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A Comprehensive Micromorphological and Anatomical Study on the Poisonous Plant *Cionura erecta* (L.) Griseb. (Apocynaceae: Asclepiadoideae)

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ABSTRACT

Cionura erecta (L.) Griseb., named as Babrik in Türkiye, is one of the widespread taxa of Apocynaceae, which is known for its toxicity as well as its common therapeutic use. This study aimed to contribute to the systematics of *C. erecta* by examining the stem, petiole, lamina and flower parts of the species in detail, for the first time, using scanning electron microscopy (SEM) and light microscopy. SEM examinations determined that the surface of vegetative parts was crisped hairy, leaf epicuticular wax was striate, and colleters were in petiolar, laminar and calycine positions, and each was defined as standard type consisting of a short, non-secretory stalk, and a conical, undivided main secretory head. The presence of collenchyma cells and scattered sclerenchyma cell groups in the stem, bicollateral vascular bundles, laticifers and druse crystals in all vegetative portions, and paracytic, anomocytic, and anisocytic stomata on the amphistomotic and equifacial leaf were determined as the most characteristic anatomical features. The obtained results revealed that anatomical and micromorphological characters could be used to contribute to the morphological characters in the identification of *C. erecta*.

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Zehirli Bitki Olan *Cionura erecta* (L.) Griseb. (Apocynaceae: Asclepiadoideae) Üzerinde Kapsamlı Bir Mikromorfolojik ve Anatomik Çalışma

ÖZET

Türkiye'de Babrik adıyla bilinen *Cionura erecta* (L.) Griseb., Apocynaceae familyasının geniş yayılışlı taksonlarından biri olup zehirli olmasının yanı sıra yaygın tedavi edici kullanımı ile de bilinmektedir. Bu çalışma ile *C. erecta*'nın gövde, petiyol, yaprak ayası ve çiçek kısımları taramalı elektron mikroskobu (SEM) ve ışık mikroskobu ile ilk kez detaylı bir şekilde incelenerek türün sistematiğine katkı sağlanması amaçlanmıştır. SEM incelemelerinde vejetatif kısımların yüzeyinin kıvrık tüylü, yaprak epikutikular mum tabakasının çizgili olduğu, colleter salgı yapılarının petiyol, yaprak ayası ve kaliksin üzerinde konumlandığı ve herbirinin kısa, salgı yapmayan bir sap ve konik, bölünmemiş salgı yapan bir baş kısmından oluşan standart tip olarak tanımlandığı belirlendi. Gövdede kollenkima hücreleri ve dağınık sklerenkima hücre gruplarının bulunması, tüm vejetatif kısımlarda bikollateral iletim demetleri, latisiferler ve druz kristallerinin varlığı, ekvifasiyal ve amfistomotik yaprakta ise parasitik, anomositik ve anizositik stomaların varlığı en karakteristik anatomik özellikler olarak belirlendi. Elde edilen sonuçlar, anatomik ve mikromorfolojik karakterlerin *C. erecta*'nın tanımlanmasında morfolojik karakterlere katkı sağlamak amacıyla kullanılabileceğini ortaya koymuştur.

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INTRODUCTION

Apocynaceae s.l., currently treated as a combined family of the two former families Apocynaceae s.str. and Asclepiadaceae (Endress & Bruyns, 2000; Endress et al., 2014), has recently been separated into five major groups: two non-monophyletic informally recognized as rauvolfioids and apocynoids grades, and three monophyletic subfamilies, Periplocoideae, Secamonoideae, and Asclepiadoideae (Endress et al., 2019). Members of the unified Apocynaceae are predominantly distributed in tropical and subtropical regions, with a more limited spread in temperate regions (Endress & Bruyns, 2000), including 378 genera and about 5000 species (Fishbein et al., 2018, Endress et al., 2019). Apocynaceae (Stearn, 1978) and Asclepiadaceae (Browicz, 1978) were recognised as two distinct families in the Flora of Türkiye and the Eastern Aegean Islands, however, recently, Güner et al. (2012) listed all the taxa belonging to the two families under the single family, Apocynaceae s.l.

Cionura Griseb. is a monotypic genus belonging to the tribe Marsdenieae Benth. in the subfamily Asclepiadoideae (Apocynaceae) (Endress et al., 2019). The genus is represented by a single species, *Cionura erecta* (L.) Griseb., which has a wide distribution in southern Europe and Asia Minor extending from the Balkan Peninsula, Aegean Islands, Crete, Cyprus, and via Anatolia to south-western Iran, northern Iraq, north-Western Syria, Lebanon, Israel, Sinai Peninsula, and Afghanistan (Browicz, 1967). *C. erecta,* known as unpleasant smelling plant with copious poisonous milky juice, is a perennial woody-based plant characterised with numerous herbaceous twining stems, opposite, ovate, and basally cordate or subcordate leaves, inflorescences in terminal or lateral with erect, branched cymes bearing many white or cream-yellow flowers, and slender ovoid follicles containing numerous seeds with silky coma at apex (Browicz, 1978; Güven & Sarıkaya, 2024; Sarıkaya & Güven, 2024). In Türkiye, members of *C. erecta* have a wide distribution except for the north-eastern Anatolia, and grow in various habitats such as conglomerate slopes, limestone cliffs, fields and river beds, beaches and waste places, from sea level to 1400 m (Browicz, 1978; Güven & Sarıkaya, 2024). The plant which is known with the name "Babrik" throughout Anatolia (Güner, 2012), and also with the local names "Halava" and "Kulf" in Şanlıurfa (Takım et al., 2022), has been considered as a poisonous plant since it was shown to be the main cause of goat and sheep poisoning in many local cases in different regions of Türkiye (Öztürk et al., 2008; Takım et al., 2022).

C. erecta, like most other Apocynaceae taxa, has medicinal value and numerous studies have been conducted to date on the phytochemical composition and biological activities of this plant such as anticancer, apoptotic, antioxidant, and enzyme inhibition (Baumann, 1996; Myrianthopoulos et al., 2007; Öztürk et al., 2008; Mirza & Navaei, 2009; Ünsal et al., 2010; Demir et al., 2011; Mozaffari et al., 2014; Takım et al., 2022; Erdogan et al., 2025). The production of chemical compounds in plants has often been associated with secretory structures. The members of Apocynaceae, known for their rich biochemical content, have been characterized with various secretory structures, such as trichomes, idioblasts, laticifers, colleters, style heads, nectaries, and osmophores in vegetative and reproductive organs (Endress & Bruyns, 2000; Demarco, 2017). Laticifers (internal glands) and colleters (external glands) were considered as the most characteristic secretory structures within the family (Metcalfe & Chalk, 1950; Demarco, 2017). Laticifers in vegetative and floral organs of the Apocynaceae members were defined as articulated or non-articulated. Phytochemical studies on Apocynaceae revealed that the latex fluid in the family members is quite rich in biochemical content playing an active role in both exhibiting various biological activities and protecting plants against microorganisms (Metcalfe & Chalk, 1950; Demarco, 2015, 2017). The other glandular structures, the family specific colleters, may be located on the basis of lamina and petiole of the leaf, and on floral parts like cotyledons, bracts, calyces, and corollas. The basic function of these structures is known to protect vegetative and generative parts against desiccation at initial stages of development, and support their natural defenses against pests (Fahn, 1990; Demarco, 2017; Ribeiro, 2017, 2021). Colleters within the Apocynaceae have generally been described as the standard type, which display cylindrical or dorso-ventrally flattened appearance, composed of a short stalk and a secretory head producing a sticky secretion of mucilage, resin, or a mixture of mucilage and lipophilic substances (Fahn, 1990; Thomas & Dave, 1991; Simões et al., 2006; Ribeiro, 2021).

As seen in the above literature, it has been determined that the studies conducted on *C. erecta* have focused more on its chemical properties. There is very limited data on anatomical and micromorphological features of *C. erecta*. Previously, the wood anatomy of *C. erecta* (=*Marsdenia erecta* R. Br.) from Türkiye was examined by Yaman & Tumen (2012), who pointed out the two basic criteria, a decreasing age-on-length graphic for vessel elements, and upright and square ray cells, for occurrence of secondarily woody. However, the researchers emphasized the need of additional taxonomic data for a clear decision about presence or absence of secondary woodiness in this species (Yaman & Tumen, 2012). More recently, fruit and seed morphology and micromorphology of *C. erecta*, together with some of the other Turkish Apocynaceae taxa, was evaluated using light and scanning electron microscopy (SEM) (Sarıkaya & Güven, 2024). Anatomical and micromorphological features of the vegetative and reproductive organs not only have a great systematic importance in distinguishing plant taxa at different taxonomic levels, but also give a good understanding of the source of phytochemical contents, physiological processes and major adaptive shifts in plants (Abid et al., 2023; Manzoor et al., 2023). Revealing the anatomical and micromorphological features

of *C. erecta* may provide a better insight into the internal and external glandular structures that may be effective in toxicity of these plants. Although several anatomical and micromorphological studies including glandular structures (laticifers and colleters) have been carried out on several members of Apocynaceae (Simões et al., 2006; Demarco, 2017; Capelli et al., 2017; Ribeiro et al., 2017, 2021; Naidoo et al., 2020; Salomé et al., 2023), to our knowladge, no detailed anatomical study including secretory structures has been found regarding *C. erecta*.

Considering the potential of *C. erecta* to be poisonous plant and its wide distribution in humid regions of Türkiye, we aimed to describe the anatomy and micromorphology of generative and vegetative organs of this species to provide additional characteristics for its identification.

MATERIAL and METHOD

Specimens

C. erecta specimens were collected from Türkiye, Adana, Feke-Saimbeyli road, 652 m, 25.05.2019, N37.86 – E36.02, in roadside, rocky places. The photograph of the investigated *C. erecta* in its natural habitat was given in Figure 1. The collected plant samples were processed using the standard herbarium techniques according to Woodland (1997), then identified using the Flora of Türkiye (Browicz, 1978), Flora of Russia (Pobedimova, 1952), Flora Europaea (Markgraf, 1972), and stored at the Herbarium of Recep Tayyip Erdogan University, Department of Biology (RUB) with the voucher number S. Güven 98. For micromorphological and anatomical examinations, at least 10 plant individuals containing stem, leaf, and flowers, were fixed in FAA (5 parts stock formalin, 5 parts glacial acetic acid, 90 parts 70% ethanol) for 24 h and stored in 70% ethanol (Johansen, 1940).

Figure 1. Natural appearance of *Cionura erecta* (S. Güven 98).
Şekil 1. Cionura erecta türünün doğadaki görünümü (S. Güven 98).

Scanning electron microscopy

Generative and vegetative parts, including colleters, were randomly sampled from fully opened flowers, young stems and leaves located between the fifth and sixth nodes above the woody base. Small pieces (5 x 5 mm) were cut superficially from the stem, petiole and lamina, and longitudinal sections were taken from the whole flower. For SEM studies, all the superficial and longitudinal sections were dehydrated in an ethanol series (50-80%), critical-point dried with CO^2 using a Quorum K 850, and then transferred on aluminum stubs, coated with gold for 4 min in a sputter-coater and observed by a JEOL-JSM 6610 microscope at the Central Research Laboratories of Recep Tayyip Erdogan University. Micromorphological examinations were carried out on 10 mature and healthy individuals per each character. In this study, SEM examinations were carried out following Ribeiro et al. (2017), and the description of stomatal type following Dilcher (1974). A detailed micromorphological description of the stem, petiole, lamina and colleters supported by SEM micrographs, were created. The micromorphological terminology followed that of Barthlott et al. (1998), Stearn (1992), Koch et al. (2008), and Prüm et al. (2012).

Anatomical protocols

Anatomical sections were obtained from the young stem, leaf blade, petiole and flower, including colleters, between the and fifth and sixth nodes above the woody base. Cross and longitudinal sections with a thickness of 20–30 µm, were taken from the stem, petiole, lamina and flower using a freezing microtome of Shandon Cryotome SME, and stained with hematoxylin for 30 min and mounted with aqua witrexia in order to obtain permanent slides. Superficial sections were taken from both adaxial and abaxial surfaces of the leaf by free hand, after being kept in hydrochloric acid for decolorization they were directly examined under light microscope, according to Güven et al. (2021). All sections were photographed from permanent slides, and anatomical traits were measured (mm) or

scored, using an Olympus BX51 light microscope (LM) equipped with the Bs200Pro analysis system software. All scores were obtained from sections taken from 10 randomly selected specimens for each anatomical character.

RESULTS

Micromorphological description of the stem, petiole, lamina and colleters

Stem (Figure 2a-b) and petiole (Figure 2c-d) surfaces are covered with crisped indumenta all around, composed of nonglandular, short curly trichomes with verrucose ornamentation, epicuticular wax is smooth. The density of the indumenta on both the stem and petiole increases from the stem base to the terminal bud. Leaf; amphistomatic with paracytic, anomocytic and anisocytic stomata; adaxial and abaxial surfaces with sparsely crisped indumenta all around, consisting of nonglandular, short curly trichomes with verrucose ornamentation; epidermal cells on both sides are polygonal with convex periclinal cell walls and channelled, straight anticlinal cell walls, epicuticular wax is densely striate (Figure 2e-h).

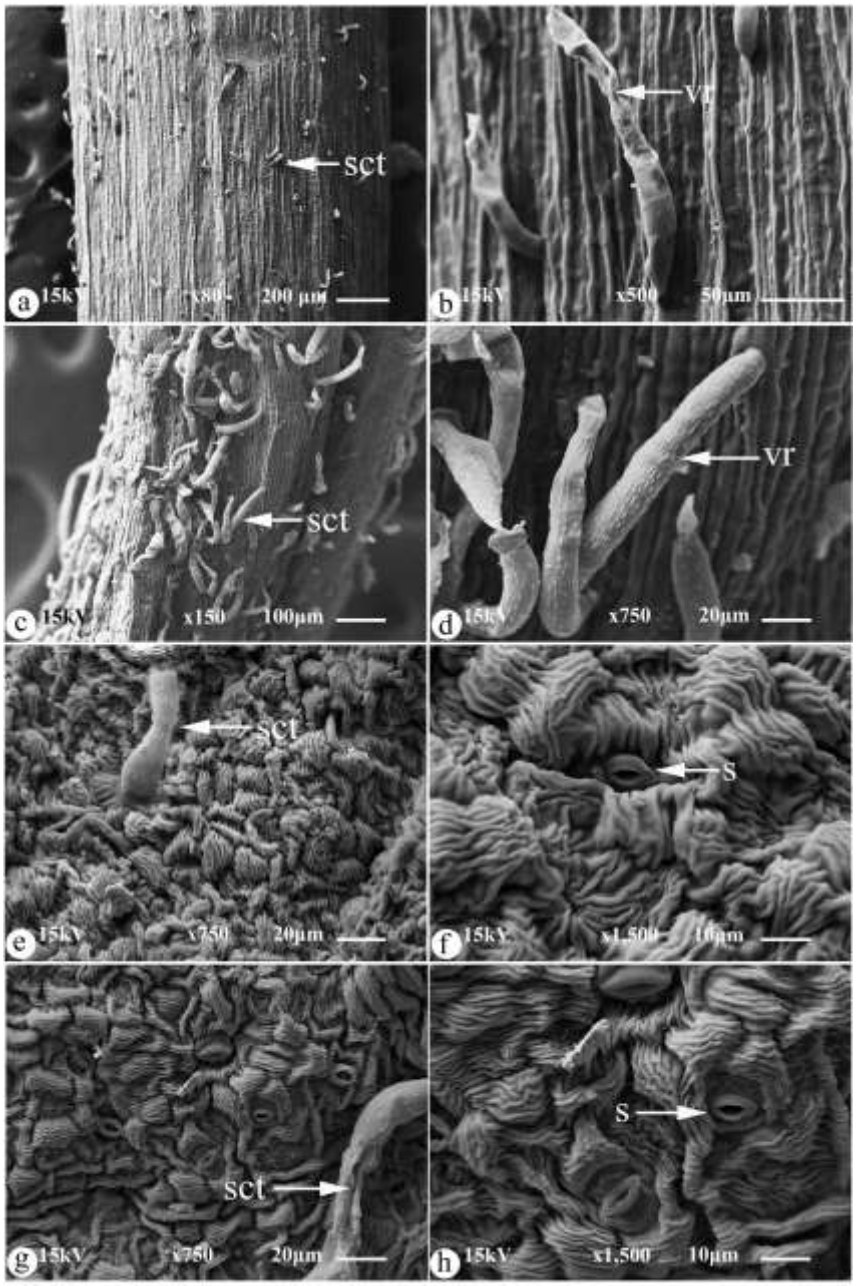

Figure 2. Scanning electron micrographs of stem, petiole and lamina surfaces of *Cionura erecta* (S. Güven 98) a-b. Stem, c-d. Petiole, e-f. Adaxial surface of the lamina, g-h. Abaxial surface of the lamina, s: stoma, sct: short curly trichome, vr: verrucose trichome ornamentation.

Şekil 2. Cionura erecta (S. Güven 98)'nın gövde, petiyol ve lamina yüzeylerine ait taramalı elektron mikroskobu görüntüleri a-b. Gövde, c-d. Petiyol, e-f. Lamina üst yüzey, g-h. Lamina alt yüzey, s: stoma, sct: kısa kıvrık tüy, vr: verrucose tüy ornamentasyonu.

Colleters are located on both floral and foliar parts (Figure 3a). Floral colleters, named as calycine colleters, occur on the adaxial connate base of sepals in alternisepalous distribution (Figure 3b-d). Foliar colleters are found on young leaves near shoot apices, occupying petiolar and laminar positions. The petiolar colleters are found at the base of the petioles, and the laminar ones on the leaf blade close to the distal portion of the petiole (Figure 3e). All the foliar (Figure 4a-d) and floral colleters (Figure 4e-h) are described as the standard type, which have a conical, undivided main secretory head placed on a short stalk at the non-secretory basal portion.

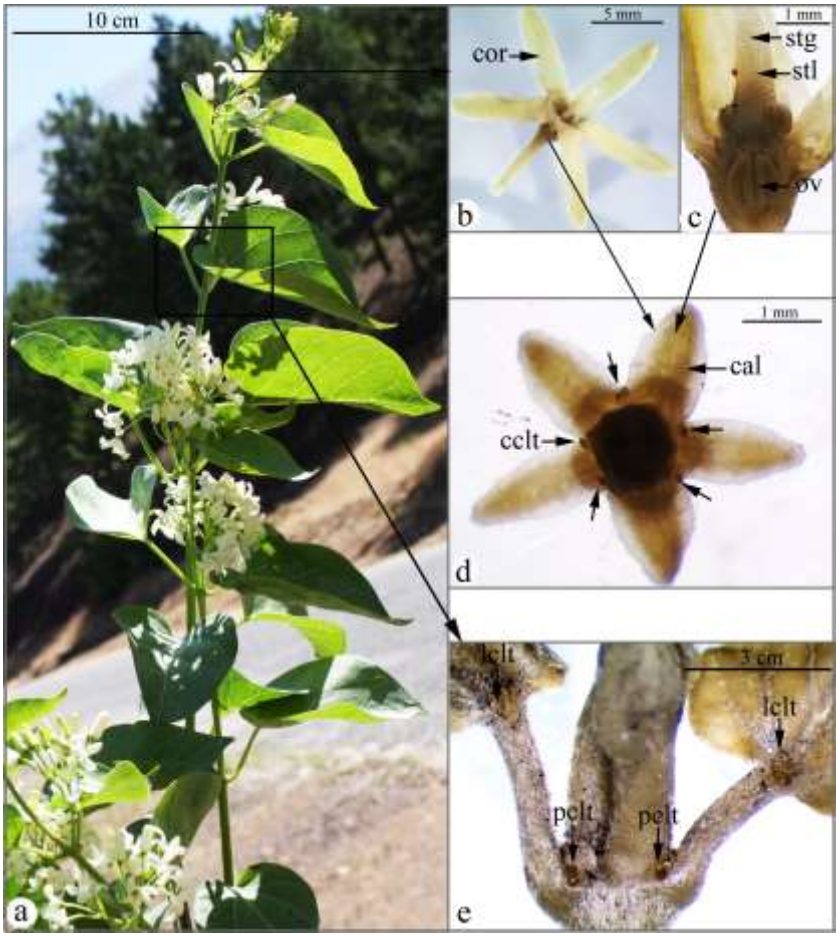

Figure 3. Position of the colleters in *Cionura erecta* (S. Güven 98) a. Young stem, b. Flower, c. Longitudinal section of the flower, d. Calyx and calycine colleters, e. Leaves on the node with petiolar and laminar colleters, cal: calyx, cclt: calycine colleter, cor: corolla, lclt: laminar colleter, ov: ovaryum, pclt: petiolar colleter, stg: stigma, stl: stylus.

Şekil 3. Cionura erecta'da (S. Güven 98) colleterlerin konumu a. Genç gövde, b. Çiçek, c. Çiçekten boyuna kesit, d. Kaliks ve calycine colleterler, e. Petiolar and laminar colleterlere sahip nod üzerindeki yapraklar, cal: kaliks, cclt: calycine colleter, cor: korolla, lclt: laminar colleter, ov: ovaryum, pclt: petiolar colleter, stg: stigma, stl: stilus.

Anatomical description of the stem, petiole, lamina and colleters

In cross-sections, the stem is circular in outline, the peridermis 100-115 µm in wide consists of 6-7 layers of rectangular, thick-walled, and occasionally fragmented cells. The cortex 400-470 µm wide comprises 3-4 layered collenchyma beneath the peridermis, and multi-layered parenchymatous tissue containing druse crystals, laticifers, and large and small groups of thick-walled sclerenchymatous fibers. The stele is composed of bicollateral vascular bundles (900-1200 µm wide) arranged in a ring containing a distinct cambium between the external phloem and the xylem, and the internal phloem is arranged in small groups adjoining the xylem. The external phloem contains dense druse crystals, and thick-walled sclerenchyma cells at the cortex border, either solitary or in pairs, or in small groups. The pith is composed of thin-walled orbicular parenchymatous cells, bearing druse crystals, and laticifers (Figure 5a-d).

In cross-sections, the petiole is circular in outline with a slightly concave adaxial side (Figure 5e), and the midrib is circular in shape (Figure 6a). Both the petiole and midrib contain a main arc shaped bicollateral bundle in the center, measured as 280-330 µm wide (Figure 5f-g) and 210-270 µm wide (Figure 6b), respectively, which is

surrounded by multi-layered, thin-walled, orbicular parenchyma cells, idioblasts containing druse crystals, and laticifers. The collenchyma, lined along the the epidermis, is 5-10 layered and 60-120 µm wide on the petiole (Figure 5e), while it is determined 4-8 layered and 50-90 µm wide beneath both the adaxial and abaxial sides of the midrib (Figure 6a). Simple, multicellular and unbranched trichomes are found on the epidermis of both the petiole and lamina. Laticifers are clearly seen in both cross and longitudinal sections of the petiole (Figure 5h).

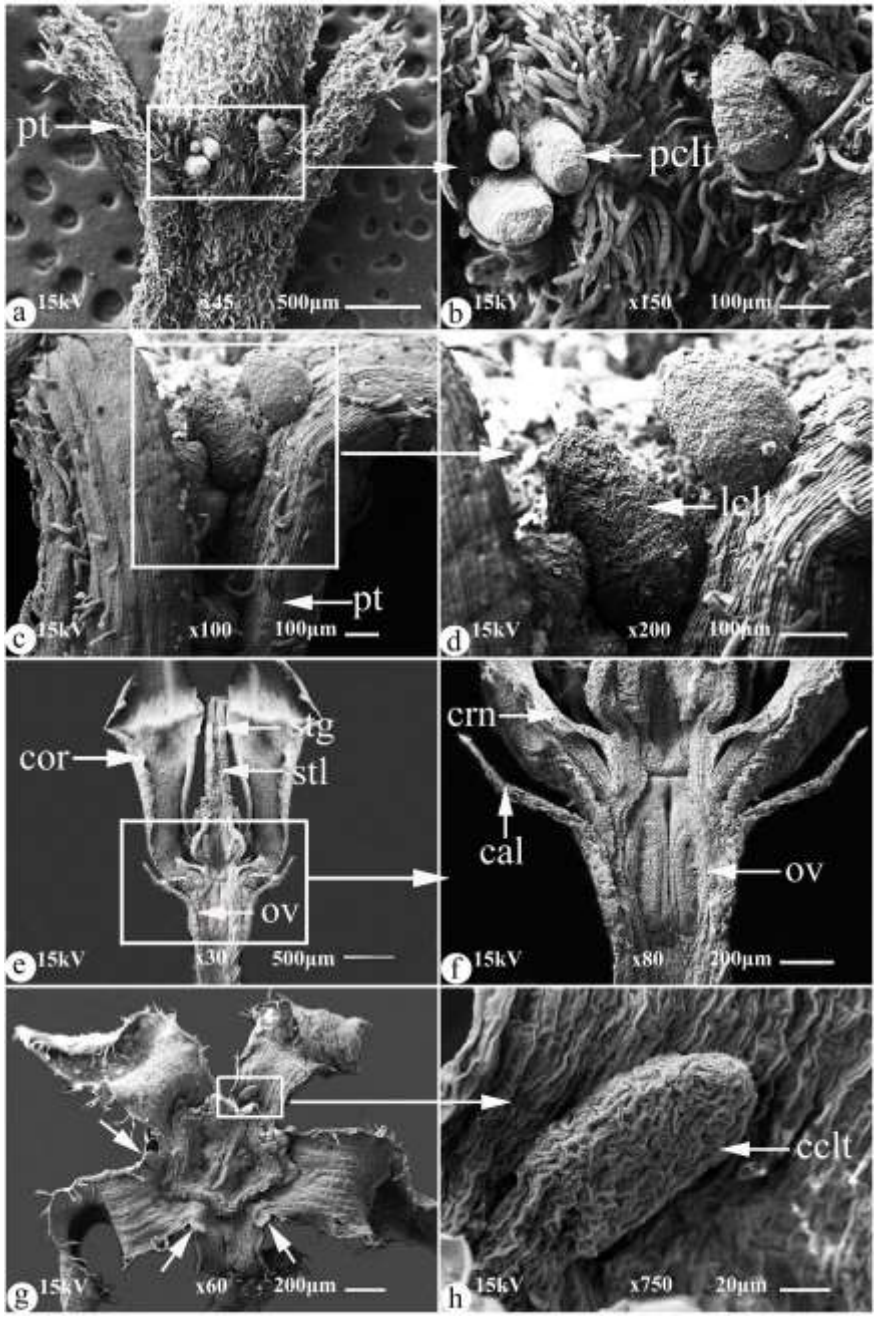

Figure 4. Scanning electron micrographs of the colleters in *Cionura erecta* (S. Güven 98) a-b. Petioles and petiolar colleters, c-d. Laminar colleters, e-f. c. Longitudinal section of the flower, g-h. Calyx and calycine colleters, cal: calyx, cclt: calycine colleter, cor: corolla, crn: corona, lclt: laminar colleter, ov: ovaryum, pclt: petiolar colleter, stg: stigma, stl: stylus.

Şekil 4. Cionura erecta (S. Güven 98)'ya özgü colleterlere ait taramalı elektron mikroskobu görüntüleri a-b. Petiyoller ve petiolar colleterler, c-d. Laminar colleterler, e-f. c. Çiçekten boyuna kesit, g-h. Kaliks ve calycine colleterler, cal: kaliks, cclt: calycine colleter, cor: korolla, crn: korona, lclt: laminar colleters, ov: ovaryum, pclt: petiolar colleter, stg: stigma, stl: stilus.

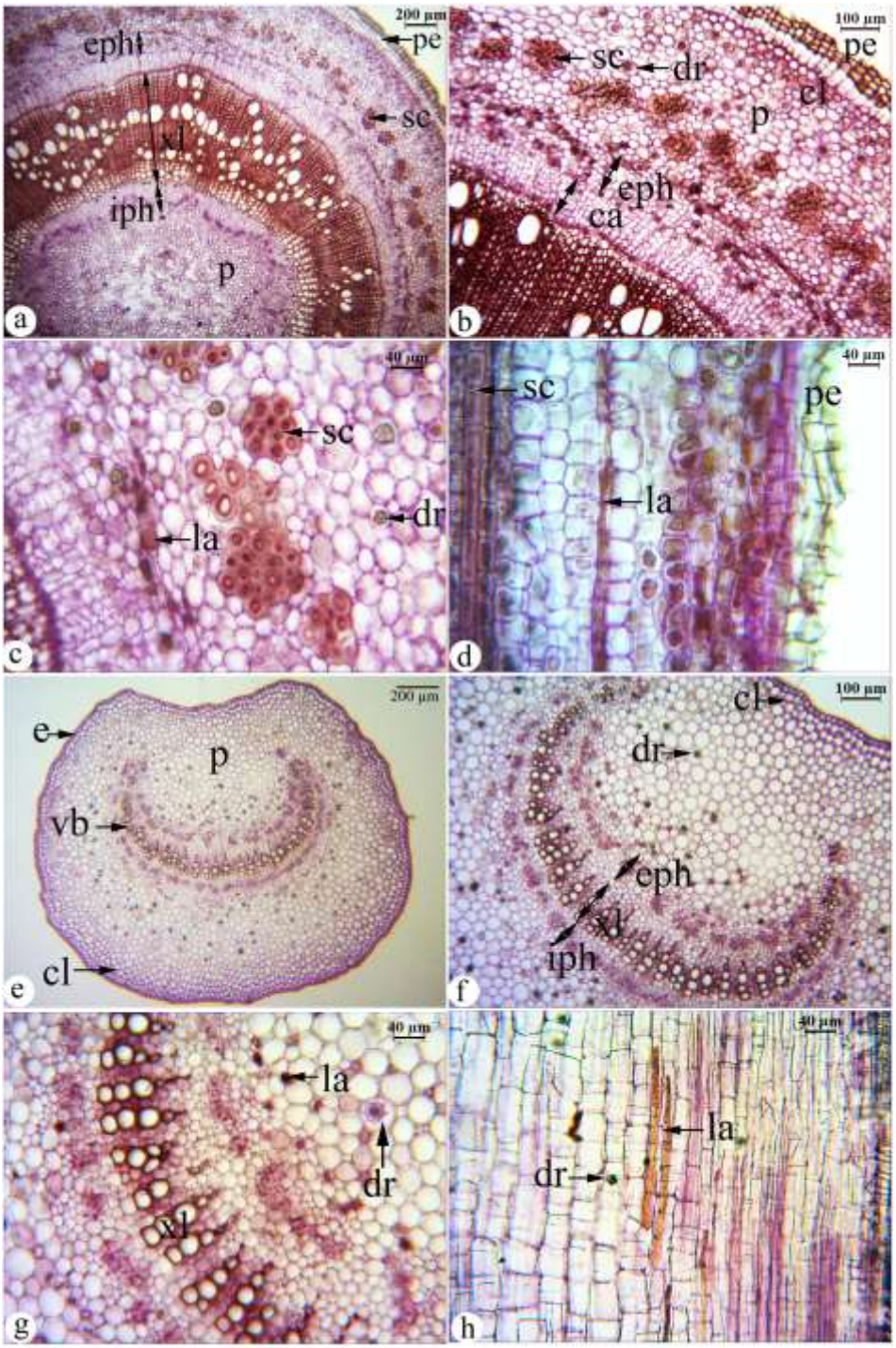

Figure 5. Stem and petiole anatomy of *Cionura erecta* (S. Güven 98) a-c. Transverse sections of the stem, d. Longitudinal section of the stem, e-g. Transverse sections of the petiole, f. Longitudinal section of the petiole, ca: cambium, cl: collenchyma, dr: druse, e: epidermis, eph: external phloem, iph: internal phloem, lt: laticifer, p: parenchyma, ph: phloem, pi: pith, pr: pericycle, sc: sclerenchyma cells, vb: vascular bundle, xl: xylem.

Şekil 5. Cionura erecta'nın (S. Güven 98) gövde ve petiyol anatomisi a-c. Gövde enine kesitleri, d. Gövde boyuna kesiti, e-g. Petiyol enine kesitleri, f. Petiyol boyuna kesiti, ca: kambiyum, cl: kollenkima, dr: druz, e: epidermis, eph: dış floem, iph: iç floem, lt: latisifer, p: parenkima, ph: floem, pi: öz, pr: perisikl, sc: sklerenkima hücreleri, vb: iletim demeti, xl: ksilem.

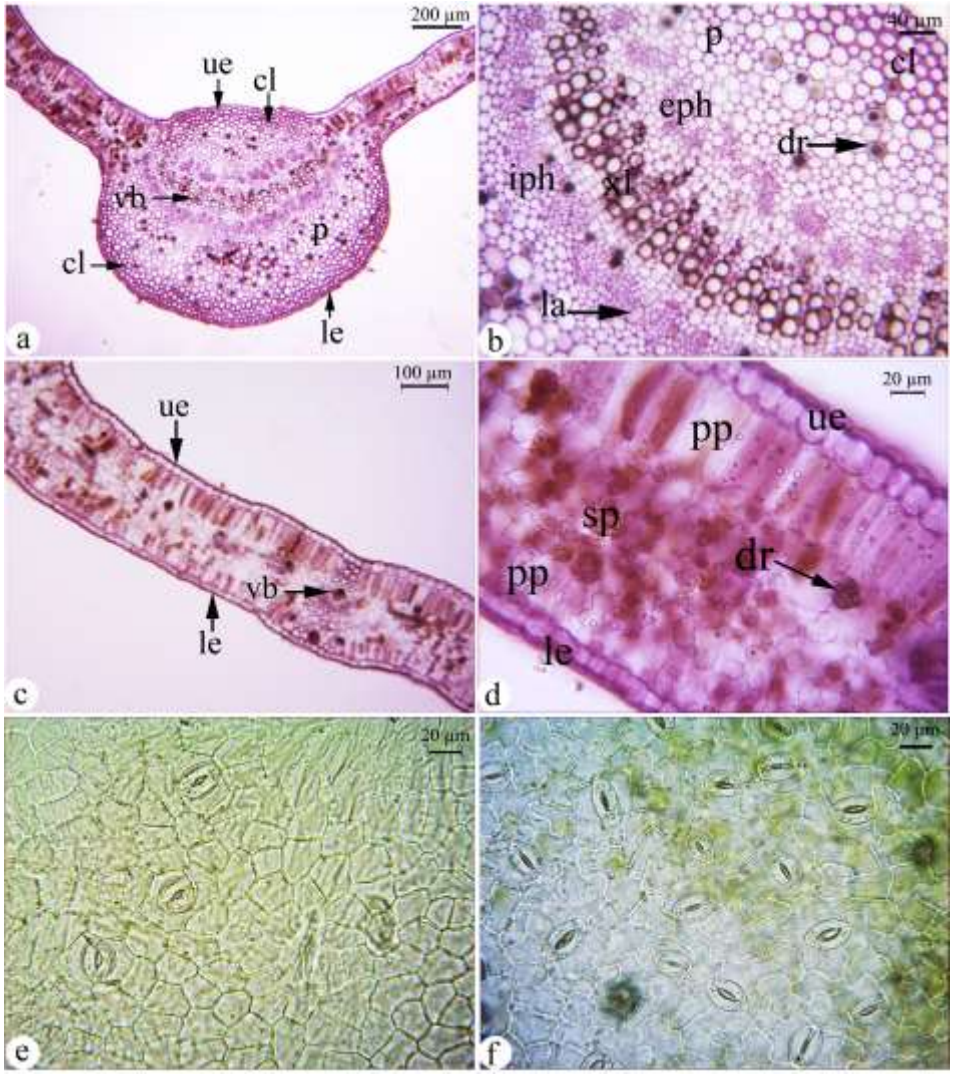

Figure 6. Leaf anatomy of *Cionura erecta* (S. Güven 98) a-b. Transverse sections of the midrib, c-d. Transverse sections of the mesophyll, e. Superficial section of the upper leaf surface, f. Superficial section of the lower leaf surface, eph: external phloem, cl: collenchyma, dr: druse, iph: internal phloem, la: laticifer, le:lower epidermis, p: parenchyma, ph: phloem, ue: upper epidermis, vb: vascular bundle, xl: xylem.

Şekil 6. Cionura erecta (S. Güven 98)'nın yaprak anatomisi a-b. Orta damarın enine kesitleri, c-d. Mezofilin enine kesitleri, e. Yaprağın üst yüzeyinin yüzeysel kesiti, f. Yaprağın alt yüzeyinin yüzeysel kesiti, eph: dış floem, cl: kollenkima, dr: druse, iph: iç floem, la: latisifer, le: alt epidermis, p: parenkima, ph: floem, ue: üst epidermis, vb: iletim demeti, xl: ksilem.

The leaf is equifacial composed of single layered palisade parenchyma cells 50-90 µm in length beneath the upper epidermis and 30-45 µm in length beneath the lower epidermis, and an 8-10 layered and 70-105 µm wide spongy parenchyma in the middle part. Small vascular bundles in regular rows and idioblasts containing druse crystals among the palisade and spongy parenchyma cells are located throughout the mesophyll tissue (Figure 6c-d).

In superficial sections, the leaf is observed as amphistomotic with paracytic, anomocytic, and anisocytic stomata on the both adaxial and abaxial surfaces. On the upper surface, stomata are 24-33 × 18-23 µm, and epidermal cells are 25-50 × 17-27 µm (Figure 6e), and per mm^2 the number of stomata is 42 and the number of epidermis cell is 2258; on the lower surface, stomata are 23-28 × 17-20 µm, and epidermal cells are 25-45 × 12-22 µm (Figure 6f), and per mm^2 the number of stomata is 286 and the number of epidermis cell is 2490. The stomatal index is determined as 2.16 for the upper surface, and as 10.24 for the lower surface.

In longitudinal sections, all the examined petiolar (Figure 7a-b), laminar (Figure 7c-d) and calycine colleters (Figure 7e-f) have a short non-secreting stalk limited to a few cell layers, and a main elongated secretory head, consisting of an axis of parenchyma covered by a uniseriate secretory palisade epidermis covered by a thin cuticle.

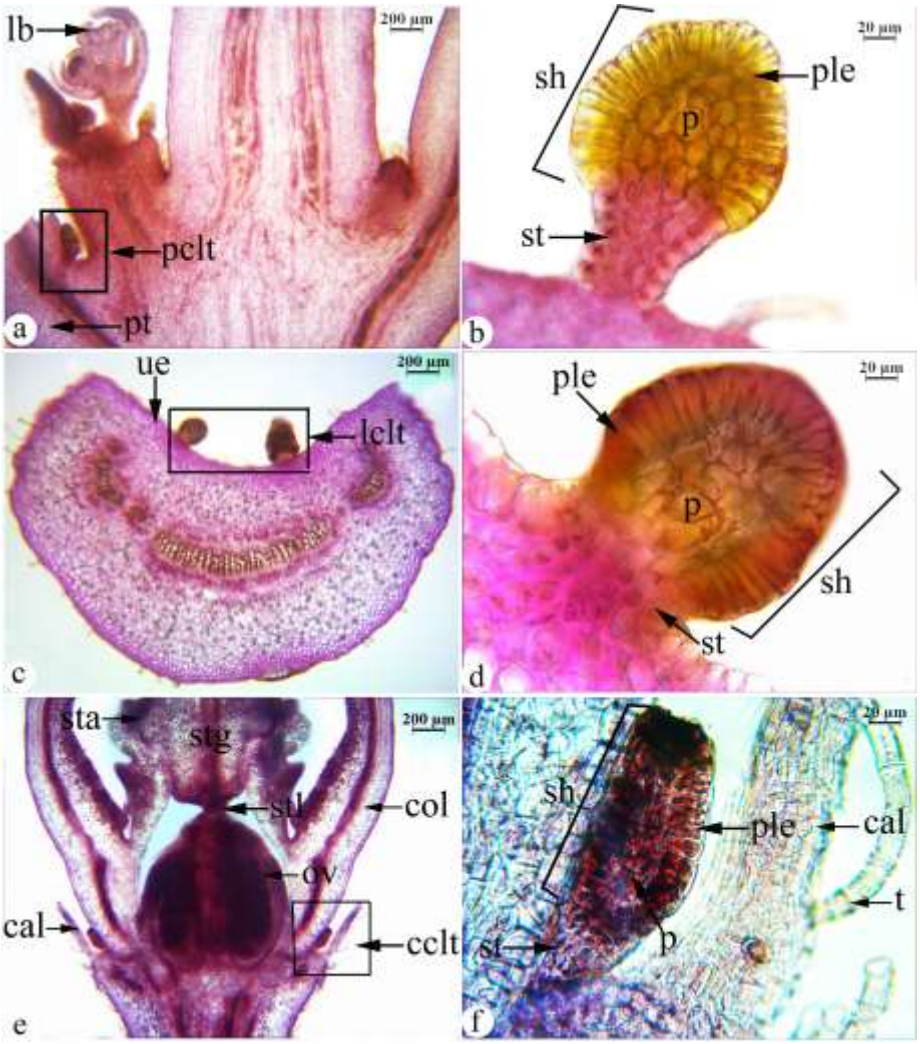

Figure 7. Anatomical sections of the colleters of *Cionura erecta* (S. Güven 98) a. Longitudinal section of the node, b. Longitudinal section of petiolar colleter, c. Cross section of the midrib, d. Longitudinal section of laminar colleter, e. Longitudinal section of the flower, f. Longitudinal section of calycine colleter, cal: calyx, cclt: calycine colleter, col: corolla, lb: lateral bud: lclt: laminar colleter, ov: ovaryum, p: parenchyma, pclt: petiolar colleter, ple: secretory palisade epidermis, sh: secretory head, st: stalk, sta: stamen, stg: stigma, stl: stylus, ue: upper epidermis.

Şekil 7. Cionura erecta (S. Güven 98)'ya özgü colleterlere ait anatomik kesitler a. Noda ait boyuna kesit, b. Petiolar colletere ait boyuna kesit, c. Orta damara ait enine kesit, d. Laminar colletere ait boyuna kesit, e. Çiçekten alınan boyuna kesit, f. Calycine colletere ait boyuna kesit, cal: kaliks, cclt: calycine colleter, col: korolla, lb: lateral tomurcuk: lclt: laminar colleter, ov: ovaryum, p: parenkima, pclt: petiolar colleter, ple: salgı yapan palizat epidermisi, sh: salgı yapan baş kısmı, st: sap, sta: stamen, stg: stigma, stl: stilus, ue: üst epidermis.

DISCUSSION

In the present SEM analyzes, the surface micro-morphology of the stem, petiole and lamina of *C. erecta*, containing colleters, was examined in detail, for the first time. The micromorphological features of vegetative organs had an important role in classification of flowering plants (Tomaszewski & Zieliński, 2014). It is well known that the epidermal surface of primary stems and leaves are covered with the cuticle, and epicuticular wax (whitish coating proper), respectively. SEM examinations on vegetative and reproductive plant organs revealed that the epicuticular wax might have a smooth appearance, or exhibite various ornamentations like striate, reticulate, or micro-papillose containing smaller crusts, platelets, granules, and tubules on the surface (Barthlott et al., 1998; Tomaszewski & Zieliński, 2014). The epicuticular wax layer of the examined *C. erecta* was determined as smooth on the stem and petiole surface, and striate on the leaf surface. Consistent with the present results, the epicuticular wax layer was determined as smooth on stems and petioles of the taxa of Turkish *Vincetoxicum* Wolf (Güven & Doğan, 2025), and as striated on leaves of many taxa belonging to Apocynaceae (Joubert, 2007; Xin-sheng, 2010; Bashir et al., 2020; Medina et al., 2021; Güven & Doğan, 2025). Epidermal cells on both sides of leaves of *C. erecta* were polygonal with convex periclinal cell walls and channelled, straight anticlinal cell walls. Similar epidermal micromorphological traits were defined for the taxa of *Vincetoxicum* (Güven & Doğan, 2025), *Cynanchum* L. (Xin-sheng, 2010) and *Cryptolepis* R.Br. (Joubert, 2007).

The stem, petiole, and lamina of *C. erecta* was covered with multicellular non-glandular, short curly trichomes. Similarly, in the tribe Marsdenieae, the indumentum of the vegetative parts was characterised with multicellular, non-glandular trichomes in some taxa of *Gymnema* R.Br. (Nikalje et al., 2013; Rajput et al., 2024), while the indumentum contained multicellular, glandular trichomes in *Dregea volubilis* (L. f.) Benth. (Biswas et al., 2018). Former researchs revealed a wide variation by means of the type of trichome within the Apocynaceae, containing unicellular to multicellular, glandular to non-glandular hairs. Although, some taxa of Apocynaceae displayed only non-glandular trichomes (Güven & Doğan, 2025), the other ones exhibited both non-glandular and glandular trichomes (Gabr et al., 2015; El-Taher et al, 2020; Medina et al., 2021).

The plant samples belonging to the examined *C. erecta* had both foliar and floral colleters, in greater numbers near shoot apices. Of the foliar ones, petiolar colleters were located at the base of petioles on nodes, and the laminar ones occured on the leaf blade close to the distal portion of the petiole. Floral colleters, named calycine colleters, were observed on the adaxial connate base of sepals in alternisepalous distribution. Anatomical and micromorphological studies including many Apocynaceae members revealed three types of foliar colleters: interpetiolar colleters (stipular colleters) distributed continuously between the petioles, petiolar colleters on the distal or the proximal position of the petiole, and laminar colleters on leaf blade (Capelli et al., 2017; Ribeiro et al., 2017, 2021). Ribeiro et al. (2021) determined that the petiolar colleters were found at the base of petioles in *Allamanda* L. and *Mandevilla* Lindl., and both at the base and in the distal portion of the petiole close to the leaf blade in *Blepharodon* Decne. and *Peplonia* Decne. Simões et al. (2006) separated calycine colleters in three groups based on their positions on sepals in seven taxa of Apocynacea: alternate (*Mandevilla pycnantha* (Steud.) Woodson, *M. tenuifolia* (J. C. Mikan) Woodson, *Secondatia densiflora* A. DC., and *S. floribunda* A. DC.), opposite (*Mandevilla scabra* (Hoffm. ex Roem. & Schult.) Schum.), and indefinitely distributed (*Macrosiphonia longiflora* (Desf.) Müll. Arg. and *Mesechites mansoanus* (A. DC.) Woodson). Although alternisepalous calycine colleters have been considered a plesiomorphic feature in Apocynaceae, they were reported to be the most common calycine colleter type within the family (Demarco, 2017).

Various types regarding colleter morphology were described for the taxa of Apocynaceae, such as standard, bifurcated, laminar and sessile (Capelli et al., 2017; Demarco, 2017; Ribeiro et al., 2017, 2021). All colleters of *C. erecta* studied here, having a short, non-secretory stalk, and a conical, undivided main secretory head composed of a parenchyma axis surrounded by an epidermis of elongated secretory cells, were defined as standard type. The standard type has been considered as the most frequent colleter type within the family (Thomas & Dave, 1991; Capelli et al., 2017; Demarco, 2017; Ribeiro et al., 2017, 2021). Although the position, structure, and number of colleters may vary among species, their secretory activities were determined to increase or decrease during similar periods for most taxa of Apocynaceae. In spring and summer, when the meristems were considered more active, the number of foliar colleters were counted as more than those of occuring in autumn and winter, when meristem activity was reduced (Ribeiro et al., 2017). However, the secretory activity was detected to continue during the entire floral development in calycine colleters, which was considered to play a significant role in protecting the plant against microorganisms (Demarco, 2017).

Cionura erecta is a woody-based perennial plant consisting of many herbaceous sprawling stems. Anatomical examinations revealed collenchyma cells beneath the peridermis and scattered sclerenchyma cell groups in the cortex for the stem, and bicollateral vascular bundles, laticifers and druse crystals in all vegetative organs. The present anatomical results regarding the general stem, petiole and leaf structure of *C. erecta* were compatible with the vegetative anatomy of the taxa in the same tribe Marsdenieae, such as *Gymnema sylvestre* (Retz) R. Br. ex. Sm (Nikalje et al., 2013; Pham et al., 2018), *G. cuspidatum* (Thunb.) Kuntze, *G. latifolium* Wall. ex Wight and *G. montanum* (Roxb.) Hook.f. (Rajput et al., 2024), *Gongronema latifolium* Benth. (Adeniran et al., 2022) and *Dregea volubilis* (Biswas et al., 2018), and also members of the other tribes in Apocynaceae from Türkiye, *Amsonia orientalis* Decne. (Akyalçin et al., 2006), *Vincetoxicum canescens* (Willdenow) Decne, *V. fuscatum* Reichenbach and *V. parviflorum* Decne (İlçim et al., 2010), and *Nerium oleander* L. (Sargın, 2021). In the examined *C. erecta*, the secondary growth in the stem displayed the presence of the periderm as reported for the other woody vine species in the tribe Marsdenieae like *D. volubilis* (Biswas et al., 2018) and *G. sylvestre* (Nikalje et al., 2013; Pham et al., 2018; Rajput et al., 2024). The young stem structure of *C. erecta* was characterised with many groups of scattered cortical sclerenchymatous fibers located near the bicollateral vascular bundles arranged in a ring, as mentioned in the above literature on the tribe Marsdenieae. The presence of sclerenchymatous cells in the stem cortex, providing firmness and mechanical support to the stem, has been considered the most common anatomical character for both the herbaceous and the woody plants in the Apocynaceae. Likewise, the collenchyma tissue, which was observed along the epidermis in the stem and petiole, and beneath both the lower and upper epidermis in the midrib, was the other main structure providing strength and support to the primary plant parts of the examined *C. erecta* samples, as in the vast majority of Apocynaceae (Metcalfe & Chalk, 1950; Schweingruber et al., 2011).

Leaves are the main organs that help plants in the three major functions for plant growth and other life processes; to produce food on their own by photosynthesis, to maintain water and heat balance through transpiration, and to convert sugars (photosynthates) back into energy by respiration (Smith et al., 1997). The foliar anatomical characters such as the presence, arrangement and dimension of palisade and sponge parenchyma in the mesophyll, the shape and size of stomata and epidermis cells, and the stomatal index on leaf surfaces have been widely used as distinguishing characters in the classification of species within the family Apocynaceae (Metcalfe & Chalk, 1950; Gabr et al., 2015; El-Fiki et al., 2019; Bashir et al., 2020; El-Taher et al., 2020). Leaves of the examined *C. erecta* were equifacial with the mesophyll containing single layered palisade parenchyma cells under both the lower and upper epidermis, and amphistomatic with paracytic, anomocytic and anisocytic stomata both on the upper and lower epidermis. However, previous anatomical studies revealed that bifacial and hypostomatic leaves were more common characteristics within the taxa of the tribe Marsdenieae, (Nikalje et al., 2013; Biswas et al., 2018), as well as the taxa in the other tribes of Apocynaceae (Gabr et al., 2015; Bashir et al., 2019; El-Fiki et al., 2019; Khaleeda & Meekiong, 2023). Otherwise, similar to *C. erecta*, it was reported that leaves were amphistomatic in *G. latifolium* (Adeniran et al., 2022), in the tribe Marsdenieae, and in *Catharanthus roseus* (L.) G. Don, *Carrisa carandas* L., *Plumeria rubra* L. and *Trachelospermum lucidum* (D.Don) K.Schum, in the other tribes of the family Apocynaceae (Bashir et al., 2020). In addition, isobilateral (equifacial) mesophyll was defined for some members of Apocynaceae such as *N. oleander*, *Leptadenia arborea* (Forssk.) Schweinf., *Calotropis procera* (Aiton) W. T. Aiton. and *Solenostemma argel* (Delile) Hayne. (Gabr et al., 2015; Sargın, 2021). Plants have adaptations in leaf orientation and external leaf morphology as well as their internal anatomy to reach more light, depending on environmental conditions. It is well known that the mesophyll structure mostly depends on the plant's degree of exposure to light. While sun leaves are generally amphistomatic with well-developed palisade layers, shade leaves are hypostomatic, and with less developed palisade layers (Smith et al., 1997). Consistent with studies drawing attention to the relationships between habitat and leaf anatomy, *C. erecta* which has stomata and palisade parenchyma on both adaxial and abaxial sides of leaves, has a wide distribution in humid and sunny habitats in Türkiye, except for central and north eastern Anatolia (Browicz, 1978; Güven & Sarıkaya, 2024).

The stoma type is an other important leaf character used to distinguish plant species (Metcalfe & Chalk, 1950). *C. erecta* was found to be characterised with paracytic, anomocytic, and anisocytic stomata on the both adaxial and abaxial surfaces. Similarly, in the tribe Marsdenieae, the stomata were determined as paracytic or anisocytic in *G. sylvestre* (Nikalje et al., 2013), anomocytic in *G. latifolium* (Adeniran et al., 2022), and paracytic and anomocytic in *D. volubilis* (Biswas et al., 2018). Moreover, several investigations regarding the taxa in the other tribes of Apocynaceae carried out that only anisocytic type of stomata or in combination with other types of stomata including anomocytic, anisocytic, cyclocytic, stephanocytic, brachyparacytic, and hemiparacytic occurred within the family (Guidoti et al., 2015; Carvalho et al., 2017; Bashir et al., 2019; El-Fiki et al., 2019; Pirolla-Souza et al., 2019; El-Taher et al., 2020; Khaleeda & Meekiong, 2023). Additonally, in Pakistan, Nisa et al. (2019) defined 10 major stomatal types including anomocytic, actinocytic, paracytic, polar paracytic, pseudoholoparacytic, anisocytic, tetracytic, laterocytic, stephanocytic, and brachyparahexacytic, and also 36 subtypes of stomata in *Vincetoxicum arnottianum* (Wight) Wight.

The stem, petiole and midrib of *C. erecta* contained numerous non-articulated laticifers, and idioblasts with druse crystals, which were among the most specific characteristics for Apocynaceae members (Metcalfe & Chalk, 1950; Akyalçin et al., 2006; İlçim et al., 2010; Nikalje et al., 2013; Gabr et al., 2015; Pham et al., 2018; Biswas et al., 2018; El-Fiki et al., 2019; Sathya et al., 2022; Abeysinghe & Scharaschkin, 2022; Rajput et al., 2024). Laticifers which are categorized as articulated and non-articulated types, are common in vegetative and floral organs of asclepiads (Metcalfe & Chalk, 1950; Demarco, 2017). The latex of Apocynaceae, milky-white or in different colors, may contain glycosides, alkaloids, sterols, cardenolides, tannins, proteins, and antimicrobial peptides, which were found to be efficient in various biological activities, such as anticancer, anti-inflammatory, analgesic, antimicrobial, antioxidant and toxic. The various compounds are considered to protect the plant against herbivores and microorganisms (Demarco, 2015, 2017). Calcium oxalate crystals were reported as another toxicologically significant secondary metabolite for the members of the family Apocynaceae. The poisonous property of both latex and calcium oxalate crystals have been considered to help adaptation of Apocynaceae plants to harsh environmental conditions by supporting to fight against herbivores and microbial invaders (Molik et al., 2025).

CONCLUSION

The present research provided the first comprehensive micromorphological and anatomical data regarding *C. erecta*. The presence of collenchyma cells and scattered sclerenchyma cell groups in the stem, bicollateral vascular bundles, laticifers and druse crystals in all vegetative portions, and paracytic, anomocytic, and anisocytic stomata on the amphistomotic and equifacial leaf were determined as the most characteristic anatomical features. *C. erecta* was characterised with petiolar (located at the base of petioles), laminar (occured on the leaf blade close to the

distal portion of the petiole) and calycine colleters (found on the adaxial connate base of sepals in alternisepalous distribution). All of the colleters were defined as standard type containing a short, non-secretory stalk, and a conical, undivided main secretory head. Although *C. erecta* has been considered to be a poisonous, various vegetative and reproductive portions of this plant have been commonly used for both folk medicine and medicinal purposes. The present results revealing the micromorphological and anatomical properties, including glandular structures like laticifers and colleters, will contribute significantly to the identification of this medicinal plant. Additionally, the present results constitiues an important data for future biochemical studies on more individuals, which may contribute to explaining the relationship between the anatomical and micromorphological structures and the therapeutic and toxic properties of plant organs.

Conflict of Interest

The author has declare no conflict of interest.

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