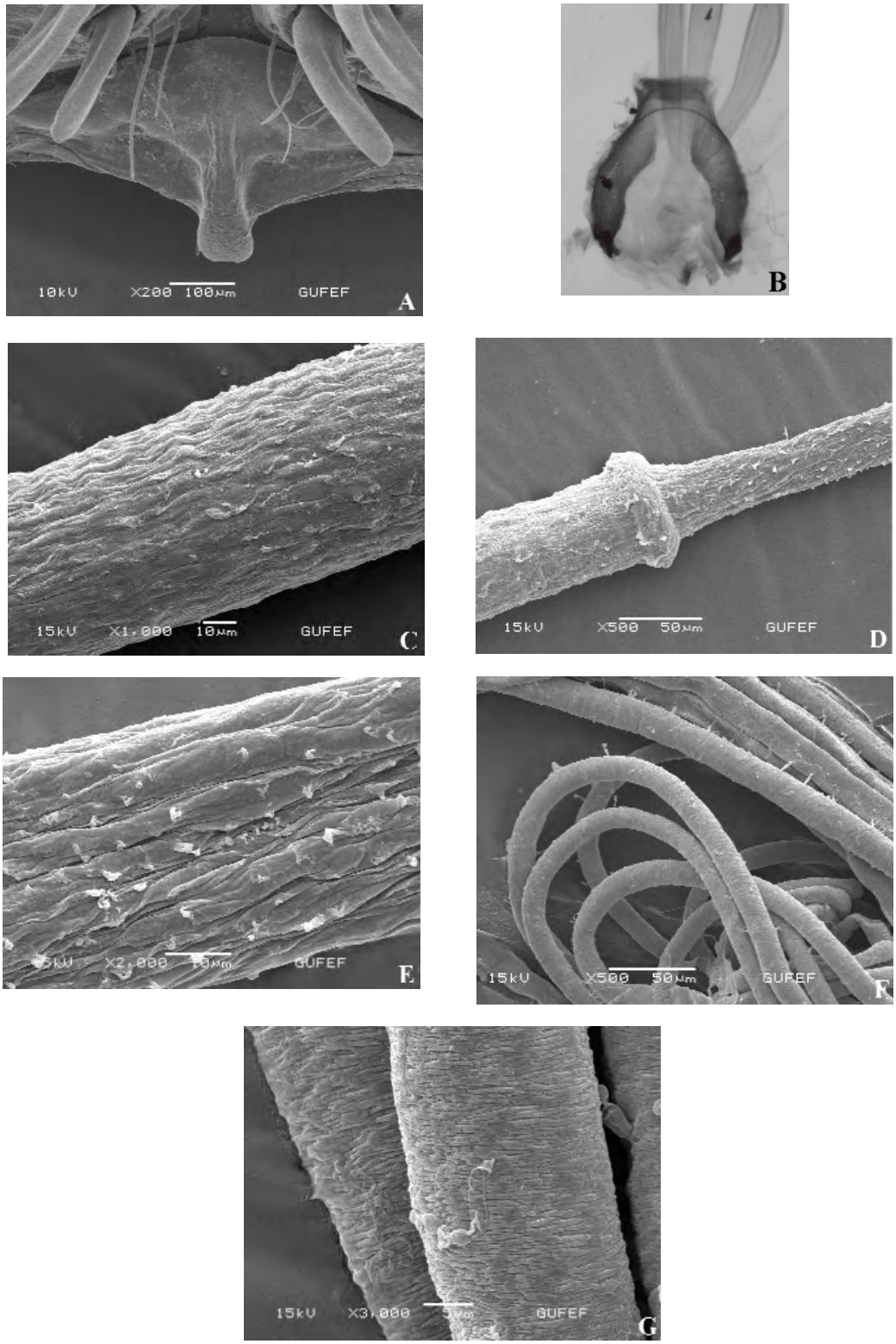

Figure 6. Spermatheca of *S. laevigatus*. A. SEM micrograph view of dorsal plate, B. Furca (light microscope view), C. SEM micrograph of sclerotized duct of ejection apparatus, D. SEM micrograph of valve, E. SEM micrograph of reservoir duct, F. SEM micrograph of reservoir, G. SEM micrograph of reservoir.

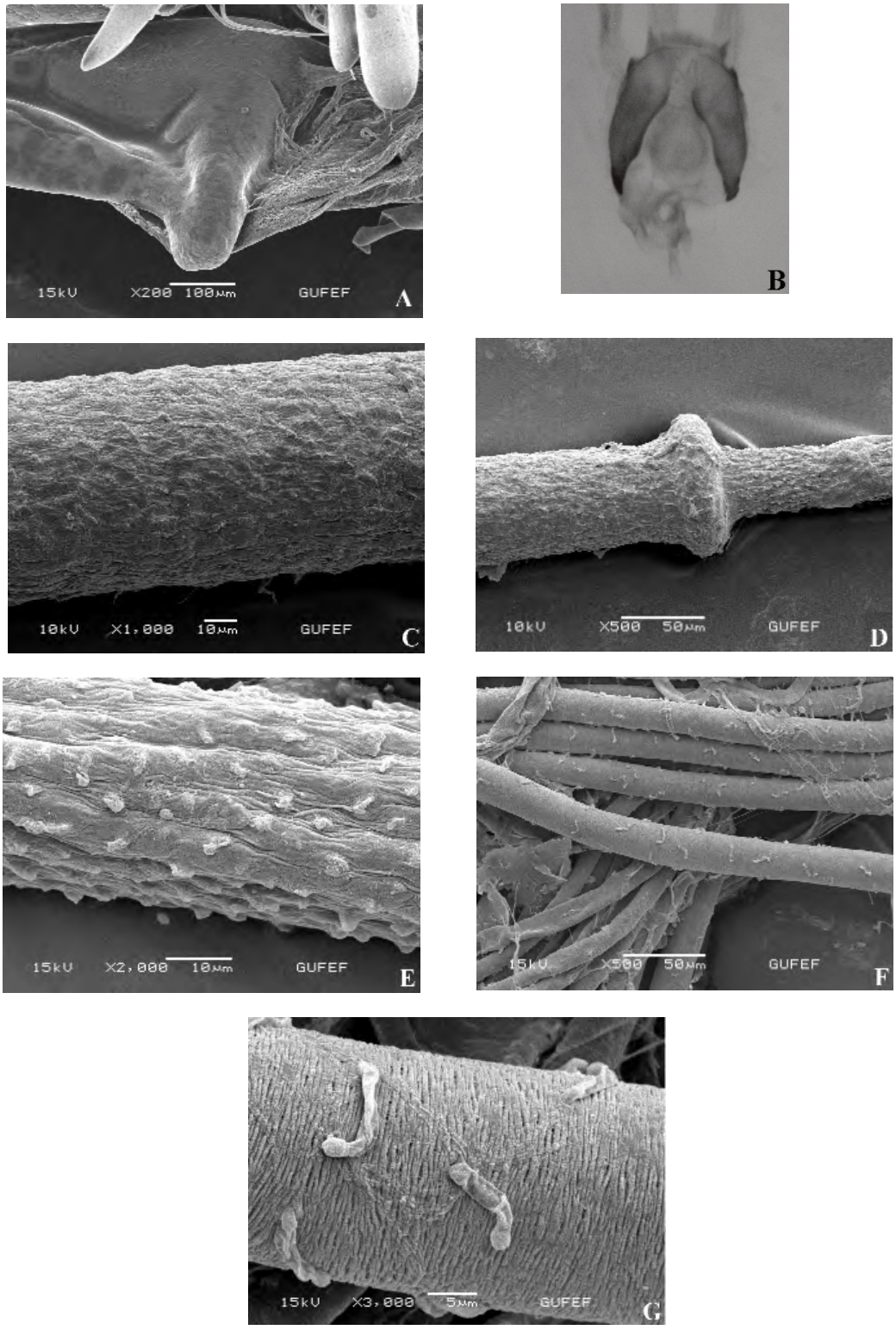

Figure 7. Spermatheca of *S. nigriventris*. A. SEM micrograph view of dorsal plate, B. Furca (light microscope view), C. SEM micrograph of sclerotized duct of ejection apparatus, D. SEM micrograph of valve, E. SEM micrograph of reservoir duct, F. SEM micrograph of reservoir, G. SEM micrograph of reservoir.

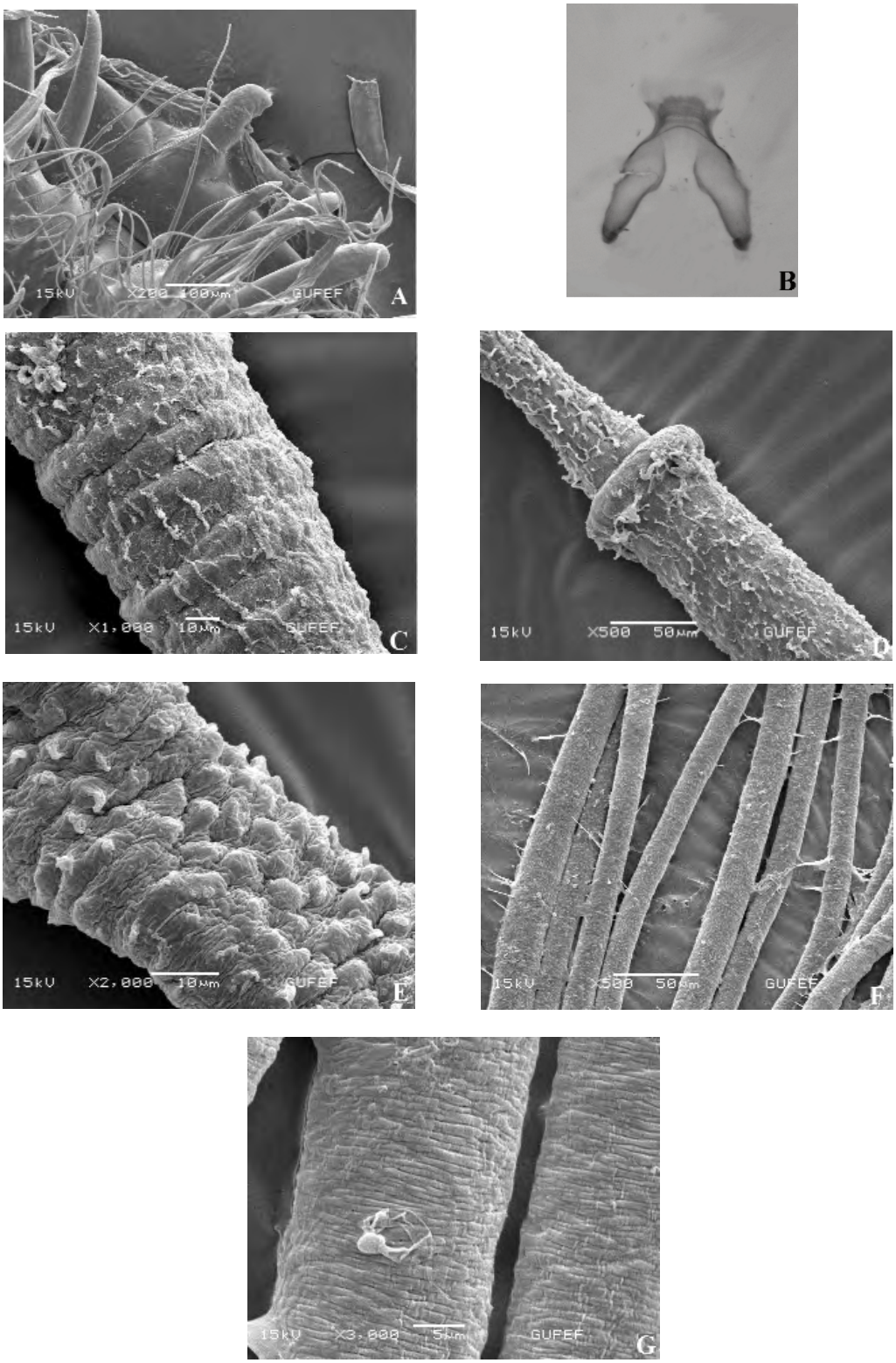

Figure 8. Spermatheca of *S. sabaudus harpax*. A. SEM micrograph view of dorsal plate, B. Furca (light microscope view), C. SEM micrograph of sclerotized duct of ejection apparatus, D. SEM micrograph of valve, E. SEM micrograph of reservoir duct, F. SEM micrograph of reservoir, G. SEM micrograph of reservoir.

wide at the base, narrow at the middle, tapering to the apex and ends with rounded apex (Figure 9A). Spermatheca extends to the middle of the 5th abdominal segment. Furca U shaped, its arms swollen at the base, and tapering towards to the apex, ends sharp. Apodeme slightly wide, rectangular, widening towards to distal, its base as wide as 2/3 width of furca at base. Proportion of apodeme length to furca length is 1/4. Groove between furca arms short, slightly wide, rectangular (Figure 9B). Common duct is short and wide. Portion surrounded by a layer of muscles of ejection apparatus approximately 2 times longer than sclerotized portion of ejection apparatus. Diameter of transverse corrugated sclerotized portion of ejection apparatus is 100 µm, which is covered with numerous papillae like canaliculi (Figure 9C). Valve slightly thickened, ring shaped, approximately 100 µm in diameter (Figure 9D). Reservoir ducts quite long, weakly sclerotized, 65 µm in diameter. Surface of the sclerotized parts of the ducts slightly transverse striate and covered with sparse and with long canaliculi (Figure 9E). Reservoirs are elliptical with many coiled tubes and 1.4 mm in diameter (Figure 9F). Approximately 16 µm diameter reservoir tubes strongly sclerotized, surface with irregular sclerotized cords, sperm openings not clearly visible (Figure 9G).

***Stenopogon junceus* (Wiedemann in Meigen, 1820)**

Character States: Ovipositor bears 7 spines on the acanthophorites. Projection of dorsal plate narrows at the middle, apex of projection wide (Figure 10A). Spermatheca extends between 3rd and 4th abdominal segments. Furca U shaped, its arms swollen at the base, and is tapered straight to the apex, ends sharp. Apodeme rectangular shaped, its base as wide as half of width of base of furca, slightly wider at the distal end. Proportion of apodeme length to furca length is 1/4. Groove between furcal arms V shaped (Figure 10B). Common duct is short. Portion surrounded by a layer of muscles of ejection apparatus approximately as long as sclerotized portion of ejection apparatus. The diameter of irregular corrugated sclerotized portion of ejection apparatus is 68 µm and covered with sparse, short canaliculi (Figure 10C). Valve wide conical, approximately 115 µm in diameter (Figure 10D). Reservoir ducts quite long, weakly sclerotized, 62 µm in diameter, sclerotization reduced on approach to the reservoir. Surface of the sclerotized parts of the ducts longitudinally striate and covered with a few and short canaliculi (Figure 10E). Reservoirs are elliptical with many coiled tubes in the abdomen, and 950 µm in diameter. Approximately 25 µm diameter reservoir tubes strongly sclerotized (Figure 10F), surface with regular sclerotized cords, sperm openings not clearly visible, sperms found in the openings formed by broadening of foldings (Figure 10G).

***Stenopogon elongatus* (Meigen, 1804)**

Character States: Ovipositor bears 7 spines on the acanthophorites. Projection of dorsal plate narrow, long, tapering to the apex (Figure 11A). Spermatheca extends to the middle of the 3rd abdominal segment. Furca U shaped, its arms swollen at the base, and tapering straight to the apex, ends blunt. Apodeme short, rectangular, widening towards distal end, its base wide as 1/3 of furca base. Proportion of apodeme length to furca length is 1/4. Groove between furca arms short, wide U shaped (Figure 11B). Common duct is short and wide. Portion surrounded by a layer of muscles of ejection apparatus approximately half of sclerotized portion of ejection apparatus. The diameter of sclerotized portion of ejection apparatus is 40 µm; fine longitudinally corrugated and covered with sparse and short canaliculi (Figure 11C). Valve conical, approximately 100 µm in diameter (Figure 11D). Reservoir ducts quite long, weakly sclerotized, 42 µm in diameter. Surface of sclerotized parts of the ducts slightly undulate and covered with sparse and short canaliculi (Figure 11E). Reservoirs are circular with many coiled tubes in the abdomen and 675 mm in diameter. Approximately 16 µm diameter reservoir tubes strongly sclerotized (Figure 11F), surface with regular sclerotized cords, sperm openings not clearly visible, sperms found in the openings formed by broadening of cords (Figure 11G).

The cluster analysis separated these taxa into 2 primary groups, A and B, on the basis of number of spirals and diameter of the reservoir. Group A, with 3 coiled reservoir tubes, included *S. coracinus, S. flavibarbis, S. schisticolor*, and *S. strateagus*. Group A was separated into sub-groups by the analysis: A1 (*S. coracinus*) separated out based on the rectangular shape of apodeme and the extension of the spermatheca to the middle of the 4th abdomen

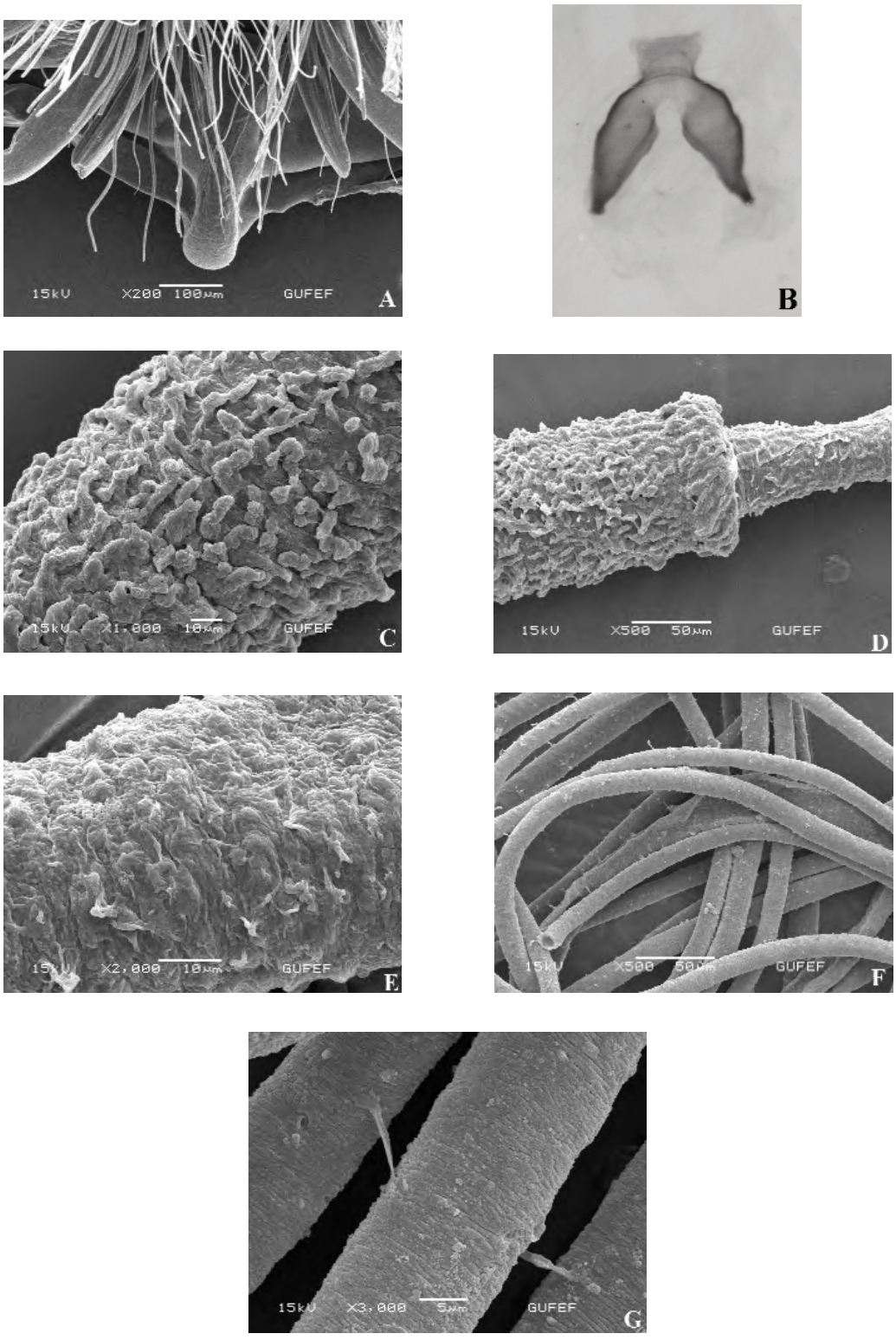

Figure 9. Spermatheca of *S. xanthotrichus xanthomelas*. A. SEM micrograph view of dorsal plate, B. Furca (light microscope view), C. SEM micrograph of sclerotized duct of ejection apparatus, D. SEM micrograph of valve, E. SEM micrograph of reservoir duct, F. SEM micrograph of reservoir, G. SEM micrograph of reservoir.

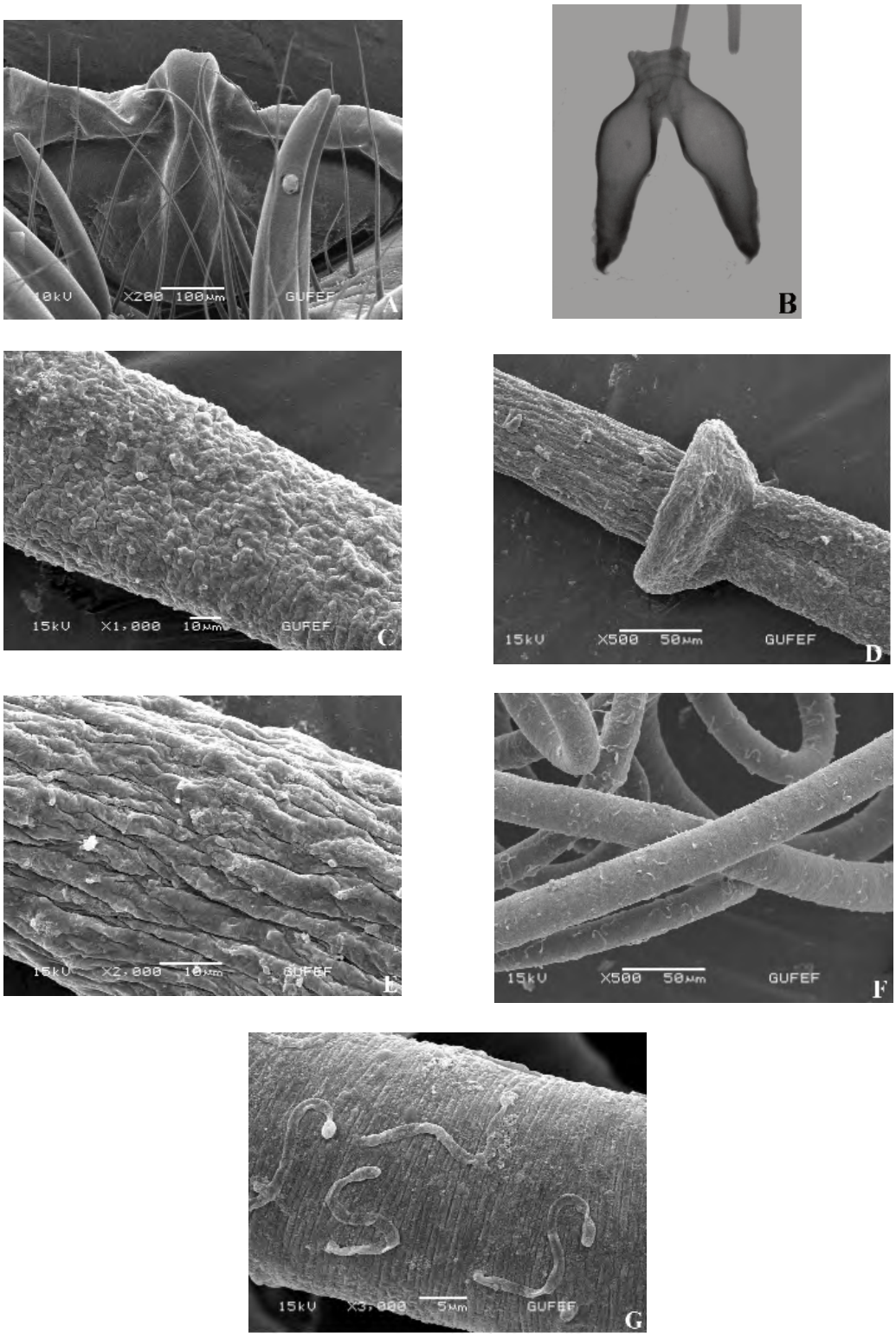

Figure 10. Spermatheca of *S. junceus*. A. SEM micrograph view of dorsal plate, B. Furca (light microscope view), C. SEM micrograph of sclerotized duct of ejection apparatus, D. SEM micrograph of valve, E. SEM micrograph of reservoir duct, F. SEM micrograph of reservoir, G. SEM micrograph of reservoir.

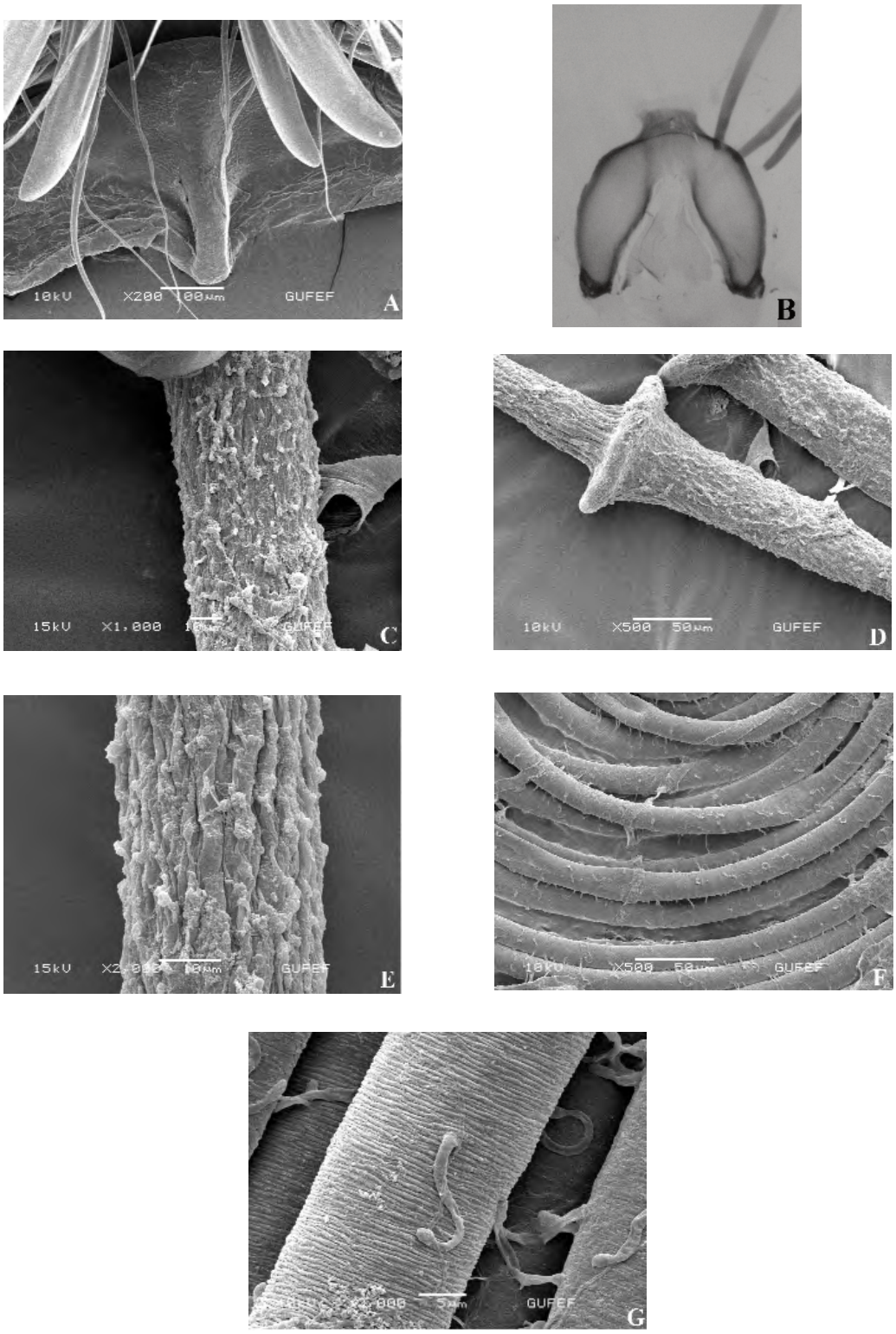

Figure 11. Spermatheca of *S. elongates*. A. SEM micrograph view of dorsal plate, B. Furca (light microscope view), C. SEM micrograph of sclerotized duct of ejection apparatus, D. SEM micrograph of valve, E. SEM micrograph of reservoir duct, F. SEM micrograph of reservoir, G. SEM micrograph of reservoir.

segment, A2 (*S. flavibarbis*) was separated out by the shape of the furca and the extension of the spermatheca to the middle of the 6th abdomen segment, A3 (*S. schisticolor* and *S. strateagus*) was separated by extension of the spermathecae to the middle of the 5th abdominal segment and the proportion of the area surrounded by a layer of muscles of the ejection apparatus to the sclerotized portion of ejection apparatus. Species of group A were shown to be monophyletic. Group B (containing a more coiled reservoir tube) included *S. laevigatus, S. nigriventris, S. junceus, S. sabaudus harpax, S. xanthotrichus xanthomelas,* and *S. elongatus*. Group B was separated into 2 sub-groups. B1 (*S. elongatus* and *S. junceus*) sub-group also separated out on the basis of the position of spermatheca in the abdomen, the proportion of the area surrounded by a layer of muscles to the sclerotized portion of the ejection apparatus and the surface characteristics of the sclerotized portion of the ejection apparatus. B2 (*S. laevigatus, S. nigriventris, S. sabaudus harpax,* and *S. xanthotrichus xanthomelas*) was separated from B1 sub-group by the thick ring shape of the valve and the elliptical shape of the reservoir. *S. xanthotrichus xanthomelas* in B2 sub-group separated from the other 3 species by the proportion of the area surrounded by a layer of muscles of ejection apparatus to the sclerotized portion of ejection apparatus and transverse striate surface of the reservoir duct. Moreover, *S. sabaudus harpax* separated out from *S. nigriventris* and *S. laevigatus* on the basis of the diameter and surface characteristics of the sclerotized portion of the ejection apparatus. Species of group B were also found to be monophyletic (Figure 12).

Discussion

The phylogenetic relationships of taxa within the monophyletic Asilidae are still very poorly known. There are many faunistic and ecological studies about the Asilidae, but phylogenetic studies of the Asilidae are very few.

The first molecular hypothesis of the higher-level phylogenetic relationships within the Asilidae based on DNA-sequence data was published by Bybee et al. (2004). The analysis is based on the 16S (mitochondrial), 18S, and 28S (both nuclear) ribosomal genes and the protein coding gene cytochrome oxidase II (mitochondrial), sequenced from 26 representative species of 10 of the currently 11 recognized subfamilies. This work provides the first molecular estimate of higher-level relationships.

Cannings (2002) published a large revision of *Lasiopogon* and included a phylogenetic analysis of 8 genera of Stichopogoninae. The hypothesis is based on 19 morphological characters from the complex male terminalia. He concluded that *Lasiopogon* is in fact a member of this monophyletic taxon and *Stichopogon*, the type genus, is a derived member of this group.

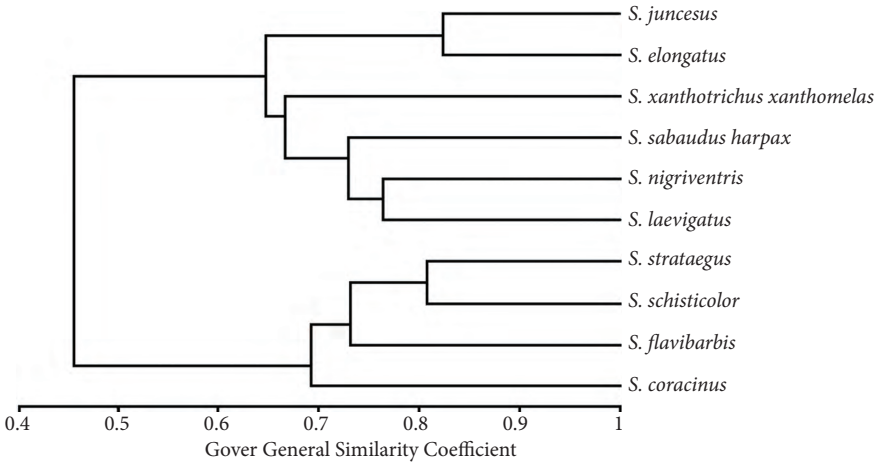

Figure 12. UPGMA dendrogram showing the relationship between taxa of *Stenopogon*.

The most comprehensive morphological analysis was published by Papavero (1973). This analysis is based on a selected number of characters of the body of the imagines.

Morphological separation of *Stenopogon* Loew, 1847 is as follows according to Lehr (1963):

R4 cell closed and stalked at the wing corner group includes *S. schisticolor*. R4 cell open at the wing corner group includes *S. strataegus, coracinus, laevigatus, sabadus harpax, elongatus, junceus, flavibarbis, xanthotrichus xanthomelas,* and *nigriventris*. R4 cell open at the wing corner group is separated into 2 subgroups. Pubescence found in the back corners of the mesopleura go to the base of the wing subgroup includes *S. strataegus* and *coracinus*. Back corner of the mesopleura without pubescence or has few pubescence group includes *S. laevigatus, sabadus harpax, elongatus, junceus, flavibarbis, xanthotrichus xanthomelas,* and *nigriventris*. A1 subgroup is separated into 2 subgroups. Arista short, 3rd segment 1/35-1/4 of the antennae subgroup includes *S. laevigatus, sabadus harpax,* and *xanthotrichus xanthomelas*. Arista long, 3rd segment 1/2-1/3 of the antennae subgroup includes *S. elongatus, junceus, flavibarbis*, and *nigriventris*.

According to cluster analysis based on the spermathecae structure group A includes *S. coracinus, S. flavibarbis, S. schisticolor*, and *S. strateagus* and group B includes *S. laevigatus, S. nigriventris, S. junceus, S. sabaudus harpax, S. xanthotrichus xanthomelas,* and *S. elongatus*. This analysis supports morphological separation except for *S. schisticolor*, *flavibarbis*, and *nigriventris*. The cluster analysis should make use of characters not normally used in identification, such as internal morphology. The study should be extended to other genera of the Asilidae after careful selection of characters.

Acknowledgements

We thank Gazi University Scientific Research Foundation for financial support and Dr. Robert Lavigne, South Australian Museum, Adelaide 5000, Australia, for inspiring us to conduct this study.

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