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Micromorphological studies on the genus *Lotus* L. (Fabaceae: Loteae) from Egypt

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Abstract: Trichomes, epidermal cell features, and seed coat sculpturing of 12 *Lotus* taxa from Egypt were studied using scanning electron microscopy. In addition, statistical analysis for the taxa under investigation was carried out. The results revealed that trichomes have high taxonomic significances between studied taxa. Anticlinal and periclinal cell walls and epidermal cell shape for the stem, leaf, and calyx for each taxon were examined. Seed coat sculpturing exhibited 4 main different surface patterns categories: rugose, reticulate, sulcate, and verrucate. These features can serve as good diagnostic characters at the species level of the genus *Lotus*.

Key words: Fabaceae, *Lotus*, seed, scanning electron microscopy, trichomes, Egypt

1. Introduction

Leguminosae (Fabaceae) is the third largest family of flowering plants, after Compositae and Orchidaceae (Christenhusz and Byng, 2016) . It contains approximately 720 genera and more than 18,000 species worldwide (Lewis et al., 2005; Escaray et al., 2012). *Lotus* (120–130 species) is the largest genus of the tribe Loteae. Based on previous studies (Gillett, 1958; Heyn, 1967; Kramina, 1999, 2006; Kramina and Sokoloff, 2004), *Lotus* is a taxonomically difficult genus. No worldwide taxonomic studies for the whole genus have been produced since the monograph of Brand (1898). A comprehensive taxonomic revision of the genus is needed (Degtjareva et al., 2006). In Egypt, *Lotus* L. is represented by 18 taxa (Boulos, 2009).

The species of *Lotus* constitute complexes of closely related groups with similar vegetative characters, including seasonal polymorphisms (Heyn, 1970), and it is difficult to distinguish among the species (Ojeda et al., 2009).

Several studies have demonstrated the use of micromorphological characters to differentiate between some taxa of Leguminosae (Lersten and Curtis, 1996; Stenglein et al., 2003; Zoric et al., 2009; Saheed and Illoh, 2010; Albert and Sharma, 2013).

Trichomes and epidermal cell features are employed to identify a particular taxon; for example, Ávalos and Salinas (2003) used leaf surface trichomes for delimiting species of *Quercus* L. They showed variations in trichome shape, intensity, size, and quantity of cells, being either glandular or nonglandular. Fayed et al. (2015) also used scanning electron microscopy (SEM) to study the importance of trichomes in the systematics of *Teucrium* L. (Lamiaceae).

Sculpturing of seed coat surfaces has been successfully employed in the identification and classification of many different taxa (Boesewinkel and Bouman, 1984; Murthy and Sanjappa, 2002; Yildiz, 2002; Vural et al., 2008; Fawzi, 2011; Dadandı and Yıldız, 2015).

Variations in seed coat surface have been used to differentiate between the species of several leguminous genera. Among the notable contributions dealing with the seed coat surface are those of Ponomarenko et al. (1990) on species of the genus *Cassia*, Chernoff et al. (1992) on species of Vicieae, Abou-El-Enain and Loutfy (1999) on the genus *Sesbania* Scop., Zareh (2005) on six species of *Hippocrepis* L., Abou-El-Enain et al. (2007) on the genus *Lathyrus*, and Zorić et al. (2010) on 38 species of the genus *Trifolium* L. Recently Mirzaei et al. (2015) and Erkul et al. (2015) published works on the genera *Colutea* and *Oxytropis*, respectively.

The aim of the present work is to describe the macro- and micromorphological characters of seeds and the types and distribution of trichomes in addition to investigating the micromorphology of epidermal cell shapes of leaves, stems, and calyces of 12 *Lotus* taxa.

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2. Materials and methods

The examined specimens originated from Egyptian material deposited at the Assiut University herbarium (ASTU) and Cairo University herbarium (CAI) (Table 1) in addition to field observations. Dry materials for SEM of seeds, stems, leaves, and calyces were prepared by mounting specimens directly onto clean stubs using double-sided adhesive tape. They were then coated with gold in a JOEL JFC1100E ion-sputtering device and examined with a JOEL JSM 5400 LV scanning electron microscope, operated at accelerated voltage of 15 kV at the Scanning Electron Microscope Unit of Assiut University.

Statistical analysis was carried out according to Rohlf (2005) using NTSYS-pc 2.0 software. A dendrogram was constructed based on similarity matrix data by using the unweighted pair-group method with arithmetic mean (UPGMA) employing sequential, agglomerative, hierarchical, and nested clustering.

The terminology of Barthlott (1981), Roe (1971), Sandral et al. (2006), Stearn (1966, 2009), and Yagueddú et al. (2009) is used to describe and classify the types of indumentum and seed coat characteristics.

2.1. Synopsis of trichomes types (Figure 1)

1. Tubular compressed curved trichomes (TCCu)
2. Tubular upright curved trichomes (TUCu)
3. Tubular upright straight trichomes (TUSt)
4. Flattened compressed straight trichomes (FCSt)
5. Flattened upright curved trichomes (FUCu)
6. Flattened upright straight trichomes (FUSt)

The abbreviations used for details of the studied stems, leaves, and calyces are: C: compressed; Cu: curved; F: flattened; PKR: papillate knobby rugose; St: straight; T: tubular; TFS: tabular flat cells with striations; TRS^{i}: tabular rugose striate, elongated cells with dense striations; TRS^{ii}: tabular rugose striate, more or less isodiametric cells with dense striations; U: upright.

3. Results and discussion

Micromorphological data play a significant role in the classification of angiosperms (Heywood, 1971). The following discussion focuses mainly upon the main aspects of seed, stem, leaf ,and calyx micromorphological attributes among the taxa under investigation. The most characteristic micromorphological criteria of *Lotus* species are summarized in Tables 2–5.

Stem anticlinal cell wall boundaries of the most studied taxa are generally raised and straight, with the exception of 3 species (*L. arenarius*, *L. ornithopodioides*, and *L. polyphyllos*) found to be recessed and straight. On the basis on the stem outer periclinal cell walls, the majority of the studied taxa are tabular (8 species). The remaining 4 species (*L. arenarius*, *L. ornithopodioides*, *L. halophilus*, and *L. polyphyllos*) are characterized by ribbed outer periclinal cell walls. Furthermore, the shape of stem epidermal cells for all the investigated taxa is tabular flat with striations (TFS) as shown in Figure 2 and Table 2.

According to Yagueddú et al. (2009), most species of Papilionaceae show epidermal cells with curved, undulate, and straight anticlinal cell walls. This matches with our results, in which the leaf anticlinal cell wall boundaries can be differentiated into two main categories. The first category is recessed while the second one is raised. The

Table 1. Plants used in the micromorphological study.

Taxa	Collection
Lotus arenarius Brot.	Ras el Hekma, 02.05.1955, *El Hadidi* s.n. (ASTU)
Lotus ornithopodioides L.	Ras el Hekmah, 03.03.1956, *Mustafa Imam* s.n. (ASTU)
Lotus halophilus Boiss.	Burg el-Arab, 02–07.04.2009, *Faried* s.n. (ASTU)
Lotus polyphyllos Clarke	Burg el Arab, 24.03.1986, *Zareh* s.n. (ASTU)
Lotus creticus L.	Maqsaba, 21.05.2003, *Mashaly & Boulos* 20208 (ASTU)
Lotus cytisoides L.	El Daba, 16.04.1972, *William Girgis* s.n. (ASTU)
Lotus glaber Mill.	El Heiz, 15.05.1978, *M. Abd El Ghani* 431 (ASTU)
Lotus schimperi Steud. ex Boiss.	Wadi Ibib, 04.03.1967, *Osborn & Helmy* s.n. (CAI)
Lotus arabicus L.	Asswan, 27.01.1971, *Abd El Aziz & Soliman* s.n. (ASTU)
Lotus hebranicus Hochst. ex Brand	Wadi Faraied, 12.02.1961, *V. Täckholm et al.* 840 (ASTU)
Lotus tetragonolobus L.	El Ameria, 25.03.1927, *G. Täckholm* s.n. (ASTU)
Lotus conjugatus L. subsp. *requienii* (Mauri ex Sanguin.) Greuter	Tarfa, 07.05.1982, Botany Department Excursion 71 (ASTU)

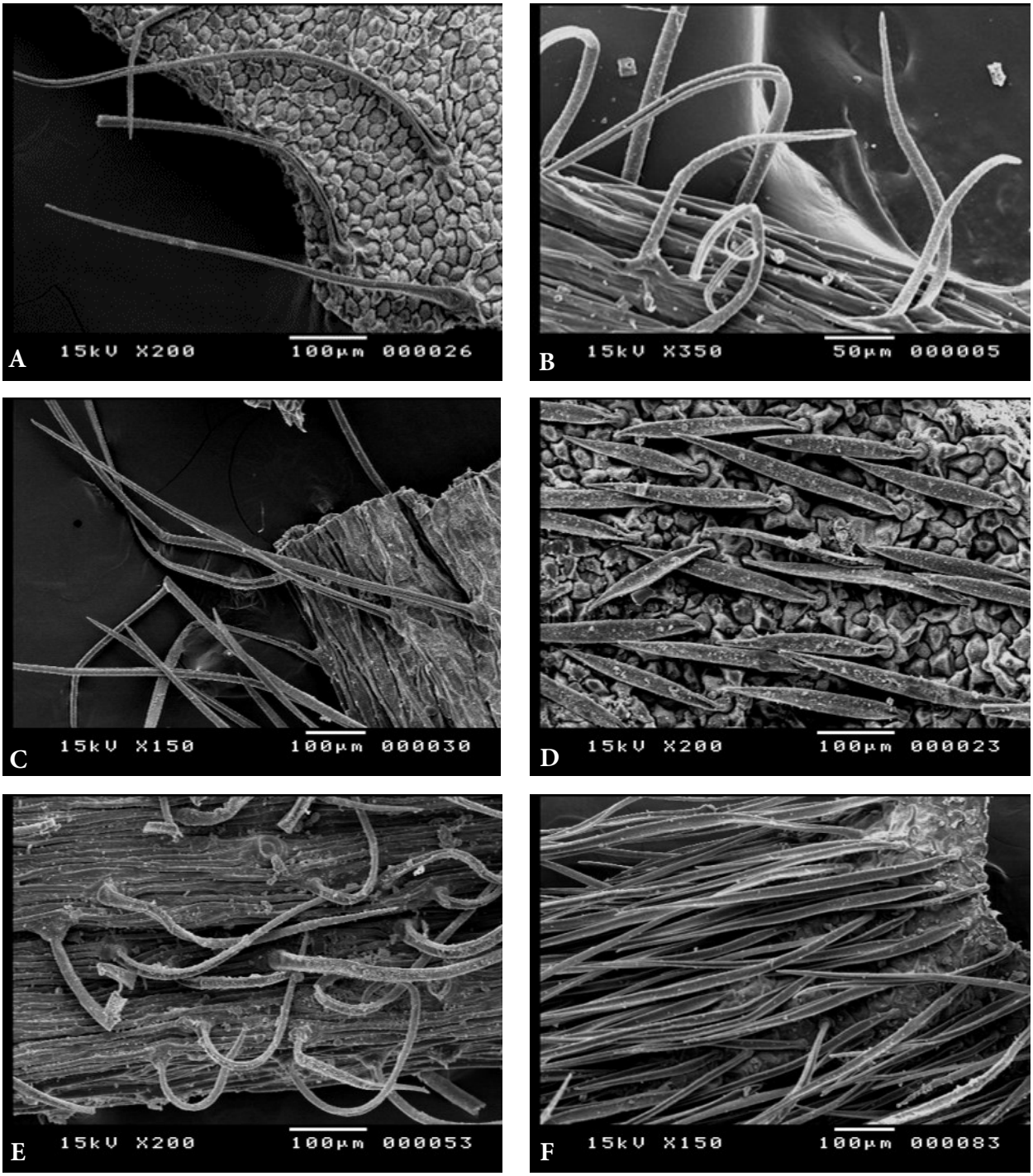

Figure 1. SEM micrographs of types of trichomes in the genus *Lotus*. A- Tubular compressed curved (TCCu); B- tubular upright curved (TUCu); C- tubular upright straight (TUSt); D- flattened compressed straight (FCSt); E- flattened upright curved (FUCu); F- flattened upright straight (FUSt).

recessed anticlinal cell walls boundaries may be straight (*L. polyphyllos*), coarsely undulate (*L. ornithopodioides*, *L. creticus*, *L. cytisoides*, and *L. schimperi*), or undulate (*L. halophilus*). Furthermore, the raised anticlinal cell wall boundaries could be either straight (*L. arenarius*, *L. glaber*, *L. arabicus*, and *L. tetragonolobus*) or undulate (*L. hebranicus* and *L. conjugatus* subsp. *requienii*). The outer periclinal cell wall was found to be tabular in 5 taxa (*L.*

Table 2. Micromorphological characters of stems for the studied taxa.

Species	Anticlinal wall	Periclinal walls	Epidermal cells shape	Trichomes		
				Presence	Shape	Length (mm)
L. arenarius	Recessed, straight	Ribbed	TFS	+	FUCu	0.2–0.4
L. ornithopodioides	Recessed, straight	Ribbed	TFS	+	FCSt	0.2–0.3
L. halophilus	Raised, straight	Ribbed	TFS	+	FCSt	0.2–0.3
L. polyphyllos	Recessed, straight	Ribbed	TFS	+	FCSt	0.4–0.9
L. creticus	Raised, straight	Tabular	TFS	+	FCSt	0.2–0.4
L. cytisoides	Raised, straight	Tabular	TFS	+	FCSt	0.2
L. glaber	Raised, straight	Tabular	TFS	–	–	–
L. schimperi	Raised, straight	Tabular	TFS	+	FUCu	0.3–0.4
L. arabicus	Raised, straight	Tabular	TFS	+	FUCu	0.2–0.3
L. hebranicus	Raised, straight	Tabular	TFS	–	–	–
L. tetragonolobus	Raised, straight	Tabular	TFS	+	FUCu	0.4–1
L. conjugatus subsp. *requienii*	Raised, straight	Tabular	TFS	+	TUCu	0.2–0.8

Table 3. Micromorphological characters of leaves for the studied taxa.

Species	Anticlinal wall	Periclinal walls	Epidermal cells shape	Trichomes		
				Presence	Shape	Length (mm)
L. arenarius	Raised, straight	Concave	TRS^{ii}	+	TCCu	0.6–0.9
L. ornithopodioides	Recessed, coarsely undulate	Tabular to convex	TRS^{ii}	+	TCCu	0.4–0.6
L. halophilus	Recessed, undulate	Convex	TRS^{i}	+	FCSt	0.3–0.5
L. polyphyllos	Recessed, straight	Convex, wrinkled	PKR	+	FCSt	0.3–0.5
L. creticus	Recessed, coarsely undulate	Tabular to convex	Isodiametric	+	FCSt	0.2–0.5
L. cytisoides	Recessed, coarsely undulate	Tabular to convex	Isodiametric	+	FCSt	0.2–0.3
L. glaber	Raised, straight	Tabular	TRS^{i}	–	–	–
L. schimperi	Recessed, coarsely undulate	Tabular to convex	Isodiametric	+	FUCu	0.2–0.3
L. arabicus	Raised, straight	Tabular	TFS	+	FUCu	0.3–0.3
L. hebranicus	Raised, undulate	Tabular	TFS	+	FUCu	0.2–0.3
L. tetragonolobus	Raised, straight	Tabular	TFS	+	TUSt	0.75–1
L. conjugatus subsp. *requienii*	Raised, undulate	Tabular	TFS	+	TUCu	0.5–0.9

Table 4. Micromorphological characters of calyces for the studied taxa.

Species	Anticlinal wall	Periclinal walls	Epidermal cells Shape	Trichomes		
				Presence	Shape	Length (mm)
L. arenarius	Raised, straight	Concave	TFS	+	FUCu	0.6–0.8
L. ornithopodioides	Recessed, undulate	Concave	TRS^{ii}	+	FUSt	0.3–0.6
L. halophilus	Raised, straight	Tabular	TFS	+	FUSt	0.3–0.5
L. polyphyllos	Raised undulate	Tabular	TFS	+	FUSt	0.4–0.6
L. creticus	Raised, undulate	Concave	TRS^{ii}	+	FUSt	0.2–0.4
L. cytisoides	Recessed, undulate	Concave	TRS^{i}	+	FCSt	0.2–0.3
L. glaber	Raised, straight	Concave	TFS	–	–	–
L. schimperi	Raised undulate	Tabular	TRS^{ii}	+	FUCu	0.5–0.6
L. arabicus	Raised, undulate	Tabular	TRS^{i}	+	FUSt	0.3–0.5
L. hebranicus	Raised, undulate	Concave	TRS^{ii}	+	FUSt	0.08–0.2
L. tetragonolobus	Recessed, undulate	Tabular	TRS^{ii}	+	TUSt	0.7–0.9
L. conjugatus L. subsp. *requienii*	Recessed, undulate	Tabular	TRS^{i}	+	TUSt	0.6–1.2

Table 5. Morphological characters of seeds for the studied taxa.

Species	Seed		
	Outline (shape)	Surface features	Dimensions (mm)
L. arenarius	Suborbicular	Rugose, foveolate	1.2 × 0.98
L. ornithopodioides	Orbicular	Sulcate	1.4
L. halophilus	Suborbicular	Rugose, foveolate	0.81 × 0.74
L. polyphyllos	Suborbicular	Rugose, foveolate	1.5 × 1.6
L. creticus	Orbicular	Reticulate, foveolate	0.99
L. cytisoides	Orbicular	Rugose, foveolate	1.2
L. glaber	Oblong	Rugose, reticulate	1.2 × 0.84
L. schimperi	Orbicular	Verrucate, foveolate	0.7
L. arabicus	Orbicular	Rugose, foveolate	1.3
L. hebranicus	Oblong	Reticulate, ribbed	1.2 × 0.7
L. tetragonolobus	Orbicular	Rugose, prolate	3
L. conjugatus subsp. *requienii*	Suborbicular	Rugose, foveolate	1.5 × 1.97

glaber, *L. arabicus*, *L. hebranicus*, *L. tetragonolobus*, and *L. conjugatus* subsp. *requienii*), tabular to convex in 4 taxa (*L. ornithopodioides*, *L. creticus*, *L. cytisoides*, and *L. schimperi*), concave in *L. arenarius*, convex in *L. halophilus*, and convex-wrinkled in *L. polyphyllos*, as shown in Figure 3.

Among the taxa of the genus *Lotus*, it was found that the leaf epidermal cells had taxonomic significance, being isodiametric in 3 taxa (*L. creticus*, *L. cytisoides*, and *L. schimperi*), tabular flat cells with striations (TFS) in 4 taxa (*L. arabicus*, *L. hebranicus*, *L. tetragonolobus*, and *L. conjugatus* subsp. *requienii*), elongated cells with dense striations (TRS^{i}) in 2 taxa (*L. halophilus* and *L. glaber*), more or less isodiametric cells with dense striations (TRS^{ii}) in 2 taxa (*L. arenarius* and *L. ornithopodioides*), and papillate knobby rugose (PKR) only in *L. polyphyllos* (Figure 3).

There is evidence that the Fabaceae members have the highest degree of variation of epidermal cell types within the flowers (Barthlott, 1990; Ojeda et al., 2009; Çildir et al., 2012). In this study, the anticlinal cell wall boundaries of calyces varied between the studied taxa (Table 4; Figure 4). It was commonly raised undulate in 5 taxa (*L. polyphyllos*, *L. creticus*, *L. schimperi*, *L. arabicus*, and *L. hebranicus*). It was recessed undulate in 4 taxa (*L. ornithopodioides*, *L. cytisoides*, *L. tetragonolobus*, and *L. conjugatus* subsp. *requienii*) and raised straight in 3 taxa (*L. arenarius*, *L. halophilus*, and *L. glaber*). The investigated taxa are distinguished by either concave or tabular outer periclinal cell walls of the calyx. Tabular periclinal cell walls were seen in 6 taxa (*L. halophilus*, *L. polyphyllos*, *L. schimperi*, *L. arabicus*, *L. tetragonolobus*, and *L. conjugatus* subsp. *requienii*). The remaining taxa showed concave outer periclinal cell walls of the calyx. The results of epidermal cell shapes revealed that the tabular flat cell (TFS) is characteristic to *L. arenarius*, *L. halophilus*, *L. polyphyllos*, and *L. glaber*. Furthermore, the tabular rugose elongate cell (TRS^{i}) distinguished *L. cytisoides*, *L. arabicus*, and *L. conjugatus* subsp. *requienii*. On the other hand, the tabular rugose isodiametric cell (TRS^{ii}) is characteristic to *L. ornithopodioides*, *L. creticus*, *L. schimperi*, *L. hebranicus*, and *L. tetragonolobus* (Figure 4).

Fabaceae species presented typical uniseriate eglandular trichomes (Metcalfe and Chalk, 1950; Uphof, 1962; Çildir et al., 2012). The present study agrees with the findings of the previous studies conducted on *Lotus*. The stem trichomes of the investigated taxa showed large variation of considerable diagnostic and systematic value. The stem was found to be hairless in both *L. glaber* and *L. hebranicus*. In addition, *L. conjugatus* subsp. *requienii* can be easily differentiated from other taxa by having the TUCu type of stem trichomes. On the other hand, the FCSt trichome type was recorded in 5 species (*L. ornithopodioides*, *L. halophilus*, *L. polyphyllos*, *L. creticus*, and *L. cytisoides*) (Table 2). Sandral et al. (2006) reported that the *Lotus arenarius*-group includes *L. arenarius*, *L. maroccanus*, and *L. eriosolen*, showing stem indumentum of curved trichomes. Our data were in accordance with Sandral et al. (2006) as *L. arenarius*, *L. schimperi*, *L. arabicus*, and *L. tetragonolobus* are distinguished by the FUCu type of stem trichomes. The length of stem trichomes ranged from 0.2 to 1 mm among the studied taxa, as shown in Table 2 and Figure 5.

The investigated taxa had different types of leaf trichomes, except for *L. glaber*, in which the trichomes are absent. The FCSt trichome type was recorded in 4 taxa (*L. halophilus*, *L. polyphyllos*, *L. creticus*, and *L. cytisoides*). The FUCu trichome type was recorded in 3 taxa (*L. schimperi*, *L. arabicus*, and *L. hebranicus*). *L. arenarius* and *L. ornithopodioides* are characterized by

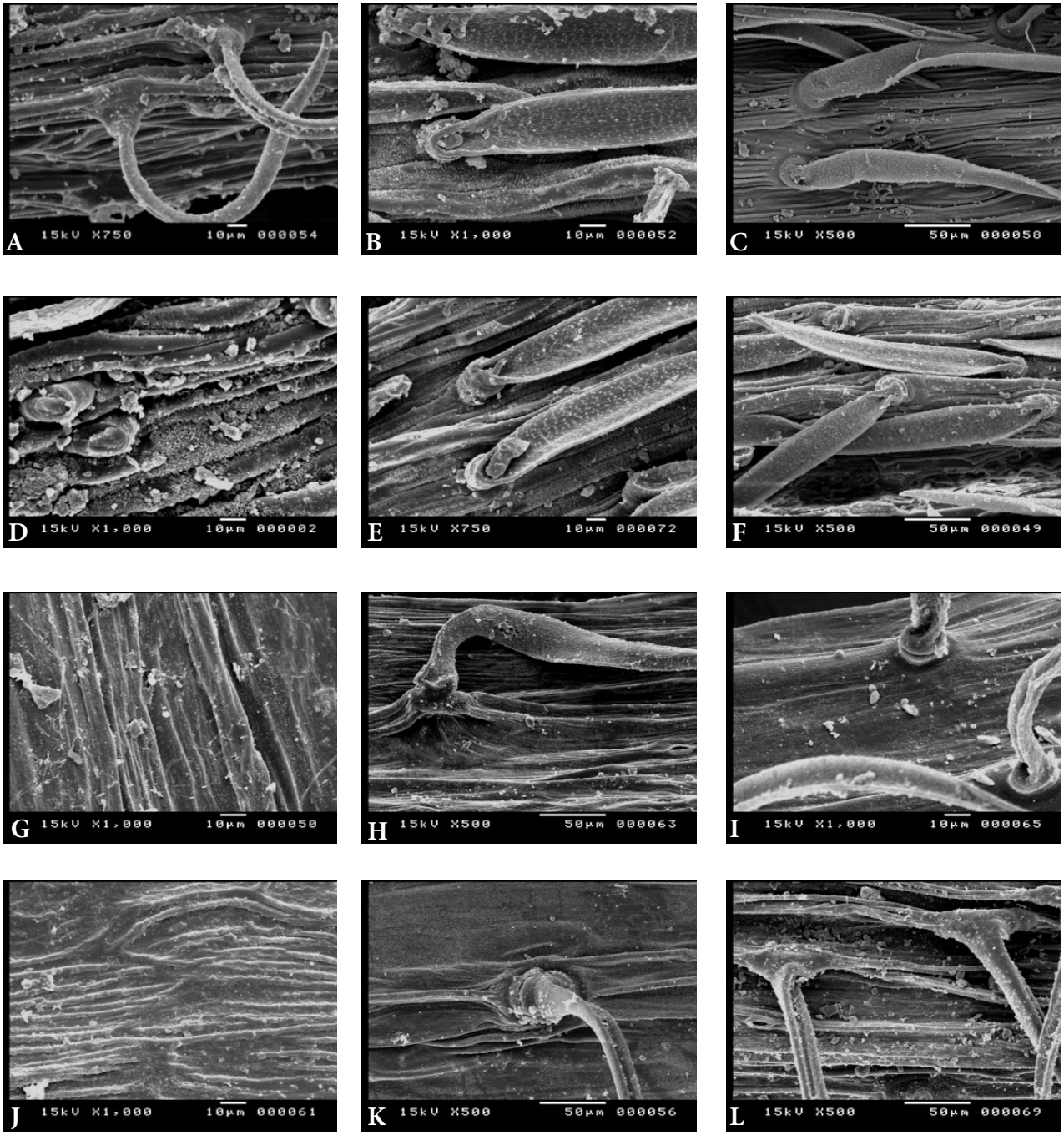

Figure 2. SEM micrographs of epidermal cells on different stems. A- *L. arenarius*; B- *L. ornithopodioides*; C- *L. halophilus*; D- *L. polyphyllos*; E- *L. creticus*; F- *L. cytisoides*; G- *L. glaber*; H- *L. schimperi*; I- *L. arabicus*; J- *L. hebranicus*; K- *L. tetragonolobus*; L- *L. conjugatus* subsp. *requienii*.

trichomes of the TCCu type, which matches the results of Sandral et al. (2006). The TUSt type was recorded only in *L. tetragonolobus*, while *L. conjugatus* subsp. *requienii* was the only taxon characterized by leaf trichomes of the TUCu type. Out of the 12 studied taxa, *L. tetragonolobus* was found to have the longest leaf trichomes (0.75–1 mm), as shown in Figure 6.

Of the 12 studied taxa, *L. glaber* can be easily distinguished by the absence of calyx trichomes; the remaining taxa showed variations in calyx trichome types. Most of the investigated taxa showed the FUSt type (6 taxa) and *L. cytisoides* is characterized by the FCSt trichome type. The TUSt type of calyx trichomes is diagnostic for both *L. tetragonolobus* and *L. conjugatus* subsp.

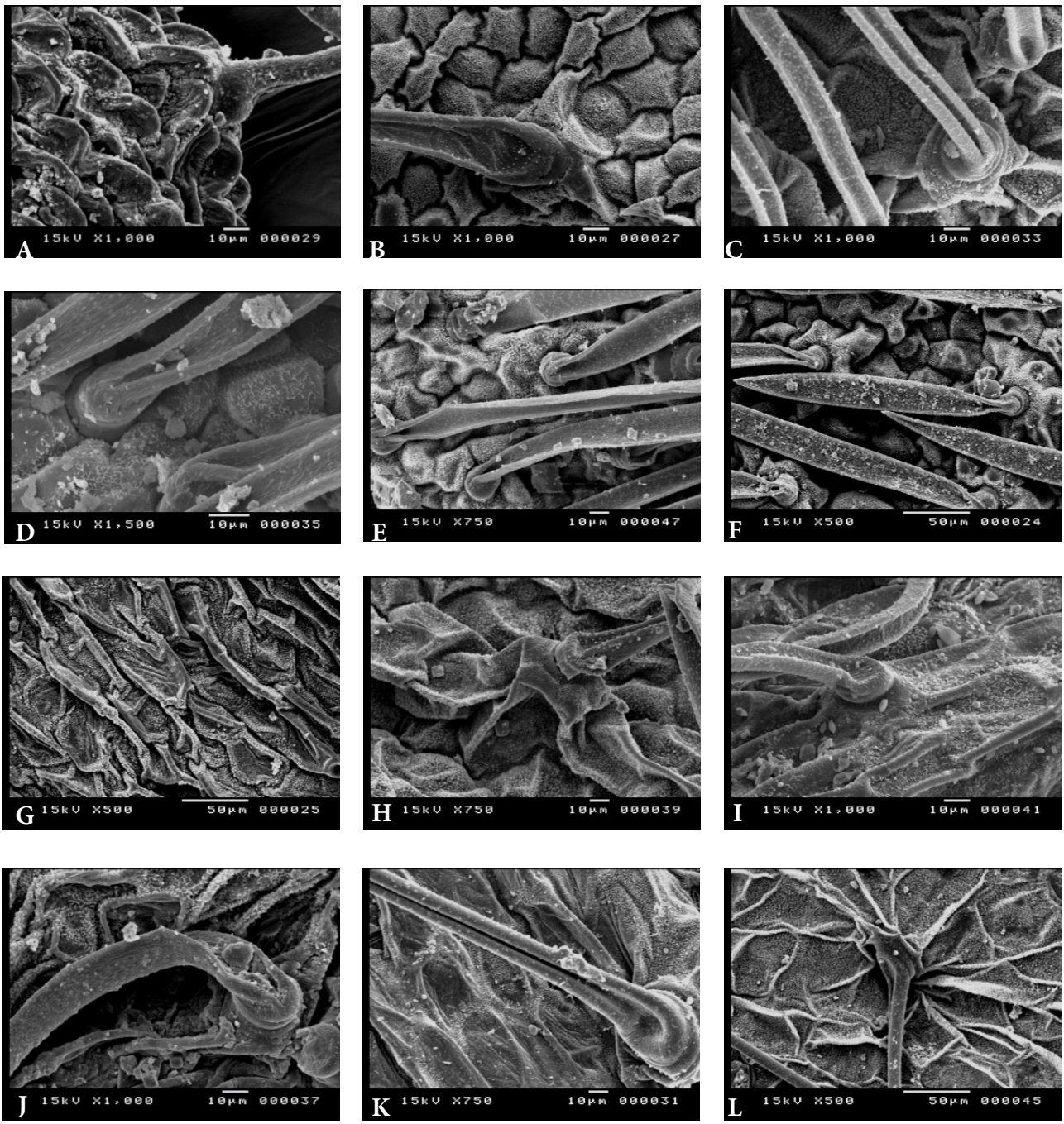

Figure 3. SEM micrographs of epidermal cells on different leaves. A- *L. arenarius*; B- *L. ornithopodioides*; C- *L. halophilus*; D- *L. polyphyllos*; E- *L. creticus*; F- *L. cytisoides*; G- *L. glaber*; H- *L. schimperi*; I- *L. arabicus*; J- *L. hebranicus*; K- *L. tetragonolobus*; L- *L. conjugatus* subsp. *requienii*.

requienii. Moreover, FUCu distinguished *L. arenarius* and *L. schimperi*, which coincides with the observations of Sandral et al. (2006). Trichome length varied greatly among the calyces of the studied taxa, ranging between 0.08 and 0.2 mm in *L. hebranicus* (the shortest) and 0.6 and 1.2 mm in *L. conjugatus* subsp. *requienii* (the longest), as shown in Table 4 and Figure 7.

Three types of seed shape were primarily recognized among the studied taxa (Table 5): orbicular, suborbicular, and oblong. The orbicular type was represented by *L. ornithopodioides*, *L. creticus*, *L. cytisoides*, *L. schimperi*, *L. arabicus*, and *L. tetragonolobus*. The suborbicular seed shape was represented by *L. arenarius*, *L. halophilus*, *L. polyphyllos*, and *L. conjugatus* subsp. *requienii*, this result

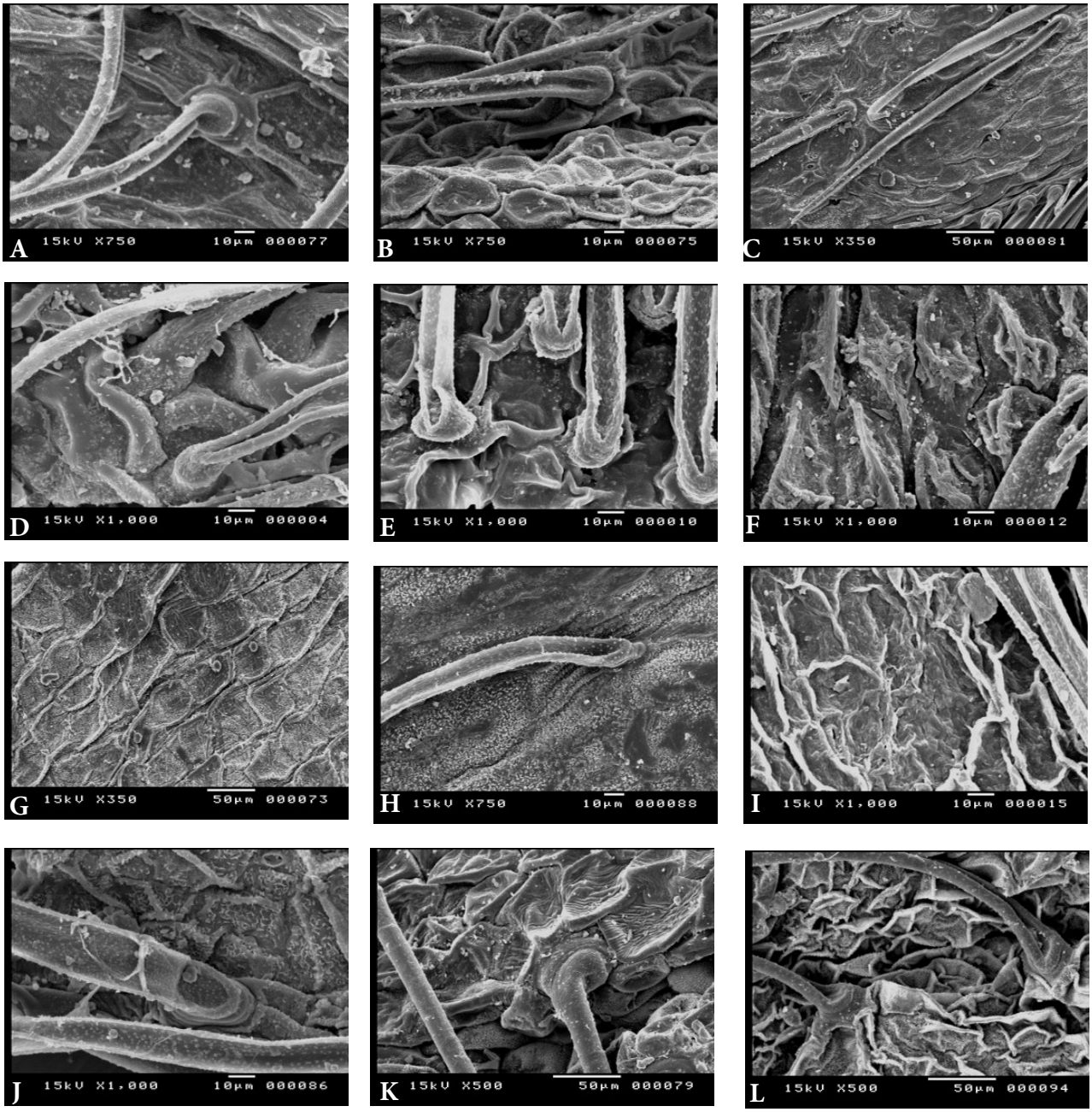

Figure 4. SEM micrographs of epidermal cells on different calyces. A- *L. arenarius*; B- *L. ornithopodioides*; C- *L. halophilus*; D- *L. polyphyllos*; E- *L. creticus*; F- *L. cytisoides*; G- *L. glaber*; H- *L. schimperi*; I- *L. arabicus*; J- *L. hebranicus*; K- *L. tetragonolobus*; L- *L. conjugatus* subsp. *requienii*.

being similar to the description of Sandral et al. (2006). The third seed shape type, oblong, can be observed in *L. glaber* and *L. hebranicus* (Figure 8). Seed size varies greatly among the studied taxa, ranging from 0.7 mm in *L. schimperi* as the smallest seed to 3 mm in *L. tetragonolobus* as the largest one.

Arambarri (1999) examined 67 seeds of *Lotus* species and proved that seed surface features can serve as good diagnostic characters for the lowest taxonomic categories. There are 4 main different categories for the seed surface: rugose, reticulate, sulcate, and verrucate (Table 5; Figure 9). The rugose type is divided into three subtypes: (A) rugose-foveolate in *L. arenarius*, *L. halophilus*, *L. polyphyllos*, *L. cytisoides*, *L. arabicus*, and *L. conjugatus* subsp. *requienii*; (B) rugose-reticulate in *L. glaber*; (C) rugose-prolate in *L. tetragonolobus*. The reticulate type of seed surface may be

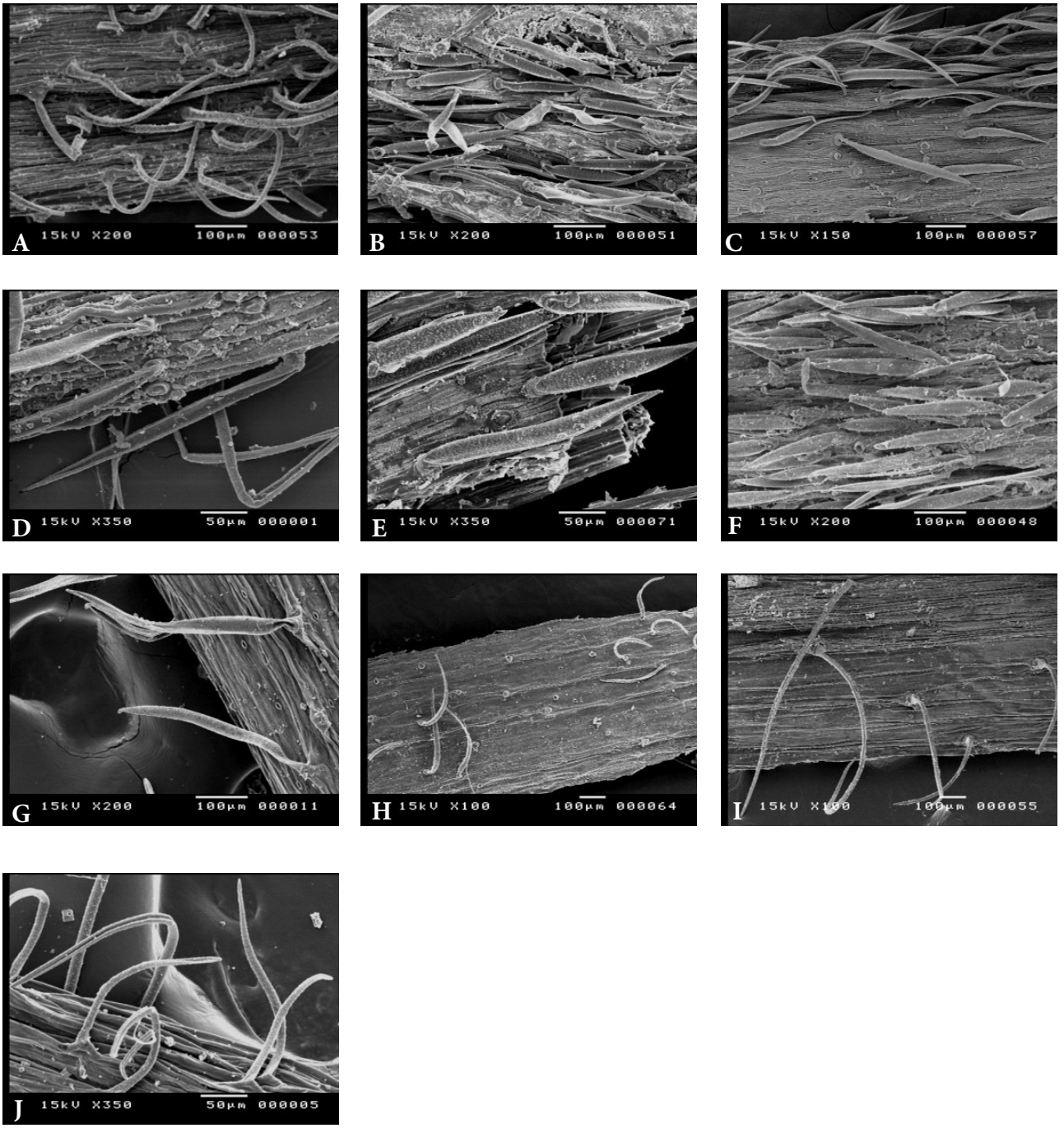

Figure 5. SEM micrographs of trichomes on different stems. A- *L. arenarius*; B- *L. ornithopodioides*; C- *L. halophilus*; D- *L. polyphyllos*; E- *L. creticus*; F- *L. cytisoides*; G- *L. schimperi*; H- *L. arabicus*; I- *L. tetragonolobus*; J- *L. conjugatus* subsp. *requienii*.

reticulate-foveolate as in *L. creticus* or reticulate-ribbed as in *L. hebranicus*. A sulcate seed surface can be observed only in *L. ornithopodioides*. The last type of seed surface category, verrucate, is characterized for only one taxon (*L. schimperi*) as verrucate-foveolate, corroborating the study of Arambarri (1999).

Table 6 shows the characters and character states applied for micromorphological investigations, including trichomes of the stem, leaf, and calyx, in addition to epidermal cells of the leaf and stem and seed features. A total of 16 characters were measured for each taxon under investigation.

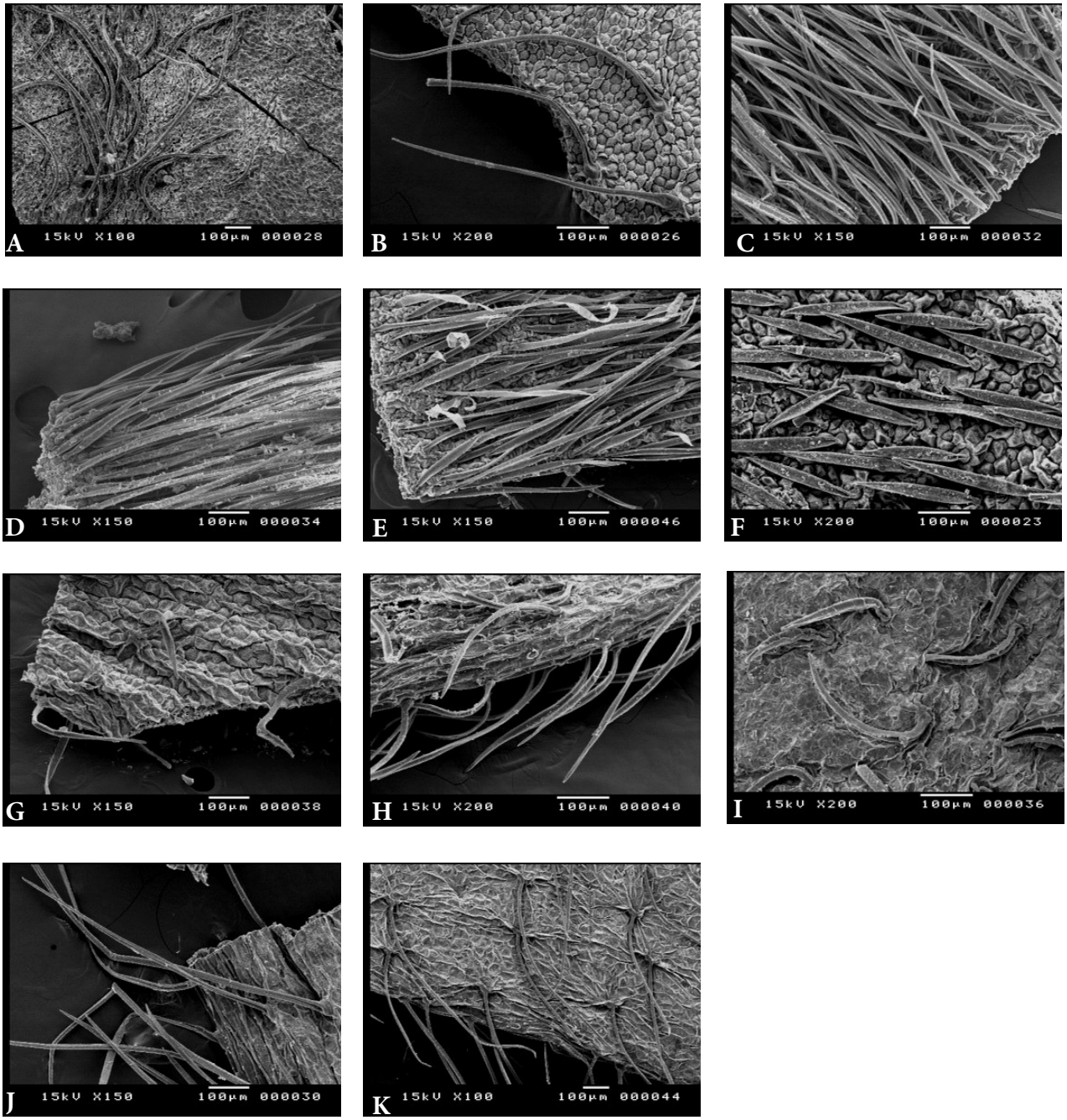

Figure 6. SEM micrographs of trichomes on different leaves. A- *L. arenarius*; B- *L. ornithopodioides*; C- *L. halophilus*; D- *L. polyphyllos*; E- *L. creticus*; F- *L. cytisoides*; G- *L. schimperi*; H- *L. arabicus*; I- *L. hebranicus*; J- *L. tetragonolobus*; K- *L. conjugatus* subsp. *requienii*.

According to UPGMA analysis, as shown in Figure 10, the micromorphological similarities among the *Lotus* taxa ranged from 0.20 to 0.75. The dendrogram topology showed two main clades. The first one represented all *Lotus* taxa except *Lotus glaber*, which was considered in the second clade. This result is in agreement with the micromorphological results; *Lotus glaber* can easily be excluded from all *Lotus* taxa by the absence of trichomes on the stem, leaf, and calyx.

The micromorphological results of the investigated taxa show high similarity between *Lotus creticus* and *Lotus cytisoides*, both having the same type of trichomes on the stem, leaf, and calyx as well as the same shape of seeds. This result was supported by statistical analysis in

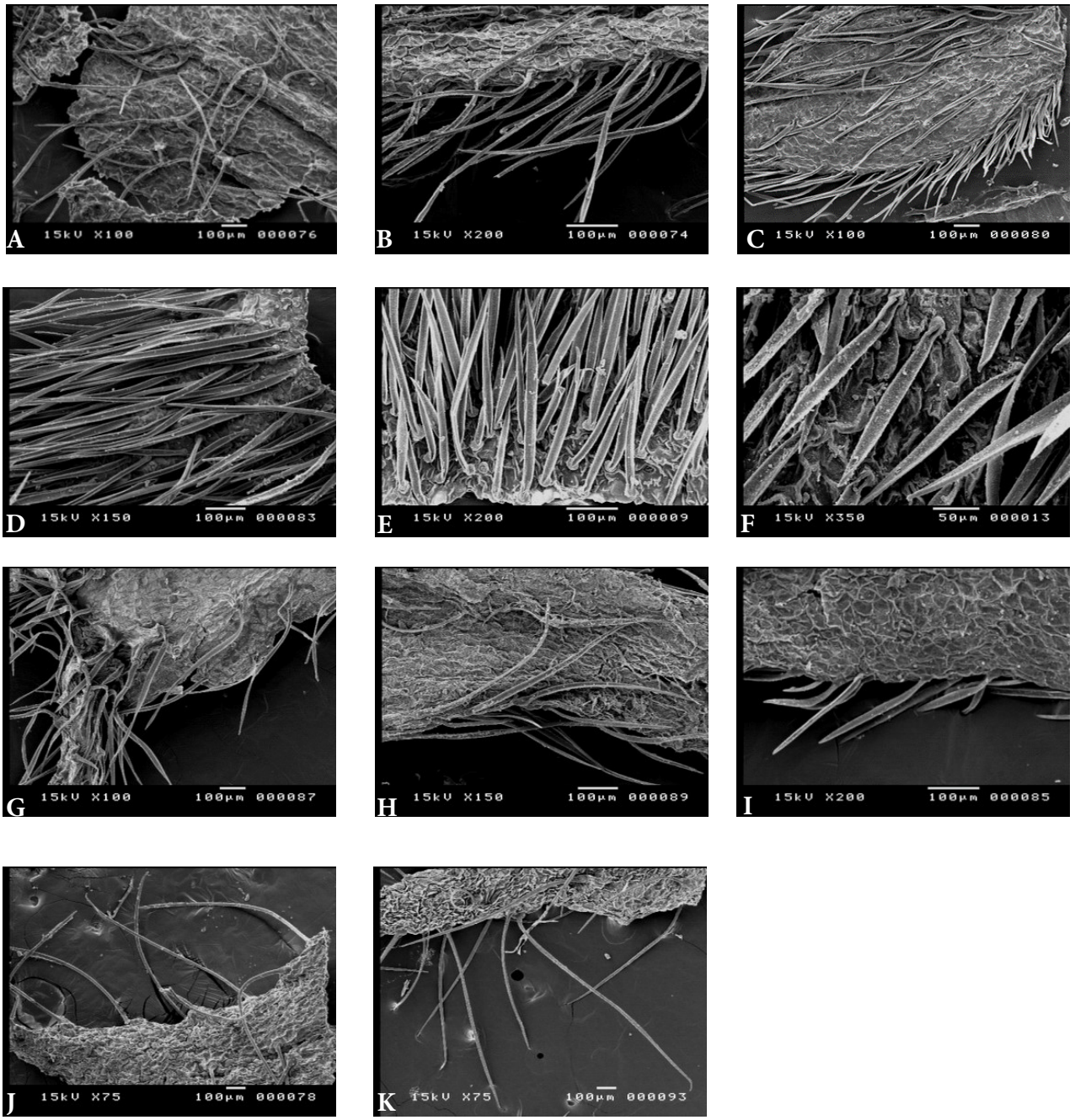

Figure 7. SEM micrographs of trichomes on different calyces. A- *L. arenarius*; B- *L. ornithopodioides*; C- *L. halophilus*; D- *L. polyphyllos*; E- *L. creticus*; F- *L. cytisoides*; G- *L. schimperi*; H- *L. arabicus*; I- *L. hebranicus*; J- *L. tetragonolobus*; K- *L. conjugatus* subsp. *requienii*.

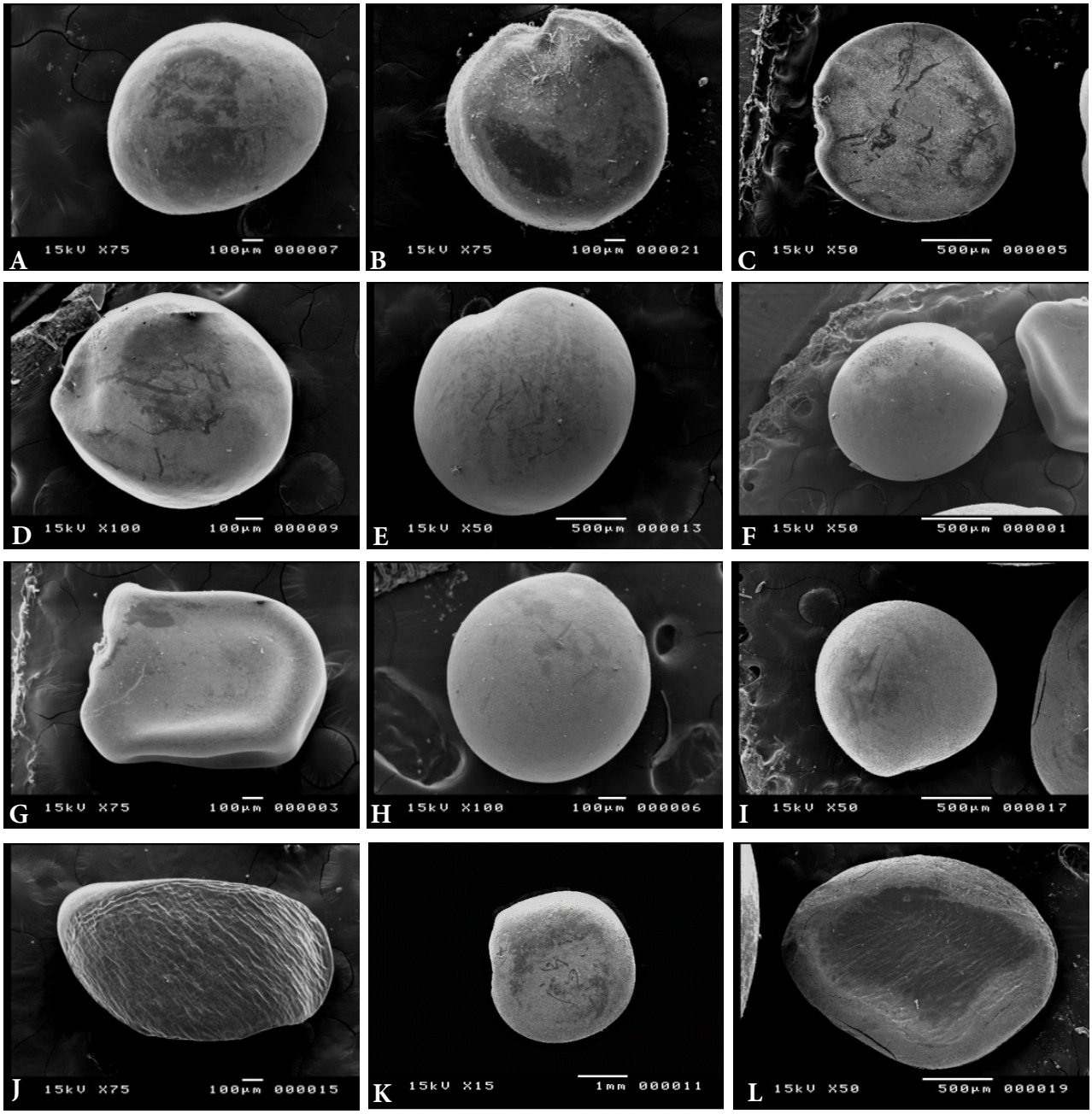

Figure 8. SEM micrographs of seed outline. A- *L. arenarius*; B- *L. creticus*; C- *L. ornithopodioides*; D- *L. halophilus*; E- *L. polyphyllos*; F- *L. cytisoides*; G- *L. glaber*; H- *L. schimperi*; I- *L. arabicus*; J- *L. hebranicus*; K- *L. tetragonolobus*; L- *L. conjugatus* subsp. *requienii*.

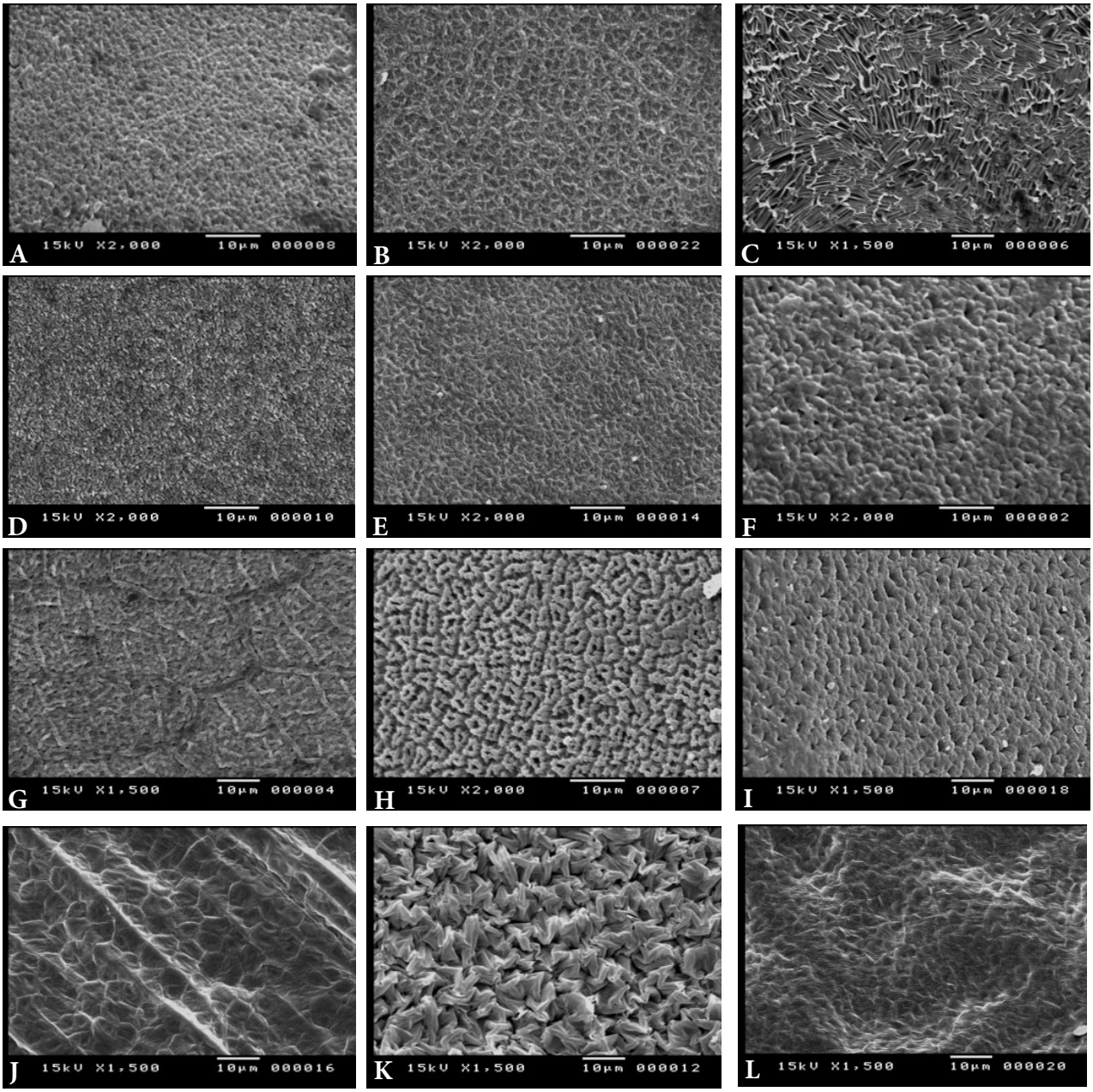

Figure 9. SEM micrographs of seed surface. A- *L. arenarius*; B- *L. creticus*; C- *L. ornithopodioides*; D- *L. halophilus*; E- *L. polyphyllos*; F- *L. cytisoides*; G- *L. glaber*; H- *L. schimperi*; I- *L. arabicus*; J- *L. hebranicus*; K- *L. tetragonolobus*; L- *L. conjugatus* subsp. *requienii*.

Table 6. Characters and character states used in micromorphological analysis of *Lotus*.

N	Character and character states
1	Stem anticlinal wall 1. Recessed, straight 2. Raised, straight
2	Stem periclinal walls 1. Ribbed 2. Tabular
3	Stem trichomes 1. Present 2. Absent
4	Shape of stem trichomes 1. N/A 2. FUCu 3. FCSt 4. TUCu
5	Leaf anticlinal wall 1. Raised, straight 2. Recessed, coarsely undulate 3. Recessed, undulate 4. Recessed, straight 5. Raised, undulate
6	Leaf periclinal walls 1. Concave 2. Tabular to convex 3. Convex 4. Convex, wrinkled 5. Tabular
7	Shape of leaf epidermal cells 1. TRS[i] 2. TRS[ii] 3. PKR 4. Isodiametric 5. TFS
8	Leaf trichomes 1. Present 2. Absent
9	Shape of leaf trichomes 1. N/A 2. TCCu 3. FCSt 4. FUCu 5. TUSt 6. TUCu
10	Calyx anticlinal wall 1. Raised, straight 2. Recessed, undulate 3. Raised, undulate
11	Calyx periclinal walls 1. Concave 2. Tabular
12	Shape of calyx epidermal cells 1. TFS 2. TRS[i] 3. TRS[ii]
13	Calyx trichomes 1. Present 2. Absent
14	Shape of calyx trichomes 1. N/A 2. FUCu 3. FUSt 4. FCSt 5. TUSt
15	Seed outline (shape) 1. Suborbicular 2. Orbicular 3. Oblong
16	Seed surface features 1. Rugose, foveolate 2. Reticulate, foveolate 3. Sulcate 4. Rugose, reticulate 5. Verrucate, foveolate 6. Reticulate, ribbed 7. Rugose, prolate

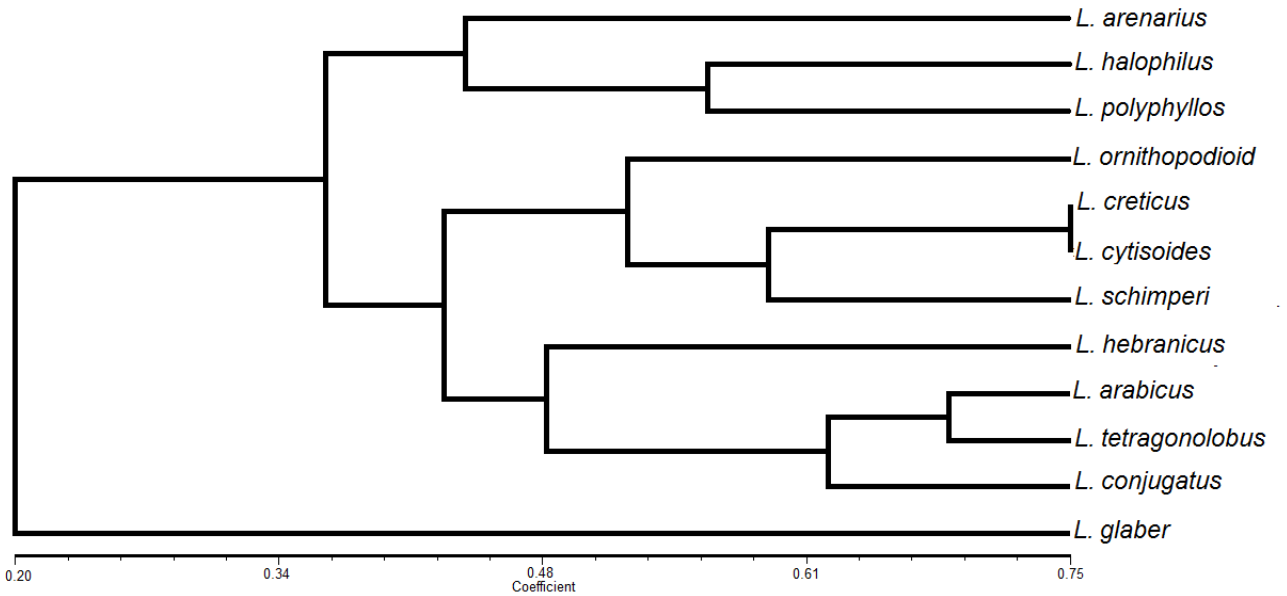

Figure 10. UPGMA dendrogram of studied taxa based on simple-matching similarity coefficients derived from micromorphological results.

which *Lotus creticus* and *Lotus cytisoides* were united in a subcluster together with a high similarity coefficient value of 0.75 (Figure 10).

In conclusion, *Lotus* is a taxonomically difficult genus. Little is known of the taxonomy and micromorphological details of this genus. Therefore, the present study should be a first step towards a better understanding of the taxonomy of this genus in the flora of Egypt. The main characteristics that help us in the recognition of these species are seed coat sculpturing and epidermal cell features including cell shape, anticlinal cell walls, and periclinal cell walls, in addition to trichome types for the stem, leaf, and calyx. These features can serve as good diagnostic characters at the species level of the genus *Lotus* in Egypt.

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