

Anatomical and micromorphological properties of some *Tanacetum* L. (Asteraceae) taxa from Turkey and their systematic implications

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Abstract – In this study, the anatomy and trichome micromorphology of *Tanacetum macrophyllum* (Waldst. & Kit.) Schultz, *T. parthenium* (L.) Schultz, *T. poteriifolium* (Ledeb.) Grierson and *T. vulgare* L. were examined by light microscopy and scanning electron microscopy. Some anatomical characters such as presence of secretory cavities and pith in root and mesophyll type in leaf provide information of taxonomical significance. In addition, the existence of a parenchymatic layer, which consists of elongated parenchymatic cells in the stem of *T. macrophyllum*, is a distinguishing character. The results obtained from scanning electron microscope studies showed that trichome micromorphology varies among examined taxa. In *T. macrophyllum*, the eglandular and glandular trichomes especially on disc florets, ligulate florets and cypselas are more sparse, whereas disc florets and cypselas of the other taxa are covered with abundant glandular trichomes. Additionally, *T. poteriifolium* and *T. parthenium* has a distinct distribution of glandular trichomes forming in a row across the entire cypselas surface.

Keywords: Asteraceae, anatomy, scanning electron microscopy, *Tanacetum*, trichome micromorphology

Introduction

Tanacetum L. is a large genus in the Asteraceae (Compositae) family containing 150–200 species (Brown et al. 1999). In the Flora of Turkey, the genus *Tanacetum* is represented by nearly 60 taxa (Fahn 1979, Güner et al. 2000). *Tanacetum* species have been used in standard and traditional medicines since medieval times (Holetz et al. 2002, Stevović et al. 2010). The essential oils and extracts of members of the *Tanacetum* genus exhibit anti-inflammatory (Brown et al. 1997), antibacterial (Stefanović et al. 1988), antifungal (Hethelyi et al. 1991, Neszmelyi et al. 1992), insecticidal (Hough-Golstein and Hahn 1992), antioxidant (Bandoniene et al. 2000, Mantle et al. 2000), antimalarial (Jansen 2006) and vasorelaxing effects (Lahlou et al. 2008). It has also been reported that the essential oil in Tansy (*T. vulgare*) helps as an indicator of plant adaptation to environmental stress conditions such as drought, high temperature, intense radiation and heavy metal content (Abu-Darwish and Abu-Dieyeh 2009, Stevović et al. 2009). There are a number of biochemical studies on *Tanacetum* (Williams et al. 1999). It has been indicated that sesquiterpene lac-

tones are a predominant compounds in this genus (Marles et al. 1995).

Some morphological and anatomical studies of the Asteraceae family have been carried out. Metcalfe and Chalk (1950) reported that due to the diversity of their habitats, Asteraceae species show various anatomical differentiations, especially in the structures of leaves. The effects of environmental factors on morphological and anatomical structure of Tansy (*T. vulgare*) have also been determined. Fahn (1979) observed that secretory structures have an important diagnostic value in Asteraceae. These secretory structures such as ducts, cavities, idioblasts, laticifers, hydathodes, extrafloral nectaries and trichomes presented diagnosis value at genus level (Castro et al. 1997).

It has been suggested that trichome micromorphology is useful in the systematics of Asteraceae (Ciccarelli et al. 2007). Napp-Zinn and Eble (1978) carried out a detailed ultrastructure study of the glandular and non-glandular hairs of twenty genera of the Anthemidae. Glandular hairs in Asteraceae may be widely distributed both on the vegetative and the floral parts (Ciccarelli et al. 2007). In several species of the Asteraceae, these trichomes have been used to

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elucidate the biosynthesis of terpenes (Neszmelyi et al. 1992, Maes et al. 2011, Majdi et al. 2011, 2013).

Micromorphological characters of cypselas have been used as discriminative characters to delimit the various taxa in the family Asteraceae (Zhu et al. 2006, Abid and Qaiser 2008a, b, Akcin and Akcin 2014). Cypselar external micromorphology and surface sculpturing in members of Asteraceae may provide useful data for the delimitation of genera (Garg and Sharma 2007, Pandey and Kumari 2007). Abid and Qaiser (2009) found the surface pattern of cypselas of the genus *Tanacetum* very useful for specific delimitation.

Regarding the medicinal and economic importance of *Tanacetum* species, studies on anatomy and trichome micromorphology of Turkish *Tanacetum* species are rather limited. So far, there have been no detailed anatomical and micromorphological studies on *Tanacetum* species naturally distributed in the Northern Anatolian region. Therefore, in this research our objective is to determine the anatomical and micromorphological characters of *T. macrophyllum*, *T. parthenium*, *T. poteriifolium* and *T. vulgare* distributed in Northern Anatolia and their contribution to the taxonomy of *Tanacetum* taxa.

Material and methods

Plant material belonging to the taxa involved was collected from North Anatolia in the year 2011, during the flowering period. The taxonomical descriptions of the specimens were made according to Grierson (1975). Voucher samples were deposited in the Herbarium of the Department of Biology of Ondokuz Mayıs University (OMUB), Samsun, Turkey. The collection data for the studied taxa are listed in Tab. 1.

Samples for anatomical studies were fixed in 70% alcohol. Anatomical studies were carried out on 30 samples. In these samples, cross sections of root, stem and leaves and surface sections of leaves were used. Sections were stained with safranin-fast green and mounted in entellan (Johansen 1944). Photographs were taken with a Nikon-Coolpix P5100 digital camera. All anatomical measurements were made with the use of Image J program on the figures. The mean and standard deviation (SD) of measurements were calculated with the use of the statistical package programme SPSS (version 10.0). The significance of these measurements was determined by Duncan test with one-way analysis of variance (ANOVA).

Micromorphological studies were carried out on stem, leaf, phyllaries, ligulate floret, disc floret and cypselas obtained from these samples. For scanning electron microscopic observations (SEM), dried samples were mounted on stubs using double-sided adhesive tape, coated with gold, examined and photographed with a JEOL-Neoscope JCM-5000 scanning electron microscope. The terminology of the cypselar characters follows Abid and Qaiser (2009).

Results

Anatomical properties

Tanacetum macrophyllum

Root anatomy – In transverse section, the periderm layer on the outermost surface is thick and multilayered. Secondary cortex is 8–9 layered and consists of parenchymatic cells. Secretory canals in cortex are present. The mean diameter of these canals, which are elliptic in shape, is $49.26 \pm 8.87 \mu\text{m}$ (Tab. 2). Cambium cells are not distinguishable. Phloem and xylem elements can be distinguished in the vascular tissue. The xylem consists of tracheary elements. In the center, there is pith composed of primary xylem elements. The mean diameter of vessel members is $19.75 \pm 6.60 \mu\text{m}$ (Tab. 2).

Stem anatomy – Epidermis consists of prismatic thick-walled cells with thick cuticle. Under the epidermis, parenchyma cells, usually one-layered, are located. A multilayered lacunar collenchyma layer is more developed at the corners of the stem. The sclerenchyma tissues are located under the cortex and are $60.13 \pm 7.65 \mu\text{m}$ in thickness. Cambium cells are distinguishable. Xylem and phloem elements are clear. The trachea diameter is $18.60 \pm 7.67 \mu\text{m}$. The pith is large and composed of parenchymatic cells with intercellular spaces in the center of stem (Fig. 1C).

Leaf anatomy – The upper and the lower epidermis are covered with a thin cuticle layer (Fig. 1F). Both epidermises consist of uniseriate, oval or rectangular cells. The leaf is of the bifacial type. The mesophyll is composed of elongated rectangular palisade parenchyma cells and isodiametric spongy parenchyma cells. Palisade parenchyma cells are $11.59 \pm 1.39 \mu\text{m}$ in width (Tab. 2). Vascular bundles are surrounded by a parenchymatic bundle sheath. The stomata are anomocytic and are found only in the lower epidermis. The dimensions of stomata are $29.76 \pm 2.65 \times 23.13 \pm 1.92 \mu\text{m}$ (Tab. 2).

Tab. 1. List of studied *Tanacetum* L. taxa and their localities and collectors.

Taxa	Specimen number and collector	Locality
<i>T. macrophyllum</i>	Ş.Dereli 132	A5 Amasya: Merzifon, Tavşan mountain, Gelinsini village, valley side, 1500 m., 09.08.2011.
<i>T. parthenium</i>	Ş.Dereli 118	A5 Amasya: Merzifon, Tavşan mountain, 10 km. from Gelinsini village to Haciveli, field sides, 1250 m., 07.08.2011.
<i>T. poteriifolium</i>	Ş.Dereli 126	A5 Samsun: Bafra, Yeraltı village, around Ağcaalan stream, 150 m., 13.07.2011. A5 Amasya: Merzifon, Tavşan mountain, Gelinsini village, road sides, 1500 m., 20.07.2011.
<i>T. vulgare</i>	Ş.Dereli 143	A5 Samsun: Bafra, around Dereler village, forest openings, 100 m., 13.07.2011. A5 Samsun: Bafra, Yer altı village, near Ağcaalan stream, meadows, 150 m., 16.07.2011.

Tab. 2. Anatomical characteristics of four *Tanacetum* L. taxa. SD – standard deviation, * denotes absence of character. Means in the column marked with the same letter are not statistically significant ($p < 0.05$).

	<i>T. macrophyllum</i>		<i>T. parthenium</i>		<i>T. poterifolium</i>		<i>T. vulgare</i>	
	Width (μm)	Length (μm)	Width (μm)	Length (μm)	Width (μm)	Length (μm)	Width (μm)	Length (μm)
	mean \pm SD		mean \pm SD		mean \pm SD		mean \pm SD	
Root								
Periderma cell	18.78 \pm 3.79b	44.89 \pm 9.33 c	12.09 \pm 3.27a	33.84 \pm 7.40b	17.47 \pm 3.23b	24.66 \pm 4.09a	13.78 \pm 2.94a	27.50 \pm 5.43a
Diameter of cortex	168.46 \pm 8.62b	–	127.47 \pm 5.25d	–	149.80 \pm 11.03c	–	212.23 \pm 18.87a	–
Thickness of phloem	58.03 \pm 7.09d	–	180.91 \pm 9.39a	–	148.07 \pm 10.43b	–	79.05 \pm 24.13c	–
Thickness of xylem	373.80 \pm 14.21c	–	577.84 \pm 23.95b	–	874.29 \pm 29.79a	–	128.13 \pm 20.54d	–
Diameter of vessel	19.75 \pm 6.60b	–	42.11 \pm 13.16a	–	12.39 \pm 3.24b	–	14.72 \pm 5.54b	–
Stem								
Epidermis cell	12.81 \pm 2.09c	14.65 \pm 3.57 d	3.72 \pm 0.64a	3.90 \pm 0.75a	10.39 \pm 1.70b	8.15 \pm 1.45b	11.18 \pm 2.57b	12.79 \pm 2.85c
Collenchyma cell	9.52 \pm 1.36b	13.99 \pm 1.66 b	3.27 \pm 0.53a	5.09 \pm 0.46a	8.66 \pm 2.69b	16.80 \pm 3.58c	15.36 \pm 2.34c	20.96 \pm 2.33d
Thickness of cortex	88.84 \pm 10.33b	–	56.68 \pm 17.95c	–	50.68 \pm 3.92c	–	184.83 \pm 72.12a	–
Thickness of sclerenchyma	60.13 \pm 7.65b	–	67.18 \pm 16.81b	–	71.13 \pm 5.64b	–	95.37 \pm 21.41a	–
Thickness of phloem	61.54 \pm 9.63b	–	65.44 \pm 15.89b	–	67.64 \pm 8.08b	–	91.18 \pm 25.75a	–
Cambium	16.33 \pm 2.68c	–	25.91 \pm 8.85b	–	39.74 \pm 7.68a	–	46.72 \pm 7.14a	–
Thickness of xylem	135.50 \pm 17.15c	–	86.30 \pm 18.17d	–	173.68 \pm 22.91b	–	209.66 \pm 21.81a	–
Diameter of vessel	18.60 \pm 7.67a	–	17.00 \pm 3.69a	–	18.61 \pm 7.58a	–	15.32 \pm 3.07a	–
Pith cell	32.99 \pm 7.42c	–	48.45 \pm 14.30c	–	75.60 \pm 17.42a	–	51.63 \pm 11.53b	–
Leaf								
Epidermis cell	19.33 \pm 5.06c	22.05 \pm 4.51ab	16.77 \pm 2.98ab	25.88 \pm 8.39b	18.19 \pm 3.94bc	23.11 \pm 6.98b	15.45 \pm 2.47a	18.26 \pm 5.04a
Palisade cell	11.59 \pm 1.39a	33.55 \pm 2.63 a	16.18 \pm 2.03b	47.45 \pm 8.09b	14.96 \pm 2.49b	47.61 \pm 6.82b	11.76 \pm 2.23a	43.97 \pm 10.52b
Spongy cell	11.34 \pm 1.56a	19.10 \pm 4.35 a	13.57 \pm 2.12b	19.95 \pm 2.86a	16.39 \pm 3.25c	22.78 \pm 5.16b	15.54 \pm 2.62c	30.94 \pm 6.58c
Thickness of phloem	38.39 \pm 6.36b	–	35.84 \pm 8.84b	–	43.80 \pm 5.73b	–	61.52 \pm 12.65a	–
Thickness of xylem	81.51 \pm 9.00a	–	89.42 \pm 9.91a	–	61.49 \pm 7.61b	–	63.20 \pm 9.01b	–
Adaxial stomata	*	*	22.17 \pm 1.48a	31.08 \pm 3.92a	*	*	25.33 \pm 2.86a	34.89 \pm 2.44a
Abaxial stomata	23.13 \pm 1.92b	29.76 \pm 2.65 b	24.35 \pm 1.44ab	30.30 \pm 3.17b	25.57 \pm 2.18a	37.17 \pm 2.01a	25.52 \pm 1.98a	33.87 \pm 3.40a

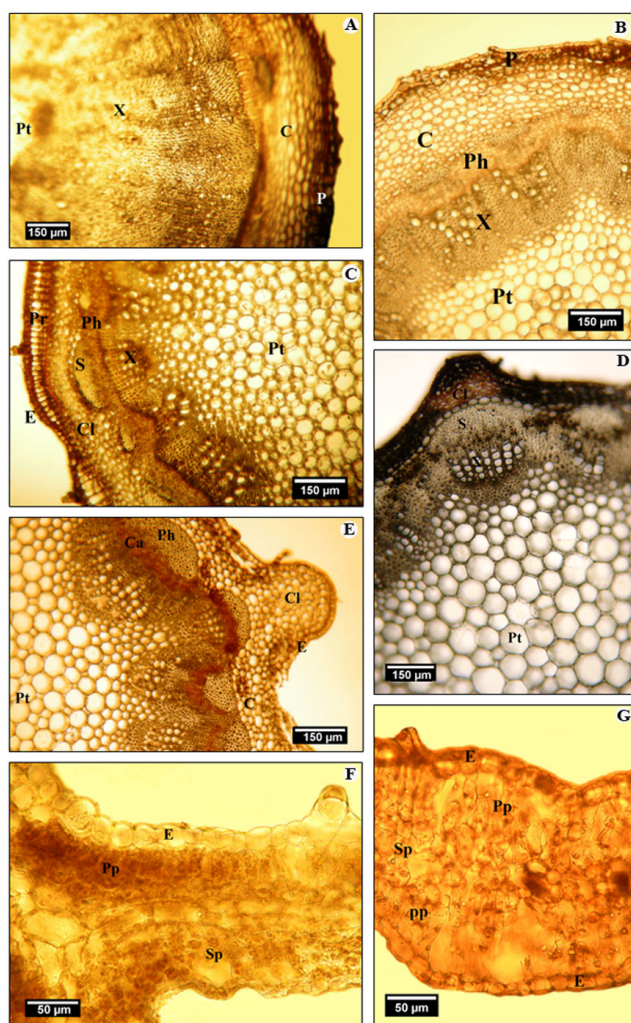


Fig. 1. Cross-sections of roots, stems and leaves of investigated *Tanacetum* taxa: (A) *T. poteriifolium* root, (B) *T. vulgare* root, (C) *T. macrophyllum* stem, (D) *T. poteriifolium* stem, (E) *T. vulgare* stem, (F) *T. macrophyllum* leaf, (G) *T. vulgare* leaf. C – cortex, Cl – collenchyma, E – epidermis, P – periderm, Ph – phloem, Pr – parenchyma, Pt – pith, Pp – palisade parenchyma, S – sclerenchyma, Sp – spongy parenchyma, X – xylem.

Tanacetum parthenium

Root anatomy – Periderm is 3–4 layered. There is a parenchymatic cortex under the periderm and the diameter of this layer is $127.47 \pm 5.25 \mu\text{m}$ (Tab. 2). Cambium cells are undistinguishable. The xylem consists of vessel members and tracheids. The trachea diameter is $42.11 \pm 13.16 \mu\text{m}$ (Tab. 2). The pith consists of primary xylem elements with pith rays in 1–4 rows.

Stem anatomy – The stem is clearly quadrangular in transverse section. The epidermis consists of oval or rectangular cells and is covered by a thick cuticula. Under the epidermis, a collenchyma layer is located at the corners of the stem. The cortex is composed of irregular oval or circular parenchymatic cells. 2–4 layered sclerenchymatic layer is present above the phloem. There is a distinguishable cambium between the phloem and the xylem. The mean trachea diameter is $17.00 \pm 3.69 \mu\text{m}$ (Tab. 2). In the whole center of the stem, there is a pith which is composed of circular, oval and hexagonal large parenchymatic cells.

Leaf anatomy – There is a single layered epidermis on the upper and lower surface of the leaf. Mesophyll consists of single layer of palisade parenchyma cells and 4–6 layers of spongy parenchyma cells with large intercellular spaces. Spongy parenchyma cells are $19.95 \pm 2.86 \mu\text{m}$ in length (Tab. 2). Vascular bundles are surrounded by a parenchymatic bundle sheath. There is one large vascular bundle in the center. The leaf surface shows stomata of the anomocytic type. Stomata occur on both epidermal surfaces and are more abundant on the lower epidermis.

Tanacetum poteriifolium

Root anatomy – Periderm is multilayered. The cortex layer is parenchymatic and the mean diameter of cortex $149.80 \pm 11.03 \mu\text{m}$ (Tab. 2). The cambium is inconspicuous. Most of the root volume is occupied by secondary xylem. Diameters of vessel members are $12.39 \pm 3.24 \mu\text{m}$ (Tab. 2). The pith consists of large parenchymatic cells that are oval or rounded in shape (Fig. 1A).

Stem anatomy – A transverse section taken from the stem showed that it was covered by a uniseriate epidermis with thick cuticle (Fig. 1D). The epidermal cells of both surfaces are more or less rectangular to oval. Seven-eight layers of lacunar collenchyma are located under the epidermis. Cortex parenchyma cells are multilayered. The endodermis is located between the cortex and the vascular tissue. The cells of the endodermis layer are one-layered. The cambium is hardly visible and undistinguishable. The size of vascular bundles at the corners is larger than the bundles between corners. The mean diameter of vessel members is $18.61 \pm 7.58 \mu\text{m}$. Pith cells are large and parenchymatic (Fig. 1D).

Leaf anatomy – The epidermis at both surfaces of the leaves is single-layered. A well developed cuticle on the surface of leaves was observed. Upper and lower epidermises consist of a single layer of rectangular cells. The mesophyll is differentiated into a 1–2 layered palisade and a 3–4 layered spongy parenchyma. Palisade parenchyma cells are elongated. Spongy parenchyma cells are $22.78 \pm 5.16 \mu\text{m}$ in length (Tab. 2). Vascular bundles are collateral and well developed. The leaves are hypostomatic. The dimensions of the anomocytic type stomata on the lower surface are $37.17 \pm 2.01 \times 25.57 \pm 2.18 \mu\text{m}$ (Tab. 2).

Tanacetum vulgare

Root anatomy – In transverse section, the root is well advanced in a secondary structure formation (Fig. 1B). Under the multilayered periderm, there is a cortex layer consisting of oval cells. The cells of this layer are $13.78 \pm 2.94 \mu\text{m}$ in width and $27.50 \pm 5.43 \mu\text{m}$ in length (Tab. 2). There are sclerenchymatic cells that form groups between the cortex and the phloem. The cambium is not distinguishable. The xylem consists of vessel members and tracheids. Vessel members are circular or hexagonal. The xylem rays are composed of 2–4 rows of rectangular cells. The pith containing parenchymatic cells occupies the center of the root (Fig. 1B).

Stem anatomy – A transverse section taken from the middle part of plants clearly showed a quadrangular shape

(Fig. 1E). Epidermal cells consist of a single layer and are orbicular or rectangular. Underneath the epidermis, there is a lacunar collenchyma layer which is more developed at the corner of the stem. The length of collenchyma cells is $20.96 \pm 2.33 \mu\text{m}$ (Tab. 2). The cortex is composed of multi-layered irregular oval or rectangular parenchymatic cells with intercellular spaces. The cambium between xylem and phloem is hardly visible. There are multilayered sclerenchymatic cells surrounding the phloem. These bundles are different sizes. The xylem and phloem elements are visible. The pith consists of large parenchymatic cells with intercellular spaces (Fig. 1E).

Leaf anatomy – In transverse section, the upper and lower epidermises are composed of uniseriate rectangular cells (Fig. 1G). Epidermis cells are covered with a cuticula layer. The leaf is of the isobilateral type. The mesophyll consists of 2 or 3 layers of upper and lower palisade cells and isodiametric spongy parenchymatic cells with large intercellular spaces. Spongy parenchyma cells are 3–4 layered and orbicular or oval in shape. The vascular bundles are collateral and surrounded by a parenchymatic bundle sheath. The stomata type is anomocytic and the stomata occur on the surfaces of both sides but are more abundant on the lower surface. The dimensions of stomata on the lower surface of leaf are $33.87 \pm 3.40 \times 25.52 \pm 1.98 \mu\text{m}$ (Tab. 2).

Micromorphology of the stem, leaf and phyllary

The micromorphological characters and distribution of the trichomes on stem, leaf and phyllaries of the *Tanacetum* taxa examined showed a considerable variation (Tab. 3). In our study, two basic types of trichomes were observed on the vegetative organs: Non-glandular and glandular trichomes. Non-glandular trichomes are acicular or curved, simple and made up from one or more cells (Figs. 2A–B), whereas glandular trichomes were peltate or capitate (Figs. 2C–F). Both these types of trichomes were common in the four species of the studied *Tanacetum* genus (Figs. 2A–F). However, glandular trichomes are absent from the stem of all *Tanacetum* taxa. In addition, these kinds of trichome are very rare on the phyllaries of *T. vulgare* (Tab. 3), although they are present on the leaves and phyllaries of the other taxa. Non-glandular trichomes especially are present in large numbers on both surfaces of leaves, phyllaries and

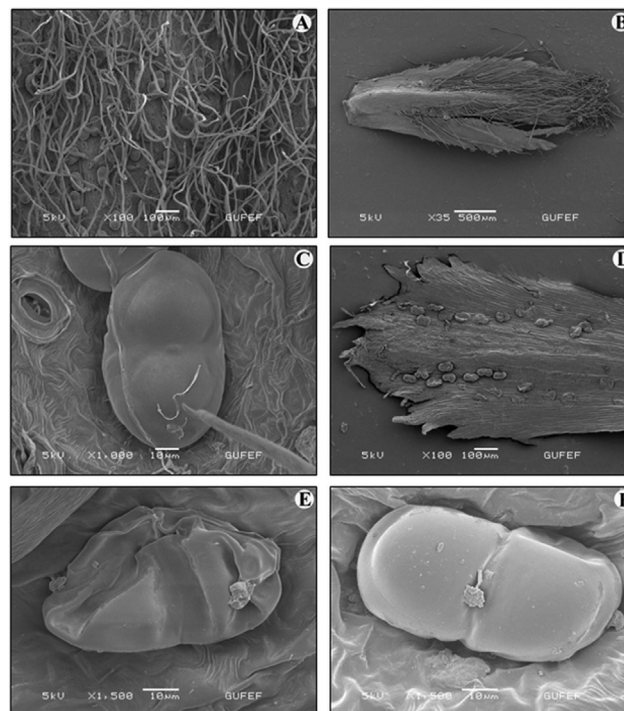


Fig. 2. Scanning electron micrographs (SEM) of trichomes of studied *Tanacetum* L. taxa: (A) eglanular trichomes on leaf of *T. macrophyllum*, (B) eglanular trichomes on phyllaries of *T. macrophyllum*, (C) glandular trichomes on the lower surface of leaf of *T. macrophyllum*, (D) glandular trichomes on phyllaries of *T. parthenium*, (E) glandular trichomes on the lower surface of leaf of *T. poteriifolium*, (F) glandular trichomes on the lower surface of leaf of *T. vulgare*.

stem in *T. macrophyllum*, *T. parthenium* and *T. poteriifolium* (Figs. 2A–B, Tab. 3).

Micromorphology of flower and cypsela

Glandular trichomes are generally abundant on the disc florets and ray florets of *Tanacetum* taxa (Figs. 3A–D). These kinds of trichome are the most common type and are observed in all investigated *Tanacetum* species. However, they are usually sparsely found on disc and ray florets of *T. macrophyllum* (Tab. 3). SEM showed that the disc florets have greater trichome density than the ray florets in *Tanace-*

Tab. 3. Distribution of glandular (Gland) and non-glandular (Egland) trichomes on different parts of studied *Tanacetum* L. species. (–) denotes absence of trichomes, (+) denotes a few trichomes, (++,+++), denote increasing presence of trichomes, (*) denotes absence of character.

Plant material	<i>T. macrophyllum</i>		<i>T. parthenium</i>		<i>T. poteriifolium</i>		<i>T. vulgare</i>	
	Gland	Egland	Gland	Egland	Gland	Egland	Gland	Egland
Stem	–	++	–	+	–	+	–	+
Adaxial leaf surface	++	+++	+++	++	++	+++	+++	+
Abaxial leaf surface	+++	+++	+++	++	+	+++	+++	+
Phyllaries	++	+++	++	+	++	+++	–	++
Disc floret	+	–	+	–	++	–	++	–
Ray floret	+	–	++	–	++	–	*	*
Cypsela	+	–	+++	–	+++	–	+++	–

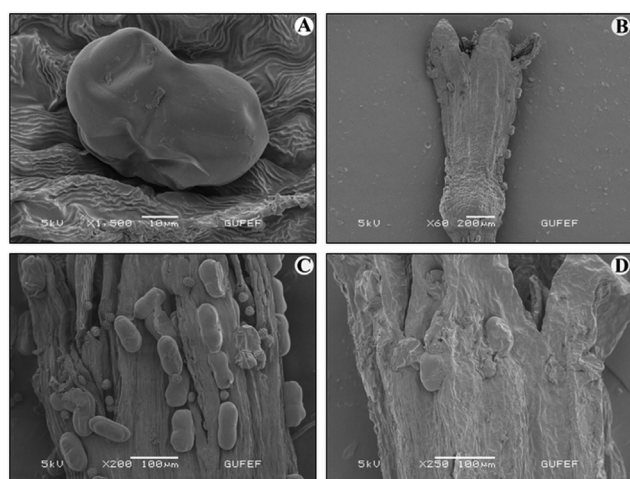


Fig. 3. Scanning electron micrographs (SEM) of glandular trichomes of studied *Tanacetum* L. taxa: (A) on ray floret of *T. parthenium*, (B) on the disc floret of *T. parthenium*, (C) on the disc floret of *T. poteriifolium*, (D) on the disc floret of *T. vulgare*.

tum (Figs. 3B–D). Trichome density on disc florets is higher on the ovary than the corolla. The non-glandular trichomes are absent from the florets of the *Tanacetum* taxa studied (Tab. 3).

Surface characteristics and ornamentation features of the cypselas of 4 Turkish *Tanacetum* species are given in Tab. 4. In cypselas of examined *Tanacetum* species there are 4–8 ribs on all sides (Figs. 4A–D). As evidenced in Tab. 4, in all species studied here, the cypselas have glandular surfaces. However, in *T. poteriifolium* and *T. parthenium* glandular trichomes on cypselas are mainly situated between the ribs especially forming in order over the cypselas surface (Figs. 4A–D, Tab. 4).

Tab. 4. Surface characteristics and ornamentation features of the cypselas of studied *Tanacetum* L. taxa.

Taxa	Surface and ornamentation characteristics
<i>T. macrophyllum</i>	4–5 ribbed, sparsely glandular on the ribs
<i>T. parthenium</i>	6–8 ribbed, densely glandular in between the ribs
<i>T. poteriifolium</i>	6–8 ribbed, densely glandular in between the ribs
<i>T. vulgare</i>	6–8 indistinct longitudinal ribbed, glandular on the ribs

Discussion

The present study provides useful information on the anatomy and micromorphology of *T. macrophyllum*, *T. parthenium*, *T. poteriifolium* and *T. vulgare*. This is the first detailed anatomical and micromorphological report on the four examined taxa of *Tanacetum*. All taxa investigated in the present study were found to have the same general characteristics as other members of the Asteraceae. Metcalfe and Chalk (1979) gave some information about general anatomical characters of the family Asteraceae. Some studies on Asteraceae reported the existence of secretory structures especially in the root (Bremer 1994, Heywood 1976). In the

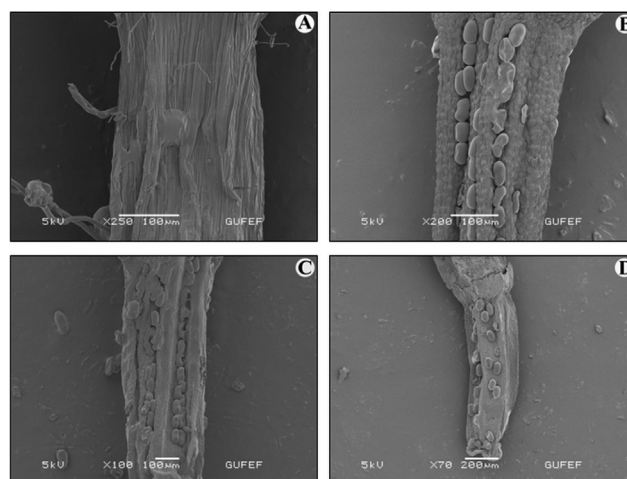


Fig. 4. Scanning electron micrographs (SEM) of cypselas of studied *Tanacetum* L. taxa: (A) *T. macrophyllum*, (B) *T. parthenium*, (C) *T. poteriifolium*, (D) *T. vulgare*.

present study, we determined that *T. macrophyllum* has secretory canals within the root cortex. Our findings are consistent with those of Metcalfe and Chalk (1979) and those of some other studies of Asteraceae (Castro et al. 1997, Milan et al. 2006). The results in the present study indicated that there was a similarity in root anatomical structure among the taxa. However, in *T. poteriifolium* and *T. vulgare*, the pith was occupied with large parenchymatic cells, whereas it was filled with primary xylem elements in the other examined taxa.

The transverse section of stem showed that the stem of investigated *Tanacetum* taxa has a well-defined collenchyma layer in the corners. Metcalfe and Chalk (1979) reported that stems of many genera and species of the family Asteraceae have distinct collenchyma. It was also determined that this layer is more distinguishable and thicker in *T. vulgare*. As well as cortex, sclerenchyma and vascular bundles are the thickest in *T. vulgare*. A distinguishable endodermis was seen between the cortex and vascular bundle. Özgüçü et al. (1991) pointed out that endodermis is usually distinguishable in the stem of Asteraceae. The parenchymatic layer, which consists of elongated cells, in the stem of *T. macrophyllum* is a useful character for distinguishing the examined species. Sclerenchyma was observed in the transverse sections of the four *Tanacetum* taxa as previously described by Melo de Pinna and Menezes (2002) and Stevović et al. (2010). In *T. poteriifolium*, pith in stem occupied a large place compared to the other species.

The studied species showed some anatomical differences in the leaf. In this study, it was seen that all the examined species have bifacial leaf types except for *T. vulgare*. Metcalfe and Chalk (1950) pointed out that there was generally bifacial mesophyll in the family Asteraceae. Whereas in *T. vulgare*, the leaf is of the isobilateral type. Stevović et al. (2010) also reported that the mesophyll in *T. vulgare* is of the isobilateral type. Our findings are consistent with those of Stevović et al. (2010). The mesophyll of the studied species varied according to the number of cell layers compos-

ing the palisade and spongy parenchyma. In this research, it was found that all examined taxa had anomocytic stomata. In addition, unlike in *T. macrophyllum* and *T. poteriifolium*, which have hypostomatic leaves, the leaves are amphistomatic in *T. parthenium* and *T. vulgare*. Metcalfe and Chalk (1950) reported that there were both anomocytic and anisocytic stomata in the family Asteraceae. These findings support those of Metcalfe and Chalk (1950).

The value of trichomes in identification of some members of Asteraceae has been previously reported by some workers (Adedeji 2004, Ciccarelli et al. 2007, Majdi et al. 2011). According to observations in the current study all the species examined here have eglandular trichomes on the stem and leaves, and glandular trichomes are not present on the stem. The glandular trichomes are absent on the phyllaries of *T. vulgare* only. Ciccarelli et al. (2007) reported that glandular hairs can be used as taxonomical characters in the Asteraceae. The secretion of terpenoids by glandular hairs was shown by several studies of this family (Pagni et al. 2003). It was suggested that some volatile terpenoids may attract pollinating insects to flowers, while other may protect the plant (Kelsey et al. 1984). In the present study, the majority of the glandular hairs were found in disc and ray florets of the all taxa. However, this type of trichome is sparser in *T. vulgare* than in the other taxa. Our results show that within the flowers of the examined *Tanacetum* taxa, the disc florets contain higher glandular trichome density. Majdi et al. (2011) reported that trichome density is highest on the disc florets, followed by leaves, while stems contain a lower density of trichomes in *T. parthenium*. These variations in the distribution of trichome forms in *Tanacetum* species can help to differentiate species.

References

- Abid, R., Qaiser, M., 2008a: Cypsel morphology of *Gnaphalium* L. and its allied genera (Gnaphalieae-Asteraceae) from Pakistan. *Pakistan Journal of Botany* 40, 25–32.
- Abid, R., Qaiser, M., 2008b: Cypsel morphology of some genera in the tribe Gnaphalieae (Asteraceae) from Pakistan. *Pakistan Journal of Botany* 40, 73–485.
- Abid, R., Qaiser, M., 2009: Taxonomic significance of the cypsel morphology in the tribe Anthemideae (Asteraceae) from Pakistan and Kashmir. *Pakistan Journal of Botany* 41, 555–579.
- Abu-Darwish, M. S., Abu-Dieyeh, Z. H. M., 2009: Essential oil content and heavy metals composition of *Thymus vulgaris* cultivated in various climatic regions of Jordan. *International Journal of Agricultural Biology* 11, 59–63.
- Adedeji, O., 2004: Leaf epidermal studies of the species of *Emilia* Cass. (*Senecioneae*, *Asteraceae*) in Nigeria. *Botanica Lithuanica* 10, 121–133.
- Akcın, T. A., Akcın, A., 2010: Morphological and anatomical characteristics and taxonomical significance of achene micromorphology of endemic *Achillea phrygia* Boiss & Bal. and *A. gypsicola* Hub-Mor. (Asteraceae) in Turkey. *Nordic Journal of Botany* 28, 65–73.
- Akcın, T. A., Akcın, A., 2014: Achene micromorphology of seven taxa of *Achillea* L. (Asteraceae) from Turkey. *Bangladesh Journal of Plant Taxonomy* 21, 19–25.
- Bandoniene, D., Pukalskas, A., Venskutonis, P. R., Gruzdiene, D., 2000: Preliminary screening of antioxidant activity of some plant extracts in rapeseed oil. *Food Research International* 33, 785–791.
- Bremer, K., 1994: *Asteraceae: cladistics and classification*, Timber Press, Portland, Oregon.
- Brown, A. M. G., Edwards, C. M., Davey, M. R., Power, J. B., Lowe, K. C., 1997: Effects of extracts of *Tanacetum* species on human polymorphonuclear leucocyte activity in vitro. *Phytotherapy Research* 11, 479–484.
- Brown, A. M. G., Edwards, C. M., Hartman, T. V. P., Marshal, J. A., Smith, R. M., Davey, M. R., Power, J. B., Lowe, K. C., 1999: Sexual hybrids of *Tanacetum*: biochemical, cytological and pharmacological characterization. *Journal of Experimental Botany* 50, 435–444.
- Castro, M. M., Leitão-Filho, H. F., Monteiro, W. R., 1997: Utilização de estruturas secretoras na identificação dos gêneros de Asteraceae de uma vegetação de cerrado. *Revista Brasileira De Biologia* 20, 163–174.
- Ciccarelli, D., Garbari F., Pagni, A. M., 2007: Glandular hairs of the ovary: a helpful character for Asteroideae (Asteraceae) taxonomy? *Annales Botanici Fennici* 44, 1–7.
- Fahn, A., 1979: *Secretory Tissues in Plants*. Academic Press, London.
- Garg, S. K., Sharma, K. C., 2007: Taxonomical significance of the morphological and scanning electron microscopic surface patterns of cypselas in some members of the tribe Heliantheae (Asteraceae). *Feddes Repertorium* 118, 165–191.
- Grierson A. J. C., 1975: *Tanacetum* L. In: Davis, P. H. (ed.), *Flora of Turkey and the East Aegean Islands*, vol. 5. Edinburgh University Press, Edinburgh.

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- Güner, A., Ozhatay, N., Ekim, T., Başer, K. H. C., 2000: Flora of Turkey and the East Aegean Islands (supplement), vol. 11. Edinburgh University Press, Edinburgh.
- Hethelyi, E., Tetenyi, P., Danos, B., Koczka, I., 1991: Phytochemical and antimicrobial studies on the essential oils of the *Tanacetum vulgare* clones by gaschromatography /mass spectrometry. *Herba Hungarica* 30, 82–90.
- Heywood, V. H., 1976: *Tanacetum*. In: Tutin, T. G., Heywood, V. H., Burges, N. A., Moore D. M., Valentine D. H., Walters S. M., Webb D. A. (eds.), *Flora Europe*, vol. IV. Cambridge University Press.
- Holetz, F. B., Pessini, G. L., Sanches, N. R., Cortez Diogenes, A. G., Nakamura, C. V., 2002: Screening of some plants used in the Brazilian folkmedicine for the treatment of infectious diseases. *Memorias do Instituto Oswaldo Cruz* 97, 1027–1031.
- Hough-Golstein, J., Hahn, S. P., 1992: Antifeedant and oviposition deterrent activity of an aqueous extract of *Tanacetum vulgare* L. on two cabbage pests. *Environmental Entomology* 21, 837–844.
- Jansen, F. H., 2006: The herbal tea approach for artemesinin as a therapy for malaria. *Transactions of the Royal Society of Tropical Medicine and Hygiene* 100, 285–286.
- Johansen, D.A., 1944: *Plant Microtechnique*. McGraw-Hill, New York.
- Kelsey, R. G., Reynolds, G. W., Rodriguez, E., 1984: The chemistry of biologically active constituents secreted and stored in plant glandular trichomes. In: Rodriguez E., Healey P. L., Metha, J. (eds.), *Biology and Chemistry of Plant Trichomes*. Plenum Press, New York.
- Lahlou, S., Tangi, K. C., Lyoussi, B., Morel, N., 2008: Vascular effect of *Tanacetum vulgare* L. leaf extract: *In vitro* pharmacological study. *Journal of Ethnopharmacology* 120, 98–102.
- Maes, L., Van Nieuwerburgh, F. C. W., Zhang, Y., Reed, D. W., Pollier, J., Vande Casteels, R. F., Goossens, A., 2011: Dissection of the phytohormonal regulation of trichome formation and biosynthesis of the antimalarial compound artemisinin in *Artemisia annua* plants. *New Phytologist* 189, 176–189.
- Majdi, M., Liu, Q., Karimzadeh, G., Malboobi, M. A., Beekwilder, J., Cankar, K., Bouwmeester, H., 2011: Biosynthesis and localization of parthenolide in glandular trichomes of feverfew (*Tanacetum parthenium* L. Schulz Bip.). *Phytochemistry* 72, 1739–1750.
- Majdi, M., Charnikhova, T., Bouwmeester, H., 2013: Genetical, developmental and spatial factors influencing parthenolide and its precursor costunolide in feverfew (*Tanacetum parthenium* L. Schultz Bip.). *Industrial Crops and Products* 47, 270–276.
- Mantle, D., Eddeb, F., Pickering, A. T., 2000: Comparison of relative antioxidant activities of British medicinal plant species *in vitro*. *Journal of Ethnopharmacology* 72, 47–51.
- Marles, R. J., Pazos-Sanou, L., Compadre, C. M., Pezzuto, J. M., Blosyk, E., Arnason, T., 1995: Sesquiterpene lactones revisited. Recent advances in the assignment of biological activities and structure relationships. In: Arnason, J. T., Mata, R., Romeo, J. T. (eds.), *Phytochemistry of medicinal plants*. Recent Advances in Phytochemistry, vol. 29. Plenum Press, New York and London.
- Melo De Pinna, G. F. A., Menezes, N. L., 2002: Vegetative organ anatomy of *Ianthopappus corymbosus* Roque & Hind (Asteraceae-Mutisieae). *Revista Brasileira de Botanica* 25, 505–514.
- Metcalf, C. R., Chalk, L., 1950: *Anatomy of the dicotyledons: leaves, stem, and wood in relation to taxonomy with notes on economic uses*. vol. 2, Clarendon Press, Oxford.
- Metcalf, C. R., Chalk, L., 1979: *Anatomy of the dicotyledons: systematic anatomy of leaf and stem, with a brief History of the subject*. vol. 1, 2nd ed, Clarendon Press, Oxford.
- Milan, P., Hayashi, A. H., Beatriz, A. D. G., 2006: Comparative leaf morphology and anatomy of three Asteraceae species. *Brazilian Archives of Biology and Technology* 49, 135–144.
- Napp-Zinn, K., Eble, M., 1978: Beiträge zur systematischen Anatomie der *Anthemideae*: die Spaltöffnungsapparate. *Plant Systematics and Evolution* 130, 167–190.
- Neszmelyi, A., Milne, G. W. A., Hethelyi, E., 1992: Composition of the essential oil of clone 409 of *Tanacetum vulgare* and 2D NMR investigation of *trans*-chrysanthenyl acetate. *Journal of Essential Oil Research* 4, 243–250.
- Özöğücü, B., Gemici, Y., Türkan, I., 1991: Karşılaştırmalı Bitki Anatomisi. İzmir: Ege Üniversitesi Fen Fakültesi Yayını (in Turkish).
- Pagni, A. M., Orlando, R., Massini, A., Ciccarelli, D., 2003: Secretory structures of *Santolina ligustica* Arrigoni (Asteraceae), an Italian endemic species. *Israel Journal of Plant Science* 51, 185–192.
- Pandey, A. K., Kumari, A., 2007: Anatomical patterns of pericarp in Asteraceae. In: Chauhan, S. V. S., Rana, A., Chauhan, S., (eds.), *Plant Reproductive Biology and Biotechnology*, Aavishkar Publishers, Distributors, Jaipur.
- Stefanović, M., Ristić, N., Vukmirović, M., 1988: Biological activities of sesquiterpene lactones. Investigations of microbial activities of lactones isolated from the Yugoslav plant species of the genus *Tanacetum* L. [*Chrysanthemum* (fam. Compositae)]. *Bulletin Academie Serbe des Sciences et des Arts Tome XCV, Classe des Sciences Mathematiques et Naturelles, Sciences Naturelles* 28, 23–43.
- Stevović S., Surčinski- Mikovilović, V., Čalić-Dragosavac, D., 2009: Environmental adaptability of tansy (*Tanacetum vulgare* L.). *African Journal of Biotechnology* 8, 6290–6294.
- Stevović, S., Mikovilović, V., Čalić-Dragosavac, D., 2010: Environmental impact on morphological and anatomical structure of Tansy. *African Journal of Biotechnology* 9, 2413–2421.
- Williams, A. C., Harborne, J. B., Geiger, H., Holut, J. R. S., 1999: The flavonoids of *Tanacetum parthenium* and *Tanacetum vulgare* and their antiinflammatory properties. *Phytochemistry* 51, 417–423.
- Zhu, S. X., Qin, H. N., Shih, C., 2006: Achene wall anatomy and surface sculpturing of *Lactuca* L. and related genera (Compositae: Lactuceae) with notes on their systematic significance. *Journal of Integrative Plant Biology* 48, 390–399.