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Flower structure in ten Asteraceae species: considerations about the importance of morpho-anatomical features at species and tribal level

Michelli Fernandes Batista[1] · Luiz Antônio De Souza[1]

Abstract Flowers of Asteraceae have conspicuous features that are important at the tribal level, such as the branches of the style, pappus, shape of the corolla, and anthers. The floral ontogeny of ten Brazilian weed species of Asteraceae was performed for *Conyza bonariensis* L., *Cosmos sulphureus* Cav., *Eclipta alba* L., *Elephantopus mollis* Kunth, *Emilia sonchifolia* L. DC., *Galinsoga quadriradiata* Ruiz and Pav., *Erechtites valerianifolius* (Link ex Spreng) DC., *Parthenium hysterophorus* L. *Praxelis clematidea* R. M. King and H. Rob, and *Sigesbeckia orientalis* L. in order to describe the morpho-anatomy, ontogenetic steps, and identify features that may support the tribeal classification. Flowers and buds were fixed in glutaraldehyde, embedded in historesin, and sectioned with a rotary microtome. Sections were stained with Toluidine Blue. The inflorescence can be heterogamous or homogamous; the pappus is absent or persistent; the corolla is tubular or ligulate; anthers have an apical appendage which is obtuse or acute; and some anatomical features are the ovary usually trapezoidal-shaped; the pappus either vascularized or not; the corolla with possible secretory structures, only two species do not show twin hair on the outer ovary epidermis;and ovarian mesophyll usually composed of two or three regions. In general, the species analyzed show several similar features, such as the anther structure, pappus, and stigma. A distinct feature was observed among the species: the absence of pappus, and twin hairs in three species: *E. alba* (Heliantheae tribe), *P.*

hysterophorus (Heliantheae tribe), and *S. orientalis* (Millerieae tribe), all belonging to "Helliantheae Alliance" clade.

Keywords Anther · Corolla · Ovary · Pappus · Twin hair

Introduction

The Asteraceae family belongs to the Asterales order and are the largest family within the eudicotyledons with approximately 1700 genera and 25000 species, also recognized as a monophyletic group. The family is divided into 43 tribes, which are often arranged into 12 subfamilies allied to the "Helliantheae Alliance" clade (Funk et al. 2009). Asteraceae are characterized by flowers grouped in the capitulum inflorescence type containing one to hundreds of individual florets (Harris 1999). The florets are usually all equal to each other (monogamous capitulum) or have differentiated rays and disks (heterogamous capitulum). The inflorescences are protected by an enclosure of bracts, and their florets have connate and introrse anthers, a modified calyx known as pappus (dispersion element), cypsela fruit type (achenes), and bifid style (Katinas et al. 2016). Some floral morphological and anatomical features are important for the classification of the species among the tribes, such as branched style, pappus form, anatomy, and shape of the corolla, and anatomy and shape of anthers (Judd et al. 2002).

This family contains several economically important species used in feeding (*Lactuta sativa* L. lettuce and *Cichoriu endivia* L. chicory), pharmaceutical industry (*Baccharis genistelloides* (Lam.)Pers. gorse), home medicine (*Matricaria recutita* L. chamomile and *Artemisia absinthium* L. wormwood), and other species are used for ornamental purposes (*Leucanthemum vulgare* (Lam.) daisy and

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Helianthus annuus L. sunflower). However, most of the genera have weedy species, and their presence causes a big problem for agriculture. For cereals and vegetables, the presence of weedy plants can cause losses of 100 %, while in cultures of forest and fruit trees, the losses are smaller (Radosevich et al. 1997).

Asteraceae have historically presented problems for ontogenetic studies (Dadpour et al. 2012), and the available information about floral structure is scarce. The knowledge of the floral biology in the widest sense is urgent (Endress 1994), and it has great importance in the understanding and knowledge of the conservation studies, systematic and ecological research, besides use and control of native or exotic plants. Thus, the main purpose of this investigation was to determine the flower morphological and anatomical aspects of weedy Asteraceae useful to separate species and contribute at tribal level.

Materials and methods

Buds and flowers of the following ten species were collected in the field (Universidade Estadual de Maringá, Brazil): *Conyza bonariensis* L., *Cosmos sulphureus* Cav.,

Eclipta alba L., *Elephantopus mollis* Kunth, *Emilia sonchifolia* L. DC., *Galinsoga quadriradiata* Ruiz and Pav., *Erechtites valerianifolius* (Link ex Spreng) DC., *Parthenium hysterophorus* L. *Praxelis clematidea* R. M. King and H. Rob, and *Sigesbeckia orientalis* L., currently recognized among the tribes according to Funk et al. (2009) (Table 1).

Material was fixed in 1 % glutaraldehyde in 0.1 M phosphate buffer (pH 7.2) and examined later after being embedded in HistoResin (Guerrits 1991) (Leica Biosystems) and sectioned transversally and longitudinally using a rotary microtome. Sections varying from 8 to 12 μm were stained with 0.05 % toluidine blue (pH 4.7) (O'Brien et al. 1964). Light microscope photographs were taken using a digital camera mounted on a Leica EZ4D Microscope and, subsequently, processed using Leica Application Suite version 1.8 software.

For the micromorphological analyses (SEM), the buds and flowers were fixed in 2.5 % glutaraldehyde in buffer solution (0.05 M sodium phosphate, pH 7.2) for 48 h, dehydrated in a graded alcohol series to 100 %, submitted to critical-point dried with liquid CO_2 (Polaron Instruments E3000), and fixed on aluminum stubs. Afterward, the samples were gold coated (Edwards Sputter Coater S150B), and subsequently examined using electron microscopy (Shymadzu SS-550 Superscan), and digital images.

Table 1 Asteraceae species collected for the studies, with their respective UEM herbarium record numbers and tribes

Species	Record number	Tribes
Conyza bonariensis	18585HUEM	Astereae
Cosmos sulphureus	18589HUEM	Coreopsideae
Eclipta alba	18587HUEM	Heliantheae
Elephantopus mollis	18588HUEM	Vernonieae
Emilia sonchifolia	18586HUEM	Senecioneae
Galinsoga quadriradiata	18590HUEM	Millerieae
Erechtites valerianifolius	21182HUEM	Senecioneae
Parthenium hysterophorus	20828HUEM	Heliantheae
Praxelis clematidea	21181HUEM	Eupatorieae
Sigesbeckia orientalis	20836HUEM	Millerieae

Results

Flower morphology

Flower morphological features are summarized in Table 2. The flowers of *E. mollis*, *E. sonchifolia*, and *P. clematidea* are arranged in homogamous capitula with perfect florets (Figs. 1, 2). In the other species, the capitula are heterogamous (Figs. 3, 4) and contain perfect disk florets and

Table 2 Flower morphological features of the investigated species

Species	Capitula	Pappus	Corolla	Apical appendage	Ovary shape
Conyza bonariensis	Heterogamous	Persists	Tubular	Acute	Trapezoidal
Cosmos sulphureus	Heterogamous	Persists	Ligulate and tubular	Acute	Trapezoidal
Eclipta alba	Heterogamous	Absent	Ligulate and tubular	Obtuse	Trapezoidal
Elephantopus mollis	Homogamous	Persists	Tubular	Obtuse	Trapezoidal
Emilia sonchifolia	Homogamous	Persists	Tubular	Acute	Trapezoidal
Galinsoga quadriradiata	Heterogamous	Persists	Ligulate and tubular	Acute	Trapezoidal
Erechtites valerianifolius	Heterogamous	Persists	Tubular	Obtuse	Trapezoidal
Parthenium hysterophorus	Heterogamous	Absent	Ligulate and tubular	Obtuse	Elliptical
Praxelis clematidea	Homogamous	Persists	Tubular	Acute	Lozenge-shaped
Sigesbeckia orientalis	Heterogamous	Absent	Ligulate and tubular	Acute	Pentagonal

pistillate ray florets. The *C. sulphureus* and *P. hysterophorus* capitula are also heterogamous, but the former species has sterile ray florets and the latter staminate disk florets.

The pappus is entirely lacking in the florets of *P. hysterophorus* and *S. orientalis* (Fig. 5), but it persists in the other species (Fig. 6). The corolla is tubular (Fig. 7) and distinctly divided into tube and limb; it is composed of five petals, except for *E. alba* and *P. hysterophorus*, which have four petals. The corolla lobation degree differs among species, with larger lobes in *E. mollis*.

Heterogamous capitula are of several types. The capitulum of *C. bonariensis* consists of numerous ray florets with a white, tubular, filiform, 2–3-lobed corolla; the disk florets are also white and 5-lobed. *Cosmos sulphureus* is composed of eight ray florets with a ligulate, yellowish, tridentate corolla; the disk florets are tubular and pentalobed. The capitulum of *E. alba* comprises approximately

Figs. 1–6 Morphologies of the inflorescences and flowers of *P. clematidea* (**1**), *E. sonchifolia* (**2**, **6**), *S. orientalis* (**3**, **5**), and *E. alba* (**4**). **1**, **2** Homogamous inflorescence. **3**, **4** Heterogamous inflorescence. **5** Tubular flower devoid of pappus. **6** Flower with persistent pappus. *Bars* 2 cm (**1**, **2**, **6**), 1 cm (**3**, **4**), 3 cm (**5**). (Color figure online)
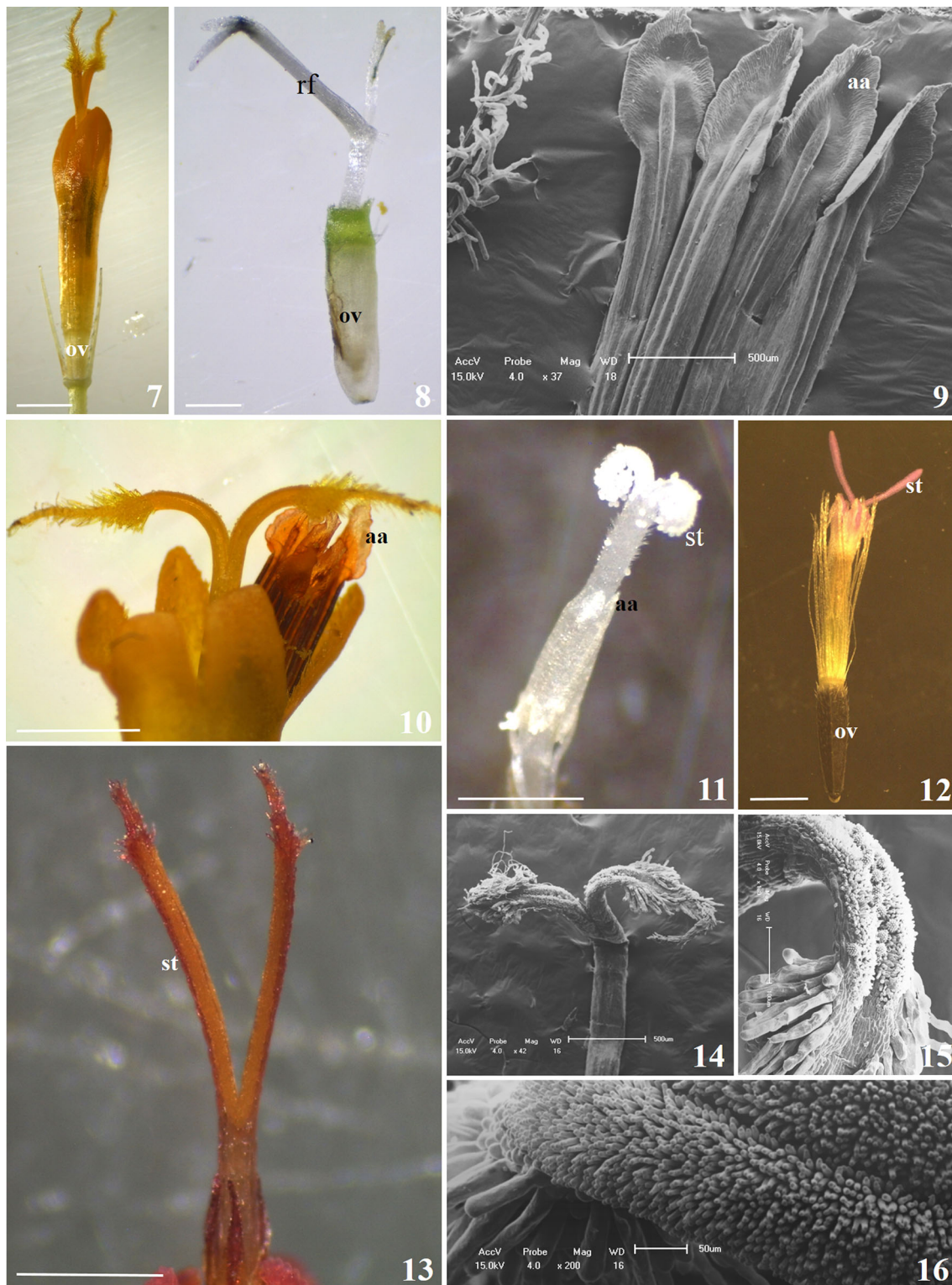
Figs. 7–16 Morphologic details of flowers. **7, 9, 10, 14–16** *Cosmos sulphureus.* **8** *Eclipta Alba.* **11** *Elephantopus mollis.* **12** *Praxelis clematidea.* **13** *Emilia sonchifolia.* **7** Tubular flower. **8** Ray flower with bidentate corolla. **9** Apical appendage of stamens (SEM). **10** Detail of acute stamen appendage. **11** Detail of obtuse stamen appendage and short, divided stigma. **12** Inferior ovary. **13** Long, divided stigma. **14–16** Detail of long stigma (SEM). *aa* Apical appendage, *ov* ovary, *pp* pappus, *rf* ray flower, *st* stigma. *Bars* 1.5 cm (**7, 8, 11, 12**), 1 cm (**10, 13**), 50 μm (**16**), 100 μm (**15**), 500 μm (**9, 14**). (Color figure online)

Table 3 Anatomical features of the investigated species

Species	Pappus shape	Pappus apex	Pappus mesophyll	Pappus vascular elements	Corrolla appex	Corrola secretory structures
Conyza bonariensis	Round	Trichomes with pointed apex	Parenchymatous	With vascular elements	Tector (ray), glabrous (disk)	Absent
Cosmos sulphureus	Round, triangular, lenticular	Papillae, trichomes with pointed apex	Sclerenchymatous	With vascular elements	Glandular (ray and disk)	Secretory ducts
Eclipta alba	Without pappus	Trichomes with pointed apex	Parenchymatous	–	Glabrous (ray and disk)	Laticifers
Elephantopus mollis	Round	Trichomes with pointed apex	Sclerenchymatous	Without vascular elements	Glabrous	Absente
Emilia sonchifolia	Round	Trichomes with pointed apex	Parenchymatous	Without vascular elements	Glabrous	Secretory ducts
Galinsoga quadriradiata	Round, lenticular	Trichomes with pointed apex	Parenchymatous	Without vascular elements	Tector (ray and disk)	Secretory ducts
Erechtites valerianifolius	Round, triangular, lenticular	Papillae trichomes with pointed apex	Sclerenchymatous	With vascular elements	Glabrous (ray), glandular (disk)	Secretory ducts
Parthenium hysterophorus	Without pappus	Trichomes with pointed apex	Parenchymatous	–	Glandular (ray), glabrous (disk)	Absent
Praxelis clematidea	Round	Trichomes with pointed apex	Parenchymatous	With vascular elements	Glabrous	Secretory ducts
Sigesbeckia orientalis	Without pappus	Trichomes with pointed apex	Parenchymatous	–	Glandular (ray), tector, and glandular (disk)	Absent

100 ray florets (Fig. 8), whose corollas are white and bidentate; disk florets are white with a 5-lobed apex. The capitulum of *E. valerianifolius* has tubular, 5-lobed lilac flowers, which usually comprise sequential 40–50 ray florets and six–ten disk florets. Five, white, tridentate ray florets allied to tubular, yellowish, 5-lobed disk florets are evident in *G. quadriradiata*. Similar to the species described above, *P. hysterophorus* has five white ray florets, but the disk florets have a 4-lobed corolla. *Sigesbeckia orientalis* contains the capitulum with five, white, pistillate, tridentate ray florets; and perfect, tubular, 5-lobed disk florets.

The stamens are connate to the inferior third of the corolla and have a sterile apical appendage (Fig. 9); there are usually five stamens composing the androecium, except for *P. hysterophorus*, which has four. The appendage is acute (Fig. 10) in *C. bonariensis, C. sulphureus, E. sonchifolia, G. quadriradiata, P. clematidea*, and *S. orientalis,* and obtuse (Fig. 11) in *E. alba, E. mollis, E. valerianifolius*, and *P. hysterophorus*.

The ovary is inferior (Figs. 7, 8, 12) and consists of two carpels. The ovary shape varies among the species: pentagonal in *S. orientalis*, elliptical in *P. hysterophorus*, lozenge-shaped in *P. clematidea*, and trapezoidal in the other species. The style (stigma) is bifurcated and commonly long-branched (Fig. 13), as seen in *C. sulphureus, E. sonchifolia*, and *P. clematidea,* or short branched, observed in *C. bonariensis, E. alba* (Fig. 14), *E. valerianifolius, E. mollis* (Fig. 11)*, P. hysterophorus*, and *S. orientalis*. The species stigmatic region occurs in two marginal lines and is highly homogeneous, except for *E. mollis*, in which the stigma surface is irregularly covered. The stigmas of *C. bonariensis, C. sulphureus, E. alba, E. sonchifolia, E. mollis, E. valerianifolius, G. quadriradiata, P. hysterophorus*, and *P. clematidea* are covered with papillae (Figs. 14–16). *Sigesbeckia orientalis* also has a stigma with a papillose surface, but clavate glandular trichomes occur in the apex of the stigma branches.

Flower anatomy

Flower anatomical features are summarized in Tables 3 and 4. As seen in cross section, the pappus members are round and lenticular in *G. quadriradiata* and *E.*

sonchifolia; round, triangular, and lenticular in *E. valerianifolius* and *C. sulphureus;* and round in *C. bonariensis, E. alba, E. mollis,* and *P. clematidea.* The pappus of *E. alba* is with a reduced and green ring-shaped structure (Fig. 8). The pappus has a uniseriate epidermis that consists of papillae (Figs. 17–19) (*C. sulphureus* and *E. valerianifolius*) or trichomes in most species (Figs. 10–12). The pappus mesophyll is usually parenchymatous, but it becomes conspicuously sclerenchymatous in *C. sulphureus, E. mollis,* and *E. valerianifolius* (Fig. 19). Vascular elements may be found in the pappus (Figs. 17, 19) of *C. bonariensis, C. sulphureus, E. valerianifolius,* and *P. clematidea,* although they are entirely lacking (Figs. 20, 22) in *E. mollis, E. sonchifolia,* and *G. quadriradiata.*

The corolla of the ray florets is composed of papillose, uniseriate, adaxial epidermis (Figs. 23, 24), and trichomes may be observed on the abaxial surface (Fig. 24). Trichomes are entirely lacking in *E. alba* corolla. The mesophyll is composed of homogeneous parenchyma (Figs. 23, 24) with collateral vascular bundles.

Disk florets have corolla with uniseriate epidermis with trichomes, although it is glabrous in *E. mollis, E. sonchifolia* (Fig. 25)*, P. hysterophorus* and *P. clematidea.* The hairy species are characterized by pluricellular and glandular trichomes, which are stalked and show a head of secretory cells (*E. alba*); nonglandular trichomes (*C. bonariensis*) (Fig. 26); multicellular nonglandular trichomes with pointed apex and clavate multicellular

Figs. 17–27 Pappus and corolla structures. **17**, **18** *Cosmos sulphureus.* **19** *Erechtites valerianifolius.* **20–22**, **25** *Emilia sonchifolia.* **23** *Parthenium hysterophorus.* **24**, **27** *Sigesbeckia orientalis.* **26** *Conyza bonariensis.* **17**, **19**, **22**, **23**, **24**, **26**, **27** Cross section. **20** Longitudinal section. **18**, **21**, **25** SEM. **17** Pappus (*black arrow*) and vascularized (*tip black arrow*). **18** Pappus. **19** Pappus with sclerified mesophyll (*white arrow*) and vascularized. **20** Pappus with elongated cells in the mesophyll. **21** Pappus. **22** Pappus nonvascularized. **23** Corolla ligulate with adaxial face epidermis papillose. **24** Corolla with epidermis of abaxial tector trichomes (*asterisk*). **25** Petals. **26** Corolla with tector trichomes (*asterisk*). **27** Corolla with tector trichomes (*asterisk*) and clavate multicellular glandular trichome. *co* Corolla, *pa* buds, *pp* pappus. *Bars* 50 μm (**17**, **19**, **20**, **23**, **24**, **26**), 100 μm (**18**, **22**, **25**), 50 μm (**21**), 40 μm (**27**). (Color figure online)

glandular trichomes (*S. orientalis*) (Fig. 27); glandular trichomes consisting of a uniseriate stalk one or two cells long, and a pluricellular head (*C. sulphureus*) (Figs. 28, 29); pluricellular glandular trichomes (*E. valerianifolius*) (Fig. 30); or extremely thin pluricellular nonglandular trichomes (*G. quadriradiata*) (Fig. 31). Both surfaces of the corolla apex are papillose, except for the abaxial epidermis of *C. sulphureus,* which is nonpapillose (Figs. 32, 33). The mesophyll is composed of homogeneous parenchyma and is restricted to the basal and apical regions of the corolla, with a larger number of layers at the apex (two–five cell layers) (Fig. 34). The homogeneous parenchyma is absent in the mesophyll middle region of all the investigated species (Fig. 35). Secretory structures are present in the

Table 4 Features of the androecium and gynoecium of the Asteraceae investigated species

Species	Twin hair (ovary)	Mesophyll (regions)	External mesophyll	Style	Abaxial epidermis (stigma)	Stigma surface
Conyza bonariensis	Present	Two regions	1-Long cells (radially)	Solid	Cylindrical cells	Papillose surface
Cosmos sulphureus	Present	Three regions	1-Long cells (radially)	Hollow	Cylindrical cells	Papillose surface
Eclipta alba	Absent	Three regions	1-Long cells (radially)	Solid	Papillose cells	Papillose surface
Elephantopus mollis	Present	Two regions	More than 1 (compact)	Solid	Papillose cells	No papillose surface
Emilia sonchifolia	Present	Two regions	2-5 layers	Solid	Papillose cells	Papillose surface
Galinsoga quadriradiata	Present	Three regions	1-Long cells (radially)	Solid	Papillae and trichomes	Papillose surface
Erechtites valerianifolius	Present	Two regions	1-Long cells (radially)	Solid	Papillose cells	Papillose surface
Parthenium hysterophorus	Absent	Three regions	1-Long cells (radially)	Solid	Cylindrical cells	Papillose surface
Praxelis clematidea	Present	Two regions	1-Long cells (radially)	Solid	Papillose cells	Papillose surface
Sigesbeckia orientalis	Absent	Three regions	2-(outer-layer cells long)	Solid	Clavate glandular trichomes	Papillose surface
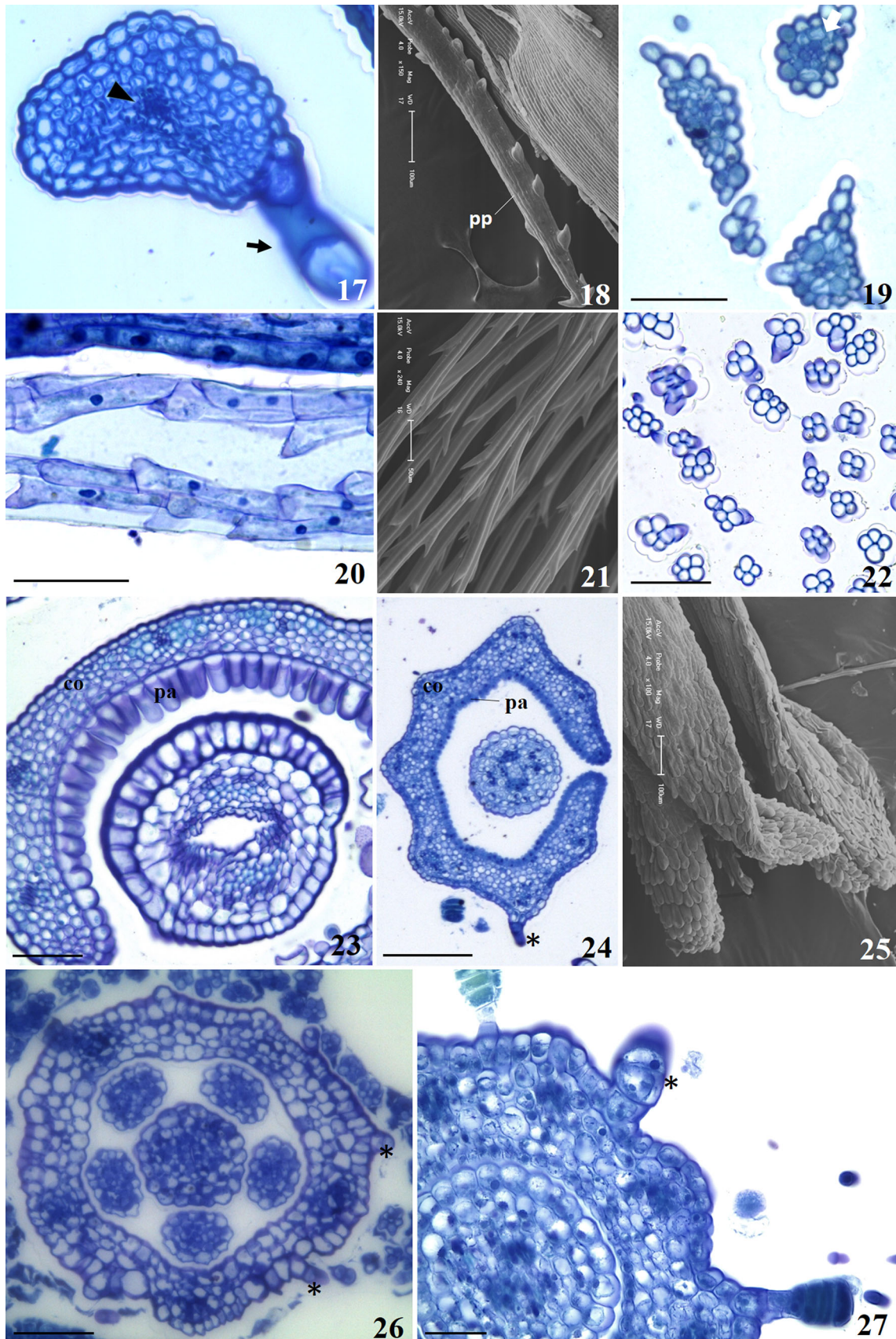
mesophyll; occurring cavities (Fig. 36) in *E. valeriani-folius, E. sonchifolia, C. sulphureus, G. quadriradiata,* and *P. clematidea*; and laticifers in *E. alba*. The corolla vasculature comprises collateral bundles.

The immature anther wall consists of epidermis with elongated and thin-walled cells, uniseriate endothecium, a single middle layer, and tapetum (Fig. 37). The tapetum cells initially remain in their original position, later penetrating into the developing pollen grains (Fig. 38). The connective is composed of parenchyma, a reduced vascular bundle, and an apical appendage (Fig. 39), which shows a V outline (Fig. 40) seen in *C. bonariensis, C. sulphureus, E. alba, E. mollis, G. quadriradiata, P. hysterophorus, P. clematidea,* and *S. orientalis* or shows a half-moon shape (Fig. 41), seen in *E. sonchifolia* and *E. valerianifolius*. The apical appendage is composed of uniseriate epidermis with a thickened cuticle layer, one to three cell layers of parenchyma, and it is devoid of vascular tissue. This appendage has secretory structures (ducts) in *E. alba* and *C. sulphureus* (Fig. 42). The mature anther exhibits epidermis and thickened endothecium (Fig. 39).

The pre-anthesis ovary consists of uniseriate outer epidermis (Fig. 43) with twin hairs, which are lacking in *E. alba, P. hysterophorus,* and *S. orientalis*. The twin hairs are composed of two basal cells and two elongate cells completely united with separated tips (Figs. 44–47). The ovary mesophyll has different tissue regions, which are more differentiated in the ovary of anthetic flowers. The inner epidermis is uniseriate with thin-walled cells (Fig. 43). The two strips of transmitting tissue are especially notable (Figs. 49, 50), one of each stigmatic branch, the strips run longitudinally along the inner surface of the ovary wall.

The anthetic flower ovary undergoes some changes in shape, size, and tissues, when compared with pre-anthetic flowers; more differentiated tissues are distinguishable in the anthetic ovary. The ovaries of the heterogamous capitula are very similar in histological structure. The outer epidermis consists of cells that are mostly tangentially elongated with twin hairs in the pubescent species (Fig. 47). The mesophyll has two or three tissue regions. The two regions of mesophyll in *E. sonchifolia* are composed of an outer layer in sclerification phase and an inner layer composed of parenchyma with intercellular spaces (Fig. 48). In *C. bonariensis, E. valerianifolia,* and *P. clematidea,* these two tissue regions have an outermost layer of mesophyll consisting of radially elongated cells, and the innermost region exhibits 3–4 layers of parenchyma (Fig. 49). The mesophyll of *E. mollis* resembles the other species, although it only consists of parenchyma (Fig. 50).

Most of the species with three regions in the mesophyll, such as *C. sulphureus, E. alba, G. quadriradiata,* and *P. hysterophorus* have an outer mesophyll consisting of a layer with large, radially elongated cells. Besides, there is a

pluriseriate parenchymatous middle mesophyll and an inner mesophyll composed of spongy parenchyma (Fig. 51). *Sigesbeckia orientalis* mesophyll differs, however, from these species with a biseriate outer mesophyll with a subepidermal layer composed of vacuolated, radially elongated cells, and an inner mesophyll partially collapsed (Fig. 52). Anthesis ovarian vasculature varies in the species, consisting of collateral bundles running longitudinally in the middle region and/or ribs of the ovary wall.

Disk floret styles are solid at the base and show a cleft particularly near the apex (Fig. 53). However, in *C. sulphureus,* a stylar canal runs longitudinally along the entire style (Fig. 54). The style consists of glabrous uniseriate epidermis and two or three parenchyma cell layers (Fig. 55), which are star-shaped in *P. clematidea* and *E. valerianifolia* (Fig. 56). Secretory ducts occur in the parenchyma of *C. sulphureus* (Fig. 57)*, P. clematidea* and *G. quadriradiata.* In the style apex, there are two strips of transmitting tissue, which run longitudinally along the canals (clef or stylar canal) (Fig. 56). The style vasculature comprises two bundles. The ray floret style is composed of asymmetric epidermis with radially elongated cells in the face extending back to the ligulate corolla (Fig. 58).

Each stigmatic branch consists of epidermis, parenchyma, one vascular bundle, and two secretory regions (Fig. 59). No glandular epidermis occurs in the outer surface of the stigmatic branch, and it consists of slightly cylindrical cells with a convex periclinal cell wall (*C. sulphureus*) (Fig. 59) and (*P. hysterophorus*), papillose cells [*E. alba, E. mollis, E. sonchifolia,* and *E. valerianifolius* (Fig. 60)], papillose cells and pluricellular trichomes (*G. quadriradiata*) (Fig. 61), and clavate glandular trichomes (*S. orientalis*) (Fig. 62). The parenchyma is composed of elongated cells or star-shaped cells (*P. clematidea* and *E. valerianifolius*). The secretory region contains papillose cells (majority of the species), slightly papillose (*E. alba*) or palisade, and nonpapillose cells (*E. mollis*).

Discussion

The pappus may have an important role in defense and dispersal (Mukherjee and Nordenstam 2008), and as a protective structure, it can protect the corolla and pollen, although not the cypsela (Robinson 1981; Stuessy and Spooner 1988). According to Judd et al. (2002) members of the tribe Heliantheae are characterized by the presence (pappus awns, scales, or bristles) or absence of pappus. The loss of pappus was considered by the same authors as a major example of synapomorphy in the Calenduleae tribe, but there is no reference in the literature notes whether the

Figs. 28–33 Corolla structure of the disk flowers. **28**, **29**, **32**, **33** *Cosmos sulphureus.* **30** *Erechtites valerianifolius.* **31** *Galinsoga quadriradiata.* **28**, **31** Cross section. **30**, **32** Longitudinal sections. **29**, **33** SEM. **28** Corolla with glandular trichomes (*white arrowhead*). **29** Disk flower with glandular trichomes (*white arrowhead*). **30** Corolla with glandular trichomes (*white arrowhead*). **31** Corolla with tector trichomes. **32** Corolla with buds just in the abaxial epidermis (*tip black arrow*). **33** Petals. *Bars* 50 μm (**28**, **30**, **31**, **32**), 100 μm (**29**), 200 μm (**33**). (Color figure online)
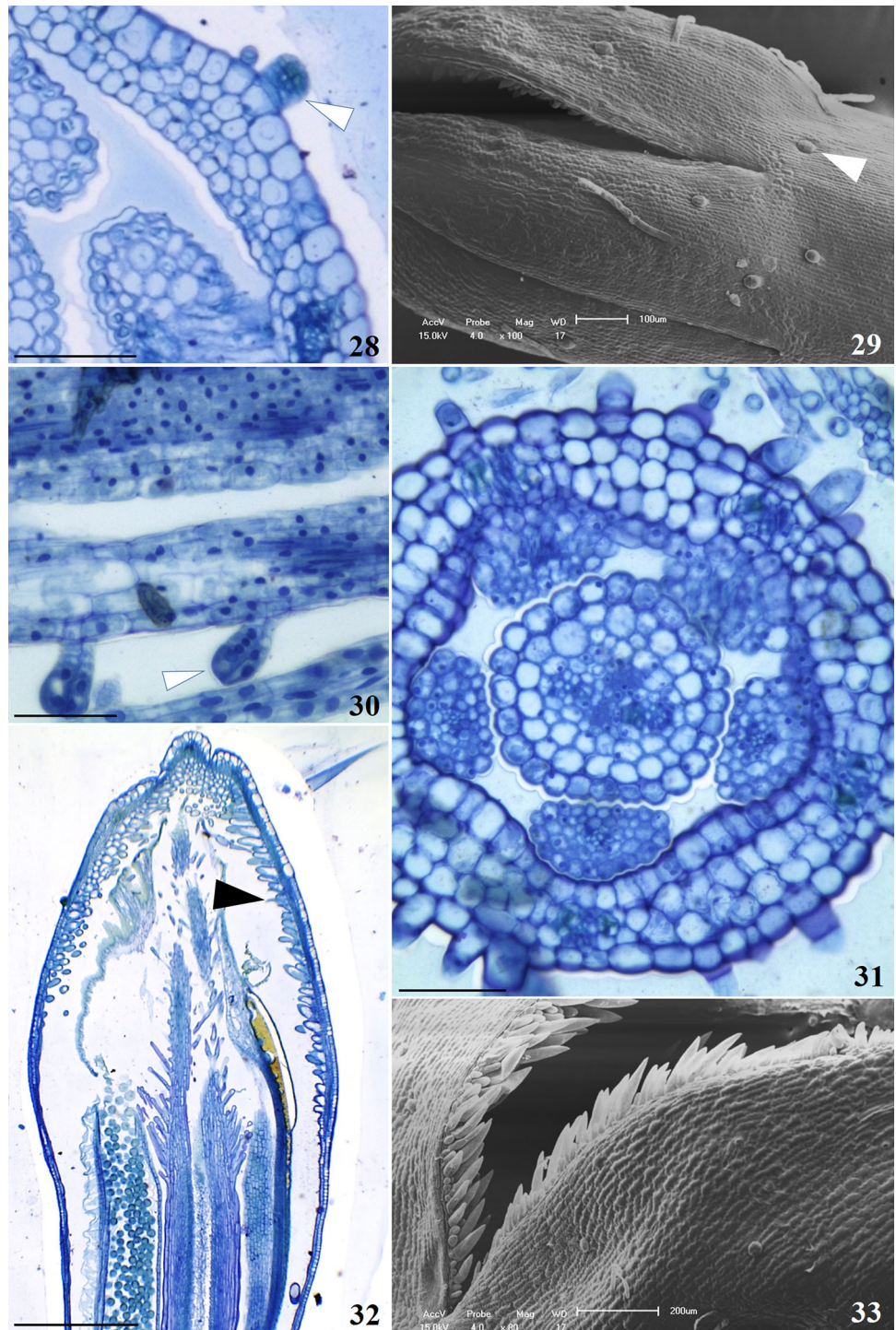

absence of pappus in some members of the "Heliantheae Alliance" clade is also synapomorphic. In *P. hysterophorus* and *S. orientalis,* both of the "Helianteheae Alliance" clade, the pappus is lacking.

The pappus of *E. mollis*, *E. sonchifolia*, and *G. quadriradiata* is devoid of typical vascular cells. Roth (1977) describes the pappus members as reduced and a less differentiated form of foliage leaves. For this author, this stage is reached in the persistent pappus in fruit (cypsela) when the mesophyll consists of sclerenchyma with few rudimentary tracheal elements.

All species investigated here have petal margins of flower buds interlocked by adhesion, a dentonection type which was proposed by Sigmond, according to Weberling (1992). The petal lobes develop from the engaging and interlocking of the pointed papilliform epidermal cells, as described by Weberling (1992).

Figs. 34–42 Structure of the corolla and anthers. **34** *Galinsoga quadriradiata.* **35, 37** *Eclipta alba.* **36, 39, 42** *Cosmos sulphureus.* **38, 41** *Erechtites valerianifolius.* **40** *Parthenium hysterophorus.* **34–41** Cross section. **42** Longitudinal section. **34** Corolla with mesophyll. **35** Corolla without mesophyll. **36** Corolla with secretory cavity in the mesophyll. **37** Anther. **38** Projections of the tapetum (*black arrow tip*). **39** Connective detail. **40** Appendage with V form. **41** Appendage with a half-moon form. **42** Secretory cavity in appendage. *Bars* 50 μm (**34–41**), 100 μm (**36, 42**). (Color figure online)
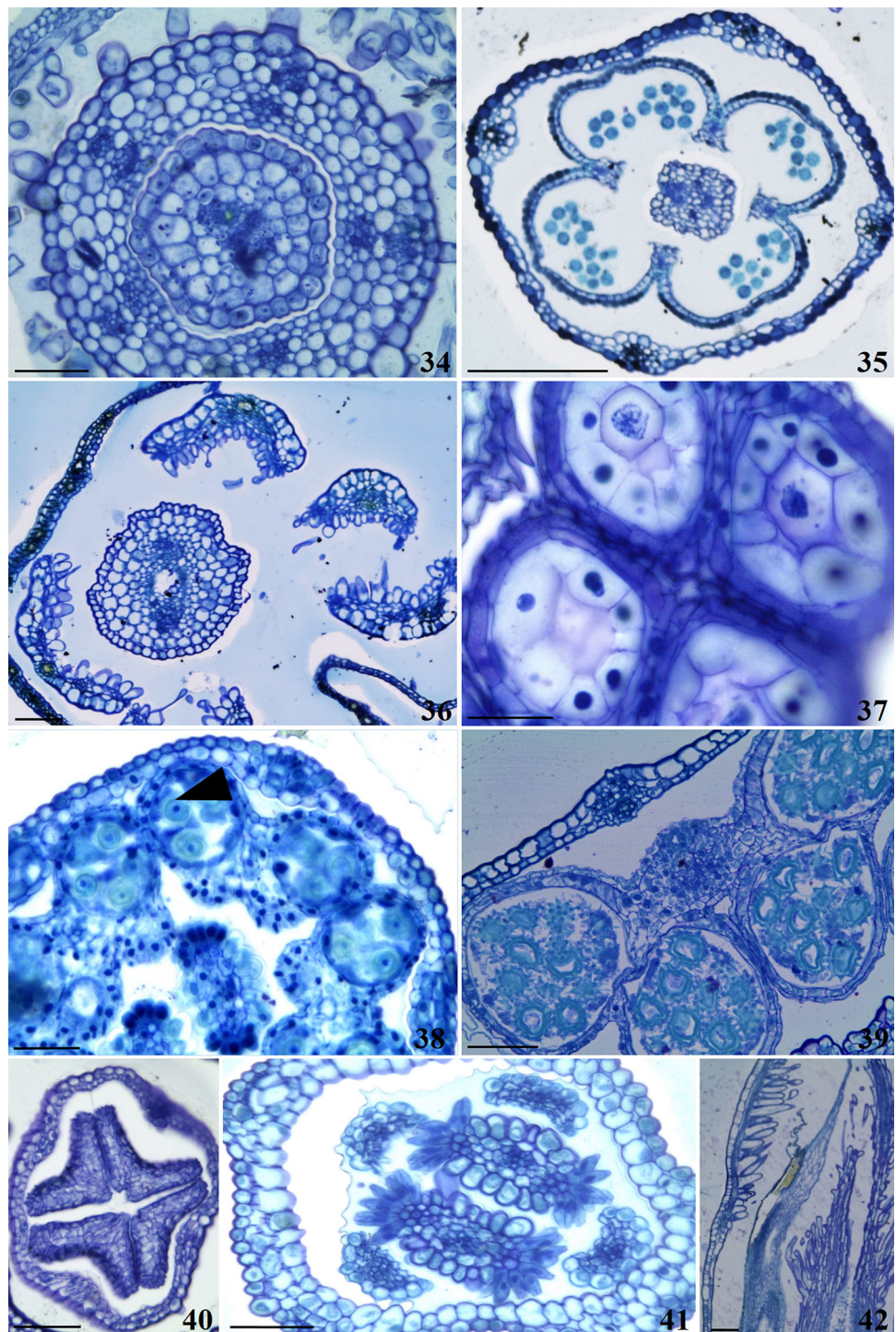

The anther wall of the Asteraceae studied species in pre-anthesis has epidermis, endothecium, a single middle layer, and tapetum. Deng et al. (2010) also find anther wall with epidermis, endothecium, middle layer, and tapetum in *Chrysanthemum multicaule* Desf. Davis (1966) either registers this type of anther wall formation as dicotyledonous or monocotyledonous for eudicotyledons. Besides, this author briefly refers to the dicotyledonous type found in Asteranae, but does not mention Asteraceae. The definitive confirmation of the anther wall formation in the investigated species depends on an ontogenetic study.

All ten Asteraceae species have an ameboid type of tapetum. Dahlgren (1991) considers Asteraceae with mainly or exclusively ameboid tapetum, although the author raises doubts when affirming that in some

Figs. 43–51 Ovary structure. **43** *Conyza bonariensis*. **44, 45, 47, 51** *Cosmos sulphureus*. **46, 49** *Erechtites valerianifolius*. **48** *Emilia sonchifolia*. **50** *Elephantopus mollis*. **43, 48, 49, 50, 51** Cross section. **44–46** Longitudinal section. **47** SEM. **43** Pre-anthetic ovary. **44–47** Twin hair (*black arrow*). **48–50** Anthetic ovaries, details of the vascular tissue (*white arrow*). **51** Anthetic ovary, details of the vascular tissue (*white arrow*). *ee* Outer epidermis, *me* mesophyll, *ei* inner epidermis, *mex* external mesophyll, *mi* internal mesophyll, *mm* average mesophyll, *ov* ovary, *SS* strips of transmitting tissue. *Bars* 50 µm (**43, 46, 48, 49, 50, 51**), 30 µm (**44, 45**). (Color figure online)
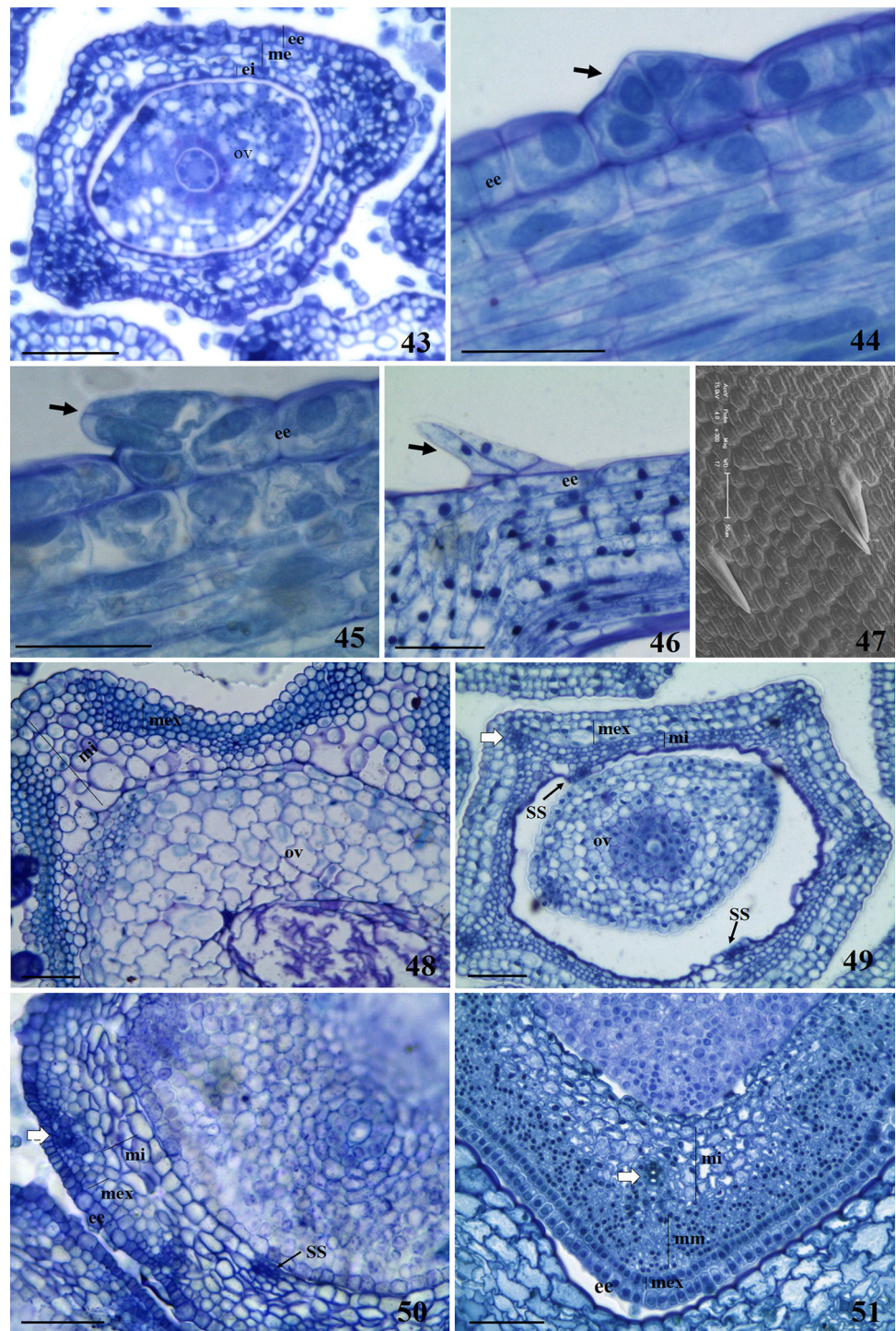
Asteraceae the tapetum is described as ameboid and as periplasmodial in others. For that author, tapetum types present terminological problems, and even if the use of more detailed distinctions could be of further taxonomic importance, it seems too difficult to achieve. The ameboid tapetum was similarly described by Horner (1977) in *H. annuus* and also found in *Ambrosia trifida* L. by Lersten and Curtis (1990).

Douglas (1944) conducted an exhaustive review about the inferior ovary, and she gives a valuable description of it for some Asteraceae species. That author reveals controversy in the interpretation of the inferior ovary nature in Asteraceae: some authors adopted the appendicular theory, whereas others decided that the ovary was axial, based on ontogenetic evidence. In the species studied here, no evidence except for fusion or inversion of the vascular bundles
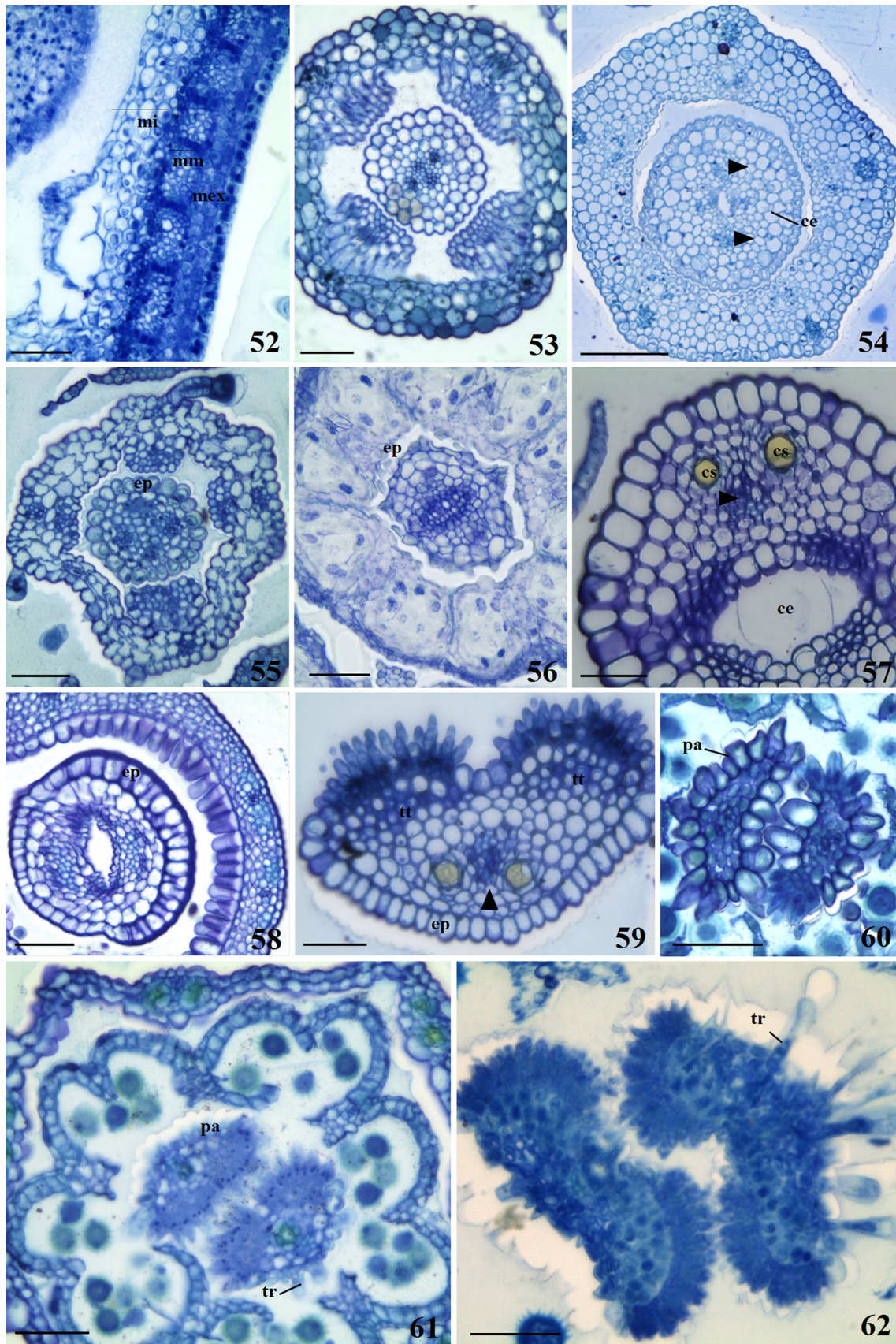
◄**Figs. 52–62** Ovarian structure, style, and stigma. **52, 55, 60** *Erechtites valerianifolius*. **53** *Eclipta alba*. **54, 57, 59** *Cosmos sulphureus*. **56** *Elephantopus mollis*. **58** *Parthenium hysterophorus*. **61** *Galinsoga quadriradiata*. **62** *Siegesbeckia orientalis*. **52–62** Cross section. **52** Ovary. **53** Solid style in the basal portion. **54** Style with detail vascular bundle (*tip black arrow*). **55, 56** Style. **57** Style detail of the secretory ducts and vascular bundle (*tip black arrow*). **58** Style and flower ligule. **59–62** Stigmatic branches with detail of the vascular tissue (*tip black arrow*). *ce* Stylar canal, *cs* secretory cavity, *ep* epidermis, *mex* external mesophyll, *mi* internal mesophyll, *mm* average mesophyll, *pa* buds, *tr* trichomes. *Scale bars* 50 μm (**52, 53, 56, 58–62**); 100 μm (**54**); 200 μm (**57**). (Color figure online)

(Roth 1977) was verified as indicators of the inferior ovary nature.

The ovarian mesophyll of the ten Asteraceae species is composed of two or three tissue regions. Most of the studies with Asteraceae fruit ontogeny describes two histological regions (Pandey and Singh 1980; Galastri and Oliveira 2010; Marzinek and Oliveira 2010; Pallone and Souza 2014) or three regions of tissues (Julio and Oliveira 2009; Pallone and Souza 2014). Compared with most eudicots, in which the ovary has a relatively simple structure, Asteraceae ovaries have a complex structure,

probably due to the difficulty in distinguishing carpellary from receptacular and/or appendicular tissue.

According to Katinas et al. (2016) the stigmatic surface and style are keys to reproduction: they represent the morphological basis of different breeding systems, are critical for optimum pollen capture, and frequently well defined features of both taxonomic and phylogenetic value. The stigmatic surfaces of many tribes are consistently continuous over the inner surface of the style branch (Mutisieae, Lactuceae, Vernonieae, Arctotideae, Eremothamneae, Cardueae); however, in other tribes, the stigmatic surface is divided into two marginal lines (Eupatorieae, Anthemideae, Astereae, most Inuleae, most Heliantheae, and most Senecioneae) (Robinson 2009 in Funk et al. 2009). The studied species have fertile areas or separate stigmatic in two marginal lines in all species, except *E. mollis* (Vernonieae) with the surface irregularly covered.

The style is hollow and with stylar canal only in *C. sulphureus*; all the other species have solid style and relatively uniform transmitting tissue. The hollow style was also registered in the disk florets of *Xeranthemum squarrosum* Boiss using epi-illumination microscopy (Dadpour et al. 2012), and in the disk and ray florets of *C. multicaule*

Table 5 Some important floral features for tribes' classification in Asteraceae

Tribes	Species	Pappus (shape/vascular elements)	Corrolla appex	Anther apical appendage (shape)	Abaxial epidermis (stigma)	Twin hair (ovary)	Mesophyll regions (ovary)
Astereae (Asteroideae)	*Conyza bonariensis*	Round/vasc.	Tector (ray), glabrous (disk)	V	Cylindrical cells	Present	Two regions
Senecioneae (Asteroideae)	*Emilia sonchilolia*	Round/without vasc.	Glabrous	Half-moon	Papillose cells	Present	Two regions
	Erechtites valerianifolius	Round/without vasc.	Glabrous (ray), glandular (disk)	Half-moon	Papillose cells	Present	Two regions
Heliantheae ("Helliantheae Alliance")	*Eclipta alba*	Without pappus	Glabrous (ray and disk)	V	Papillose cells	Absent	Three regions
	Parthenium hysterophorus	Without pappus	Glandular (ray), tector and glabrous (disk)	V	Cylindrical cells	Absent	Three regions
Coreopsideae ("Helliantheae Alliance")	*Cosmos sulphureus*	Round/vasc.	Glandular (ray and disk)	V	Cylindrical cells	Present	Three regions
Millerieae ("Helliantheae Alliance")	*Galinsoga. quadriradiata*	Round/without vasc.	Tector (ray and disk)	V	Papillae and trichomes	Present	Three regions
	Sigesbeckia orientallis	Without pappus	Glandular (ray), Tector and glandular (disk)glabrous (disk)	V	Clavate glandular trichomes	Absent	Three regions
Vernonieae (Cichorioideae)	*Elephantopus mollis*	Round/without vasc.	Glabrous	V	Papillose cells	Present	Two regions
Eupatorieae ("Helliantheae Alliance")	*Praxelis clematidea*	Round/vasc.	Glabrous	V	Papillose cells	Present	Two regions

(Deng et al. 2010). According to Endress (1994), a gynoecium with many ovules tends to have a several-layered pollen tube transmitting tract, and a gynoecium with a single ovule, which is the Asteraceae case, usually has a single-layered tract. In fact, this seems to be the case of the Asteraceae species investigated here, although it seems appropriate to emphasize the presence in these species of a subepidermal tissue with secretory appearance. In the classification of the pollen tube transmitting tracts suggested by Endress (1994), in which the Asteraceae species have one cell layer transmitting tissue, *C. sulphureus* is distinguished from other species, mainly from other Heliantheae species, by its stylar canal.

The Asteraceae ducts attract the researchers interest due to their form and mainly for their probable content: latex, resins or acetylenes. In Asteraceae, the latex is usually associated with ducts, and their presence was detected in Lactuceae, Liabeae (Robinson 1983) and Vernonieae (Lewinsohn 1991). The presence of associated latex with ducts is still a seemingly restricted character to Cichorioideae (Funk et al. 2009). Ducts were found in *C. sulphureus* (Coreopsideae), *P. clematidea* (Eupatorieae), and *G. quadriradiata* (Millerieae), but the exudate composition was not investigated.

The flower features that are potentially significant in the characterization and separation of Asteraceae species are summarized in Tables 2, 3, and 4. In general, the study shows the usefulness of the floral characters to distinguish species. Morphological features that are taxonomically important at tribal level and that had already been widely revised by Bremer (1994), Judd et al. (2002) and Funk et al. (2009) were confirmed for the flowers of the Asteraceae analyzed here. Judd et al.'s (2002) morphological features of tribes were registered in the studied species (Table 5). Here, it has been noted that *P. clematidea* is the only species of the clade "Heliantheae Alliance" devoid of three regions in the ovary wall. In addition, some species belonging to the same clade (*E. alba*, *P. hysterophorus,* and *S. orientalis*) have no pappus and twin hairs. These characters can be useful to separate the species, although they can probably be critical for understanding the morphology and anatomy of the clade and its diversity.

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