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A morphological, anatomical and palynological study of *Aethionema lepidioides* (Brassicaceae) – an endangered species endemic to Turkey

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Abstract – *Aethionema lepidioides* Hub.-Mor. is an endangered endemic species in Turkey with a very narrow natural distribution area. The present study aims to reveal the structural features of this species. For this purpose, its unknown morphological features – such as leaf, petal, sepal dimensions and shapes, filament and anther lengths, as well as seed dimensions, shape, colour and micromorphology – have been studied for the first time. To reveal the pollen characteristics of *Ae. lepidioides*, light microscope and SEM studies were conducted. Additionally, in the present study the anatomical features of *Ae. lepidioides* were also studied for the first time. The *Ae. lepidioides* leaves were found to be linear-oblanceolate, with an entire margin and a subacute to obtuse apex. The seeds were found to be dark brown and oval-shaped, with reticulated surface ornamentation. Anatomical studies found roots in the secondary growth stage, with xylem-filled pith. The stem was in the primary growth stage, featuring a multi-layer cortex under its outermost single-layered epidermis, a pronounced endodermis and a central cylinder beneath. The leaves were thick, amphistomatic and covered with a prominent wax layer. Their mesophyll was equifacial, and their stoma type was anisocytic. A stomatal index of 26 was found for the upper epidermis, while a corresponding index of 28.4 was found for the lower epidermis. The pollens were monad, radially symmetrical and isopolar. The pollen type was colpate, and the pollen shape was found to be prolate-spheroidal with a P/E ratio of 1.08.

Keywords: *Aethionema lepidioides,* anatomy, Cruciferae, endemic, morphology, pollen, Turkey

Introduction

The family Brassicaceae is one of the largest families of the dicotyl group, which includes 321 genera and 3,660 species worldwide (Al-Shehbaz 2012). The major Brassicaceae distribution centres are the Mediterranean, Irano-Turanian and Saharo-Sindian regions (Hedge 1976). Owing to its 653 native species belonging to 61 genera, Turkey is among the countries richest in Brassicaceae family members (Al-Shehbaz et al. 2007).

Aethionema S.L. is an Irano-Turanian genus that belongs to the tribe Aethionemeae and has about 60 species worldwide. The main diversity centres of the tribe Aethionemeae and genus *Aethionema* are Anatolia and Iran (Hedge 1965, Mohammadin et al. 2017, Moazzeni et al. 2018). The genus *Aethionema* includes 42 species that grow naturally in Turkey, and 19 of these species are endemic to the country at 45% endemism (Moazzeni et al. 2018). The taxa of this genus are known as 'Kayagülü' in Turkish (Ertuğrul 2012). *Aethionema* presents some taxonomic difficulties, such as flower features and fruit morphology, chromosome number and habit variety; additionally, with the absence of ripe fruit, species identification can be very difficult (Hedge 1965, 1968, Al-Shehbaz et al. 2006, Pınar et al. 2007). For this reason, new macro- and micro-morphological and anatomical studies aimed at determining the diagnostic characters of allied species are more important for the genus *Aethionema* than many other genera of the family Brassicaceae.

A literature survey revealed a very limited number of studies of the genus *Aethionema*, excepting genus morphology. After a study by Hedge (1965), Ertuğrul (1989) studied the morphology of the *Aethionema* taxa naturally distributed across Inner Anatolia. However, *Aethionema lepidioides* Hub.-Mor. was not among the species included in that study. Some recent studies have examined both the morphology and the anatomy of the Turkish *Aethionema* taxa. Ceter et al. (2018) studied the pollen morphology of some *Aethionema* species. Karaismailoğlu (2018, 2019), meanwhile, studied the

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seed mucilage content, as well as seed anatomy and morphology, of some *Aethionema* species. Pınar et al. (2007) studied seed-coat microsculpturing in some Turkish *Aethionema* species. Atçeken et al. (2016) studied the morphology, anatomy and palynology of some *Aethionema* species (*Ae. arabicum* [L.] Andrz. ex DC., *Ae. cordatum* Boiss., *Ae. armenum* Boiss. and *Ae. karamanicum* K.Ertuğrul & Beyazoğlu). Birgi and Sezer (2019) studied the morphology and anatomy of Turkish-endemic *Ae. turcicum* H.Duman & Aytaç. Additionally, Dural and Çıtak (2020) recently investigated the anatomical characteristics of another Turkish endemic species, *Ae. dumanii* Vural & Adıgüzel.

Ae. lepidioides was discovered and first described by Huber-Morath (1963) (Fig. 2B). Its morphological description is weak because it is based on a limited sample and no detailed morphological study was performed prior to the current study. For example, Flora of Turkey and the East Aegean Islands, included only very limited detail on flower characteristics in its morphological description of *Ae. lepidioides*, failing to mention its leaf and seed characteristics at all (Hedge 1965). *Ae. lepidioides* is called 'tere kayagülü' in Turkish and is only known to grow in three calcareous localities in Turkey's Sivas and Malatya provinces (Figs.1, 2A). Its populations are weak and cover a narrow area. According to the International Union for Conservation of Nature (IUCN) Red List of Threatened Species criteria, *Ae. lepidioides* is an 'endangered' endemic species on the verge of extinction (Ekim et al. 2000). Therefore, the current study is aimed at determining the unknown anatomical and seed-micromorphological features of *Ae. lepidioides* and contributing to the taxonomy of the genus by completely examining its hitherto little-known morphological features.

Materials and methods

Samples of *Ae. lepidioides* were collected from its natural habitats in Sivas Province during its flowering and fruiting periods. These plant samples' locality, collection-date and collector data were: Kangal to Gürün district, 8th km, calcareous area, 39° 07′ 52.2″ N, 37° 14′ 33.5″ E; 1,536 m, M. Tekin 1283, 30.06.2012; ibid. M. Tekin 1461, 18.06.2013; ibid. M. Tekin 1561, 05.06.2014; ibid. M. Tekin 1684, 24.06.2015. Some collected samples were dried, according to standard herbarium techniques, and stored in the Trakya University Faculty of Pharmacy Herbarium. Hedge's study (1965) was used to diagnose and identify plant specimens. For morphological measurements, herbarium and fresh plant samples were used, and at least 20 measurements were taken for each character from as many different individual plants as possible. The plant samples used in the anatomical studies of the current research were selected from the individuals which best represented the field population. Then, these samples were fixed in 70% ethyl alcohol in their natural habitat and, subsequently, stored in a refrigerator at +4 °C until their use in sectional uptake.

In the anatomical studies of the current research, cross sections of the plant samples' roots and stems, as well as both cross and superficial leaf sections, were obtained manually using a razor. These cross sections were placed in a mixture of 3:2 Alcian blue (Sigma) and Safranin O (Sigma) dyes, except for the superficial leaf sections, in order to stain pectic or lignin-substance-rich tissues differently through contrast (Davis and Barnett 1997). The cross sections were kept in this double dye until they were stained (approximately 5–10 minutes), and then they were kept in a mixture of distilled water and glycerine at a ratio of 1:1 for 10 minutes to remove excess stain from the tissues. A solution of 60 °C glycerine-gelatine was dropped onto a clean slide. Next, well-stained thin cross sections were placed into this drop and covered with a coverslip, preparing permanent microscope slides (Jensen 1962). The cross sections in these slides were examined using an Olympus BX21 light microscope. Photomicrographs were taken with an Olympus BX51 light microscope, which was connected with an Olympus DP70 digital camera. Statistical analysis of the obtained anatomical measurements was conducted with the SPSS package program (ver. 15). Stomatal indices were calculated according to the study by Meidner and Mansfield (1968). For the current research project's palynological studies with a light microscope, microscope slides were prepared using the Wodehouse method (Wodehouse 1959). The terminology of Punt et al. (2007) was used in this project's palynological studies. For this project's scanning electron microscope (SEM) study, pollen samples and mature seeds were placed onto lead staples using double-sided tape. Then, the samples were covered with gold and examined. Micrographs were obtained with the LEO 440 SEM using different magnifications.

Results

Morphological properties

Ae. lepidioides was found to be perennial, with a strongly ± woody base and, rarely, simply or generally branched, with many twiggy stems at its base. Its stems are erect, 12–40 cm, glabrous, and glaucous. Its leaves are thick, 11.8–26 × 2–7.7 mm, simple, sessile, alternate, margin-entire, linear to narrowly oblanceolate with a subacute to obtuse apex. Inflorescence is small, elongating during fruiting. The sepals are 2.0–2.5 × 1.1–1.3 mm and elliptic-obovate, with a membranous margin and a subacute apex. The petals of *Ae. lepidioides* are 4–4.9 × 2–2.4 mm, obdeltoid with a truncate apex. Its petals are pink when the flowers are budding and white when the flowers are blooming – though sometimes, the veins alone are pink. The androecium is tetradynamous, with a filament of short stamens that are 1.3–1.8 mm and a filament of long stamens of 1.5–2 mm. The anthers of *Ae. lepidioides* are 0.5–0.7 mm, with an equal length of long and short stamens. Its ovary is 1–1.4 × 0.7–1 mm, subglobose to broadly ellipsoidal, and unilocular, with two ovules. Its styles are 0.1–0.3 mm with a capitate stigma. The fruiting pedicels are 2–3 mm. Its siliculae are 2.2–4.8 × 2–3.9 mm, elliptic, slightly flattened, entire-margin, dehiscent, greenish straw-coloured, and one-seeded. Its seeds are 1.6–2 ×

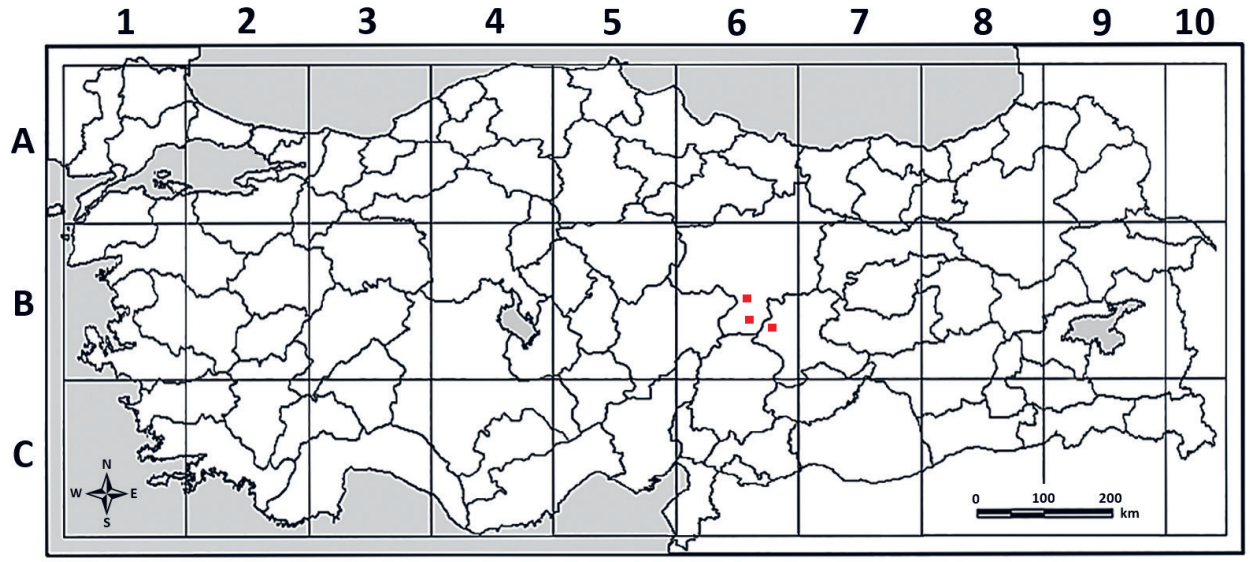

Fig. 1. Geographic distribution area of *Aethionema lepidioides* (■) in Turkey.

Tab. 1. Anatomical measurement results of *Aethionema lepidioides* (Min – minimum, Max – maximum, SD – standard deviation, N = 100).

	Width (μm)		**Length** (μm)	
	Min–Max	Mean ± SD	Min–Max	Mean ± SD
Root				
Cortex cells	8.48–43.71	21.34 ± 8.52	4.12–18.26	12.03 ± 4.24
Fascicular cambium cells	3.08–7.56	5.06 ± 1.36	1.22–3.48	1.46 ± 0.59
Interfascicular cambium cell	6.32–18.45	11.20 ± 2.52	2.27–6.75	3.90 ± 1.14
Trachea diameter	7.70–22.56	14.48 ± 5.60	–	–
Stem				
Cuticle thickness	0.55–1.01	0.71 ± 0.12	–	–
Epidermis cells	10.28–32.37	16.80 ± 4.46	5.33–25.74	14.00 ± 5.32
Cortex cells	4.33–25.74	10.90 ± 5.22	5.36–18.17	10.06 ± 2.50
Endodermis cell	11.16–37.48	21.13 ± 6.07	5.41–18.35	10.21 ± 2.50
Trachea diameter	9.39–28.15	18.14 ± 5.07	–	–
Pith cells	10.20–36.78	21.06 ± 7.30	10.15–28.07	18.16 ± 5.87
Leaf				
Upper cuticle thickness	1.04–1.31	1.15 ± 0.12	–	–
Upper epidermis cell	6.43–22.62	14.46 ± 4.53	14.07–30.50	21.46 ± 4.23
Palisade parenchyma cell	16.27–52.72	31.03 ± 10.48	5.41–20.14	12.83 ± 4.24
Spongy parenchyma cell	13.74–79.52	39.98 ± 24.35	15.77–49.13	29.42 ± 14.79
Lower epidermis cell	16.33–33.71	22.16 ± 5.07	8.22–28.32	15.53 ± 4.48
Lower cuticle thickness	0.83–1.33	1.06 ± 0.13	–	–
Mesophyll thickness	400.90–610.30	531.04 ± 40.02	–	–
Leaf thickness	452.33–655.51	593.47 ± 44.14	–	–

0.9–1.5 mm, dark brown, and ovoid-shaped, with reticulate surface ornamentation. The flowering period is on June to July. *Ae. lepidioides* grows in stony slopes and calcareous habitat range from 1,050 to 1,550 m (Fig. 2A–B).

Anatomical properties

Root: The root of *Ae. lepidioides* was in its secondary growth stage when the current study's cross section was obtained. It features a multi-layered periderm comprising flattened, tight and tile-like brown cells that align on top of one another at the outermost layer of the root cross section. The periderm layer was torn in some places and removed from the root (Fig. 4A). Beneath the periderm is a cortex layer comprising 6–10 cell rows of oval or irregularly shaped cells with intercellular spaces between these cortex cells. Under the cortex are vascular bundles that are separated from each other by large pith rays from the outside of the cross section to almost the centre of the cross section. The fascicular and interfascicular cambium layers are ordered as one to four cell rows, and they generally comprise flattened irregularly

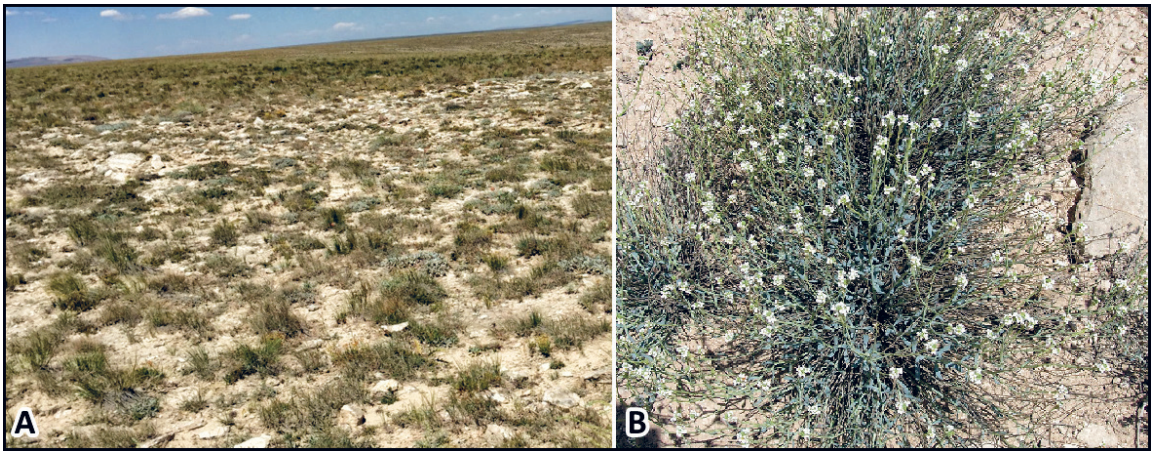

Fig. 2. Calcareous natural habitat of *Aethionema lepidioides* (A) and general appearance of *Ae. lepidioides* in its habitat (B).

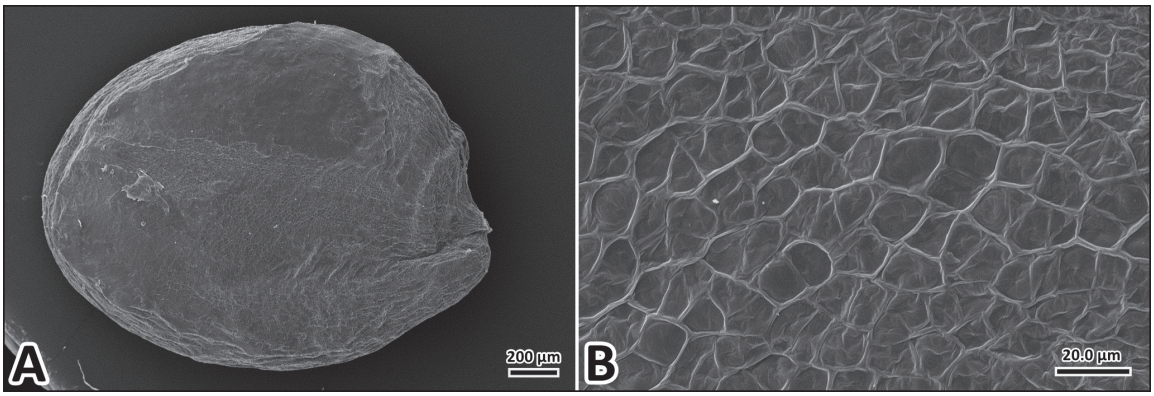

Fig. 3. Micrographs of *Aethionema lepidioides* seed. A – general view of the seed, B –ornamentation of the seed-coat surface.

shaped cells or flattened rectangular cells (Fig. 4C). The pith area is occupied by xylem elements (Fig. 4A–C, Tab. 1).

Stem: In this study's cross section, the plant's stem was in its primary stage of development. Outside this section is a single-cell-row epidermis layer whose cells are square, rectangular or ovally rectangular. The stomata in the epidermis are level with the epidermis cells, and epidermal hairs are lacking. The cortex layer under the epidermis comprises seven to nine cell rows whose cells are circular or oval. While the cortex cells close to the epidermis contain many small chloroplasts, the cortex cells close to the endoderm contain fewer large chloroplasts. Underneath the cortex is a single-cell-row endodermis layer whose cells are oval (Fig. 4F). Under the endodermis is a sclerenchymatous fibre layer, which can comprise one to four cell rows, placed corresponding to the vascular bundles and just above the phloem. Xylem is under the phloem, and no cambium is apparent between these two layers. The vascular bundles are collateral-type (Fig. 4F). The area between these vascular bundles is filled with sclerenchymatous fibres, which surround the stem in the form of an inner ring. The pith region comprises circular, oval or sometimes polygonal parenchymatous pith cells (Fig. 4D–F, Tab. 1).

Leaf: The leaf cross section revealed single-cell-row upper and lower epidermises with a square, rectangular or oval cell shape. No epidermal hair was observed on the upper or lower epidermises. A thick wax layer covers the leaf's lower and upper epidermises (Fig. 6D, F). The well-developed leaf mesophyll comprises palisade and spongy parenchyma and vascular bundles. In the mesophyll layer, the boundaries between the palisade and spongy parenchyma cells are not easily distinguishable (Fig. 5B). Palisade parenchyma is adjacent to the lower and upper epidermises of the leaf mesophyll, and spongy parenchyma is located between the two palisades. Thus, the leaf is equifacial. While the lower and upper palisade parenchymas comprise three to five rows of cells that are usually cylindrical but sometimes rectangular, the spongy parenchyma cells, with one to three cell lines, are oval, ovally rectangular or irregularly shaped. The midrib is located at the middle area of the leaf mesophyll (Fig. 5). The leaves are amphistomatic (Fig. 6A–B), mesomorphic (Fig. 5B) and stomata are anisocytic (Fig. 6A–B), and its stomatal indices were calculated as 26 for the upper epidermis and 28.4 for the lower epidermis. Therefore, the number of stomata in the lower epidermis exceeds the corresponding number in the upper epidermis (Figs. 5, 6, Tab. 1).

Palynological properties

The pollens of *Ae. lepidioides* are monad, radially symmetrical and isopolar. The pollen type is colpate, and its

shape is prolate-spheroidal with a P/E ratio of 1.08. The pollen outline is elliptic or circular from an equatorial view (Fig. 7A–D) and circular from a polar view (Fig. 7E–G). The length of their polar axis is 15.05–20,43 µm (mean: 18.65 µm), and the length of their equatorial axis is 14.66–20.16 µm (mean: 17.19 µm). Their colpus is long, stretching along the polar axis. Their colpus length is 13.29–16.33 µm (mean: 15.11 µm), and their colpus width is 4.22–9.07 µm (mean: 7.04 µm). Colpus borders are evident (Fig. 7B, F–H). The exine thickness is 0.81–1.43 µm (mean: 1.22 µm), and the exine ornamentation is reticulate, with large lumina in both the equatorial and the polar sides (Fig. 7).

Discussion

Morphological studies related to the genus *Aethionema* are much more numerous than anatomical and palynological studies on the genus. Some morphological and taxonomical studies have been conducted on the genus *Aethionema* (Hedge 1965, Ertuğrul 1989, 2012, Atçeken et al. 2016, Moazzeni et al. 2018). Like some other Turkish Brassicaceae species, a very weak morphological description of *Ae. lepidioides* was presented in the Flora of Turkey and the East Aegean Islands (Davis 1965, Hedge 1965). However, in Hedge (1965), the morphological description of *Ae. lepidioides* was also the weakest among all species of the genus since this species had not been studied with sufficient plant samples bearing all of the plant's organs due to its very narrow distribution area. The current study, by contrast, examined and revealed all of the species' morphological features, comparing them with the very limited data in Hedge's study (1965). Differences were found in some generative characters. For example, while Hedge (1965) found the plant's stylus and fruiting pedicel lengths to be 0.1–0.2 mm

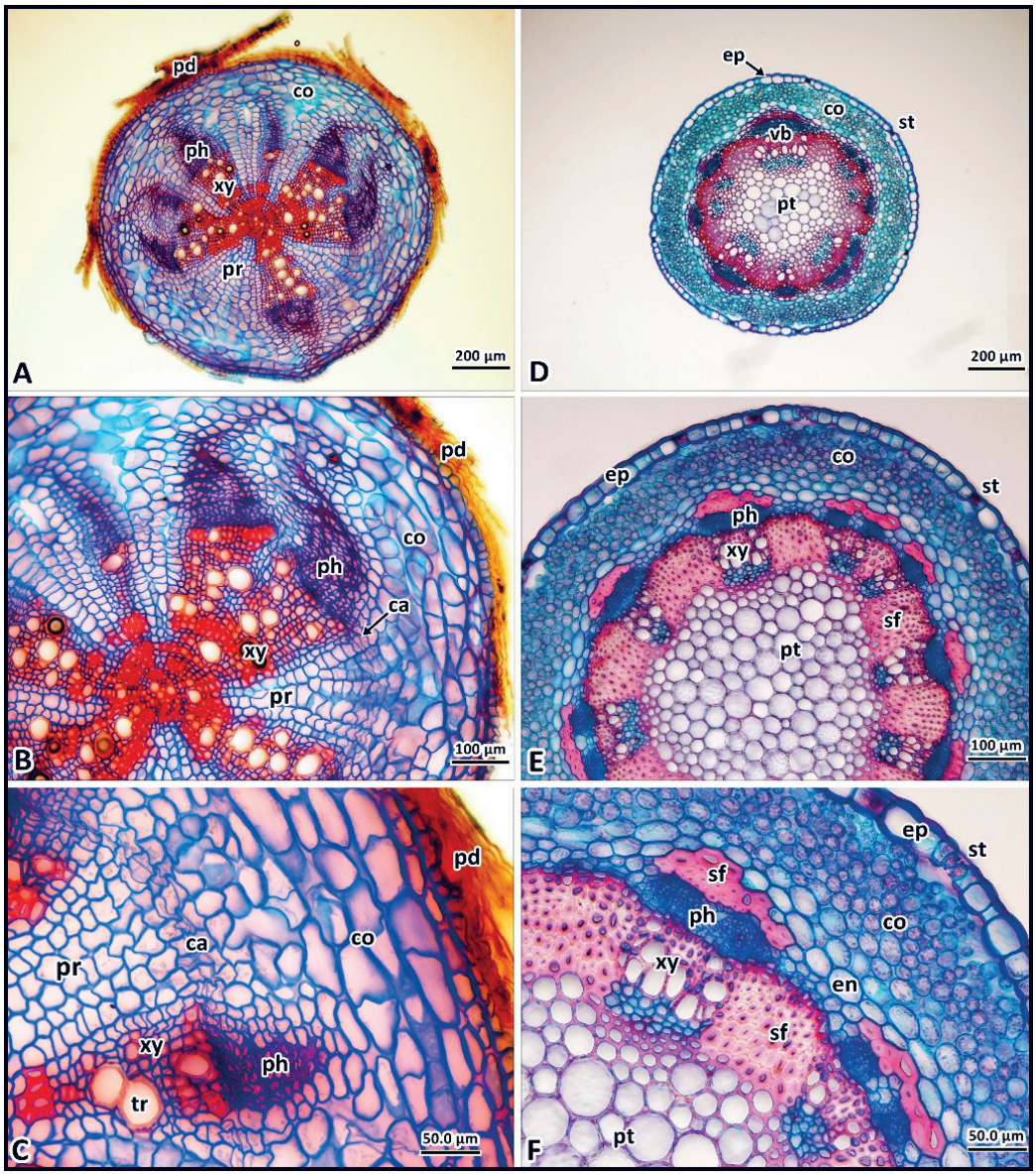

Fig. 4. Photomicrographs of root (A–C) and stem (D–F) cross sections of *Aethionema lepidioides*. Abbreviations: ca – cambium, co – cortex, en – endodermis, ep – epidermis, pd – periderm, ph – phloem, pr – pith ray, pt – pith, st – stoma, sf – sclerenchymatous fibers, tr – trachae, vb – vascular bundle, xy – xylem.

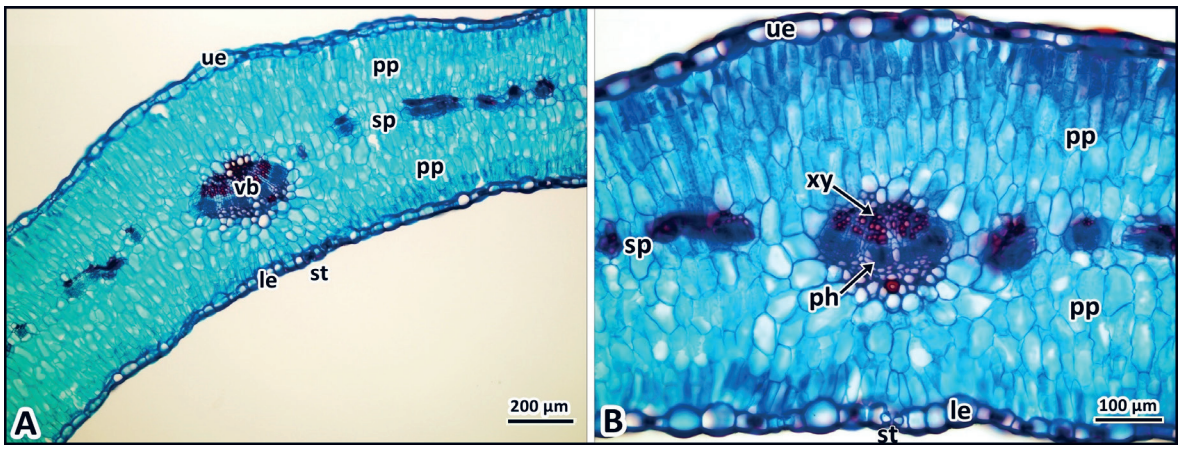

Fig. 5. Photomicrographs of a leaf-cross section of *Aethionema lepidioides*. A – general view of the leaf lamina, B – detailed view of mesophyll and midrib. Abbreviations: le – lower epidermis, ph – phloem, pp – palisade parenchyma, sp – spongy parenchyma, st – stoma, ue – upper epidermis, vb – vascular bundle, xy – xylem.

and 2.5–3 mm, respectively, the present study identified these values as 0.1–0.3 mm and 2–3 mm, respectively. Also, while Hedge (1965) specified the fruit dimension of *Ae. lepidioides* as 4 × 3 mm, the present study identified the plant's fruit dimension as 2.2–4.8 × 2–3.9 mm. Atçeken et al. (2016) studied the fruit morphology of four *Aethionema* – specifically, *Ae. arabicum*, *Ae. cordatum*, *Ae. karamanicum* and *Ae. armenum* – reporting fruit dimensions for these species of 3.99–8.64 × 3.94–7.48 mm, 3.7–7.24 × 2.94–5.91 mm, 4.8–6.39 × 4.16–5.96 mm and 4.36–5.6 × 4.37–4.8 mm, respectively. According to these results, *Ae. lepidioides* has the smallest fruits of the four species examined. *Ae. lepidioides* differs from these four other *Aethionema* species not only by the size of its fruit but also by its shape being elliptical and its colour greenish to straw-coloured. These differences were significant in that Atçeken et al. (2016) found the fruits of *Ae. arabicum* to be broadly ovate and green-yellow, while they found the fruits of *Ae. cordatum* to be ovate and green-brown, the fruits of *Ae. karamanicum* to be obovate or orbicular and green-brown and the fruits of *Ae. armenum* to be ovate-obovate and green-brown.

For the family Brassicaceae, seed-coat structure and ornamentation are considered important systematic characters. These characters are mostly used to solve classification problems between close relatives and other groups (Kaya et al. 2011). Additionally, many researchers have stated that seeds' macro- and micro-characteristics are important for separation of taxa within the family Brassicaceae (Buth and Roshan 1983, Koul et al. 2000, Khalik and Maesen 2002, Tantawy et al. 2004, Kaya et al. 2011, Karaismailoğlu 2016, Karaismailoğlu and Erol 2018).

Brochmann (1992) reported that seed colour is important for distinguishing between the genera *Draba* L. and *Bolboschoenus* (Asch.) Palla of the family Brassicaceae. Using this approach, in the present study the seed features and micromorphology of *Ae. lepidioides* were studied using SEM, and its seeds were found to be ovoid with reticulate ornamentation and a dark brown colour. Pınar et al. (2007) studied the seed features and seed-coat sculpturing of 17 Turkish *Aethionema*, excluding *Ae. lepidioides*. They specified three different seed colours amongst the species they studied: eight dark brown, five light brown and four dark green. Dark brown – the dominant colour in the Pınar et al. (2007) study – was found to be the seed colour of *Ae. lepidioides* in the present study.

Pınar et al. (2007) also determined that four different major seed surface ornamentations occur in the genus *Aethionema*, namely: reticulate, ruminate, reticulate-verrucate and verrucate. Reticulate seed surface ornamentation was found in *Ae. iberideum* (Boiss.) Boiss., *Ae. stylosum* DC., *Ae. eunomioides* (Boiss.) Bornm. and *Ae. dumanii*. Similarly, the seed surface ornamentation of *Ae. lepidioides* was found to be reticulate in the current study (Fig. 3B). Atçeken et al. (2016) studied the seed micromorphology of *Ae. arabicum*, *Ae. cordatum*, *Ae. karamanicum* and *Ae. armenum*, finding all seeds to be ovoid-shaped, with a yellowish-brown colour and verrucate ornamentation. Comparing the results, the current study found that *Ae. lepidioides* differs from all four of these other species vis-à-vis surface ornamentation and seed colour – although *Ae. lepidioides* was found to share an identical seed shape with these other species (Fig. 3A).

Karaismailoğlu (2019) studied the seed micromorphology of *Ae. syriacum* Bornm., *Ae. froedinii* Rech. f., *Ae. fimbriatum* Boiss., *Ae. speciosum* Boiss. subsp. *speciosum*, *Ae. speciosum* subsp. *compactum* Hartvig & Strid, *Ae. saxatile* (L.) W.T.Aiton, *Ae. oppositifolium* Boiss., *Ae. grandiflorum* Boiss. & Hohen., *Ae. arabicum*, *Ae. eunomioides*, *Ae. iberideum* and *Ae. armenum*. He reported that the seed shapes of *Ae. eunomioides*, *Ae. speciosum* subsp. *compactum*, *Ae. iberideum*, *Ae. armenum* and *Ae. grandiflorum* were ovoid and identical with *Ae. lepidioides*, while the other examined species were found to have elliptical seed shapes. The seed colours of *Ae. syriacum, Ae. oppositifolium* and *Ae. grandiflorum* were found to be dark brown, identical with *Ae. lepidioides*, and the seed-surface ornamentation of *Ae. froedinii* and

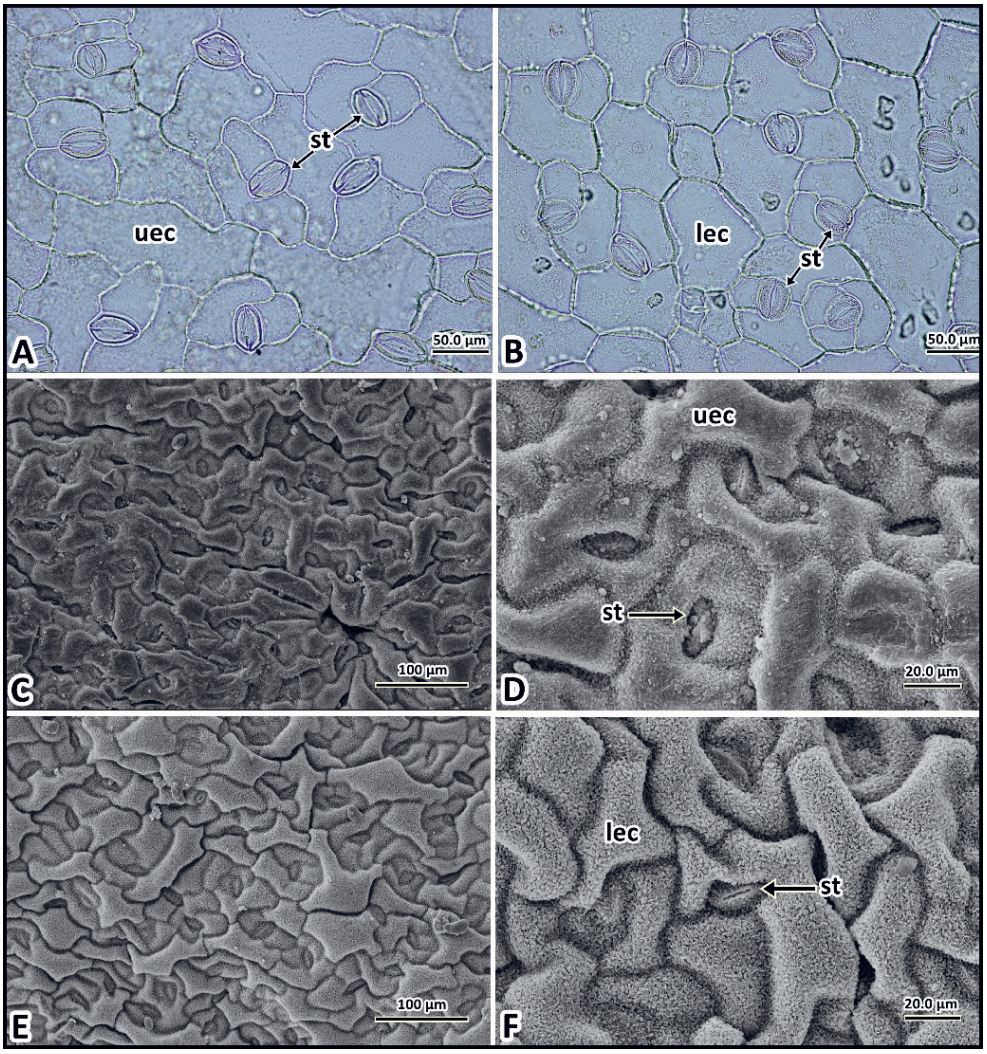

Fig. 6. Photomicrographs of leaf superficial sections (A–B) and micrographs of the leaf surface of *Aethionema lepidioides* (C–F). A, C, D – upper surface, B, E, F – lower surface; lec – lower epidermis cell, st – stoma, uec – upper epidermis cell.

Ae. iberideum was found to be reticulate, similar to *Ae. lepidioides*.

Metcalfe and Chalk (1950) conducted general anatomical investigations of the family Brassicaceae. According to Metcalfe and Chalk (1950), idioblastic cells containing myrosin are widely distributed throughout the family's taxa. In the present study, no idioblastic cells were observed in any organ tissues of *Ae. lepidioides*. Metcalfe and Chalk (1950) reported that the leaf mesophyll layer can be bifacial or equifacial, that its stomata are cruciferous (anisocytic) and that its wax layer is mostly present on the epidermis surface of the leaves in the family's taxa. These results are aligned with the current study's findings regarding *Ae. lepidioides*' equifacial mesophyll, anisocytic stomata and fairly distinct wax layer on both the upper and lower epidermis surface (Figs. 5, 6). Regarding stem anatomy, Metcalfe and Chalk (1950) reported that the endodermis is well defined in many different species of the genera Brassicaceae; accordingly, the endodermis of *Ae. lepidioides* was evident in the current study (Fig. 4F). Moreover, Metcalfe and Chalk (1950) reported the pith region of stems in the taxa Brassicaceae is parenchymatous, occupying a large proportion of the stems' total diameter. Similarly, the current study found the stem pith region of *Ae. lepidioides* to be filled by parenchymatous cells; however, unlike Metcalfe and Chalk's (1950) findings, this study found the pith region of *Ae. lepidioides* not to constitute a large part of the stem's diameter (Fig. 4D, E).

Birgi and Sezer (2019) studied the root, stem and leaf anatomy of the other rare *Aethionema* species endemic to Turkey, *Ae. turcicum*, reporting anatomical results that generally align with the current study's findings on *Ae. lepidioides*. A principal anatomical difference, however, is the presence of bifacial mesophyll in the leaves of *Ae. turcicum* versus the equifacial leaves of *Ae. lepidioides* (Fig. 5). Dural and Çıtak (2020) studied the anatomical features of *Ae. dumanii*, which is also endemic to Turkey, and Atçeken et al. (2016) studied the anatomical features of four *Aethionema* species (*Ae. arabicum*, *Ae. cordatum*, *Ae. karamanicum* and *Ae. armenum*). In comparing the anatomical features of *Ae. lepidioides* with the reported anatomical results of the five species studied by Dural and Çıtak (2020) and Atçeken et al. (2016), the current study identified a general similarity in anatomical structure despite some important differences

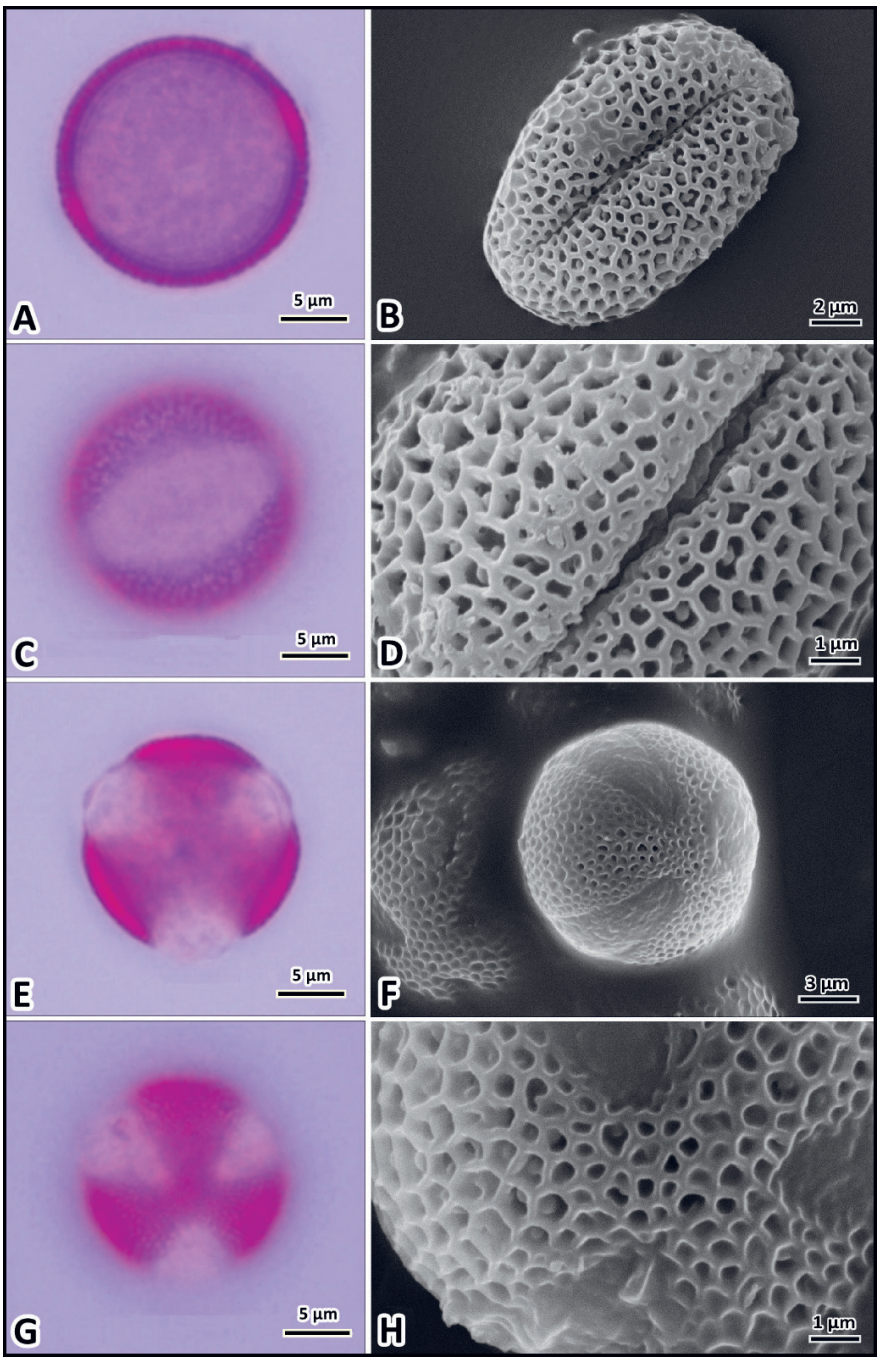

Fig. 7. Pollen grains photomicrographs (A, C, E, G) and micrographs (B, D, F, H) of *Aethionema lepidioides*. A, B – an equatorial view, C, D – detailed apertures and exine ornamentation in an equatorial view, E, F – a polar view, G, H – detailed apertures and exine ornamentation in a polar view.

for *Ae. lepidioides*. For example, while *Ae. lepidioides* has a root cortex of six to 10 cell rows, the four species studied by Atçeken et al. (2016) had corresponding cortices of five to eight cell rows, versus 10–11 corresponding cell rows for *Ae. dumanii* (Fig. 4A–C). Atçeken et al. (2016), moreover, stated that *Ae. arabicum* and *Ae. karamanicum* have single-cell-rowed stem epidermises, while *Ae. cordatum* and *Ae. armenum* have two-to-three-cell-rowed stem epidermises. Dural and Çıtak (2020) reported that the stem epidermis of *Ae. dumanii* is single-cell-rowed. The current study found *Ae. lepidioides*, which has a single-cell-rowed stem epidermis, to be similar to *Ae. arabicum*, *Ae. karamanicum* and *Ae. dumanii* regarding this feature (Fig. 4D, F). While a stem endodermis layer is present in *Ae. lepidioides* (Fig. 4F) and the four species studied by Atçeken et al. (2016), Dural and Çıtak (2020) did not specify its presence in *A. dumanii*. Additionally, the present study found the leaves of *Ae. lepidioides* to be equifacial (Fig. 5). The leaves of *Ae. cordatum*, *Ae. armenum* and *Ae. karamanicum* were also reported to be equifacial, while the leave of *Ae. arabicum* were found to be unifacial (Atçeken et al. 2016).

The family Brassicaceae are a stenopalynous family the members of which usually have tricolpate-type pollens with reticulate exine ornamentation. Pollen shape varies more or

less among the genera of the same tribe; however, such diversity is rare amongst different species of the same genus (Erdtman 1972, Reile 1992, Abdel Khalik et al. 2002, Tekin and Martin 2017). The pollens of *Ae. lepidioides* were found to be tricolpate, with reticulate exine ornamentation, by light microscope and SEM – identical to the general pollen structure of the Brassicaceae family (Erdtman 1972) (Fig. 7).

Some studies have examined the pollen morphology of the *Aethionema* genus. İnceoğlu and Karamustafa (1977) studied the pollen properties of *Ae. arabicum* and *Ae. armenum* with a light microscope. Atçeken et al. (2016) studied the pollens of *Ae. arabicum*, *Ae. cordatum*, *Ae. karamanicum* and *Ae. armenum*. Ceter et al. (2018) also studied the pollen morphology of 23 *Aethionema* species, including *Ae. lepidioides*. Both Atçeken et al. (2016) and Ceter et al. (2018) reported such pollen features as symmetry, polarity, pollen type and exine ornamentation to be identical to the corresponding features found in the present study for *Ae. lepidioides* (Fig. 7). However, some differences were also observed. Atçeken et al. (2016) found the pollen shapes of *Ae. cordatum* and *Ae. karamanicum* to be prolate, observing the corresponding shapes for *Ae. armenum* and *Ae. arabicum* to be subprolate and prolate-spheroidal, respectively. According to the present study's findings, *Ae. lepidioides* – which has a prolate-spheroidal pollen shape with a P/E ratio of 1.08 – is similar to *Ae. arabicum*. Exine thicknesses were found to be similar, varying between 0.91 µm and 1.06 µm in the four species studied by Atçeken et al. (2016), versus 1.22 µm for *Ae. lepidioides*.

Ceter et al. (2018) found the mean polar and equatorial axis lengths of *Ae. lepidioides* pollens to be equal at 16.6 µm; therefore, the corresponding P/E ratio was 1, and the pollen shape was also spheroidal. However, the current study found the polar and equatorial axis lengths of *Ae. lepidioides* pollens to be 18.65 µm and 17.19 µm, respectively, with a P/E ratio of 1.08; therefore, the pollen shape was determined to be prolate-spheroidal. Additionally, another important difference was found with respect to colpus dimensions. While Ceter et al. (2018) reported the mean pollen colpus length and width of *Ae. lepidioides* as 11.4 µm and 4.2 µm, respectively, the present study identified these values as 15.11 µm and 7.04 µm, respectively (Fig. 7C, F). Moreover, while Ceter et al. (2018) determined an exine thickness of 1 µm, the current study found an exine thickness of 1.22 µm for *Ae. lepidioides*. Consequently, the present study and the study by Ceter et al. (2018) found significant differences in the same species' pollen features – such as shape, colpus length and width, and exine thickness.

Thus, the current study offers the most comprehensive morphological research on *Ae. lepidioides*. Many of the hitherto unknown morphological features of the species' vegetative and generative organs were revealed in this study and discussed via comparisons with the limited literature available. Moreover, this research constitutes the first anatomical and seed-micromorphological study on the species *Ae. lepidioides*. The palynological features of *Ae. lepidioides* were also studied, and pollen photomicrographs and micrographs of the plant were published for the first time. Additionally, the narrow spread and endangered status of *Ae. lepidioides* increases this study's importance. The author believes that many structural features determined by structural studies on *Ae. lepidioides* will contribute to similar future studies on both the taxa of the genus *Aethionema* and the taxa of the other genera of the family Brassicaceae.

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