

INVESTIGATIONS ON A MONOTYPICAL GENUS: *PENTAPLEURA* HAND.-MAZZ. (LAMIACEAE)

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Abstract

The genus *Pentapleura* Hand.-Mazz. (Lamiaceae) is represented by only one species *Pentapleura subulifera* Hand.-Mazz and distributed in southeast of Turkey and north Iraq. The stem and leaf of the species were examined for anatomical studies. The stems are rectangular in shape and the leaves are equifacial type. The micromorphologic properties of trichomes, nutlets and pollens were described. Trichomes can be divided into two types as nonglandular and glandular. Also, glandular trichomes are of two types as peltate and capitate type. The capitate trichomes are divided into four subtypes. The nutlets are obovoid shaped and they have the prominent undulate-ridged ornamentation. The pollen grains of the species are hexacolpate, suboblate to subprolate in shape and they have perforated and microreticulate exine ornamentation.

Introduction

The genus *Pentapleura* Hand.-Mazz. (Lamiaceae, Nepetoideae), represented by only one species, *Pentapleura subulifera* Hand.-Mazz. is distributed in southeast of Turkey and northern Iraq (Handel-Mazzetti 1913, Davis *et al.* 1982, Harley *et al.* 2004).

There is a study on the chemical characteristics of the species in Turkey by Başer *et al.* (2005), however, there has been no comprehensive investigation involving different aspects of *P. subulifera* as yet. Moon *et al.* (2008) investigated pollen of the two other subtribes Salviinae and Nepetinae, on the systematic significance of pollen characters in tribe Menthae. Also a comparative micromorphological study of leaves was carried out on 102 species of Menthae where 61 species were selected for the anatomical study by Moon *et al.* (2009).

Trichomes morphology can be meaningful regarding the systematic relationships among the species of Lamiaceae (Özkan 2008). The morphology, distribution and frequency of glandular trichomes are used as determinative characters at sub-family level in Lamiaceae (Ascensão *et al.* 1995).

The importance of scanning electron microscopy (SEM) for the study of nutlet surfaces and the taxonomic value of nutlet characters has been described for many genera of Lamiaceae (Husain *et al.* 1990, Demissew and Harley 1992, Marin *et al.* 1996, Budantsev and Lobova 1997, Jamzad *et al.* 2000).

Until now, the anatomical, palynological and nutlet characters of *P. subulifera* have not been studied. Also, some characters including indumentum of leaves, calyx and corolla and nutlets, were not included in Flora of Turkey. This study was undertaken to find out important anatomical and palynological characters which play an important role in the taxonomy of *P. subulifera*.

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Materials and Methods

Pentapleura subulifera specimens were collected from Şırnak province in Turkey. Voucher specimens were deposited in the Herbarium of the Faculty of Education of Balıkesir University in Turkey. Collection information: Turkey C9 Şırnak: Şırnak to Hakkari, 45 km, 37°21'700"N, 043°09'700"E, 965 m, 17.08.2008, B.Yıldız (16937), T. Dirmenci & M. Fırat.

For bright-field microscopy, fixed materials from the specimens were prepared. Mature stems and leaves of living materials were fixed in FAA for 24 hrs. Then, materials were stored in 70% alcohol for anatomical studies. Transverse sections of stems and leaves were prepared manually and dehydrated in a graded ethanol series, and mounted in glycerine-gelatine. Olympus BX 51 phase contrast binocular microscope with drawing tube were used in anatomical drawings and observations. Specimens and pollen grains were mounted onto SEM stubs using double-sided adhesive tape and coated with gold. Nutlet micromorphology of *P. subulifera* was studied by Tabletop Scanning Electron Microscopy.

Results and Discussion

The stem is rectangular (Fig. 1) and covered with glandular and non-glandular trichomes. The epidermis is composed of a single layer of compactly arranged paranchymatous cells. Collenchyma tissue, consisting of regular cells, is located immediately under the epidermis. There are 4 - 5 layers of collenchyma in the corners and 2 - 3 layers in between the corners of the stem sections. The paranchyma tissue is 1 - 2 layered under the collenchyma.

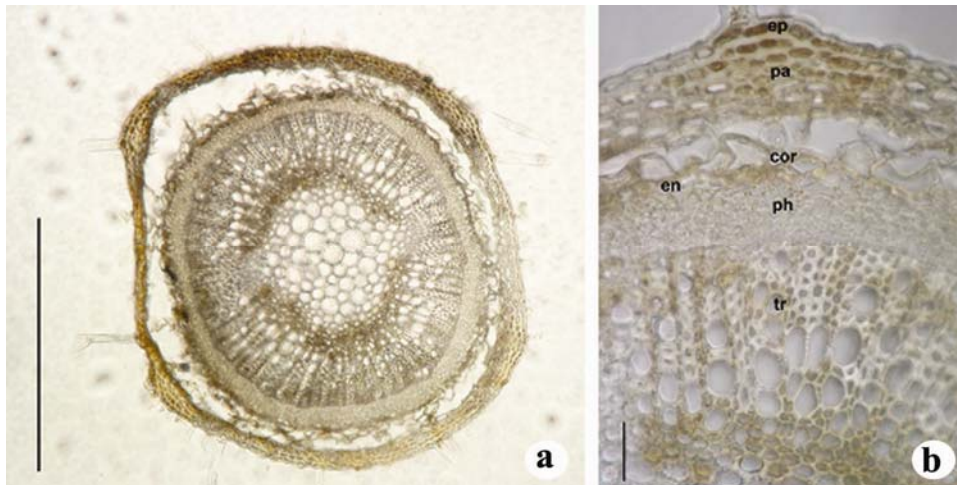


Fig. 1. Stem anatomy of *P. subulifera* (ep: epidermis, pa: parenchyma, col: collenchyma, cor: cork, en: endodermis, ph: phloem, tr: vessel. Bar: a: 500 μ m, b: 100 μ m).

Endodermis and pericycle are not distinguishable. The phloem is 5-7-layered and consists of irregular or rectangular cells. The xylem comprises large vessel and tracheids. The vessels are orbicular or ovoid while the tracheids are polyhedral. The pith is composed of large orbicular or polygonal parenchymatous cells.

Leaf epidermal cells are long and irregularly shaped, with sinuous anticlinal walls. The cuticle is smooth/striated on both the adaxial and abaxial surfaces. Both the adaxial and the abaxial

epidermis consist of uniseriate oval and rectangular cells in transverse section. The upper walls of the epidermis are thicker than the lateral and lower walls. Cells of the adaxial epidermis are larger than those of the abaxial epidermis. Both the adaxial and the abaxial epidermis are covered with a relatively thick cuticle and is covered with trichomes. Trichomes are both of nonglandular and glandular types. The stomata are diacytic and occur both surface of epidermis (amphistomatic). They are hygromorph type and located a little higher than the epidermal level. The leaf is the equifacial type. Mesophyll tissue consists of one layered palisade parenchyma in the both surfaces. Spongy parenchyma is two or four layered and located between palisade parenchyma tissue (Fig. 2).

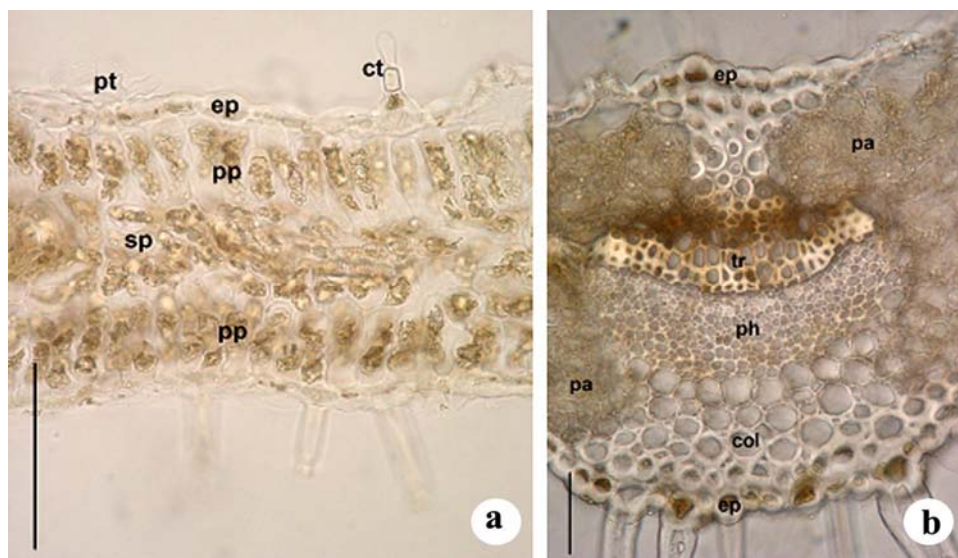


Fig. 2. Leaf anatomy of *P. subulifera*. a: Lamina, b: Midribregion (pt: peltate trichome, ct: capitata trichome, ep: epidermis, pp: palisade parenchyma, sp: spongy parenchyma, col: collenchyma, pa: parenchyma, tr: vessel, ph: phloem. Bar: 100 μ m).

The midrib region forms a projecting part. The shape of the vascular bundle is elliptic. It is collateral type, occurring over a large area. The xylem towards the adaxial surface is with the phloem towards the abaxial surface of epidermis. The phloem is surrounded by several layers of collenchymatous tissue. Also, the collenchymatous cells are located below both the adaxial and the abaxial epidermis in the midrib region (Fig. 2).

Trichomes micromorphology: Trichomes in *P. subulifera* can be divided into two main types: nonglandular and glandular (Fig. 3).

Nonglandular hairs are present on the stems, leaves and calyces (Fig. 4). They are found on both surfaces in leaf (Fig. 4 c-h). They are scarce on the adaxial surfaces, and are mainly restricted to the ribs on the abaxial surfaces. The nonglandular trichomes are covered by micro-papillae and composed of a basal epidermal cell while additional cells are uniseriate and tapering towards the apex. They usually consist of two to seven (rarely ten) cells and are straight or curved (Fig. 3).

In addition, glandular trichomes can be divided into two types, peltate and capitates (Fig. 3). The peltate glandular trichomes consist of a basal cell, a short unicellular stalk and a broad head of 12 secretory cells. From these cells, eight large and peripheral, four are small and occupy the

central area of the head. Capitulate trichomes are more common than peltate ones and two types of capitulate trichomes, differing in structure and size, were observed:

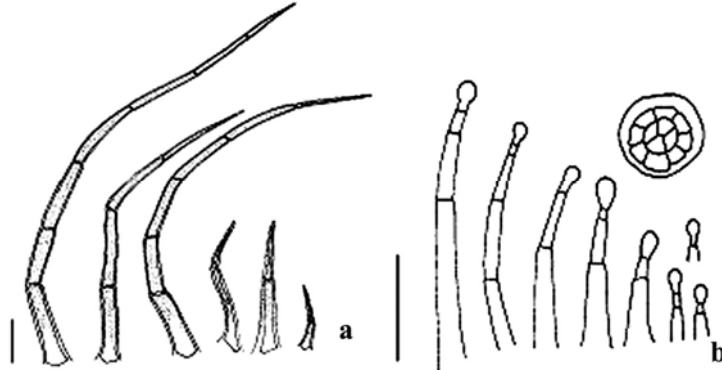


Fig. 3. Trichomes types in *P. subulifera*. a: Nonglandular, b: Glandular trichomes (Bar: 50 μm).

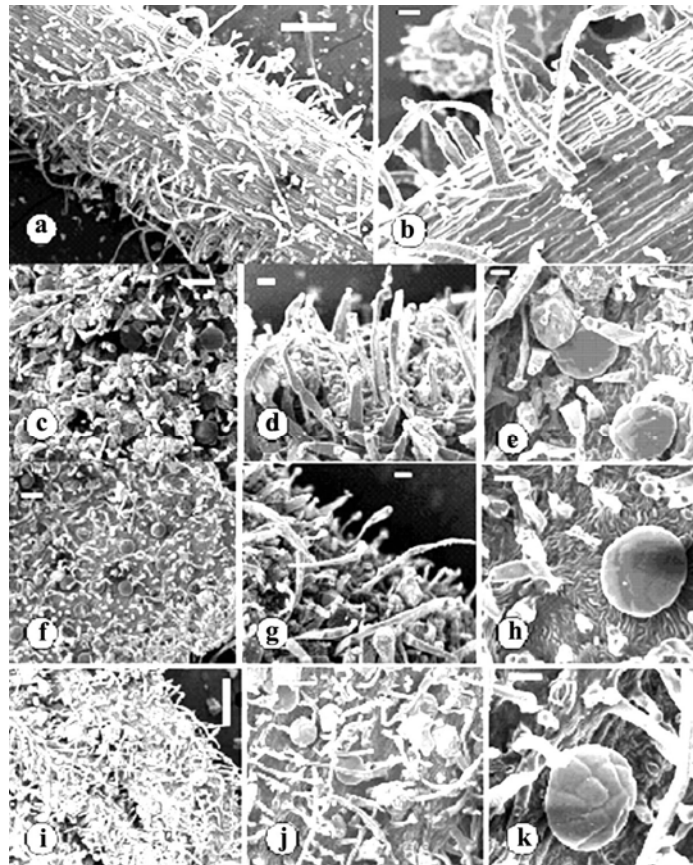


Fig. 4. Scanning electron micrographs of stem (a, b), leaf (c-e: adaxial, f-h: abaxial) and calyx (i-k) show trichomes in *P. subulifera* (a, i: 200 μm ; b, d, e, g, h, k: 20 μm ; c, f, j: 100 μm).

Type I consists of short or long a stalk cell and a globose unicellular head.

(a) Subtype Ia: Presence of neck below the head, (b) Subtype Ib: Absence of neck below the head

Type II consists of a short or a long stalk cell and a pear-shaped unicellular head. (a) Subtype IIa: Presence of neck below the head, (b) Subtype IIb: Absence of neck below the head.

Glandular trichomes are more common than the nonglandular ones. Capitate trichomes are more common than peltate trichomes and they are found on the stems, calyces and leaves (Fig. 4).

Nutlet micromorphology: The nutlets are dark brown in colour. The basic shape of nutlets is obovoid and trigonous. The apex is obtuse or obtuse-rounded. The areole is bilobed, white coloured and it is V-shaped. Nutlet size ranged from 1.4 to 1.5 mm in length and from 0.67 to 0.69 mm in width. The nutlet ornamentation of *P. subulifera* is of smooth type which consists of prominent undulate-ridged pattern (Fig. 5).

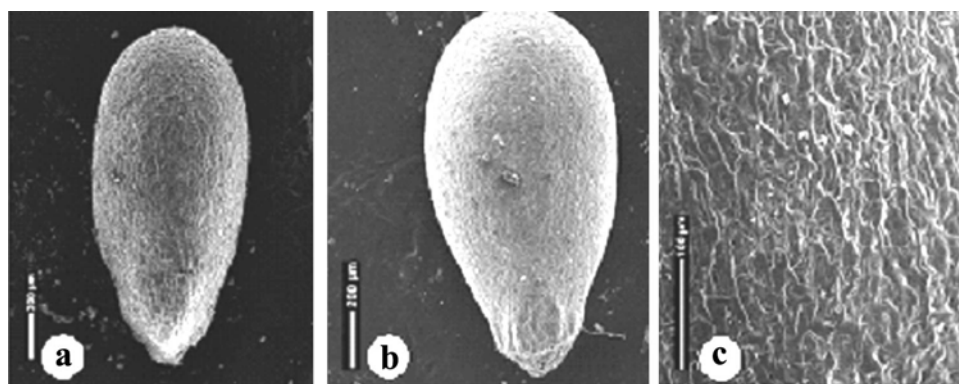


Fig. 5. Scanning electron micrographs of nutlet in *P. subulifera* (a: dorsal, b: ventral, c: surface).

Palynological studies: Pollen grains are shed as monads and hexacolpate. In SEM, ranges of dimensions are: polar axis (P) 42-48 μm , equatorial axis (E) 23-36 μm , colpus length (Cl) 30-42 μm and colpus width (Clg) 0.5-1 μm ; the shape of dry pollen grains is subprolate (P/E 1.33-1.82 μm) or P 20-25 μm , E. 24.4-30.6 μm , Cl 17.6-20.8 μm , Clg 2.8-5 μm ; the shape of dry pollen grains is suboblate (P/E 0.81 μm). The basic exine ornamentation is perforated and microreticulate. The thick colpus membranes are beset with granules in suboblate pollen grains. The outline in polar view is circular in suboblate pollens or elliptical in subprolate pollens (Fig. 6).

According to Davis *et al.* (1982), the genus *Pentapleura* is similar to *Cyclotrichium* in habit, but shrubbier and very distinct in calyx structure. The genus *Pentapleura* is an isolated genus in tribe *Mentheae* by its calyx structure with 5 main veins along crest of narrow wings and 5 weak veins in lower half of membranous inter-costal furrows (Davis *et al.* 1982, Harley *et al.* 2004). Also, *Pentapleura* is closely related to the genus *Zataria* than *Origanum* and *Cyclotrichium* with respect to phylogenetic properties in tribe *Mentheae* (Davis *et al.* 1982, Brauchler *et al.* 2010).

Anatomical features typical of the Lamiaceae (Metcalf and Chalk 1950) such as the rectangular cross-section of the stem, well-developed collenchyma in each corner and diacytic stomata, were also observed.

In tribe *Mentheae*, *Pentapleura* is close to the genus *Origanum* with phylogenetic and morphological properties. But, there are some anatomical differences. Cork layer is not observed in the stem of *Origanum onites* L., but present in *P. subulifera*. The vessels and tracheids in the

stems of *P. subulifera* are larger than those of *O. onites* and *O. vulgare* which have bifacial leaves, but *P. subulifera* has equifacial leaves (Gönüz and Özgörgücü 1999).

As in most Lamiaceae species, *P. subulifera* bears both glandular with peltate, capitate type and nonglandular trichomes. Glandular trichomes are more common than nonglandular ones. Nonglandular trichomes were observed in all parts of the plant and 2-4-celled trichomes were common than others. They are straight or curved and covered by cuticular micro-papillae. Same trichomes have been reported in *Cyclotrichium* (Boiss.) Manden. & Scheng. (Kaya *et al.* 2000, Satıl *et al.* 2011) and *Satureja* L. (Satıl and Kaya 2007), *Origanum rotundifolium* Boiss., *O. vulgare* L. subsp. *viride* (Boiss.) Hayek and *Teucrium hircanum* L. (Özcan and Eminađaođlu 2014).

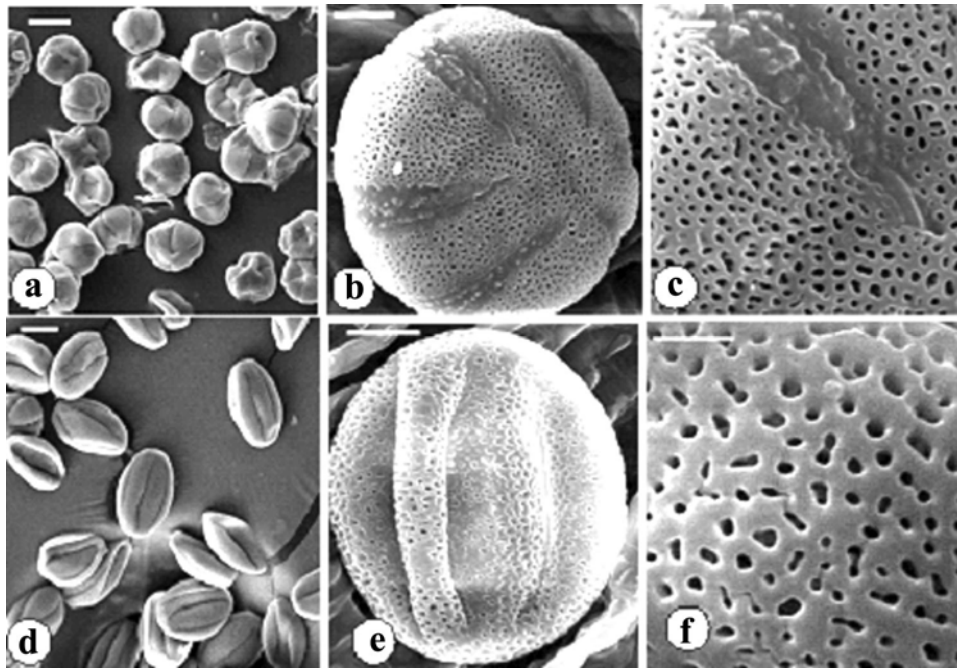


Fig. 6. SEM of pollen grains in *P. subulifera* (a: suboblate pollen grains, b: pollen shape in the polar view, c: microreticulate exine ornamentation, d: subprolate pollen grains, e: pollen shape in the equatorial view, f: microreticulate exine ornamentation, a,d: 20 μ m, b,e: 6 μ m, c,f: 2 μ m).

Capitate trichomes are widespread in the Lamiaceae, but they vary greatly in structure and size (Werker *et al.* 1985). The capitate hairs of *P. subulifera* consists of short or long stalk cell with a globose or a pear-shaped unicellular head and four subtypes of these capitate trichomes are also present. Five types of capitate trichomes were observed by Satıl *et al.* (2011) in *Cyclotrichium*. Type I-IV of these trichomes are similar to capitate trichomes in *P. subulifera*. On the other hand, eight types of capitate trichomes were determined in *Salvia halophila* Hedge (Kaya *et al.* 2007) and type III, IV and VI of capitate trichomes are similar to our samples. A short unicellular or two-cellular stalks and a globose or pear-shaped head cell capitate trichomes have been also recorded in *Ziziphora* species (Kaya *et al.* 2013). According to Davis *et al.* (1982), the genus *Pentapleura* resembles *Cyclotrichium* in habit. Trichome properties of *P. subulifera* supports this similarity.

Kaya and Dirmenci (2008) examined the nutlet surfaces of the *Nepeta* L. species. The smooth nutlets consists of three subtypes as undulate-ridged, cellular and reticulate. The undulate ridged subtype is the same to that of *P. subulifera*.

P. subulifera has hexacolpate pollen and belongs to the subfamily Nepetoideae. In the present study, the shape of the pollen grains is mainly suboblate or subprolate in dry grains (SEM). The sculpturing of the exine in *P. subulifera* is perforated and microreticulate. The shape of *P. subulifera* pollens in study of Moon *et al.* (2008) is oblate, suboblate, oblate spheroidal, prolate spheroidal and subprolate types and the basic exine ornamentation is recognized as microreticulate. The present results support the study of Moon *et al.* (2008) In addition, pollen of *Acinos arvensis* (Lam.) Dandy and *A. rotundifolius* Pers. (Kaya and Kutluk 2007) bear clearly defined thickening of the colpus membrane while *P. subulifera* also possess similar structure.

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