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Research Article

Morphological, Anatomical and Palynological Features of *Alyssum strigosum* s. l. (Brassicaceae)

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Abstract

Alyssum strigosum Banks & Sol. subsp. *strigosum* and *A. strigosum* Banks & Sol. subsp. *cedrorum* (Schott & Kotschy) Dudley, which are two subspecies of *Alyssum strigosum* with their morphological, anatomical, palynological traits, were presented in detail with this study. The transverse sections of roots showed that two subspecies have similar anatomical characters as with primer structure. The sections of stem were reflected the typical dicotyledonous stem anatomical structure and were determined that species have oval shaped cortex parenchyma. The vascular bundles in stem were arranged in one row both examined species. The mesophyll type was detected as equifacial. The examined species have monad, isopolar, prolate and tricolpate pollen grains. For two examined species, exine sculpture were observed as reticulate. As a conclusion, the characteristics of anatomical and palynological in *Alyssum strigosum* have been showed the supporting the separation of subspecies.

Keywords: Alyssum, Anatomy, Cruciferae, Palynology, Morphology

Alyssum strigosum s.l. (Brassicaceae)'un Morfolojik, Anatomik ve Palinolojik Özellikleri

Öz

Alyssum strigosum Banks & Sol. subsp. *strigosum* ve *A. strigosum* Banks & Sol. subsp. *cedrorum* (Schott & Kotschy) Dudley taksonlarının morfolojik, anatomik ve palinolojik özellikleri, bu çalışma ile ayrıntılı olarak ortaya konulmuştur. Köklerin enine kesitleri, iki alt türün, primer yapısında olduğu gibi benzer anatomik karakterlere sahip olduğunu gösterdi. Gövde bölümleri tipik dikotiledon gövde anatomik yapısını yansıtmış ve türlerin oval şekilli korteks parankimine sahip oldukları belirlenmiştir. Gövdede bulunan damar demetleri, incelenen türlerin her ikisinde de tek sıra halinde düzenlenmiştir. Mezofil türü eş yüzeyli olarak tespit edildi. İncelenen türler monad, isopolar, prolat ve trikolpat polen tanelerine sahiptir. İncelenen iki tür için ekzin heykel retikülat olarak gözlenmiştir. Sonuç olarak, Alyssum strigosum'daki anatomik ve palinolojik özellikler, alttürlerin ayrılmasını destekleyici niteliktedir.

Anahtar Kelimeler: Alyssum, Anatomi, Cruciferae, Palinoloji, Morfoloji

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1. Introduction

For the family Brassicaceae, the main distribution areas comprise the Mediterranean, Irano-Turanian, and Saharo-Sindian regions (Hedge, 1976). In Turkey, which is one of the richest countries with regards to Brassicaceae abundance, there are 686 native species, which belong to 91 genera (Güner et al.,2007).

Members of section *Alyssum* L. are characterised by monomorphic sepals free; unilaterally or bilaterally winged, dentate, or appendaged filaments; dehiscent monomorphic or dimorphic indumentum or, rarely glabrous and valves equally or unequally inflated fruits; and biovulate loculi (Dudley, 1964a, 1964b, 1965; Karabacak et al., 2016). Section *Alyssum*, of the family Brassicaceae, which is one of the largest genera, comprises approximately 230 species. The main distribution areas for section *Alyssum* are eastern Europe and Turkey (Al-Shehbaz et al.,2006). This genus is the largest among the genera in Turkey and is represented by 54 species, 25 of which are endemic (Karabacak et al., 2016).

There have been karyological (Şirin et al., 2020), micromorphological (Bülbül et al., 2018; Şirin, 2019), palynological (Başer et al., 2018; Karaismailoğlu, 2019a), and anatomical (Orcan and Binzet, 2003; Karaismailoğlu, 2020) studies conducted of some *Alyssum* species growing in Turkey; however, this is the first study regarding *A. strigosum* Banks & Sol..

A. strigosum subsp. *strigosum* spreads in Tekirdağ, İstanbul, Bolu, Gümüşhane, İzmir, Afyon, Ankara, Adana, Erzincan, Erzurum, Muğla, Antalya, Konya, İçel and Maraş and *A. strigosum* subsp. *cedrorum* in Gümüşhane, Kayseri, Adana, Konya, Muğla, Isparta, İçel and Kahramanmaraş (Dudley, 1965). The Turkish name of *A. strigosum* is "dökük kuduzotu" while that of *A. strigosum* subsp. *cedrorum* is "kaya kuduzotu" (Mutlu, 2012).

In this study, the morphological (some of them), anatomical, and palynological characters of *A. strigosum* were revealed for the first time and contributed to the classification at the subspecies level.

2. Material and Methods

A. strigosum was collected from Konya (Table 1) and collected specimens are deposited in the Herbarium of the Science Faculty of Selçuk University (KNYA). The morphometric analyses were performed with ten specimens.

Table 1. Localities of the studied taxa

Taxa	Localities
A. strigosum subsp. *strigosum*	C4 Konya: Seydişehir, *Abies cilicica* openings, 1550 m, 11.04.2018, *E. Şirin* 693 *& H. Günal* (KNYA)
A. strigosum subsp. *cedrorum*	C4 Konya: Çumra–Bozkır road 15. km, fields-side, 1020 m, 08.04.2018, *E. Şirin 680* & H. Günal (KNYA)

The herbarium specimens were used both anatomical and palynological studies. The vegetative parts of species were put on directly to 70% etyhl-alcohol solution and later they were treated with NaCl solution. The cross sections were taken by hand and they were stained with Phloroglucinol-HCL (Vardar, 1987). The best sections were chosen and photographed at magnifications of 10x, 20x, and 40x aid of a light microscope (Leica DM 1000). The measurements, which were made with the Kameram 21 software programme, were based on at least 30 or more cells per specimen. The mean values of the measurements of all of the investigated taxa were given. The root, stem and, leaf were used for anatomical studies. The palynological slides were made according to Wodehouse (1935). The pollen slides were observed using a Leica DM 1000 light microscope (LM) (Leica Microsystems, Wetzlar, Germany), and measured using Kameram 21 software (Argenit, Istanbul, Turkey). The measurements were based on at least 30 or more pollen grains from each specimen. All anatomical and palynological slides were photographed with aid of a Canon EOS 450 D camera attached to the light microscope.

3. Results

Morphological analysis

A. strigosum subsp. *strigosum*

Annual herb, 7.5–14 cm, stem densely stellate hairs with 5–10 rayed, cauline and sterile shoot leaves similar in shape, sterile shoot leaves oblanceolate or obovate–spathulate, 3–5 × 1–2 mm, acute, lower and upper surfaces greenish (yellowish when dry) white stellate hairs with 5–11 rayed, 3–7 × 1–3 mm, acute, lowermost fruiting pedicels 3–6 mm, densely stellate hairs, 4–10 rayed, cauline leaves oblanceolate or obovate–spathulate, 3–7 × 1–3 mm, acute. Inflorescence raceme, 14–28 flowered, sepals early deciduous, petals attenuate, entire or emarginate or bilobed, 2–3 × 0.5–1 mm, filaments of long stamens 2.5 mm, appendages connate for about half of their length, anthers c. 0.5 mm, yellow, styles rigid 0.5–1 mm, Figure 1. General view of *A. strigosum* subsp. *strigosum* (a) and *A. strigosum* subsp. *cedrorum* (b) glabrous or with sparse indumentum, green. Fruits with dimorphic indumentum of coarse, unequally bifurcate, slightly tuberculate hairs and adpressed stellate hairs, seed wing 0.2–0.3 mm wide (Figure 1).

Figure 1. General view of *A. strigosum* subsp. *strigosum* (a) and *A. strigosum* subsp. *cedrorum* (b)

A. strigosum* subsp. *cedrorum

Annual herb, 9–19 cm, stem densely stellate hairs with 4–10 rayed, cauline and sterile shoot leaves similar in shape, sterile shoot leaves oblanceolate or obovate–spathulate, 3–5 × 1–2 mm, acute, lower and upper surfaces greenish (yellowish when dry) white stellate hairs with 4–9 rayed, 3–6 × 1.5–3 mm, acute, lowermost fruiting pedicels 3–6 mm, densely stellate hairs, 4–10 rayed, cauline leaves oblanceolate or obovate–spathulate, 3–7 × 1–3 mm, acute. Inflorescence raceme, 15–30 flowered, sepals early deciduous, petals attenuate, entire or emarginate or bilobed, 2–3 × 0.5–1 mm, filaments of long stamens 2.5 mm, appendages connate for about half of their length, anthers c. 0.5 mm, yellow, styles rigid 1–1.5 mm, glabrous or with sparse indumentum, green. Fruits with dimorphic indumentum of coarse, unequally bifurcate, slightly tuberculate hairs and adpressed stellate hairs, seed wing 0.2–0.3 mm wide (Figure 1).

Anatomical Analysis

Root anatomy

The cross-sections of the root had epidermis as protective tissue in studied taxa (Figure 2). The cortex parenchyma was on a small area of the roots with spindle shaped in *A. strigosum* subsp. *cedrorum*, oval-shaped in *A. strigosum subsp. strigosum*. The phloem and xylem were well developed in cross sections. The pith region was filled with xylem elements. The vessels were quite organised in taxa.

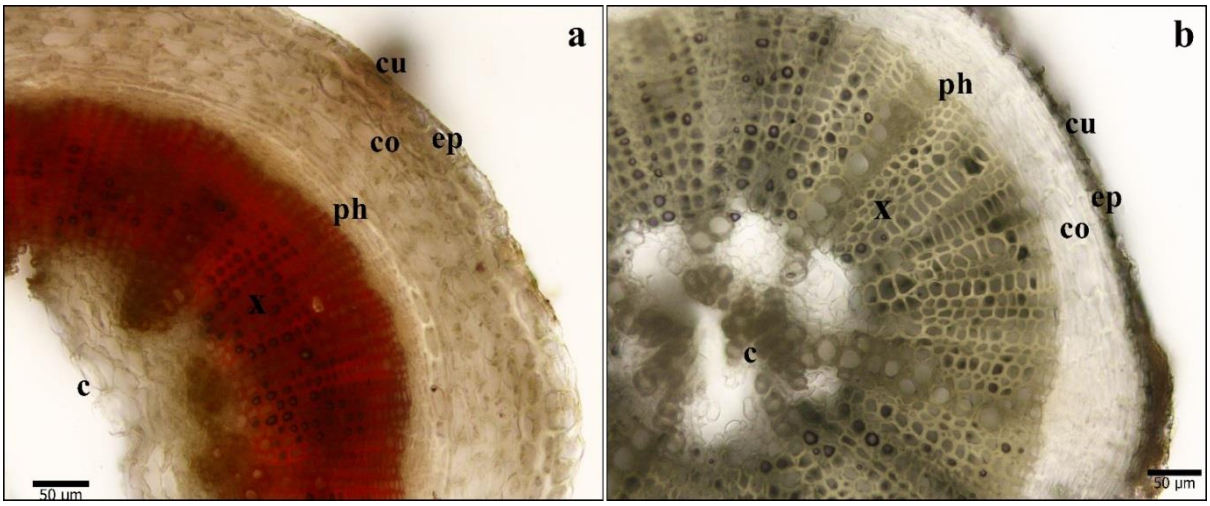

Figure 2. The root cross sections of *Alyssum strigosum*. a. *A. strigosum* subsp. *strigosum* b. *A. strigosum* subsp. *cedrorum* cu:cuticle, ep: epidermis, co: cortex parenchyma, ph: phloem, x: xylem, c: center of root.

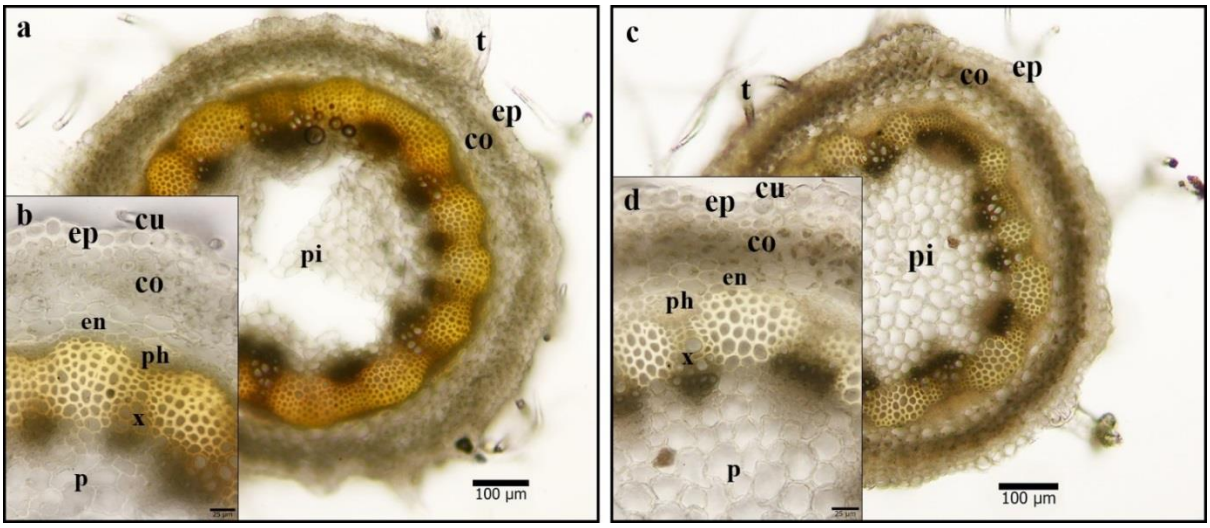

Figure 3. The stem cross sections of *Alyssum strigosum*. a-b. *A. strigosum* subsp. *strigosum* c-d. *A. strigosum* subsp. *cedrorum* ep: epidermis, co: cortex parenchyma, ph: phloem, x: xylem, cu: cuticle, pi:pith region, en: endodermis, p: parenchyma, t: trichome.

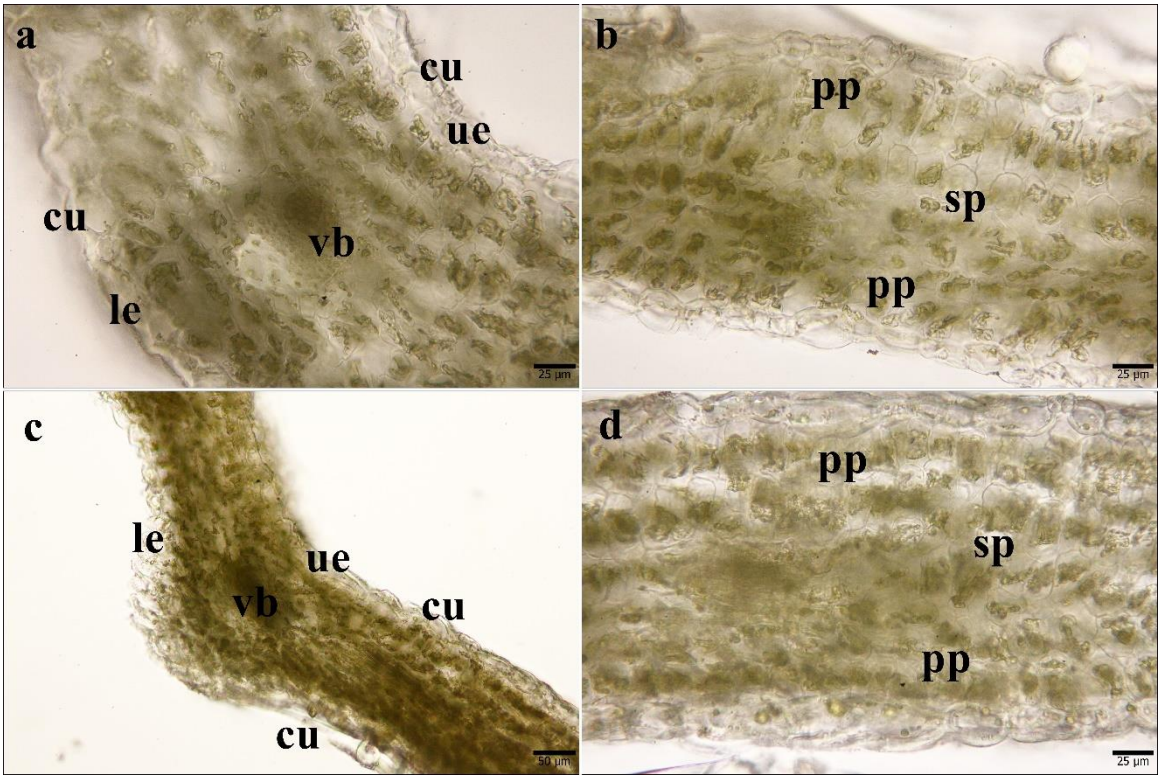

Figure 4. The leaf cross sections of *Alyssum strigosum* sl.. a-b. subsp. *strigosum* c-d. subsp. *cedrorum* ue: upper epidermis, le: lower epidermis, vb: vascular bundle, , cu: cuticle, pp: palisade parenchyma, sp: spongy parenchyma.

Stem Anatomy

The stem cross sections were composed of an epidermis, cortex, and vascular bundles and a pith region towards the centre in the examined taxa. The epidermis was single-layered with oval–rectangular-shaped cells and trichomes over them with thin cuticle. The cortex parenchyma had chlorophyll pigments with oval-shaped cells in a small area. Endodermis was composed of a single layered. Xylem and phloem elements were found continuously in the stem. Oval-shaped parenchymatic cells were covered the pith region (Figure 3, Table 2).

Leaf Anatomy The general view of the leaf cross-section showed the presence of an epidermis, mesophyll, and vascular bundles. The epidermis was limited to the leaves from the abaxial and adaxial sides. The midrib shape was linear in subsp. *strigosum*, v-shaped in subsp. *cedrorum.* The cylindrical-shaped palisade parenchyma cells were observed. They were arranged in 2 or 3 layers on both sides of the leaves (Figure 4, Table 2).

Palynological Analysis

A. strigosum subsp. strigosum

The pollen grains of subsp. *strigosum* was determined as monad, isopolar, and symmetrical. Aperture type was tricolpate. The polar and equatorial axis were measured as 33.1-38.92 µm (Mean 36.69±2.08 µm), 18-26.14 µm (22.36±3.38 µm), respectively. P/E ratio was calculated 1.64 and the pollen shape was determined as prolate. Colpus length was 36.06 µm and colpus width was 4.19 µm. The exine and intine thickness were measured 1.44 µm, and 0.79 µm, respectively. Apocolpium was 4.01 µm. The sculpture of exine were observed as reticulate. The mean of lumina width was 1.87 µm.

A. strigosum subsp. *cedrorum*

The pollen grains of subsp. *cedrorum* were determined as monad, isopolar, and symmetrical. Aperture type was tricolpate. The polar and equatorial axis were measured as 29.71-41.44 µm (Mean 35.83±4.58 µm), 17.58-25.1 µm (22.92±3.26 µm), respectively. P/E ratio was calculated 1.56 and the pollen shape was determined as prolate. Colpus length was 37.18 µm and colpus width was 4.12 µm. The exine and intine thickness were measured 1.53 µm, and 0.70 µm, respectively. Apocolpium was 3.14 µm. The sculpture of exine were observed as reticulate. The lumina width was 1.47 µm.

Table 2. The anatomical measurements of subsp. *strigosum* and subsp. *cedrorum* (μm).

Species/Anatomic characters		***A. strigosum* subsp. *strigosum***				***A. strigosum subsp. cedrorum***			
		Length		**Width**		**Length**		**Width**	
		Min-Max	**Mean±SD**	**Min-Max**	**Mean±SD**	**Min-Max**	**Mean±SD**	**Min-Max**	**Mean±SD**
Root	Epidermis	25.56-31.42	29.16±2.25	21.91-35.07	26.38±4.68	9.59-20.20	13.54±3.93	10.89-23.32	17.61±4.35
	Cortex	-	-	25.59-40.26	31.34±5.07	8.63-14.77	10.83±2.04	21.23-59.37	37.82±14.51
	Vessel	-	-	12.82-17.78	15.58±2.12	-	-	18.61-25.46	22.15±2.29
Stem	Cuticle	-	-	4.68-6.00	5.62±0.50	-	-	3.96-7.36	5.57±1.48
	Epidermis	12.65-17.76	14.58±2.03	12.87-23.21	18.64±2.87	12.52-18.45	15.21±2.22	15.47-22.85	18.14±2.41
	Cortex	-	-	14.57-23.03	18.92±2.92	-	-	14.53-27.14	18.95±3.20
	Vessel	-	-	8.91-15.84	13.15±2.34	-	-	10.45-14.69	11.47±1.40
	Pith cell	-	-	31.17-48.42	40.19±5.59	-	-	28.10-44.87	35.88±5.23
Leaf	Upper epidermis	15.20-27.52	20.61±4.01	27.03-37.47	31.58±4.09	15.41-19.24	18.31±2.19	20.03-38.98	27.22±6.66
	Lower epidermis	14.37-20.79	17.47±2.76	19.49-36.07	29.80±6.36	17.32-27.93	23.38±6.08	15.60-28.99	22.26±5.25
	Palisade parenchyma	26.31-35.92	30.00±3.20	14.18-20.91	18.17±2.67	18.10-33.23	27.64±5.44	13.47-21.20	17.03±2.73
	Spongy parenchyma	-	-	23.39-35.52	28.08±4.84	-	-	14.34-31.39	24.99±5.41

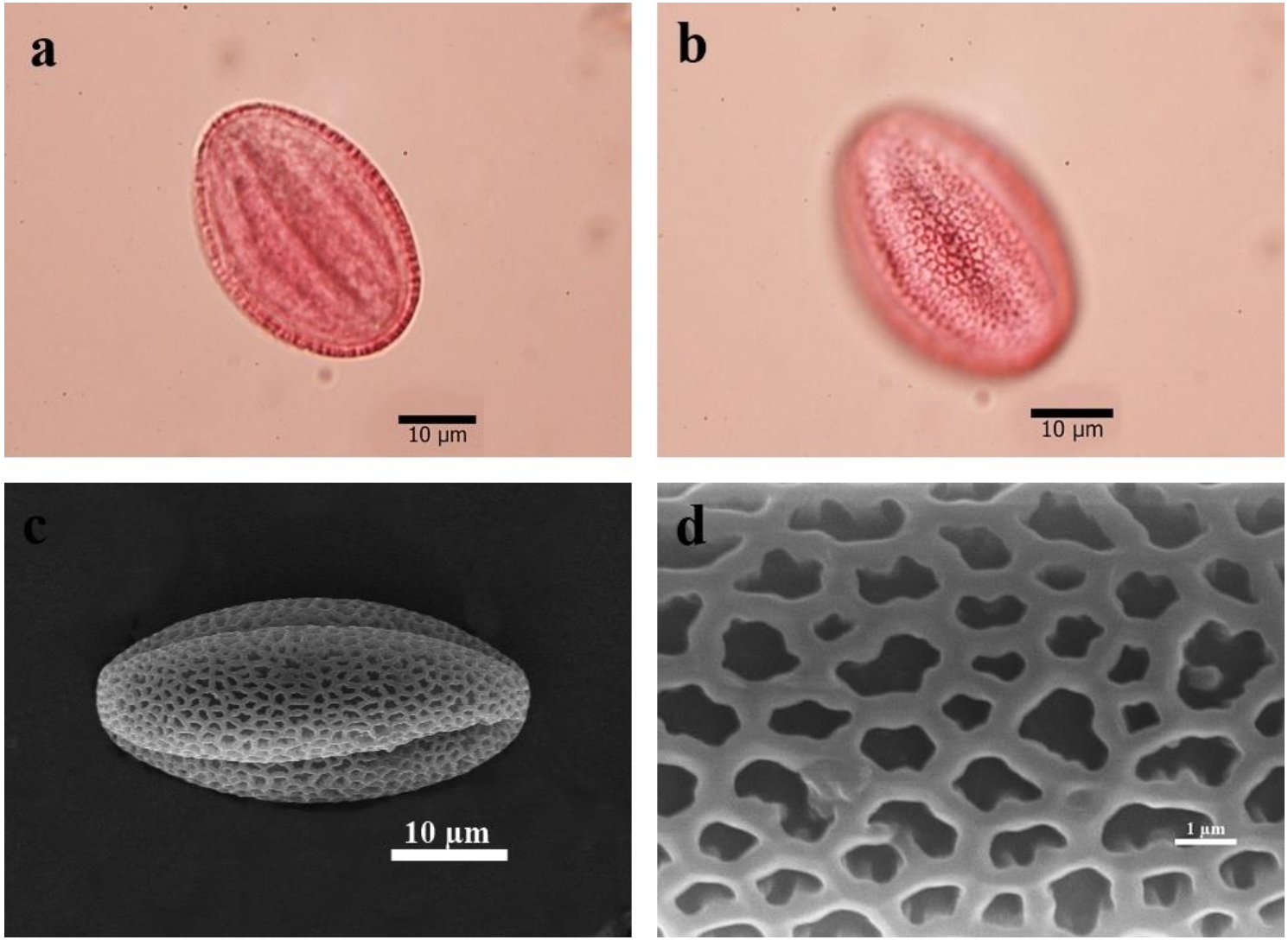

Figure 5. a-b. Light microscope (LM) photographs of *A. strigosum* subsp. *strigosum c*-d. Scanning electron microscope (SEM) photographs of *A. strigosum* subsp. *strigosum*

4. Discussion

A. strigosum subspecies are separated from each other by sepals deciduous / persistent, styles length, fruits measurements (Dudley, 1965) and seed measurements (Şirin, 2019). Sterile shoot morphology, stem and leaves indumentum, inflorescence flowered number, anther and filaments length are the first morphological characters studied for *A. strigosum*

A. strigosum seed coat pattern was interpreted by Şirin (2019) as rugose-foveate while Bulbul et al. (2019) as reticulate-foveate. This difference is due to the different interpretation of the seed coat pattern by the authors.

The anatomical properties for the investigation and designation of the family Brassicaceae have been based on research by Metcalfe and Chalk (1950). In current study, the diagnostic anatomical characters were observed and determined as the midrib shape, via the vessel diameter in the root. The stem shape was rectangle, circular, rounded, semi-rounded, or irregular in the family Brassicaceae (Orcan and Binzet, 2003; Atçeken et al., 2016; Qader, 2018, Çıtak and Dural, 2020).

The stem shape in the investigated *Alyssum* taxa was circular. The cortex cells were characterised by parenchyma that covered the small area in the cross-sections of the stem. The observations and investigation of the stem cross-sections showed that most of species were characterised by vascular bundles that were placed and arranged in a ring. The leaf anatomy has been used to characterise tribes and some genera in the family Brassicaceae (Orcan and Binzet, 2003; Selvi and Paksoy, 2013; Çıtak and Dural, 2020; Karaismailoğlu, 2019b; Karaismailoğlu and Erol, 2020; Karaismailoğlu and Şirin, 2020; Çıtak et al. 2021; Yıldırım et al. 2021). In the present study, the equifacial leaves were observed in the studied species, which were declared in a previous study (Orcan and Binzet, 2003) and were generally characteristic of xerophytic plants, according to Yentür (2003).

Brassicaceae is considered as a stenopalinous family (Erdtman, 1966). The sculpturing of the exine of the pollen grains plays an important role within the tribe, family, and between species in the same genus of Brassicaceae (İnceoğlu and Karamustafa, 1977; Khalik et al., 2002; Atçeken, 2016; Başer et al., 2018; Çıtak, 2019).

The family Brassicaceae was separated into 2 (reticulate and perforate) and 3 (microreticulate with lumina less than 1 μm, reticulate lumina between 1 and 2 μm, and coarsely reticulate with lumina more than 2 μm) pollen types according to the exine sculpturing by Anchev and Deneva (1997) and Khalik et al. (2002).

According to their classification system, the 2 studied varieties had a reticulate exine ornamentation. However, these 2 species were evaluated to have quite different lumina widths, which was measured as 1.87 µm in subsp. *strigosum* and 1.47 µm in subsp. *cedrorum* so they can be considered in the second type (reticulate lumina between 1 and 2 µm). Orcan and Binzet (2003) reported that *Alyssum obtusifolium* had prolate-shaped pollen grains, with a similar pollen shape in the investigated *Alyssum* species in the current study.

With the current investigation, it was clearly shown that the morphological, anatomical, and palynological features of the A. *strigosum* subspecies were stable within the same subspecies and these properties provided useful information in separating the taxa.

In conclusion, these two subspecies can separate each other according to anatomical (midrib shape), and palynological (lumina width).

Ethics in Publishing

There are no ethical issues regarding the publication of this study.

Conflict of Interest

The authors have no conflicts of interest to declare.

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