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Review

Floral glands in asclepiads: structure, diversity and evolution

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ABSTRACT

Species of Apocynaceae stand out among angiosperms in having very complex flowers, especially those of asclepiads, which belong to the most derived subfamily (Asclepiadoideae). These flowers are known to represent the highest degree of floral synorganization of the eudicots, and are comparable only to orchids. This morphological complexity may also be understood by observing their glands. Asclepiads have several protective and nuptial secretory structures. Their highly specific and specialized pollination systems are associated with the great diversity of glands found in their flowers. This review gathers data regarding all types of floral glands described for asclepiads and adds three new types (glandular trichome, secretory idioblast and obturator), for a total of 13 types of glands. Some of the species reported here may have dozens of glands of up to 11 types on a single flower, corresponding to the largest diversity of glands recorded to date for a single structure.

Keywords: anatomy, Apocynaceae, Asclepiadoideae, diversity, evolution, flower, secretory structures

Introduction

Apocynaceae is an extremely diverse family in morphological terms, represented by trees, shrubs, herbs and climbers, with single leaves usually opposite, rarely alternate or whorled, with stipules modified in colleters in several species (Endress & Bruyns 2000; Capelli *et al*. 2017) and with various secretory structures in vegetative and reproductive organs of recognized importance in taxonomy, phylogeny and/or ecology (Thomas & Dave 1991; Demarco 2008). Due to their highly elaborate flowers, the family stands out among the eudicotyledons, especially when considering its most derived subfamily Asclepiadoideae.

The close relationship between the former families Apocynaceae and Asclepiadaceae has always been recognized since its establishment as "Apocineae" by Jussieu (1789). Although Brown (1810) divided it into two families and this separation had been maintained in the subsequent taxonomic studies until recently (Cronquist 1981), many researchers have found a gradation in the morphology of the complex reproductive organs between the two families. Phylogenetic studies carried out mainly during the 1990s have shown that the two families form a monophyletic group, thus constituting a single family (Judd *et al*. 1994;

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Struwe *et al*. 1994; Endress *et al*. 1996; Sennblad & Bremer 1996; 2002). As a result, Endress & Bruyns (2000) proposed a new classification for Apocynaceae *s.l*., including Asclepiadaceae, based mainly on morphological evidence, and grouped the current 366 genera (Endress *et al*. 2014) into five subfamilies: Rauvolfioideae (=Plumerioideae), Apocynoideae, Periplocoideae, Secamonoideae and Asclepiadoideae.

The members of Asclepiadoideae, also known as asclepiads, are recognized for having the most complex and elaborate flowers of all eudicots (Endress 1994; 2016). Some characteristics are so distinct from the most basal Apocynaceae that only with the joint evaluation of the other subfamilies is it possible to understand how asclepiads reached this degree of complexity (Endress & Bruyns 2000). They have an unusual flower synorganization that led to the origin of new organs. From the corolla and the androecium, the corona and a complicated system of channels for secondary presentation of nectar evolved. From the androecium and gynoecium, the gynostegium was formed through postgenital adnation of the anther to the base of the style head, and the pollinarium was formed by the pollinia plus the translator, which is secreted by the epidermis of the style head (Endress 1994).

Asclepiadoideae consist of ca. 3000 species (Rapini 2012) occurring in diverse areas ranging from deserts and open vegetation to swamp and shaded areas in tropical and subtropical regions and with centers of diversity in Africa (about 35% of species) and South America (about 20% of species), becoming less diverse and abundant in temperate regions (Rapini 2000). The members of this subfamily can be distinguished from other Apocynaceae by the presence of pollinaria carrying only two pollinia (Kunze 1994; Swarupanandan *et al*. 1996; Endress & Bruyns 2000; Demarco 2014).

Asclepiads have received the attention of researchers for centuries because of their complex pollination system, but few anatomical studies have attempted to unravel the complex floral morphology of the group. The first comprehensive anatomical studies were made by Brown (1810), who described the formation of the translator in *Asclepias syriaca* L., and Corry (1883), Gager (1902) and Frye (1902), all of which described many flower characteristics of *Asclepias* species, especially the gynostegium. Despite the great diversity of species and the long period of study, there is little information available on the anatomy of the species of this group.

The complex floral pollination mechanism of Asclepiadoideae is only comparable to orchids (Endress 2016). These two families present a series of evolutionary convergences that allowed the production and dispersion of pollen aggregate into pollinia. Apparently, a high degree of synorganization of the floral organs seems to have been necessary to allow the evolution of pollinia. In Apocynaceae, the presence of corona has greatly increased the morphological complexity of the flowers (Fig. 1). In addition, the highly complex pollination mechanism seems to have influenced mainly the diversity of clades bearing pollinia in Apocynaceae and Orchidaceae since these clades represent more than half of the species of both families (Endress 2016). The relation of at least some glands with pollination resulted in a large diversity of floral secretory structures and, theoretically, the greater the complexity and/or the specificity of the pollination mechanism, the greater the number of glands that provide this interaction.

Secretory structures

Among the anatomical characters reported for Apocynaceae, only three are present in Asclepiadoideae and all other members of the family: amphiphloic siphonostele, laticifers and style head. Of these three, the latter two are secretory, and Metcalfe & Chalk (1950) considered the occurrence of laticifers as one of the most important characteristics demonstrating the close relationship between the former Apocynaceae and Asclepiadaceae. In addition, one of the diagnostic features of the family is the style head, which has a secretory epidermis (Judd *et al*. 2002).

The floral secretory structures found in this group are extremely diverse and distinguish asclepiads as the group with the largest number of glands in a single flower among the angiosperms, which is related to a large extent to the complex reproductive system of this group. The glands reported up to now added to those described in this review are the following: colleters, glandular trichomes, laticifers, secretory idioblasts, nectaries (primary and secondary), osmophores, style head, tapetum, staminal wing gland, extragynoecial compitum, stylar canal and obturator. These structures are detailed later.

Floral glands in asclepiads

The secretory structures of asclepiads occur in vegetative and/or reproductive organs and are involved in the production of different compounds of the secondary metabolism. They may be classified as protective glands, which play a defensive function, or nuptial glands, associated with pollination.

The protective function is performed by external and internal glands of the flowers, which are also frequently found in the stem and/or leaves; the defensive function is also necessary for vegetative organs. On the other hand, the nuptial glands of asclepiads are exclusive to flowers and serve to attract or provide nutritional resources for the pollinator. In some cases, they are also related to pollen removal and/or pollen adhesion to the stigma, as a stimulus for pollen germination, a guide and nourisher for pollen tubes, etc. All these functions and others are found in the flowers of asclepiads.

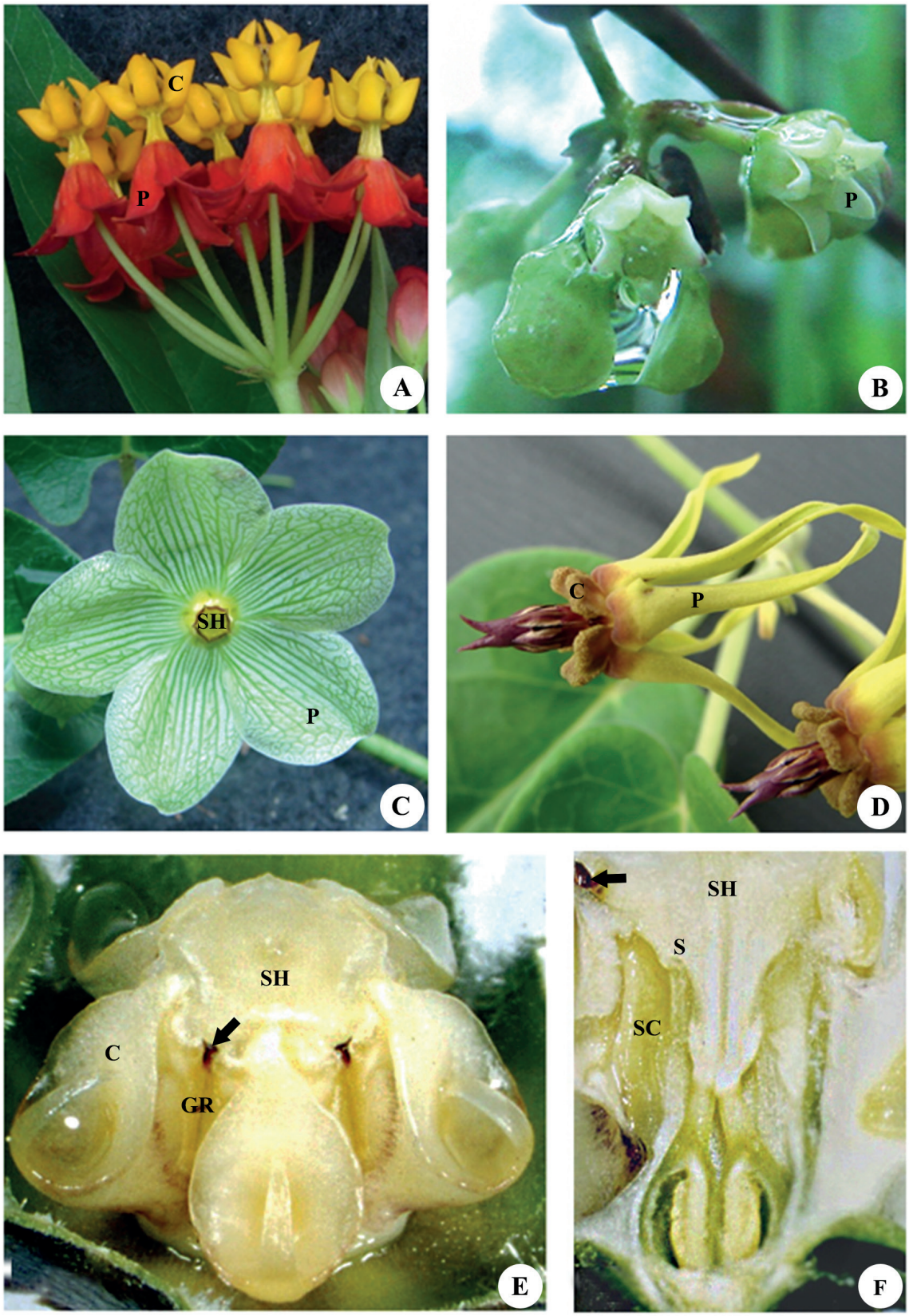

Figure 1. Flowers of Asclepiadoideae. (A) *Asclepias curassavica* L. (B) *Peplonia axillaris* (Vell.) Fontella & Rapini. (C) *Matelea denticulata* (Vahl) Fontella & E.A. Schwarz. (D) *Oxypetalum banksii* subsp. *banksii* Roem. & Schult. (E-F) *Blepharodon bicuspidatum* E. Fourn. (F) Longitudinal section of the flower. Abbreviations: C, corona; GR, guide rail; P, petal; S, stigma; SC, stigmatic chamber; SH, style head; arrow, translator.

External protective glands

Colleters (Fig. 2)

Colleters are widespread in Apocynaceae (Endress & Bruyns 2000) and occur in flowers of all Asclepiadoideae. In this family, they are calycine emergences (Fig. 2) that produce a viscous secretion which protects the meristems against desiccation (Thomas 1991) and can also protect the flowers against fungal proliferation (Ribeiro *et al*. 2017).

The position of colleters may be variable (Woodson & Moore 1938), but the asclepiads have colleters alternating with the sepals (Frye 1902; Rao & Ganguli 1963; Tiagi & Dixit 1965; Valente *et al*. 1973; Silva *et al*. 1975; Valente 1983; 1984; 1995; Pereira & Schwarz 1983; Endress & Bruyns 2000; Valente & Costa 2005; Demarco 2008), except in *Oxystelma esculentum* R.Br., which have opposite colleters (Rao & Ganguli 1963). Although alternisepalous colleters have been considered a plesiomorphic feature in Apocynaceae (Woodson & Moore 1938), they occur in almost all members of Asclepiadoideae, which is the most derived subfamily.

Morphologically, the calycine colleters are much more constant than those in the leaf (Demarco 2005). In general, they are classified as the standard type in the family (Thomas 1991), being cylindrical or dorso-ventrally flattened, persistent (Thomas *et al*. 1989; Thomas & Dave 1989a; b; c; 1991; Thomas 1991; Appezzato-da-Glória & Estelita 2000; Schwarz & Furlan 2002; Demarco 2005; Simões *et al*. 2006; Martins *et al*. 2010; Martins 2012) (Fig. 2C), and may be found at the base of fruits (Thomas 1991; Thomas & Dave 1991; 1994). The most frequent variations observed are the presence or absence of peduncles (Fig. 2D) and the number of colleters per flower (Rao & Ganguli 1963; Ramayya & Bahadur 1968; Silva *et al*. 1975; Stevens 1975; 1988; Pereira & Schwarz 1983; Thomas & Dave 1989a; Schwarz & Furlan 2002; Demarco 2005; Rio *et al*. 2005; Simões *et al*. 2006; Martins *et al*. 2010; Martins 2012). Colleters have taxonomic significance for the family (Woodson & Moore 1938; Thomas 1991; Simões *et al*. 2006) and their occurrence, type and/or position have been used as diagnostic characters in identification keys at the genus and species level (Barroso 1986; Rio & Kinoshita 2005; Rio *et al*. 2005).

Colleters are formed early in the ontogeny of sepals, originating from the adaxial side of the connate portion of the calyx (Fig. 2A-B), just below the sinus. Immediately after their formation in the floral meristem, the colleters begin secreting. The secretory portion is composed of a uniseriate palisade epidermis covering a non-secretory parenchyma (Demarco 2005; 2008) (Fig. 2C-D). More than one layer of secretory epidermis has been observed in a few species of other subfamilies of Apocynaceae (Ramayya & Bahadur 1968; Thomas *et al*. 1989). Secretory cells have dense cytoplasm, and the secretion is accumulated in a periplasmic space before it is released to the outside through the cell wall and cuticle (Ribeiro *et al*. 2017). According to Fahn (1990), secretion release in colleters usually occurs due to cuticle rupture, but this was not observed in my study nor in several species recently investigated (Appezzato-da-Glória & Estelita 2000; Rio *et al*. 2002; Demarco 2005; 2008; Simões *et al*. 2006; Martins *et al*. 2010; Martins 2012; Canaveze & Machado 2015).

Calycine colleters are always avascularized in asclepiads (Woodson & Moore 1938), but vascularized colleters have already been recorded in flowers of other subfamilies (Woodson & Moore 1938; Rao & Ganguli 1963; Dave *et al*. 1987; Thomas & Dave 1989c) and crystalliferous idioblasts and laticifers are often found in many species (Ramayya & Bahadur 1968; Arekal & Ramakrishna 1980; Fjell 1983; Murugan & Inamdar 1987a; b; Thomas & Dave 1989a; b; Subramanian *et al*. 1989; Thomas *et al*. 1989; Thomas & Dave 1991; Appezzato-da-Glória & Estelita 1997; 2000; Schwarz & Furlan 2002; Demarco 2005; 2008; Martins *et al*. 2010).

Calycine colleters remain in secretory activity during the entire floral development and maintain their shape during the post-secretory phase in post-anthetic flowers, unlike the leaf colleters (Demarco 2005; 2008). Among the asclepiads analyzed histochemically to date, the production of a heterogeneous secretion composed of mucilage and lipidic compounds seems to be predominant (Fig. 2E-H), with the occurrence of exclusively mucilaginous secretion found only in *Peplonia* (Ribeiro *et al*. 2017). Proteins, phenolic compounds and fatty acids have been detected in the secretion of calycine colleters, as well as several alkanes. The distinct components of secretion confer different functions to the colleters. While the mucilage protects against desiccation, the lipophilic compounds provide an antifungal property (Ribeiro *et al*. 2017).

Glandular trichomes (Fig. 3)

Glandular trichomes have a restricted occurrence in Apocynaceae and have been reported for only eight genera of Asclepiadoideae: *Araujia*, *Dischidia*, *Fischeria*, *Gongronema*, *Gonolobus*, *Marsdenia*, *Matelea* and *Sarcostemma* (Solereder 1908; Woodson 1941; Metcalfe & Chalk 1950; Stevens 1975; 1988; Murphy 1986; Morillo 1998). Among these genera, the presence of mixed indumentum composed of long tector trichomes and short glandular trichomes in *Fischeria* and *Matelea* is unique and shows the relation of these genera (Woodson 1941), both grouped in the subtribe Gonolobinae (Endress *et al*. 2014).

These glandular trichomes have never been studied anatomically and are described for the first time in the present work for *Matelea denticulata*. In this genus, glandular trichomes are present on the pedicel and abaxial side of the sepals (Stevens 1975; 1988) (Fig. 3A-B). In *M. denticulata*, they are multicellular, uniseriate with lignified peduncle (Fig. 3C-D) and an apical secretory cell with a dilated base

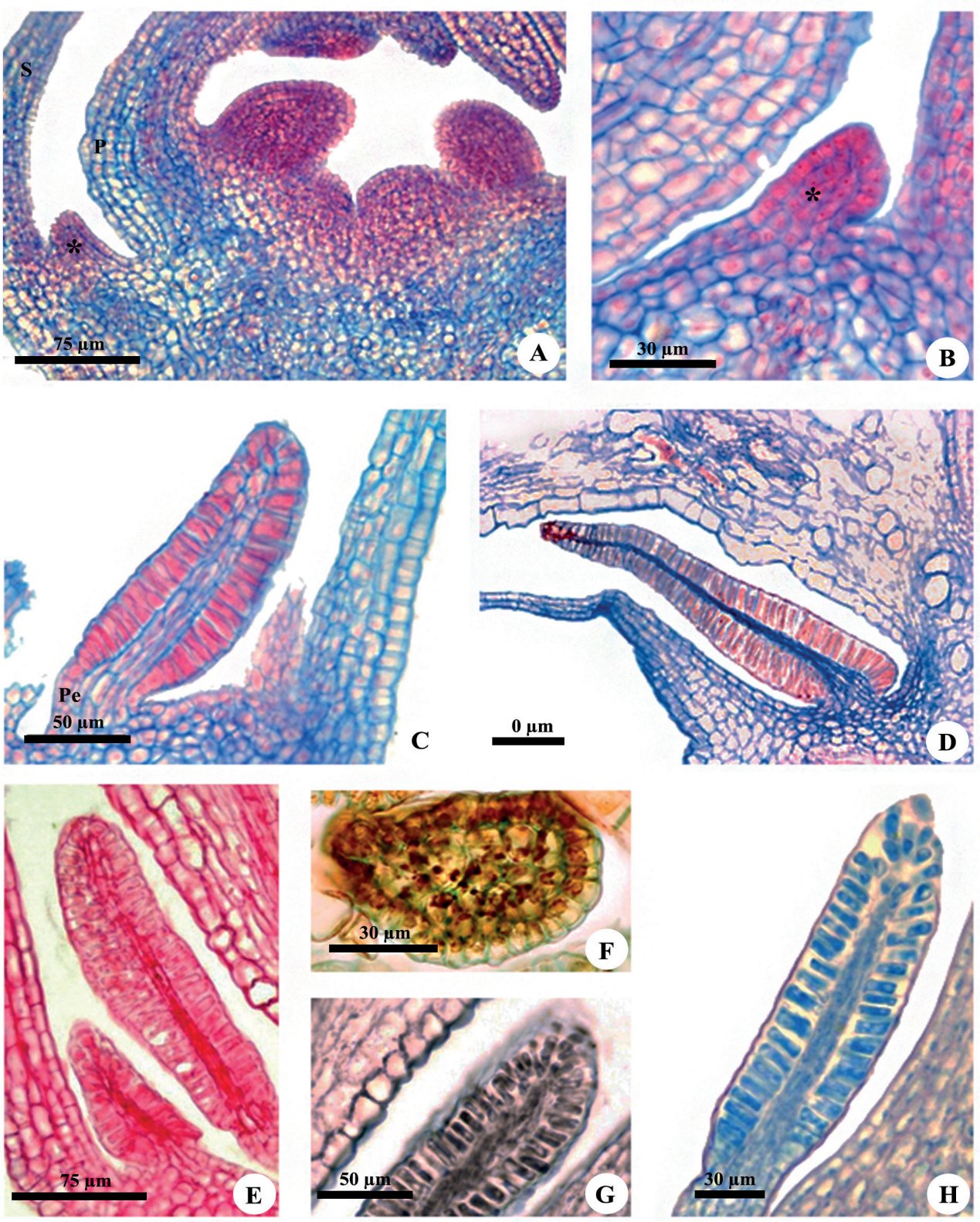

Figure 2. Calycine colleters in Asclepiadoideae. (A) *Matelea denticulata* (Vahl) Fontella & E.A. Schwarz. (B, F) *Oxypetalum banksii* subsp. *banksii* Roem. & Schult. (C, E, G-H) *Asclepias curassavica* L. (D) *Blepharodon bicuspidatum* E. Fourn. (A-B) Colleter initiation in floral buds (asterisk). (C-D) Mature colleters formed by palisade secretory epidermis and a parenchyma axis. (C) Colleter with peduncle (standard type). (D) Sessile colleter. (E) Detection of acidic mucilage with ruthenium red. (F) Identification of starch grains using safranin, astra blue and iodine-potassium iodide. (G-H) Lipids detected with Sudan black B (G) and Nile blue (H). Abbreviations: P, petal; Pe, peduncle; S, sepal.

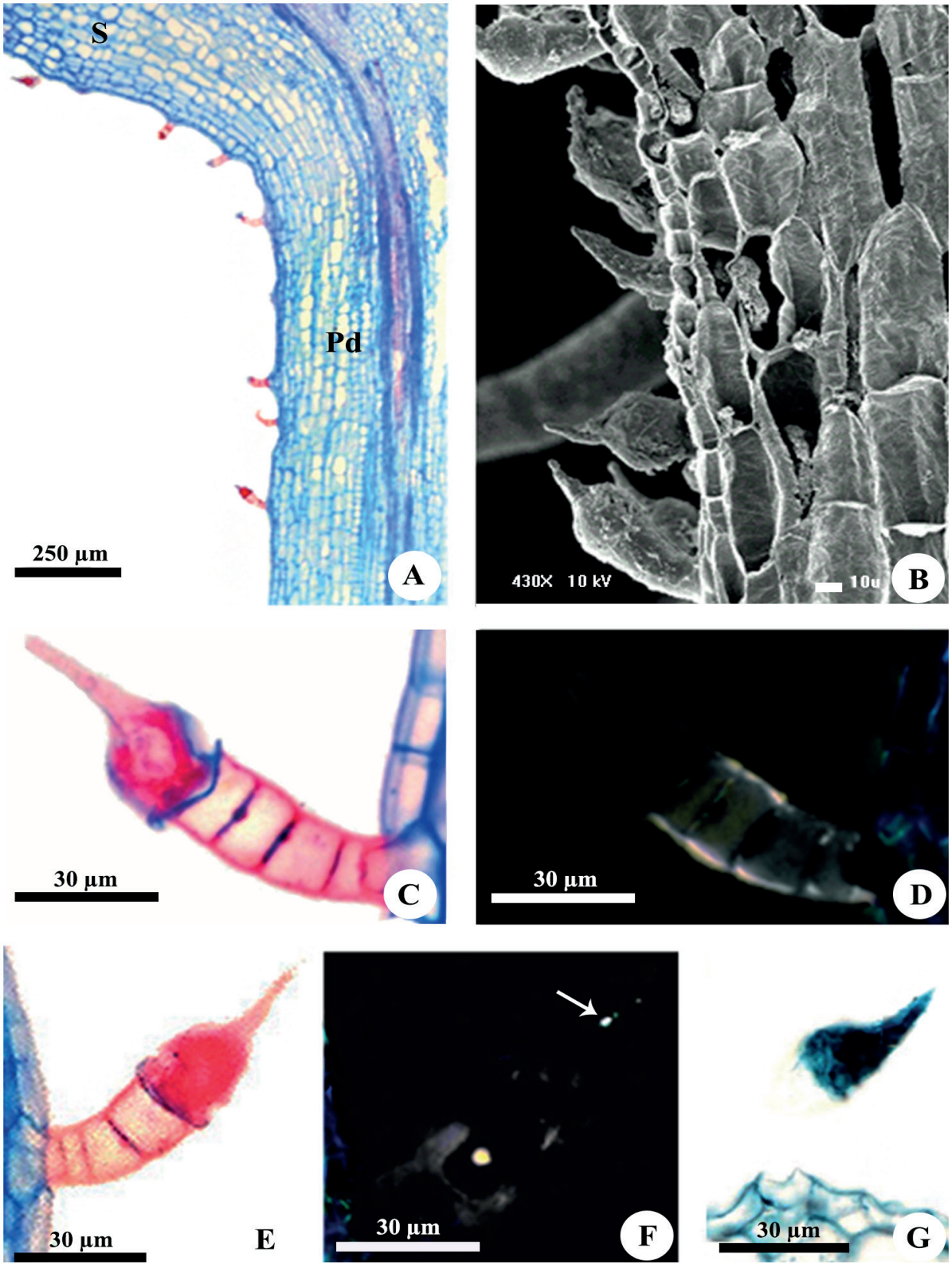

Figure 3. Glandular trichomes in flowers of *Matelea denticulata* (Vahl) Fontella & E.A. Schwarz. (A-B) General view of the glandular trichomes in light microscopy (A) and scanning electron microscopy (B). (C, E) Mature trichomes. (D, F) Identification of secondary cell walls. Polarization microscopy of 3C and 3E respectively. (D) Secondary walls in the peduncle cells. (F) Crystal in the apex of the glandular cell (arrow). (G) Detection of proteins with aniline blue black. Abbreviations: Pd, pedicel; S, sepal.

and an elongated, acuminate upper portion (Fig. 3C, E). This cell has a rounded tip with a constriction just below it where crystals are located, providing a mechanical rupture (Fig. 3F). The secretion is composed exclusively of amino acids and/or proteins (Fig. 3G). The morphology of the trichome, composition of the secretion and its mechanism of release to the outside resemble those of stinging trichomes (Thurston & Lersten 1969; Thurston 1974; 1976; Fahn 1979).

Internal protective glands

Laticifer (Fig. 4)

Laticifers are ubiquitous in Apocynaceae (Metcalfe & Chalk 1950) and are found in all vegetative and floral organs of asclepiads, absent only in the ovules (Demarco *et al*. 2006). Although those laticifers are generally interpreted as non-articulated type in the family (Chauveaud 1891; Solereder 1908; Metcalfe 1967; Mahlberg 1993), recent developmental studies of laticifers indicate that possibly all the vegetative and floral laticifers of Apocynaceae are articulated anastomosing (Fig. 4A-C) with early dissolution of the terminal walls, a fact that led many authors to misclassify them (Demarco *et al*. 2006; Demarco & Castro 2008; Gama *et al*. 2017, and references therein).

Laticifers branch by lateral fusion in the meristematic regions, forming a system that likely interconnects most laticifers of the adult plant (Demarco *et al*. 2006; Demarco & Castro 2008; Lopes *et al*. 2009; Canaveze & Machado 2016; Gama *et al*. 2017) (Fig. 4D). Cell walls are dissolved from the center to periphery, followed by the fusion of protoplasts, resulting in a continuous multinucleated protoplast throughout the laticifer system (Gama *et al*. 2017). They are found in the fundamental and vascular systems of all organs (Groom 1889; Blaser 1945; Milanez 1960/1961; 1966; 1977; Mahlberg 1963; Valente 1977; 1984; 1995; 1996; Murugan & Inamdar 1987a; b; Appezzato-da-Glória & Estelita 1997; Sacchetti *et al*. 1999; Valente & Costa 2005; Demarco *et al*. 2006; Demarco & Castro 2008) (Fig. 4E-F) and ultrastructural analyses have demonstrated the impossibility of intrusive growth of these laticifers (Gama *et al*. 2017).

The laticifer cell walls are exclusively primary and highly hydrated, especially in the young portion, where their acidic characteristic (Fig. 4G) makes them more flexible, allowing the increase of cell diameter (Demarco *et al*. 2006). Immunocytochemical studies of laticifers in *Asclepias speciosa* Torr.(Serpe *et al*. 2001; 2002) have shown that the pectin composition of the cell wall in the mature portions of the laticifers is different from that of the younger portions.

Latex is observed from the younger region of the laticifer and corresponds to its protoplast (Demarco 2015). Some vesicles and small vacuoles with secretion fuse to the central vacuole, transferring their contents and increasing its volume, restricting the cytoplasm to a thin parietal layer (Gama *et al*. 2017). According to Giordani (1978), Fahn (1979) and Fineran (1983), the protoplast can remain intact or degenerate at maturity. However, the protoplast disarrangement is apparently due to an artifact during the plant collection and fixation caused by the destabilization of the turgor pressure, modifying all laticifer content.

The latex of Apocynaceae may have different colors (Solereder 1908), but the few latices described for flowers to date have all been milky-white (Appezzato-da-Glória & Estelita 1997; Demarco *et al*. 2006; Demarco & Castro 2008; Demarco 2015). While the latex is generally described as having predominantly lipids (Fig. 4H-I), especially terpenes (Die 1955; Warnaar 1982; Giordani 1996), many other compounds have been detected in the latex of the family, such as triterpenes and polyisoprenes, steroids, fatty and aromatic acids, polysaccharides (Fig. 4J), cardenolides and proteins (Fig. 4K), including enzymes, phenolic compounds and alkaloids (Die 1955; Rao & Malaviya 1966; Wilson *et al*. 1976; Yoder & Mahlberg 1976; Baas *et al*. 1981; Groeneveld & Made 1982; Warnaar 1982; Allen & Nessler 1984; Eilert *et al*. 1985; Murugan & Inamdar 1987b; Giordani & Lafon 1993; Giordani 1996; Appezzato-da-Glória & Estelita 1997; Sacchetti *et al*. 1999; Giordani *et al*. 2000; Castro & Demarco 2008; Demarco 2015). The various compounds protect the plant against herbivores and microorganisms as well as seal wounds (Fahn 1979; 1990; Farrel *et al*. 1991; Hunter 1994; Demarco 2015).

Secretory idioblasts (Fig. 5)

There have been few reports of secretory idioblasts in Apocynaceae, and almost all are restricted to vegetative organs (Solereder 1908; Metcalfe & Chalk 1950; Baas & Gregory 1985; Endress & Bruyns 2000; Demarco 2005). In Asclepiadoideae, secretory idioblasts have been reported for the tribes Ceropegieae and Asclepiadeae (Solereder 1908; Metcalfe & Chalk 1950; Endress & Bruyns 2000), but their presence varies, even in the same subtribe. All of these reports referring to vegetative organs and floral secretory idioblasts are described in asclepiads for the first time in this review.

Oil idioblasts have been identified in flowers of *Peplonia axillaris* (Fig. 5A). The production of oil by idioblasts has been reported for 12 genera of Apocynaceae, but none of them belongs to the subfamily Asclepiadoideae (Metcalfe & Chalk 1950). The idioblasts of *P. axillaris* occupy the most outer region of the pedicel cortex (Fig. 5B) and are found beneath the epidermis of sepals (Fig. 5C) and petals (Fig. 5D-E). Their shape varies from cubic to elongated and have trilamellar walls with a median suberin lamella between two cellulosic portions of the cell wall (Fig. 5F-G), as is normally observed in oil idioblasts (Postek & Tucker 1983) with the oil occurring as droplets in the periphery of the vacuole (Fig. 5H-I).

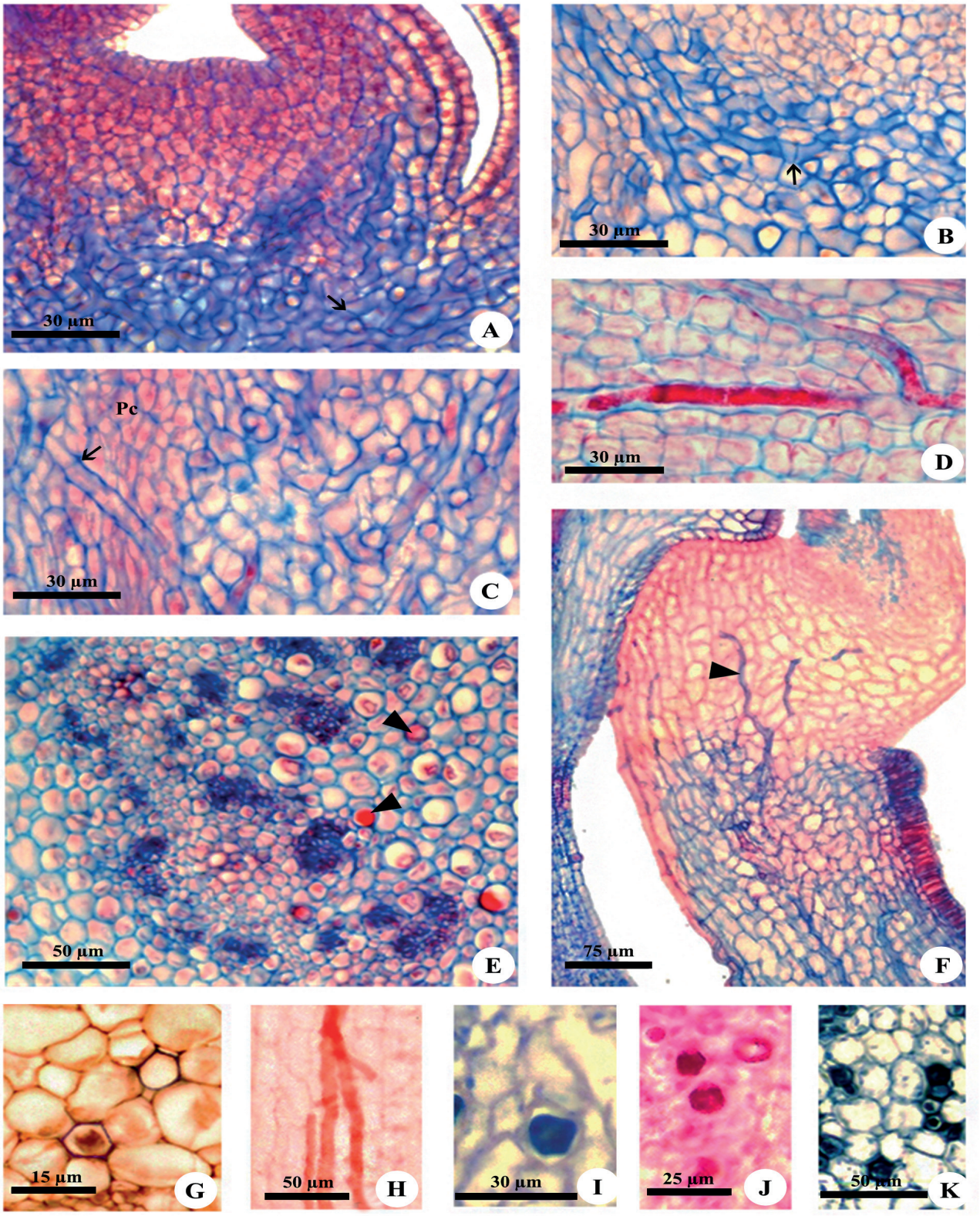

Figure 4. Articulated anastomosing laticifers in flowers of Asclepiadoideae. (A, I-J) *Matelea denticulata* (Vahl) Fontella & E.A. Schwarz. (B, F, K) *Peplonia axillaris* (Vell.) Fontella & Rapini. (C, G) *Asclepias curassavica* L. (D-E, H) *Oxypetalum banksii* subsp. *banksii* Roem. & Schult. (A-C) Origin of laticifers in the floral buds. (D) Branched mature laticifer. (E) Laticifers in the pedicel. (F) Laticifers in the stamen. (G) Acidic character of the laticifer walls (violet) detected with triple Flemming's staining. (H-K) Histochemical identification of latex components. (H-I) Identification of lipids using Sudan IV (H) and Nile blue (I). (J) Polysaccharides identified with PAS reaction. (K) Proteins stained with aniline blue black. Abbreviations: Pc, procambium; arrow, terminal wall of the laticifer cells; arrowhead, laticifer.

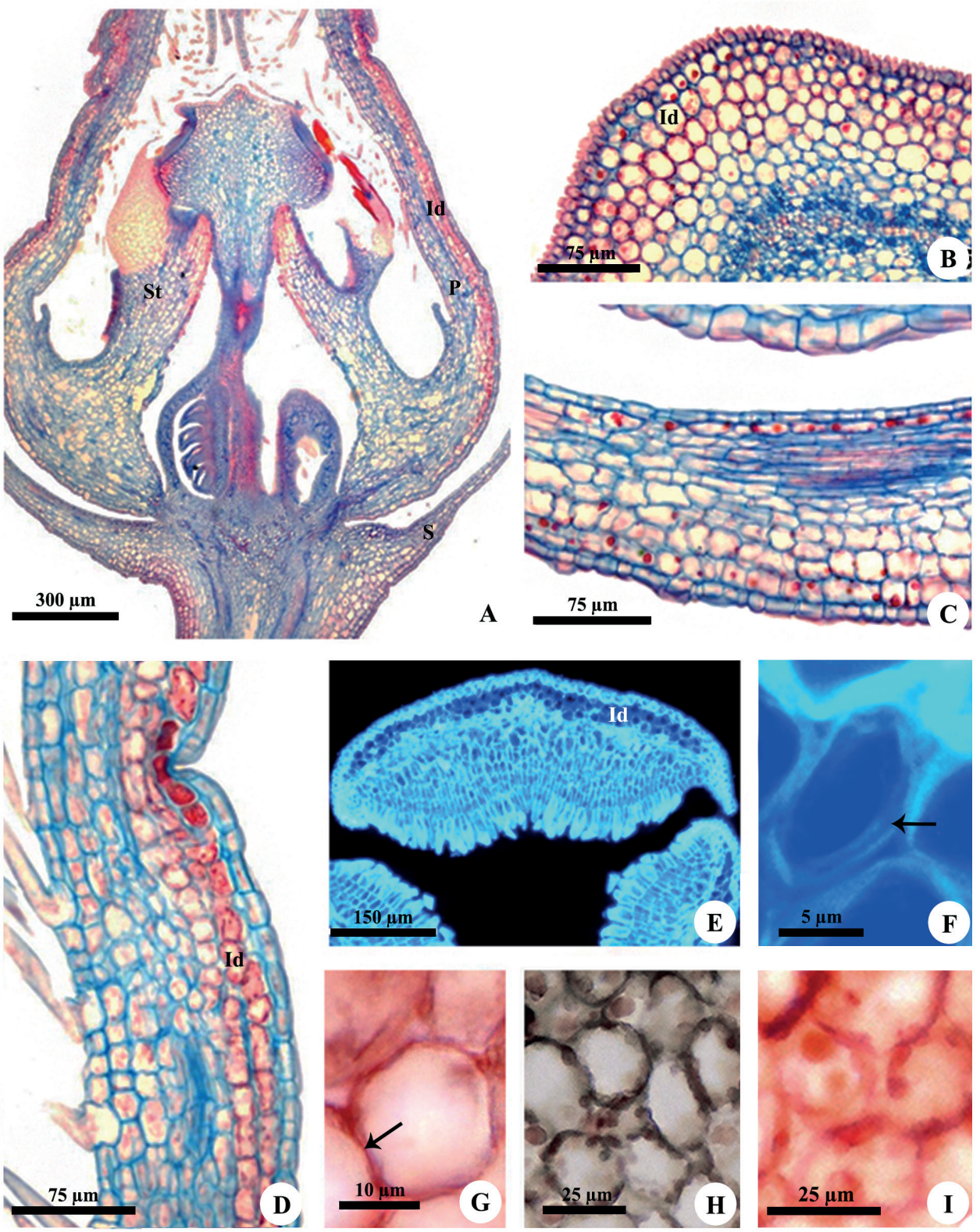

Figure 5. Oil idioblasts in flowers of *Peplonia axillaris* (Vell.) Fontella & Rapini. (A) General view of the flower in longitudinal section. (B-D) Oil idioblasts in the pedicel (B), sepal (C) and petal (D) with the secretion stained red. (E-F) Detection of cellulose with calcofluor white under UV. (F) Idioblast trilamellar wall. Note the absence of cellulose in a median lamella inside the cell wall (arrow). (G) Presence of suberin in the median lamella inside the cell wall (arrow) detected with Sudan IV. (H-I) Detection of oil using Sudan black B (H) and Sudan IV (I). Abbreviations: Id, oil idioblast; P, petal; St, stamen.

Nuptial glands

Nectary (Figs. 6-7)

Nectaries occur exclusively in flowers of Apocynaceae. Although there have been reports of extrafloral nectaries in the group, in actuality, these reports misinterpreted the colleters (Thomas 1991, and references therein).

The position of the nectaries is controversial in asclepiads, often due to the terminology applied in the description of flowers and to the inaccuracies in relation to the complex floral morphology. However, all species of this subfamily have primary nectaries in the filament tube (Galil & Zeroni 1965; Christ & Schnepf 1985; Kunze 1991; 1995; 1997; Kunze & Liede 1991; Endress & Bruyns 2000; Vieira & Shepherd 2002; Demarco 2005; Monteiro & Demarco 2017) (Fig. 6A), which corresponds to the secretory epidermis of the stigmatic chamber (Galil & Zeroni 1965; Valente 1977; 1984; 1995; Schnepf & Christ 1980; Valente & Silva 1984; Kunze 1991; 1995; 1999; Kunze & Liede 1991; Vieira & Shepherd 2002; Demarco 2005; Valente & Costa 2005; Monteiro & Demarco 2017) (Fig. 6B-E). In general, it is assumed that only the primary nectary is secretory and the nectar flows through an intricate capillary system to the nectar holder (Galil & Zeroni 1965; Kunze 1997). However, nectariferous tissue has been described in the corona of some genera (Rao & Ganguli 1963; Valente & Silva 1984; Bruyns 1993; Kunze 1995; 1999; Valente 1995; Demarco 2005; Monteiro & Demarco 2017) (Fig. 7), in this case referred to as a secondary nectary. The nectariferous tissue of the stigmatic chamber is usually composed of a uniseriate epidermis (Fig. 6C, E). However, the secondary nectary may be composed of only epidermis in the staminal corona (Fig. 7A-D) or epidermis and several layers of nectariferous parenchyma in a ring-shaped corona (Monteiro & Demarco 2017).

The nectar also has varied composition depending on the type analyzed. The nectar of all primary nectaries (stigmatic chambers) studied thus far is composed of carbohydrates (Fig. 6E), including glucose and mucilage, in addition to lipids (Christ & Schnepf 1985; Monteiro & Demarco 2017); this also holds true for the secondary nectary of *Peplonia axillaris*, but in *Matelea denticulata*, the secondary nectary exudes exclusively carbohydrates (Monteiro & Demarco 2017). The difference detected between the nectars may be related to distinct functions. The nectar in the stigmatic chamber may have a dual function: a resource for pollinators and an inducer of pollen germination (Galil & Zeroni 1965; Eisikowitch 1986; Kunze 1991) (Fig. 6E). However, flowers with two types of nectaries may divide these functions with the secondary nectary producing nectar for the pollinator and the primary nectary acting exclusively as an inducer of pollen germination (Monteiro & Demarco 2017).

Osmophore (Fig. 8)

The osmophore (or scent gland) is a structure secreting volatile substances of variable composition (Jürgens *et al*. 2008; 2010) and having the function of attracting pollinators over long distances (Vogel 1990). Except for the study of Vogel (1990) with *Ceropegia elegans* Wall., the osmophores of Apocynaceae have been studied structurally only in two other species (Plachno *et al*. 2010). In the rest of family, this gland is only mentioned without any structural corroboration (Endress 1994; Demarco 2005). The main reports have been the description of the scent in taxonomic studies and a few chemical analyses (Stevens 1988; Vogel 1990; Rohrbeck *et al*. 2006; Jürgens *et al*. 2008; 2010; Setzer 2014). In *Ceropegia*, the osmophore was found at the tip of petals, consisting of secretory epidermis and subepidermal layers, as well as in *Ditassa* (Fig. 8A-F) and *Boucerosia*, but *Orbea* has two types of secretory epidermal cells with distinct structural characteristics (Vogel 1990; Plachno *et al*. 2010).

The main components of the scents are terpenes and phenolic compounds of low molecular weight (Jürgens *et al*. 2010), which produce a sweet aroma in some species and a fetid aroma in others (Stevens 1988; Vogel 1990; Wolff *et al*. 2008). The different types of scents are often associated with the corolla color, as in the sapromyophilic flowers of Ceropegieae, which have dark brown, red or yellow corollas and release a characteristically putrid aroma (Meve & Liede 1994). In general, sweet scents are related to white corollas (Vogel 1990), as is the case with *Ditassa gracilis* (present study).

Style head (Fig. 9)

The style head is present in all Apocynaceae and corresponds to the upper portion of the styles (Figs. 1E-F, 9A), which fuses postgenitally and dilates. With the exception of Rauvolfioideae, in all other members of the family the style head is adnate to the anthers through the retinaculum, forming the gynostegium (Rao & Ganguli 1963; Fallen 1986; Endress 1994; Swarupanandan *et al*. 1996; Endress & Bruyns 2000). The style head in the family is covered by a secretory epidermis (Walker 1975; Fallen 1986; Kunze 1993; 1994; Galetto 1997; Lin & Bernardello 1999; Demarco 2014) (Fig. 9A-B), and its secretion is related to the process of pollen transportation. Initially, it helps adhere the pollen to the pollinator and then assists in the capture of pollen by the stigma or the guide rail/stigmatic chamber of another flower. The secretory surface is present only on the lateral side of the style head and alternate with the anthers in asclepiads (Fig. 9B-C), but in Rauvolfioideae and Apocynoideae it covers almost the entire surface of this dilated portion of the style apex. This is one of the differences that led Fallen (1986) to describe four basic types of style head and define a morphological progression of this structure for the family.

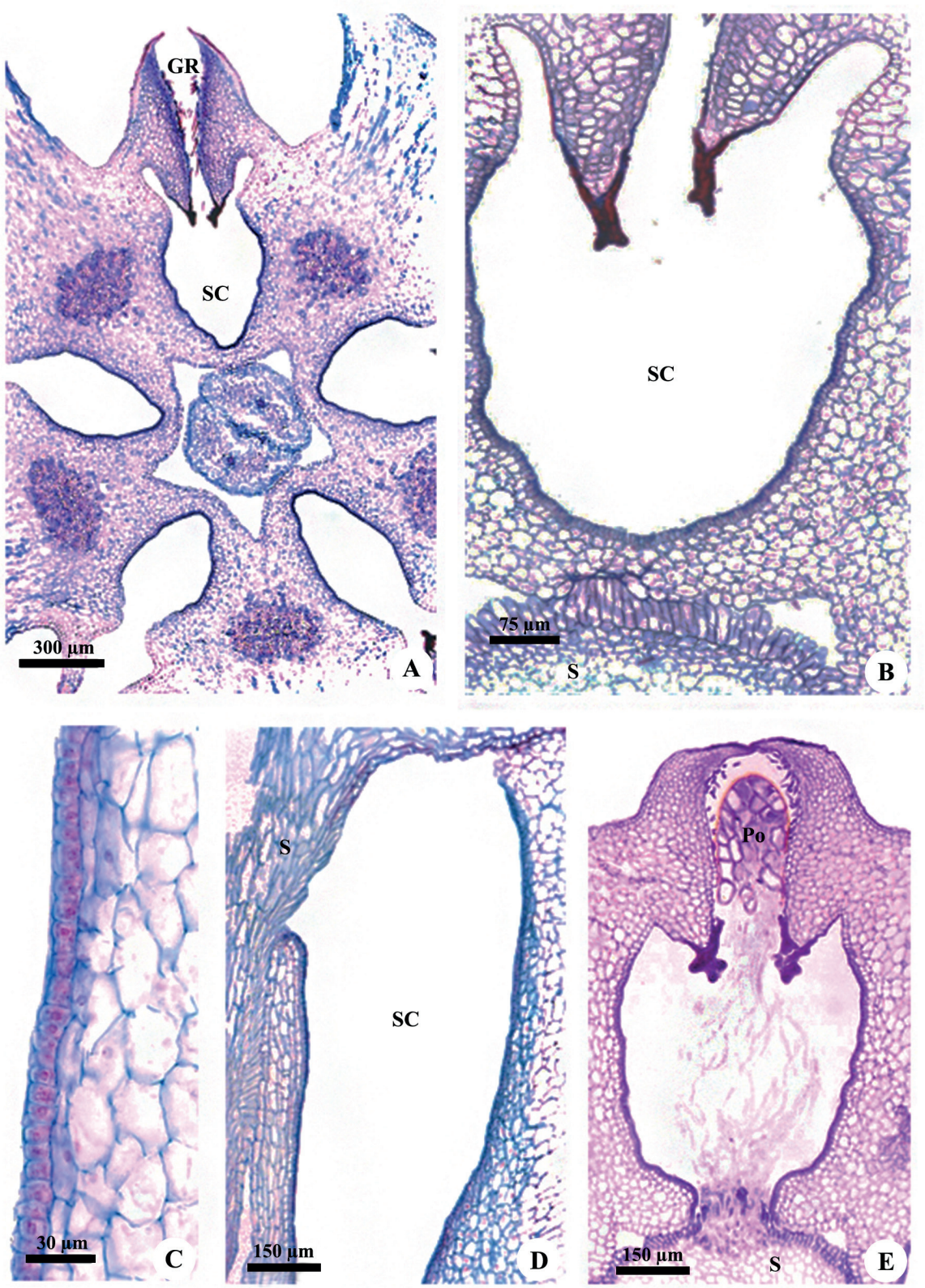

Figure 6. Primary nectaries in flowers of *Blepharodon bicuspidatum* E. Fourn. (A) General view of the primary nectaries (stigmatic chambers) behind the guide rail in transversal section. (B) Detail of the nectary which is formed by the nectariferous epidermis of the stigmatic chamber. (C) Detail of the nectariferous epidermis. (D) Longitudinal view of the stigmatic chamber and its opening at the stigma level. (E) Pollinium inserted into the guide rail and germinated due to the presence of nectar in the stigmatic chamber. Note the entrance of pollen tubes into the stigma (section stained with PAS reaction). Abbreviations: GR, guide rail; Po, pollinium; S, stigma; SC, stigmatic chamber.

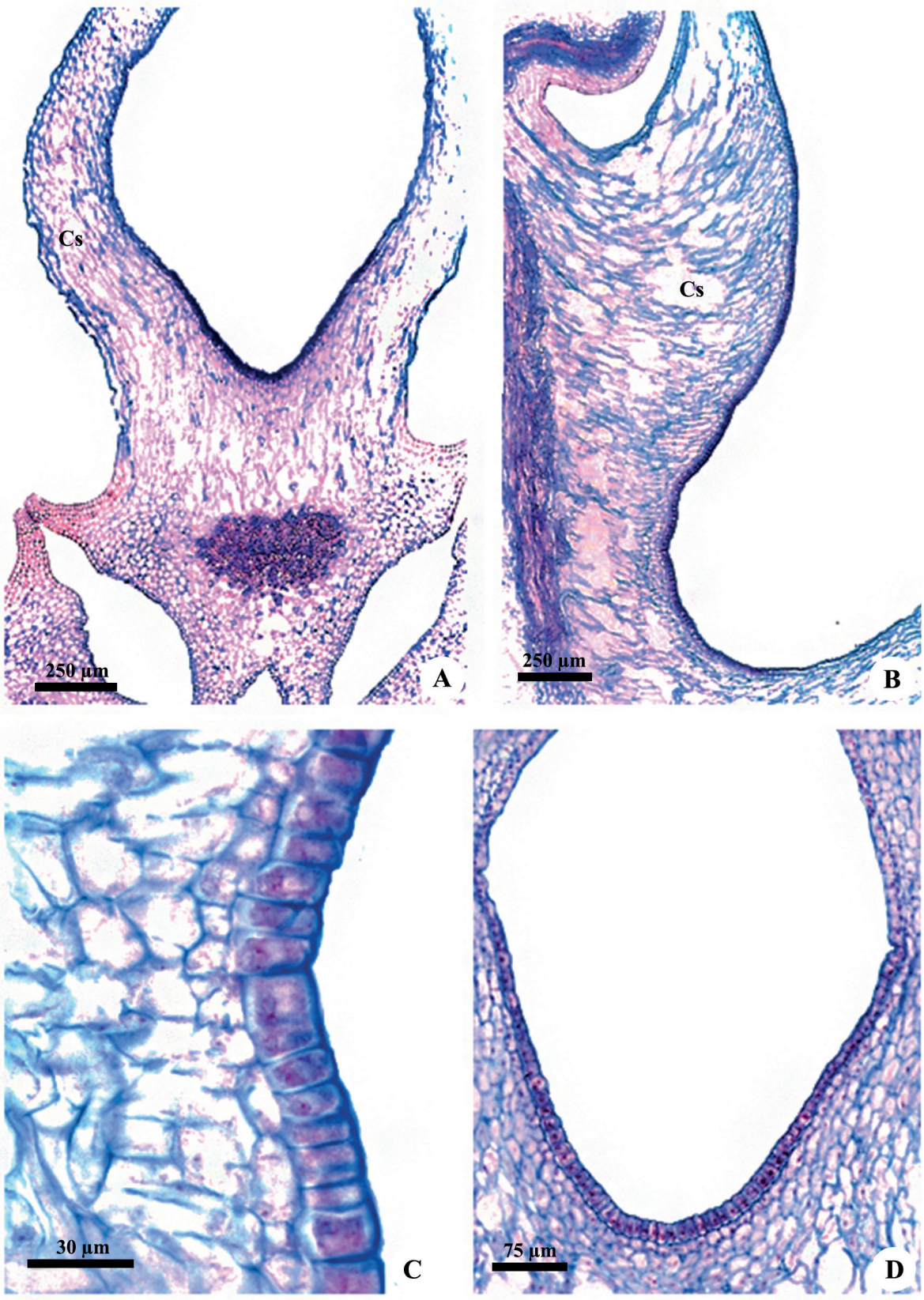

Figure 7. Secondary nectaries in flowers of *Blepharodon bicuspidatum* E. Fourn. (A) General view of the secondary nectary in the staminal corona. (B) Longitudinal view of the nectariferous epidermis of the corona. (C) Detail of B. (D) Nectariferous tissue composed exclusively of epidermis. Abbreviation: Cs, staminal corona.

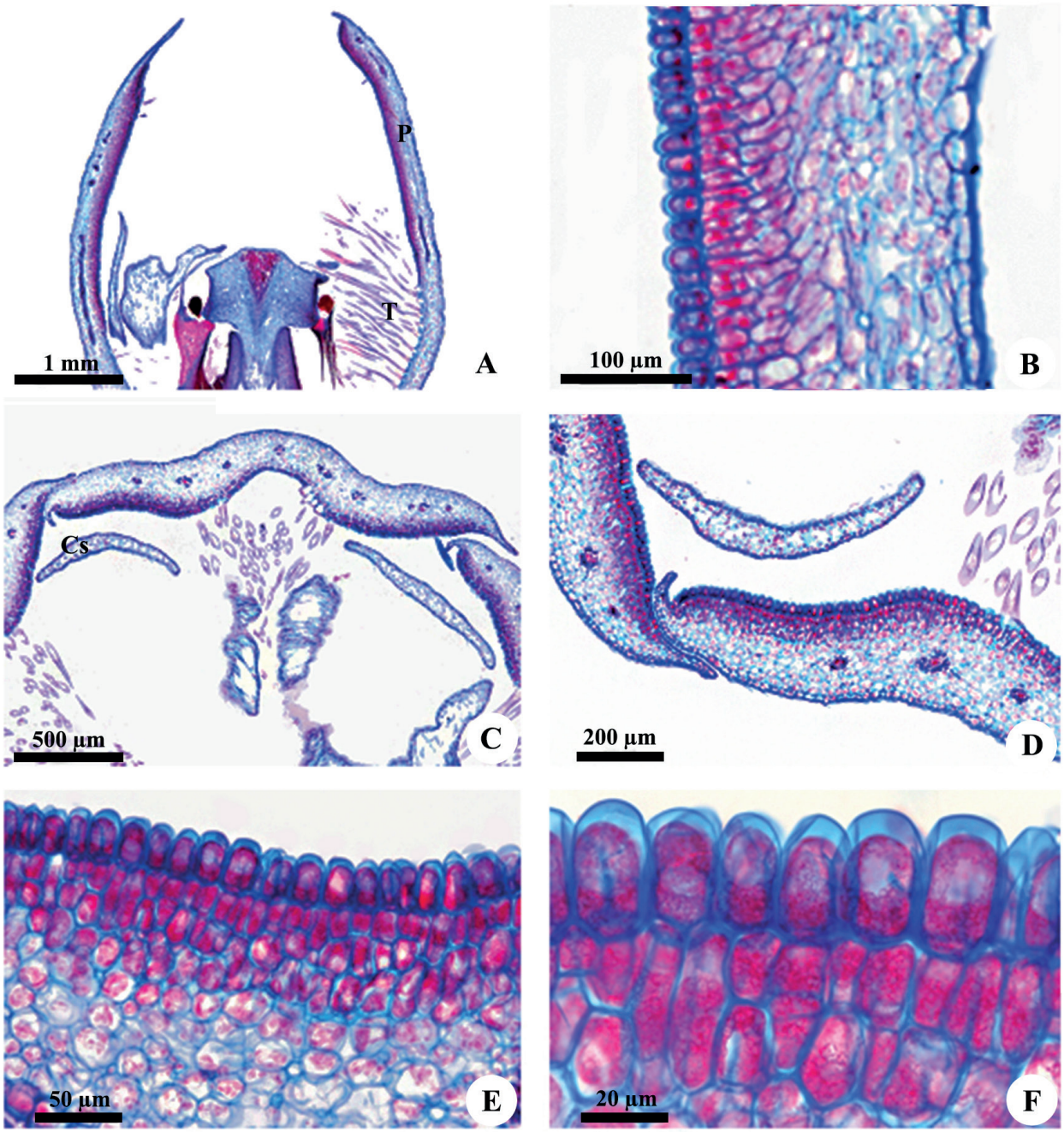

Figure 8. Osmophore in flowers of *Ditassa gracilis* Hand.-Mazz. (A) General view of the flower in longitudinal section. Note the presence of osmophore in the upper part of the petal above the trichomatous zone. (B) Detail of the osmophore in the adaxial face of the petal. (C) Secretory tissue occurs in the free portion of petals from the gamopetalous corolla. (D) Main portion of the osmophore is located in the lateral sides of the petal, except in the margins. (E) Osmophore is composed of epidermis and two to three layers of parenchyma. (F) Presence of multiple vesicles in the secretory cells and a prominent vacuole. Abbreviations: Cs, staminal corona; P, petal; T, trichomes.

In Asclepiadoideae, the formation of the style head occurs during the beginning of floral development and is an indication of its complexity and importance in the later stages (Fallen 1986; Endress 1994; Swarupanandan *et al*. 1996; Demarco 2014). The secretory tissue is responsible for the secretion of the translator in Periplocoideae, Secamonoideae and Asclepiadoideae (Brown 1810; Corry 1883; Rao & Ganguli 1963; Vijayaraghavan & Cheema 1977; Dicko-Zafimahova 1980; Valente & Silva 1984; Endress 1994; Kunze 1994; Valente 1995; Endress & Bruyns 2000;

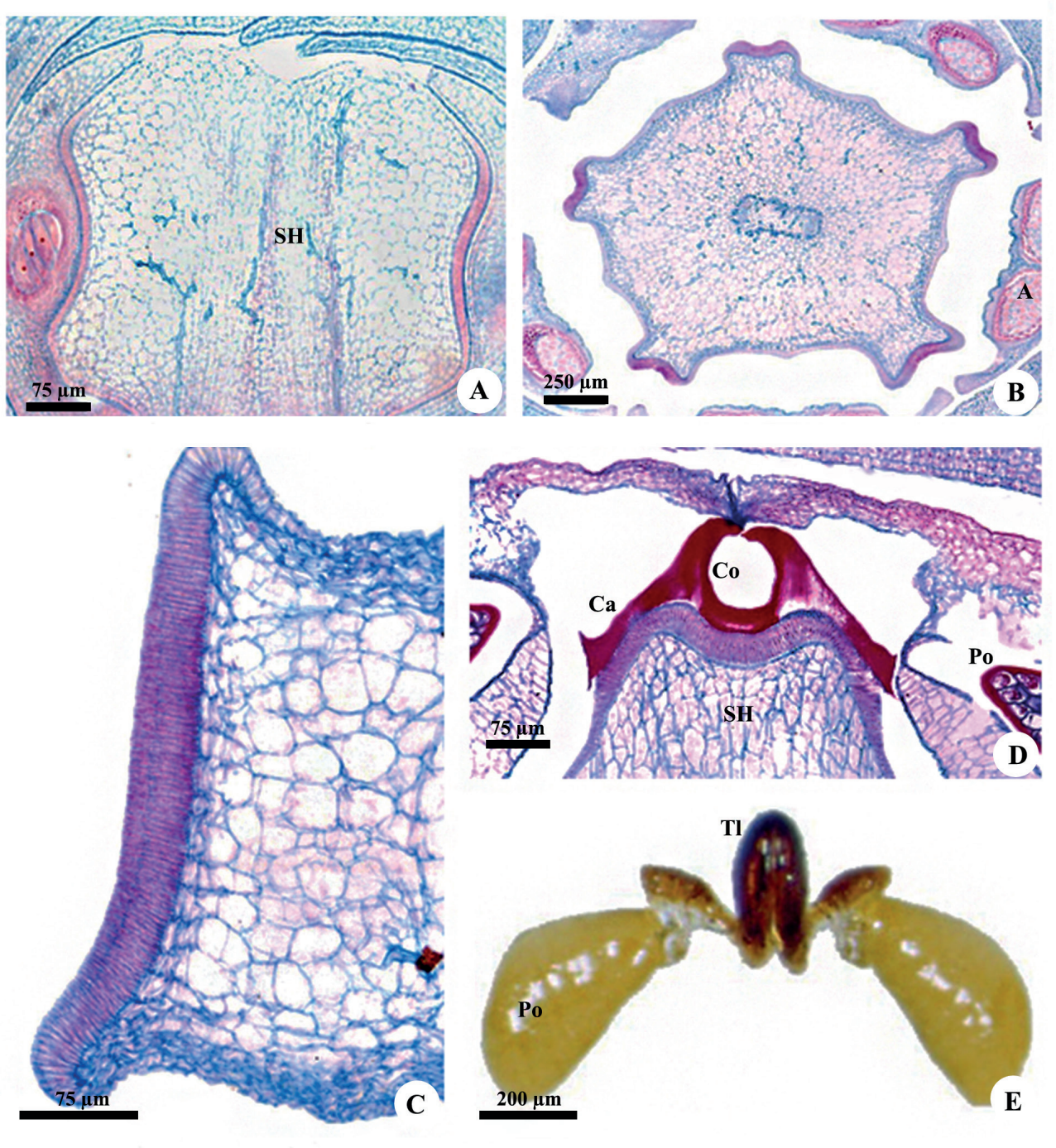

Figure 9. Style head of *Blepharodon bicuspidatum* E. Fourn. flowers. (A) General view of the style head in longitudinal section. (B) Style head is pentagonal in transverse section and present five secretory regions alternate to anthers. (C) Secretory portion of the style head constituted of a palisade epidermis. (D) Translator composed of corpusculum and two caudicles produced by the style head. (E) Morphology of the pollinarium formed by the translator and two pollinia. Abbreviations: A, anther; Ca, caudicle; Co, corpusculum; Po, pollinium; SH, style head; Tl, translator.

Valente & Costa 2005; Demarco 2014) (Fig. 9D).

In asclepiads, the translator is a thick secretion composed of a corpusculum and two caudicles which attach to two pollinia of adjacent anthers, forming the pollinarium (Brown 1810; Corry 1883; Schumann 1895; Valente 1977; 1984; 1995; Valente & Silva 1984; Kunze 1993; 1994; Endress 1994; Swarupanandan *et al*. 1996; Valente & Costa 2005; Demarco 2014) (Fig. 9D-E). The caudicles are absent only in *Fockea* and *Cibirhiza* (Kunze 1993; 1994; Swarupanandan *et al*. 1996), genera placed in Marsdenieae, the most basal

tribe of Asclepiadoideae (Endress & Bruyns 2000). The morphogenesis of the translator begins in the early stages of floral development, and its specific shape is mainly due to the differential secretory activity of the cells and the undulated outline of the secretory surface of the style head (Kunze 1994; Demarco 2014). Serbanescu-Jitariu & Tarnavschi (1976) observed that the structure of the pollinarium provides useful characters for the identification and classification of this subfamily, which is still used in taxonomic and phylogenetic studies today (Endress & Bruyns 2000; Rapini *et al*. 2003; Rapini 2012).

Secretory cells produce different amounts of secretion and distinct types of compounds in each region of the style head. The translator is composed mainly of lipids, but the composition of the corpusculum and caudicles is different (Woodson 1954; Safwat 1962; Vijayaraghavan & Cheema 1977; Schnepf *et al*. 1979; Demarco 2014). Fatty acids, phenolic compounds, mucilage and proteins have been detected in the corpusculum and only neutral lipids and mucilage in the caudicles (Vijayaraghavan & Cheema 1977; Demarco 2014). This difference in the composition of the translator parts is related to their different functions. The corpusculum must adhere to the pollinator's body, while the caudicles dehydrate after pollinarium removal from the flower and often contracts, moving the pollinia to the correct position for their insertion in the stigmatic chamber (Vijayaraghavan & Cheema 1977; Kunze 1991; Demarco 2014). Although there is a high probability that the corpusculum and caudicles differ from each other in relation to the chemical composition in the majority of asclepiads, the shape of some pollinaria does not change after dehydration of the caudicles (Wiemer *et al*. 2012).

Tapetum (Fig. 10)

Although the tapetum is usually not considered a secretory structure, all Apocynaceae have tapetum of the secretory type (Pacini *et al*. 1985), which plays an important role in the formation of pollinia and pollinarium as a whole in all asclepiads (Woodson 1954; Linskens & Suren 1969; Schnepf *et al*. 1979; Demarco 2014) (Fig. 10A-H).

The pollinium corresponds to the aggregation of all pollen grains from a pollen sac, and this grain aggregation is increased by the secretion by the tapetum cells of a pellicle, which surrounds externally the entire pollinium (Fig. 10G) and internally every pollen grain. This pellicle is mainly composed of lipids (sporopollenin) (Vijayaraghavan & Shukla 1976; Schill & Jäkel 1978; Schill & Dannenbaum 1984; Pacini & Hesse 2005; Wyatt & Lipow 2007; Demarco 2014), and the secretion produced by tapetum may help the caudicle adhere to the pollinium apex (Schnepf *et al*. 1979). In *Matelea*, the tapetum produces a projection of the pellicle (hyaline crest) in a sterile portion of the anther which will attach to the caudicle after anther dehiscence (Demarco 2014).

Staminal wing gland (Fig. 11)

In most Apocynaceae, there is a transfer of the pollen capture area from the gynoecium to the androecium, which seems to have also occurred in Apocynoideae (Fallen 1986). In Asclepiadoideae, the capture function is performed by the guide rail (Figs. 6E; 11A-B), which guides the insect to the nectar holder in an interstaminal position at the base of the flower (Fig. 1E) and retains the pollinium brought by the pollinator inside the stigmatic chamber (primary nectary). The nectar present in this chamber induces pollen germination (Bookman 1981; Kunze 1991; Endress 1994; Vieira & Shepherd 2002; Demarco 2017; Monteiro & Demarco 2017) (Fig. 6E). The upward movement of the pollinator, directed by the guide rail, also leads the proboscis or leg of the pollinator to the corpusculum of the translator (Fig. 1E), promoting the removal of the whole pollinarium (Kunze 1991; Wiemer *et al*. 2012; Demarco 2014).

Recently, an ontogenetic study of Asclepiadeae flowers has shown that the origin of the wings, which compose the guide rail, is variable. In *Asclepias*, *Oxypetalum* and *Peplonia*, they are formed by lateral projections of the anther and filament and, therefore, should be designated staminal wings (Demarco 2017) and not anther wings, as reported by several authors (Frye 1902; Rao & Ganguli 1963; Valente 1977; 1980; 1983; 1995; Valente & Silva 1984; Swarupanandan *et al*. 1996; Endress & Bruyns 2000; Vieira & Shepherd 2002; Valente & Costa 2005). In the intermediate stages of the floral bud development, two glands are formed along the staminal wings: one at the outer margin and the other at the inner margin of the guide rail (Fig. 11C-E). The secretory tissue is composed exclusively of the epidermis which secretes mucilage and lipids. These glands senesce and necrose before pre-anthesis (Demarco 2017) (Fig. 11F).

During pollination, the secretion exuded by the staminal wing gland in the floral bud is present inside the guide rail and might play an important role as lubricant, facilitating the entrance of the insect's proboscis or leg in this slit and/or assisting in the removal of pollinium or part of the pollinarium adhering to the insect. The disintegration of the gland before anthesis is also related to the introduction of proboscis and/or pollinium into the guide rail due to the increase of the chamber area without the glands (Demarco 2017).

Extragynoecial compitum (Fig. 12)

All asclepiads have an apocarpic bicarpelar gynoecium with partially free styles, united only in the apical region where the style head and a subterminal stigma are formed (Fallen 1986; Endress & Bruyns 2000) (Fig. 1F). Since the stigma is located below the style head, it has been proposed that one of the functions of this connate region may be to act as a compitum (Fallen 1986). However, the compitum is formed by the union of the transmitting tissue tract from

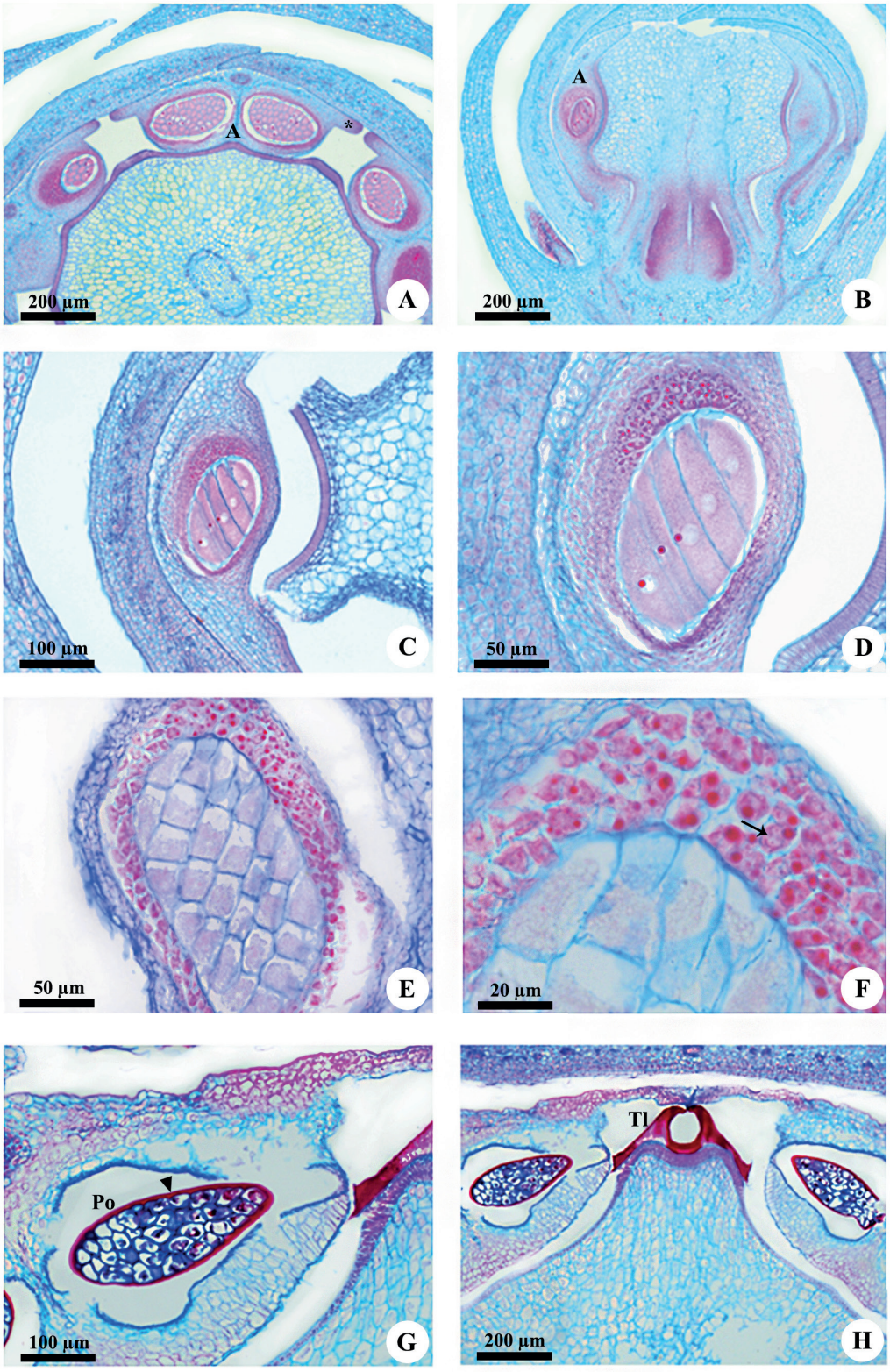

Figure 10. Tapetum in flowers of *Blepharodon bicuspidatum* E. Fourn. (A-F) Floral buds. (G-H) Mature flower. (A) Bithecal, bisporangiate anther with initial projection of the staminal wing from its dorsolateral side (asterisk). (B) Longitudinal section of the young anther. (C) Secretory tapetum surrounding elongated microspore mother cells. (D) Presence of secretory globules in the tapetum cells. (E) Tapetum in secretory activity around the microspores. (F) Detail of the tapetum with secretory globules and vacuoles with heterogenous content (arrow). (G) Mature anther without tapetum containing pollinia covered by a pellicle (arrowhead) secreted by tapetum. (H) Pollinarium formed by translator and two pollinia from adjacent anthers. Abbreviations: A, anther; Po, pollinium; Tl, translator.

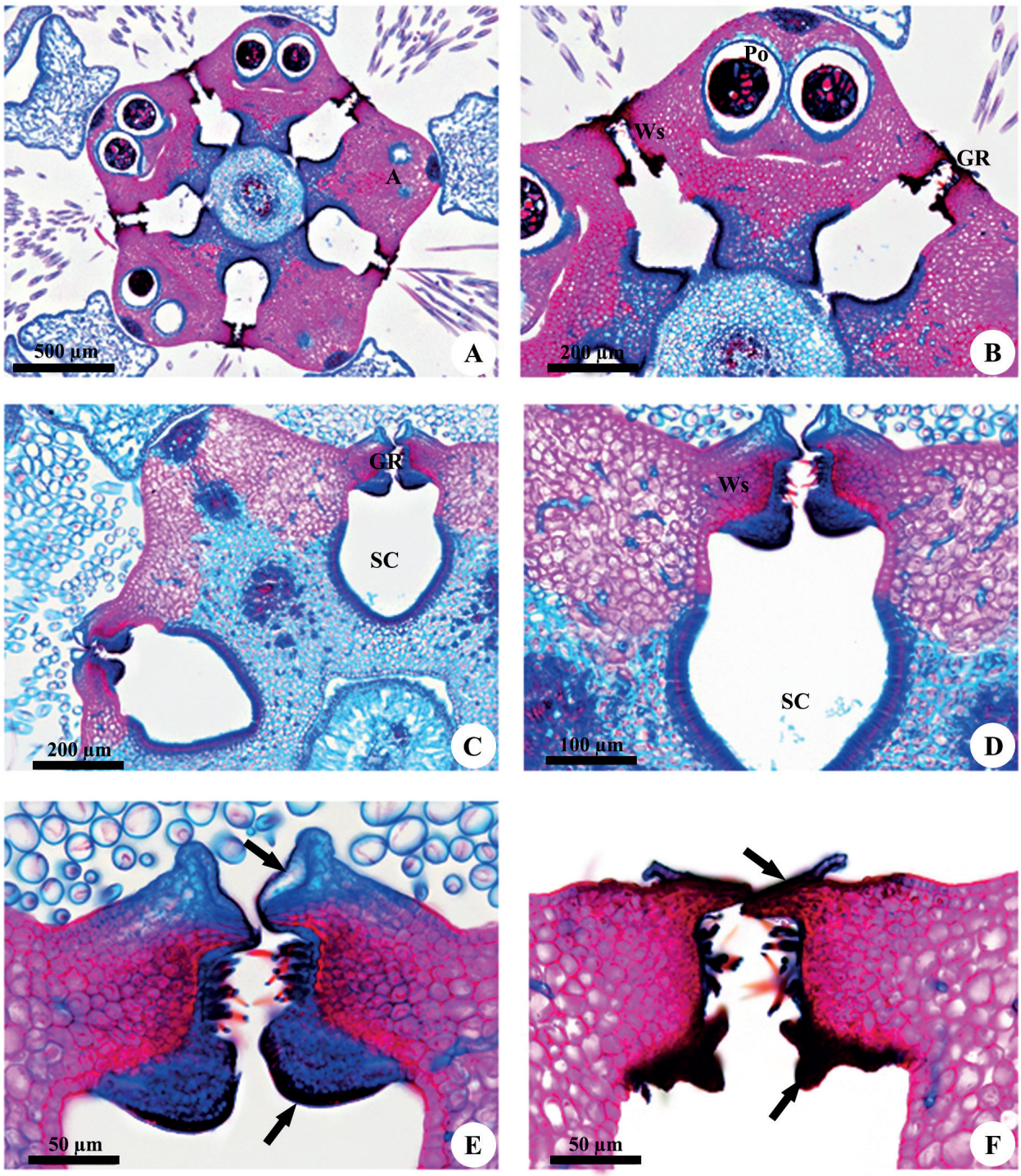

Figure 11. Staminal wing gland of *Ditassa gracilis* Hand.-Mazz. flowers. (A-B, F) Anthetic flowers. (C-E) Floral buds. (A) General view of anthers in transverse section. (B) Anther with two pollen sacs and two lateral wings. Staminal wings of two adjacent anthers form the guide rail. (C) Guide rail with glands in front of the stigmatic chamber. (D) Staminal wing glands in the outer and inner margins of the guide rail. Note staminal wings completely lignified, except in the glandular areas. (E) Detail of the glands with palisade secretory epidermis. Note the presence of lignified trichomes in the middle region of the guide rail. (F) Necrotic wing glands in a mature flower. Abbreviations: A, anther; GR, guide rail; Po, pollinium; SC, stigmatic chamber; Ws, staminal wing; arrow, wing gland.

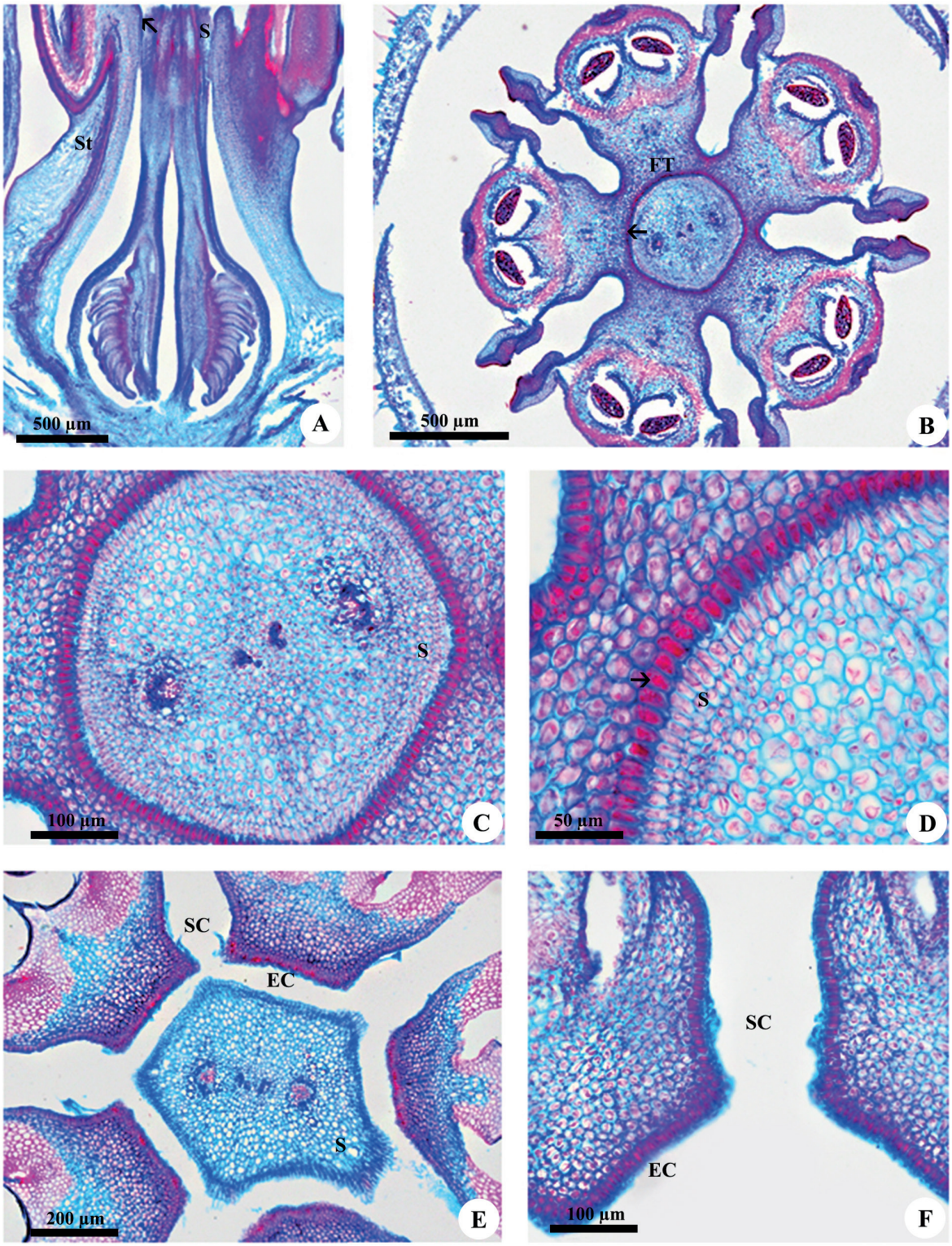

Figure 12. Extragynoecial compitum in flowers of *Oxypetalum banksii* subsp. *banksii* Roem. & Schult. (A) Longitudinal section. (B-F) Transverse sections. (A-B) Extragynoecial compitum formed by the secretion produced by the inner epidermis of the filament tube in its upper portion. (C) Secretory epidermis around the dry stigma. (D) Detail of C. (E) Continuity between the secretory epidermis of the stigmatic chamber and extragynoecial compitum at stigma level. (F) Secretory portion of the extragynoecial compitum composed exclusively of epidermis. Abbreviations: EC, extragynoecial compitum; FT, filament tube; S, stigma; SC, stigmatic chamber; St, stamen; arrow, epidermis of the extragynoecial compitum.

each carpel at the level of style, which allows pollen tubes to reach the ovules of different carpels (Carr & Carr 1961) independently from where they entered the stigma. The analysis of the connate region of the style below the stigma in asclepiads shows that the two strands of transmitting tissue are independent in most species and the pollination by a single pollinium generally forms one single follicle (Kunze 1991). Among the asclepiads, compitum has been identified only in *Tylophora* and *Matelea* (Kunze 1991; Demarco 2008).

In addition to this type of gynoecial compitum, some species have a secretion involving the stigma (or stigmata) that allows the entrance of pollen tubes through different regions in order to reach all the free ovaries (Endress 1980). This secretion functions as an extragynoecial compitum. In Apocynaceae, the production of twin follicles from a single pollinium demonstrates the presence of a compitum in the asclepiad *Oxypetalum banksii*, and the anatomical study revealed the presence of an extragynoecial compitum formed by the mucilage produced by epidermal cells of the inner surface of the filament tube around the stigma (Vieira & Shepherd 2002) (Fig. 12A-F). This is the only report for the family.

In addition, the mucilage of some Monimiaceae flowers not only acts as an extragynoecial compitum but also serves as a primary pollen receptor and has been given the name hyperstigma (Endress 1979; 1980; 1982). Although the mucilage present in the primary nectar within the stigmatic chamber (Monteiro & Demarco 2017) does not have the function of pollinium capture in Asclepiadoideae and the concept may not be applied in the same sense, this secretion has previously been considered a hyperstigma in *Oxypetalum* (Vieira & Shepherd 2002).

Stylar canal (Fig. 13)

The general description of the pollen tube paths through the gynoecium in asclepiads begins with the germination of pollen grains in the stigmatic chamber promoted by the nectar (Eisikowitch 1986; Kevan *et al*. 1989; Valente 1994; Wyatt & Broyles 1994; Sage & Williams 1995; Monteiro & Demarco 2017). Pollen tubes penetrate the dry stigma and grow through a non-secretory transmitting tissue, reaching a canal lined with a secretory epidermis (Sage *et al*. 1990; Kunze 1991; Sage & Williams 1995; Demarco 2008) (Fig. 13A-F). The pollen tubes grow through the canal and obliterate it, as occurs with the adjacent parenchyma cells (Sage *et al*. 1990; Sage & Williams 1995; Vieira & Shepherd 2002). In the ovary, pollen tubes grow in the ovarian locule on the surface of the placenta to the ovule micropyle (Sage & Williams 1995; Vieira & Shepherd 2002).

Along the pathway through the style, pollen tubes grow, digesting the cells of the transmitting tissue strand at first but then grow immersed in the secretion of the stylar canal at a later stage (Sage & Williams 1995; Vieira & Shepherd 2002; Demarco 2008). Stylar canals, occurring in all asclepiads (Kunze 1991), are very narrow (Fig. 13B, F) and promote a place of strong pollen tube competition, increasing the male gametophyte selection (Kunze & Liede 1991). The secretory activity starts in pre-anthetic flowers, and the secretion is composed of mucilage and lipids, which will nourish and direct the pollen tubes towards the ovarian locule (Demarco 2008) (Fig. 13B).

Obturator (Fig. 14)

Upon reaching the ovary, the pollen tubes are directed through the locule to the micropyle of the ovules by an obturator (Fig. 14A-B). The obturator was first reported and described for Apocynaceae in *Aspidosperma* (Rauvolfioideae; Demarco 2005); later, its presence was also confirmed for Asclepiadoideae (Demarco 2008), and recently, the obturator was also identified in another species of Rauvolfioideae (Morokawa *et al*. 2015). It is possible that it is present in all Apocynaceae. In some Rauvolfioideae, the obturator is composed of secretory placental trichomes (Demarco 2005; Morokawa *et al*. 2015), but in asclepiads it is formed by secretory cubic cells on the surface of placenta and at the base of funiculus (Fig. 14C-F), which is described for the first time for asclepiads in this review.

The aperture of the stylar canal is continuous with the ovary locule (Fig. 14A), and the secretory epidermis of this canal is continuous with the secretory epidermis of the placenta and funiculus (Fig. 14B). The secretion produced by the obturator fills the entire locule and has the same components as the secretion of the stylar canal: mucilage and lipids. Therefore, the pollen tubes grow inside a continuous layer of secretion from the style to the ovary until they fertilize the ovules (Demarco 2008).

Evolution and ecological importance of the glands

Some secretory structures found in the flowers of asclepiads reveal their relationship with other members of Apocynaceae due to their conservative nature, such as the presence of the style head and articulated anastomosing laticifers in the entire family (Tab. 1). On the other hand, the huge diversity of floral glands in Asclepiadoideae and their much more elaborate and synorganized flowers emphasize their derived condition in the family and highlight the asclepiads as the group with the largest diversity of floral glands among the angiosperms. In the 13 types of glands described in this review, *Matelea* (Gonolobinae, Asclepiadeae; Endress *et al*. 2014) has 11 in the same flower (colleters, glandular trichomes, laticifers, primary nectaries, secondary nectaries, osmophores, style head, tapetum, staminal wing gland, stylar canal and obturator; Demarco 2008). The two remaining secretory structures have restricted occurrence. Secretory idioblasts have been observed only in *Peplonia* (Metastelmatinae, Asclepiadeae) and extragynoecial

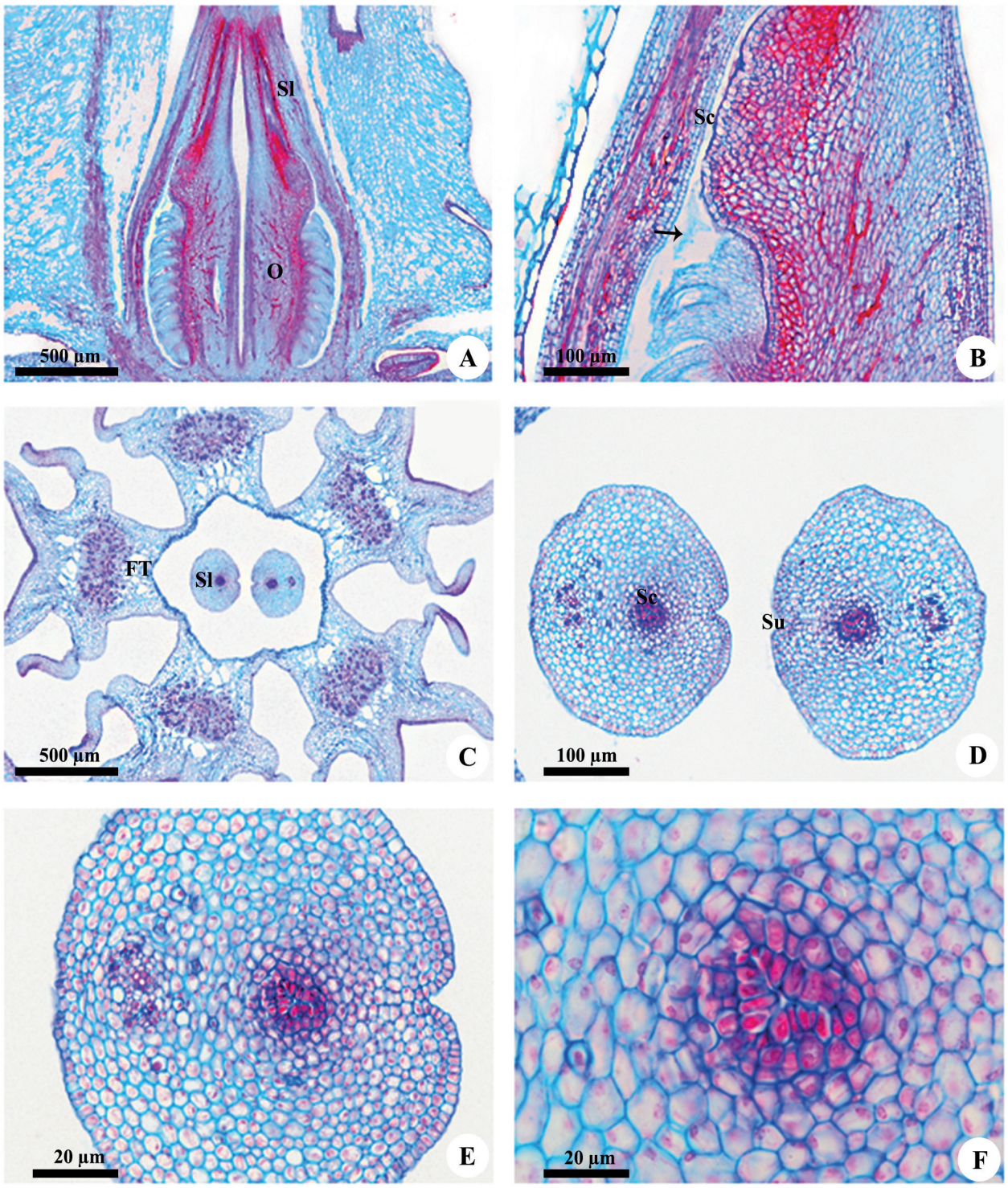

Figure 13. Stylar canal in flowers of *Blepharodon bicuspidatum* E. Fourn. (A) General view of the gynoecium with two free ovaries and the free portion of the styles in longitudinal section. (B) Secretory stylar canal with opening in the ovarian locule. Note the presence of secretion (arrow). (C) Stylar canals in the free portion of the styles surrounded by the filament tube. (D) Stylar canal occurs in the adaxial side of the style at the suture line. (E) Stylar canal originated from the adaxial epidermis of the folded style. (F) Stylar canal with a very narrow lumen lined by a secretory epidermis. Abbreviations: FT, filament tube; Sc, stylar canal; Su, suture; O, ovary; Sl, style.

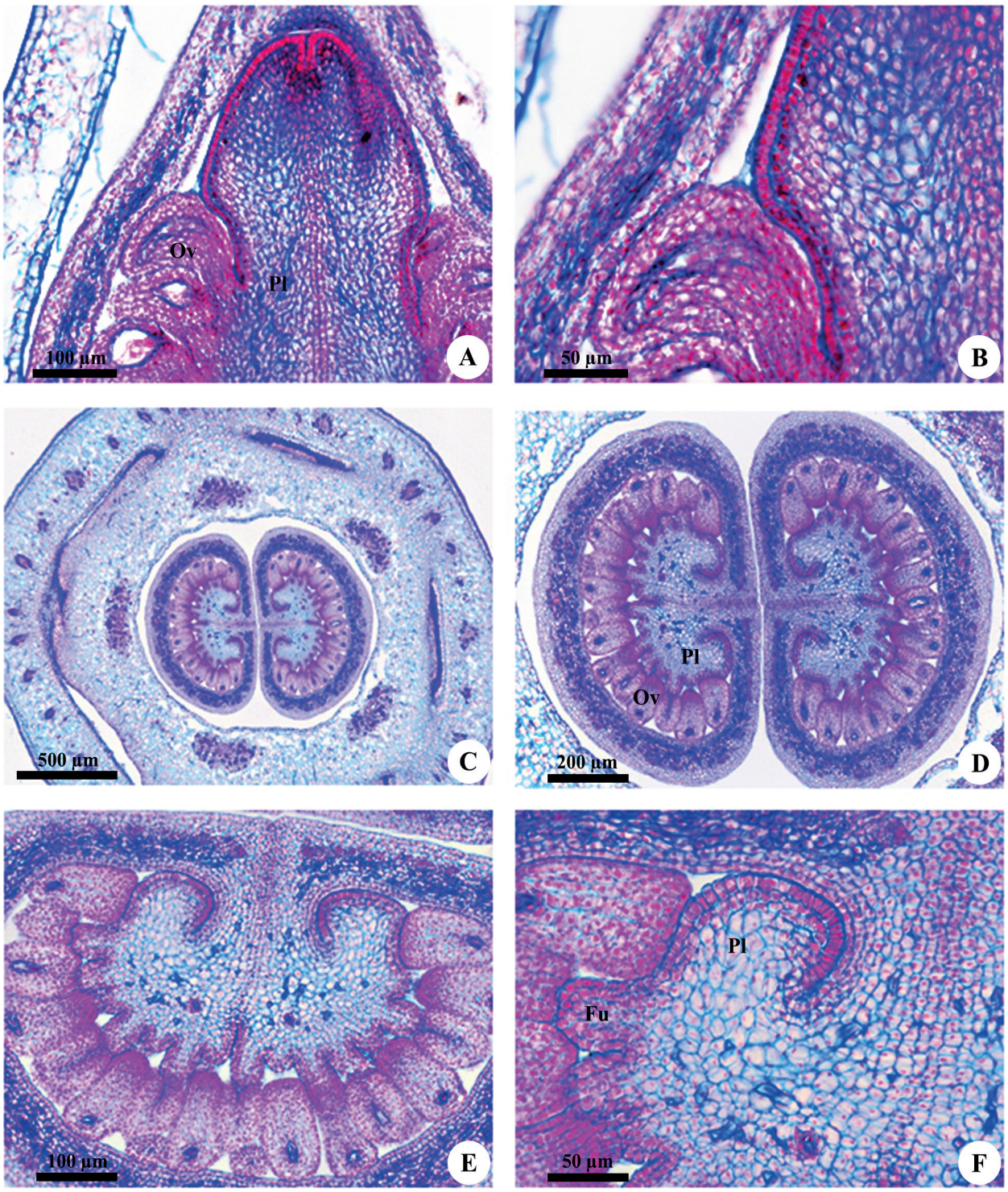

Figure 14. Placental-funicular obturator in flowers of Asclepiadoideae. (A-B, E-F) *Ditassa gracilis* Hand.-Mazz. (C-D) *Blepharodon bicuspidatum* E. Fourn. (A-B) Longitudinal sections. (C-F) Transverse sections. (A) Continuity between the secretory epidermis of the stylar canal and obturator. (B) Detail of A. (C) General view of the ovaries. (D-E) Obturator composed of the secretory epidermis of placenta and funiculus base. (F) Detail of the obturator. Abbreviations: Fu, funiculus; Ov, ovule; Pl, placenta.

Table 1. Distribution of floral glands in the tribes and subtribes of Asclepiadoideae (*sensu* Endress *et al*. 2014).

Tribe	Subtribe	Glands already registered
Fockeeae		L, PN, Os (2), SH, T
Eustegieae		L, PN, SH, T
Marsdenieae		C, GT (3), L, PN, SN (1), Os (5), SH, T, SC
Ceropegieae	Heterostemminae	C, L, PN, SH, Os (1), T, SC
	Leptadeniinae	C, L, PN, SH, T, SC
	Anisotominae	C, L, PN, SH, T, SC
	Stapeliinae	C, L, PN, SN (3), Os (13), SH, T, SC
Asclepiadeae	Astephaninae	C, L, PN, SH, T, SC
	Asclepiadinae	C, L, PN, SN (2), SH, T, WG (1), SC, Ob (1)
	Cynanchinae	C, GT (1), L, PN, SN (2), Os (1), SH, T, SC
	Tylophorinae	C, L, PN, SN (1), Os (1), SH, T, SC
	Pentacyphinae	C, L, PN, SH, T, SC
	Diplolepinae	C, L, PN, SH, T, SC
	Orthosiinae	C, L, PN, Os (2), SH, T, SC
	Metastelmatinae	C, L, SI (1), PN, SN (3), Os (2), SH, T, WG (3), SC, Ob (3)
	Tassadiinae	C, L, PN, SH, T, SC
	Oxypetalinae	C, GT (1), L, PN, Os (2), SH, T, WG (1), EC (1), SC, Ob (1)
	Gonolobinae	C, GT (3), L, PN, SN (1), Os (2), SH, T, WG (1), SC, Ob (1)

Note. The number in parenthesis represents the quantity of genera where the gland has already been registered and its absence indicates the ubiquitous occurrence of the gland in the group. The references for this data survey are found in the description of each gland in this review. C = colleter; GT = glandular trichome; L = laticifer; SI = secretory idioblast; PN = primary nectary; SN = secondary nectary; Os = osmophore; SH = style head; T = tapetum; WG = staminal wing gland; EC = extragynoecial compitum; SC = stylar canal; Ob = obturator.

compitum only in *Oxypetalum* (Oxypetalinae, Asclepiadeae; Tab. 1). In absolute numbers, if we consider the quantities of each gland type, we could count dozens of glands in the same flower, not to mention the countless laticifers, idioblasts and/or glandular trichomes.

Among the Apocynaceae already studied, the genus with the lowest number of floral glands is *Aspidosperma* (Aspidospermateae, Rauvolfioideae; Endress *et al*. 2014) with three types of glands (laticifers, style head and tapetum; Demarco 2005), the one with the largest number being *Matelea* (Gonolobinae, Asclepiadeae, Asclepiadoideae). When the position of these genera is analyzed in the phylogeny, Aspidospermateae is the most basal tribe of the family and Asclepiadeae the most derived (Yang *et al*. 2016). The exponential increase in the number of gland types per flower in the family is directly related to the greater complexity of pollination types in asclepiads, mainly due to their dispersion of pollen grains in pollinia.

If we consider the origin of the secretory tissues in the glands, we noticed that 10 of the 13 gland types present in the flowers have secretory tissue originating exclusively from the protoderm or mainly from this meristem. The large number of postgenital connations and adnations occurring during flower development in this group is also due to the fusion of protodermal surfaces and related to the floral evolution of the group. Studies focusing on protoderm are needed to better understand the evolution of epidermal tissues which exude completely different compounds with distinct functions in so many parts of the flower.

Two evolutionary trends may be noted within asclepiads in relation to the glands: 1) redundancy in protection with external and internal glands protecting the flowers against herbivory, microorganism proliferation, meristem desiccation, etc. (*e.g.*, colleters, trichomes, laticifers and idioblasts) and 2) functional division between glands (*e.g.*, primary and secondary nectaries) or between cells of a same secretory tissue (*e.g.*, style head).

The redundancy in relation to the protection of glands is reflected in the low predation rate of these plants, but the division of functions between glands or between cells of the same gland is related to the evolution of the secretory structures in the family. When we analyze the secretory epidermis of the style head from the most basal genera, all cells produce the same type of secretion. On the other hand, in Periplocoideae, Secamonoideae and Asclepiadoideae the thick secretion (translator) produced by this tissue has specific morphology and distinct chemical composition in each part, demonstrating differences in the secretory activity of the cells of the style head.

In spite of the great diversity of glands in the flowers of asclepiads and the occurrence of some specific secretory structures in some genera, the glands related to pollination are relatively constant throughout the group (Tab. 1), making the general description of the pollination more uniform. The pollinator is often attracted by the scent produced by osmophores or by the accumulation of nectar in cups formed by staminal corona. When collecting the nectar in the corona or in the interstaminal position,

the insect introduces the proboscis or leg in the guide rail and can only withdraw it by making a movement forward and upward. The corpusculum secreted by the style head and located above the guide rail adheres to the part of the pollinator's body, thus removing the entire pollinarium from the flower. When collecting nectar from another flower, the insect is again caught by the guide rail and, by making the movement forward and upward, introduces the pollinium into the guide rail or the stigmatic chamber by its basal aperture. The insertion of the pollinium or part of the pollinarium into the guide rail is facilitated by the secretion of the wing gland and the primary nectar, present in the stigmatic chamber, stimulating the germination of the pollen grains, which will grow through the nectar of the chamber to the stigma located below the style head. When the pollen tubes penetrate the gynoecium, they are directed by transmitting tissues to the stylar canal, where they grow immersed in the secretion of the canal until the ovary and then grow immersed in the secretion of the placental-funicular obturator, fertilizing the ovules.

Future perspectives

The floral glands of asclepiads have been poorly studied in structural terms and, despite their simple tissue composition often containing only a secretory epidermis, recent studies have shown that their secretion may be much more complex and heterogeneous than previously thought, demonstrating a high metabolic complexity of their cells. Therefore, new studies are still necessary to verify the actual composition of some secretions, how the exudates are produced by the organelles, the process of secretion release to the outside and the ontogenetic factors related to the formation of the different glands in an evolutionary perspective.

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References

Allen RD, Nessler CL. 1984. Cytochemical localization of pectinase activity in laticifers of *Nerium oleander* L. Protoplasma 119: 74-78.

Appezzato-da-Glória B, Estelita MEM. 1997. Laticifers systems in *Mandevilla illustris* and *M. velutina* Apocynaceae. Acta Societatis Botanicorum Poloniae 66: 301-306.

Appezzato-da-Glória B, Estelita MEM. 2000. Development, structure and distribution of colleters in *Mandevilla illustris* and *M. velutina* (Apocynaceae). Revista Brasileira de Botânica 23: 113-120.

Arekal GD, Ramakrishna TM. 1980. Extrafloral nectaries of *Calotropis gigantea* and *Wattakaka volubilis*. Phytomorphology 30: 303-306.

Baas P, Gregory M. 1985. A survey of oil cells in the dicotyledons with comments on their replacement by and joint occurrence with mucilage cells. Israel Journal of Botany 34: 167-186.

Baas WJ, Warnaar F, Niemann GJ. 1981. Investigations on *Hoya* species. VI. Latex composition and leaf phenolics and their taxonomic significance. Acta Botanica Neerlandica 30: 257-263.

Barroso GM. 1986. Sistemática de angiospermas do Brasil. Vol. 3. Viçosa, Universidade Federal de Viçosa, Imprensa Universitária.

Blaser HW. 1945. Anatomy of *Cryptostegia grandiflora* with special reference to the latex system. American Journal of Botany 32: 135-141.

Bookman SS. 1981. The floral morphology of *Asclepias speciosa* (Asclepiadaceae) in relation to pollination and a clarification in terminology for the genus. American Journal of Botany 68: 675-679.

Brown R. 1810. On the Asclepiadeae, a natural order of plants separated from the Apocineae of Jussieu. Memoirs of the Wernerian Natural History Society 1: 12-78.

Bruyns P. 1993. A revision of *Hoodia* and *Lavrania* (Asclepiadaceae-Stapelieae). Botanische Jahrbücher für Systematik 115: 145-270.

Canaveze Y, Machado SR. 2015. Leaf colleters in *Tabernaemontana catharinensis* (Apocynaceae, Rauvolfioideae): structure, ontogenesis, and cellular secretion. Botany 93: 1-10.

Canaveze Y, Machado SR. 2016. The occurrence of intrusive growth associated with articulated laticifers in *Tabernaemontana catharinensis* A.DC., a new record for Apocynaceae. International Journal of Plant Sciences 177: 458-467.

Capelli NV, Rodrigues BA, Demarco D. 2017. Stipules in Apocynaceae: an ontogenetic perspective. AoB Plants 9: plw083. doi: https://doi.org/10.1093/aobpla/plw083

Carr SGM, Carr DJ. 1961. The functional significance of syncarpy. Phytomorphology 11: 249-256.

Castro MM, Demarco D. 2008. Phenolic compounds produced by secretory structures in plants: a brief review. Natural Product Communications 3: 1273-1284.

Chauveaud MLG. 1891. Recherches embryogèniques sur l'appareil laticifère des Euphorbiacèes, Apocynées et Asclépiadées. Annales des Sciences Naturelles. Botanique et Biologie Vegetale (ser. 7) 14: 1-161.

Christ P, Schnepf E. 1985. The nectaries of *Cynanchum vincetoxicum* (Asclepiadaceae). Israel Journal of Botany 34: 79-90.

Corry TH. 1883. On the structure and development of gynostegium and the mode of fertilisation in *Asclepias cornuti*. Transactions of the Linnean Society of London 2: 173-207.

Cronquist A. 1981. An integrated system of classification of flowering plants. New York, Columbia University Press.

Dave Y, Thomas V, Kuriachen PM. 1987. Structure and development of colleters in *Aganosma caryophyllata* G. Don. Pakistan Journal of Botany 19: 243-248.

Demarco D. 2005. Estruturas secretoras florais e coléteres foliares em espécies de cerrado de *Aspidosperma* Mart. e *Blepharodon* Decne. (Apocynaceae *s.l.*). MSc Thesis, Universidade Estadual de Campinas, Campinas.

Demarco D. 2008. Glândulas de órgãos vegetativos aéreos e florais de espécies de Asclepiadoideae (R.Br.) Duby (Asclepiadoideae, Apocynaceae) de Mata Atlântica do estado de São Paulo. PhD Thesis, Universidade Estadual de Campinas, Campinas.

Demarco D. 2014. Secretory tissues and the morphogenesis and histochemistry of pollinarium in flowers of Asclepiadeae (Apocynaceae). International Journal of Plant Sciences 175: 1042-1053.

Demarco D. 2015. Micromorfología y histoquímica de los laticíferos de órganos vegetativos de especies de Asclepiadoideae (Apocynaceae). Acta Biológica Colombiana 20: 57-65.

Demarco D. 2017. Staminal wing gland: a novel secretory structure of asclepiads. Botany 95: in press. doi: 10.1139/cjb-2016-0239

Demarco D, Castro MM. 2008. Laticíferos articulados anastomosados em espécies de Asclepiadeae (Asclepiadoideae, Apocynaceae) e suas implicações ecológicas. Revista Brasileira de Botânica 31: 699-711.

Demarco D, Kinoshita LS, Castro MM. 2006. Laticíferos articulados anastomosados – novos registros para Apocynaceae. Revista Brasileira de Botânica 29: 133-144.

Dicko-Zafimahova L. 1980. Ultrastructure des parois des pollinies de *Calotropis procera*. Adansonia 17: 455-463.

Die J. 1955. A comparative study of the particle fractions from Apocynaceae latices. Annales Bogorienses 2: 1-124.
Eilert U, Nesbitt LR, Constabel F. 1985. Laticifers and latex in fruits of periwinkle, *Catharanthus roseus*. Canadian Journal of Botany 63: 1540-1546.
Eisikowitch D. 1986. Morpho-ecological aspects on the pollination of *Calotropis procera* (Asclepiadaceae) in Israel. Plant Systematics and Evolution 152: 185-194.
Endress ME, Bruyns PV. 2000. A revised classification of Apocynaceae *s.l.* The Botanical Review 66: 1-56.
Endress ME, Liede-Schumman S, Meve U. 2014. An updated classification for Apocynaceae. Phytotaxa 159: 175-194.
Endress ME, Sennblad B, Nilsson S, *et al*. 1996. A phylogenetic analysis of Apocynaceae *s.str*. and some related taxa in Gentianales: a multidisciplinary approach. Opera Botanica Belgica 7: 59-102.
Endress PK. 1979. Noncarpellary pollination and "hyperstigma" in an angiosperm (*Tambourissa religiosa*, Monimiaceae). Experientia 35: 45.
Endress PK. 1980. Ontogeny, function and evolution of extreme floral construction in Monimiaceae. Plant Systematics and Evolution 134: 79-120.
Endress PK. 1982. Syncarpy and alternative modes of escaping disadvantages of apocarpy in primitive angiosperms. Taxon 31: 48-52.
Endress PK. 1994. Diversity and evolutionary biology of tropical flowers. Cambridge, University Press.
Endress PK. 2016. Development and evolution of extreme synorganization in angiosperm flowers and diversity: a comparison of Apocynaceae and Orchidaceae. Annals of Botany 117: 749-767.
Fahn A. 1979. Secretory tissues in plants. London, Academic Press.
Fahn A. 1990. Plant anatomy. 4th edn. Oxford, Pergamon Press.
Fallen ME. 1986. Floral structure in the Apocynaceae: morphological, functional and evolutionary aspects. Botanishe Jahrbücher für Systematik 106: 245-286.
Farrell BD, Dussourd DE, Mitter C. 1991. Escalation of plant defense: do latex/resin canals spur plant diversification? American Naturalist 138: 881-900.
Fineran BA. 1983. Differentiation of non-articulated laticifers in poinsettia (*Euphorbia pulcherrima* Willd.). Annals of Botany 52: 279-293.
Fjell I. 1983. Anatomy of the xeromorphic leaves of *Allamanda neriifolia*, *Thevetia peruviana* and *Vinca minor* (Apocynaceae). Nordic Journal of Botany 3: 383-392.
Frye TC. 1902. A morphological study of certain Asclepiadaceae. Botanical Gazette 34: 389-413.
Gager CS. 1902. The development of the pollinium and sperm-cells in *Asclepias cornuti*, Decaisne. Annals of Botany 16: 123-148.
Galetto L. 1997. Flower structure and nectar chemical composition in three Argentine Apocynaceae. Flora 192: 197-207.
Galil J, Zeroni M. 1965. Nectar system of *Asclepias curassavica*. Botanical Gazette 126: 144-148.
Gama TSS, Rubiano VS, Demarco D. 2017. Laticifer development and its growth mode in *Allamanda blanchetii* A.DC. (Apocynaceae). Journal of the Torrey Botanical Society 144: in press.
Giordani R. 1978. Autophagie cellulaire et differenciation des laticifères non articulés chez une Asclepiade. Biologie Cellulaire 33: 253-260.
Giordani R. 1996. Les lipids du latex chez *Asclepias curassavica* et *Lactuca sativa*: nature, origine, localisation subcellulaire et rôle. Oleagineux Corps Gras Lipides 3: 89-94.
Giordani R, Lafon L. 1993. A b-D-fucosidase from *Asclepias curassavica* latex. Phytochemistry 33: 1327-1331.
Giordani R, Tolla D, Regli P, Buc J. 2000. Role of terpenes from *Asclepias curassavica* latex for antifungal activity. Journal de Mycologie Medicale 10: 34-38.
Groeneveld HW, Made LA. 1982. Cardenolide and triterpene synthesis in the laticifers of *Asclepias curassavica* L. Acta Botanica Neerlandica 31: 5-10.
Groom P. 1889. On the function of laticiferous tubes. Annals of Botany 3: 157-169.
Hunter JR. 1994. Reconsidering the functions of latex. Trees 9: 1-5.
Judd WS, Campbell CS, Kellogg EA, Stevens PF, Donoghue MJ. 2002. Plant systematics: a phylogenetic approach. 2nd edn. Sunderland, Sinauer Associates.
Judd WS, Sanders RW, Donoghue MJ. 1994. Angiosperm family pairs: preliminary phylogenetic analyses. Harvard Papers in Botany 5: 1-51.
Jürgens A, Dötterl S, Liede-Schumann S, Meve U. 2008. Chemical diversity of floral volatiles in Asclepiadoideae-Asclepiadeae (Apocynaceae). Biochemical Systematics and Ecology 36: 842-852.
Jürgens A, Dötterl S, Liede-Schumann S, Meve U. 2010. Floral scent composition in early diverging taxa of Asclepiadoideae, and Secamonoideae (Apocynaceae). South African Journal of Botany 76: 749-761.
Jussieu AL. 1789. Genera Plantarum. Zürich, Viduam Herissant.
Kevan PG, Eisikowitch D, Rathwell B. 1989. The role of nectar in the germination of pollen in *Asclepias syriaca* L. Botanical Gazette 150: 266-270.
Kunze H. 1991. Structure and function in asclepiad pollination. Plant Systematics and Evolution 176: 227-253.
Kunze H. 1993. Evolution of the translator in Periplocaceae and Asclepiadaceae. Plant Systematics and Evolution 185: 99-122.
Kunze H. 1994. Ontogeny of the translator in Asclepiadaceae *s.str*. Plant Systematics and Evolution 193: 223-242.
Kunze H. 1995. Floral morphology of some Gonolobeae (Asclepiadaceae). Botanische Jahrbücher für Systematik 117: 211-238.
Kunze H. 1997. Corona and nectar system in Asclepiadinae (Asclepiadaceae). Flora 192: 175-183.
Kunze H. 1999. Pollination ecology in two species of *Gonolobus* (Asclepiadaceae). Flora 194: 309-316.
Kunze H, Liede S. 1991. Observations on pollination in *Sarcostemma* (Asclepiadaceae). Plant Systematics and Evolution 178: 95-105.
Lin S, Bernardello G. 1999. Flower structure and reproductive biology in *Aspidosperma quebracho-blanco* (Apocynaceae), a tree pollinated by deceit. International Journal of Plant Science 160: 869-878.
Linskens HF, Suren ML. 1969. Die Entwicklung des Polliniums von *Asclepias curassavica*. Berichte der Deutschen Botanischen Gesellschaft 82: 527-534.
Lopes KLB, Thadeo M, Azevedo AA, Soares AA, Meira RMSA. 2009. Articulated laticifers in the vegetative organs of *Mandevilla atroviolacea* (Apocynaceae, Apocynoideae). Botany 87: 202-209.
Mahlberg PG. 1963. Development of nonarticulated laticifer in seedling axis of *Nerium oleander*. Botanical Gazette 124: 224-231.
Mahlberg PG. 1993. Laticifers: an historical perspective. The Botanical Review 59: 1-23.
Martins FM. 2012. Leaf and calycine colleters in *Odontadenia lutea* (Apocynaceae - Apocynoideae – Odontadenieae): their structure and histochemistry. Brazilian Journal of Botany 35: 59-69.
Martins FM, Kinoshita LS, Castro MM. 2010. Coléteres foliares e calicinais de *Temnadenia violacea* (Apocynaceae, Apocynoideae): estrutura e distribuição. Revista Brasileira de Botânica 33: 489-500.
Metcalfe CR. 1967. Distribution of latex in the plant kingdom. Economic Botany 21: 115-127.
Metcalfe CR, Chalk L. 1950. Anatomy of the dicotyledons: leaves, stem and wood in relation to taxonomy with notes on economic uses. Vol. 2. Oxford, Claredon Press.
Meve U, Liede S. 1994. Floral biology and pollination in stapeliads – new results and a literature review. Plant Systematics and Evolution 192: 99-116.
Milanez FR. 1960/1961. Contribuição ao conhecimento anatômico de *Cryptostegia grandiflora* - II. Sobre os laticíferos da estrutura primária (Asclepiaceae). Rodriguésia 35/36: 99-128.
Milanez FR. 1966. Contribuição ao conhecimento anatômico de *Cryptostegia grandiflora* – III. Nota sobre a estrutura secundária. Rodriguésia 25: 335-350.
Milanez FR. 1977. Ontogênese dos laticíferos contínuos de *Neridium* (*Nerium) oleander* L. Trabalhos do XXVI Congresso Nacional de Botânica, Rio de Janeiro 1975: 343-379.
Monteiro MM, Demarco D. 2017. Corona development and the floral nectaries in Asclepiadeae (Asclepiadoideae, Apocynaceae). Acta Botanica Brasilica 31: 420-432.
Morillo G. 1998. *Matelea gracieae* Morillo, a new species from French Guiana, and *Cynanchum gortsianum* Morillo, a new record for Suriname. Brittonia 50: 296-300.

Morokawa R, Mayer JLS, Simões AO, Kinoshita LS. 2015. Floral development of *Condylocarpon isthmicum* (Apocynaceae). Botany 93: 769-781.

Murphy H. 1986. A revision of the genus *Fischeria* (Asclepiadaceae). Systematic Botany 11: 229-241.

Murugan V, Inamdar JA. 1987a. Studies in the laticifers of *Vallaris solanacea* (Roth) O. Ktze. Phytomorphology 37: 209-214.

Murugan V, Inamdar JA. 1987b. Organographic distribution, structure and ontogeny of laticifers in *Plumeria alba* Linn. Proceedings of the Indian Academy of Sciences (Plant Sciences) 97: 25-31.

Pacini E, Franchi GG, Hesse M. 1985. The tapetum: its form, function, and possible phylogeny in Embryophyta. Plant Systematics and Evolution 149: 155-185.

Pacini E, Hesse M. 2005. Pollenkitt - its composition, forms and functions. Flora 200: 399-415.

Pereira JF, Schwarz EA. 1983. Contribuição ao estudo das Asclepiadaceae brasileiras. XX. Uma nova espécie de *Gonioanthela* Malme. Atas da Sociedade Botânica do Brasil 1: 71-74.

Plachno BJ, Swiatek P, Szymczak G. 2010. Can a stench be beautiful? Osmophores in stem-succulent stapeliads (Apocynaceae-Asclepiadoideae-Ceropegieae-Stapeliinae). Flora 205: 101-105.

Postek MT, Tucker SC. 1983. Ontogeny and ultrastructure of secretory oil cells in *Magnolia grandiflora* L. Botanical Gazette 144: 501-512.

Ramayya N, Bahadur B. 1968. Morphology of the "squamellae" in the light of their ontogeny. Current Science 18: 520-522.

Rao AR, Malaviya M. 1966. The non-articulated laticifers and latex of *Tabernaemontana coronaria* Willd. Proceedings of the National Institute of Sciences of India 32: 233-242.

Rao VS, Ganguli A. 1963. The floral anatomy of some Asclepiadaceae. Proceedings of the Indian Academy of Sciences (B) 57: 15-44.

Rapini A. 2000. Asclepiadaceae ou Asclepiadoideae? Conceitos distintos de agrupamento taxômico. Hoehnea 27: 121-130.

Rapini A. 2012. Taxonomy "under construction": advances in the systematics of Apocynaceae, with emphasis on the Brazilian Asclepiadoideae. Rodriguésia 63: 75-88.

Rapini A, Chase MW, Goyder DJ, Griffiths J. 2003. Asclepiadeae classification: evaluating the phylogenetic relationships of New World Asclepiadoideae (Apocynaceae). Taxon 52: 33-50.

Ribeiro JC, Ferreira MJP, Demarco D. 2017. Colleters in Asclepiadoideae (Apocynaceae): protection of meristems against desiccation and new functions assigned. International Journal of Plant Sciences 178: in press.

Rio MCS, Castro MM, Kinoshita LS. 2002. Distribuição e caracterização anatômica dos coléteres foliares de *Prestonia coalita* (Vell.) Woodson (Apocynaceae). Revista Brasileira de Botânica 25: 339-349.

Rio MCS, Kinoshita LS. 2005. *Prestonia* (Apocynaceae) no sul e sudeste do Brasil. Hoehnea 32: 233-258.

Rio MCS, Kinoshita LS, Castro MM. 2005. Anatomia foliar como subsídio para a taxonomia de espécies de *Forsteronia* G. Mey. (Apocynaceae) dos cerrados paulistas. Revista Brasileira de Botânica 28: 713-726.

Rohrbeck D, Buss D, Effmert U, Piechulla B. 2006. Localization of methyl benzoate synthesis and emission in *Stephanotis floribunda* and *Nicotiana suaveolens* flowers. Plant Biology 8: 615-616.

Sacchetti G, Ballero M, Serafini M, Romagnoli C, Bruni A, Poli F. 1999. Laticifer tissue distribution and alkaloid location in *Vinca sardoa* (Stearn) Pign. (Apocynaceae), an endemic plant of Sardinia (Italy). Phyton 39: 265-275.

Safwat FM. 1962. The floral morphology of *Secamone* and the evolution of the pollinating apparatus in Asclepiadaceae. Annals of the Missouri Botanical Garden 49: 95-129.

Sage TL, Broyles SB, Wyatt R. 1990. The relationship between the five stigmatic chambers and two ovaries of milkweed (*Asclepias amplexicaulis* Sm.) flowers: a three-dimensional assessment. Israel Journal of Botany 39: 187-196.

Sage TL, Williams EG. 1995. Structure, ultrastructure, and histochemistry of the pollen tube pathway in the milkweed *Asclepias exaltata* L. Sexual Plant Reproduction 8: 257-265.

Schill R, Dannenbaum KC. 1984. Bau und Entwicklung der Pollinien von *Hoya carnosa* (L.) Br. (Asclepiadaceae). Tropische und Subtropische Pflanzenwelt 48: 1-54.

Schill R, Jäkel U. 1978. Beiträge zür Kenntnis der Asclepiadaceen-Pollinarien. Tropische und Subtropische Pflanzenwelt 22: 1-122.

Schnepf E, Christ P. 1980. Unusual transfer cells in the epithelium of the nectaries of *Asclepias curassavica* L. Protoplasma 105: 135-148.

Schnepf E, Witzig F, Schill R. 1979. Über Bildung und Feinstruktur des Translators der Pollinarien von *Asclepias curassavica* und *Gomphocarpus fruticosus* (Asclepiadaceae). Tropische und Subtropische Pflanzenwelt 25: 1-33.

Schumann K. 1895. Asclepiadaceae. In: Engler A, Prantl K. (eds.) Die natürlichen Pflanzenfamilien. Vol. 4. Leipzig, Wilhelm Engelmann. p. 189-306.

Schwarz EA, Furlan A. 2002. Coléteres foliares de *Oxypetalum* R.Br. (Asclepiadoideae, Apocynaceae) - aspectos ultraestruturais e anatômicos úteis à taxonomia das espécies do Paraná (Brasil). Acta Biológica Paranaense 31: 79-97.

Sennblad B, Bremer B. 1996. The familial and subfamilial relationships of Apocynaceae and Asclepiadaceae evaluated with *rbc*L data. Plant Systematics and Evolution 202: 153-175.

Sennblad B, Bremer B. 2002. Classification of Apocynaceae *s.l.* according to a new approach combining Linnaean and phylogenetic taxonomy. Systematic Biology 51: 389-409.

Serbanescu-Jitariu G, Tarnavschi IT. 1976. Observations regarding the structure of the pollinaria of some representatives of the family Asclepidaceae. Bulletin de la Société d'Historie Naturelle de l'Afrique du Nord 67: 19-41.

Serpe MD, Muir AJ, Driouich A. 2002. Immunolocalization of b-D-glucans, pectins, and arabinogalactan-proteins during intrusive growth and elongation of nonarticulated laticifers in *Asclepias speciosa* Torr. Planta 215: 357-370.

Serpe MD, Muir AJ, Keidel AM. 2001. Localization of cell wall polysaccharides in nonarticulated laticifers of *Asclepias speciosa* Torr. Protoplasma 216: 215-226.

Setzer WN. 2014. Chemical composition of the floral essential oil of *Tabernaemontana longipes* from Monteverde, Costa Rica. Americal Journal of Essential Oils and Natural Products 1: 16-18.

Silva NMF, Valente MC, Alencastro FMMR, Pereira JF, Sucre BD. 1975. Contribuição ao estudo das Asclepiadaceae brasileiras. X. Estudos taxonômico e anatômico de: *Gonioanthela odorata* (Decne.) Malme e *Gonioanthela hilariana* (Fourn.) Malme. Revista Brasileira de Biologia 35: 745-756.

Simões AO, Castro MM, Kinoshita LS. 2006. Calycine colleters of seven species of Apocynaceae (Apocynoideae) from Brazil. Botanical Journal of the Linnean Society 152: 387-398.

Solereder H. 1908. Systematic anatomy of the dicotyledons. Vol. 2. Oxford, Clarendon Press.

Stevens WD. 1975. Notes on the genus *Matelea* (Apocynaceae *s.l.*). Phytologia 32: 387-406.

Stevens WD. 1988. A synopsis of *Matelea* subg. *Dictyanthus* (Apocynaceae: Asclepiadoideae). Annals of the Missouri Botanical Garden 75: 1533-1564.

Struwe L, Albert A, Bremer B. 1994. Cladistics and family level classification of Gentianales. Cladistics 10: 175-206.

Subramanian RB, Murugan V, Mohan JSS, Inamdar JA. 1989. Optical microscopic studies on the structure and secretion of resin glands in some Apocynaceae. Proceedings of Indian Academy Sciences (Plant Sciences) 99: 423-429.

Swarupanandan K, Mangaly JK, Sonny TK, Kishorekumar K, Chand Basha S. 1996. The subfamilial and tribal classification of the family Asclepiadaceae. Botanical Journal of the Linnean Society 120: 327-369.

Thomas V. 1991. Structural, functional and phylogenetic aspects of the colleter. Annals of Botany 68: 287-305.

Thomas V, Dave Y. 1989a. Histochemistry and senescence of colleters of *Allamanda cathartica* L. (Apocynaceae). Annals of Botany 64: 201-203.

Thomas V, Dave Y. 1989b. The colleters of *Alstonia scholaris* L. (Apocynaceae). Indian Botanical Contactor 6: 25-29.

Thomas V, Dave Y. 1989c. Structure, origin, development and senescence of colleters in *Nerium indicum* Mill. (*N. odorum* Soland., Apocynaceae). Korean Journal of Botany 32: 163-172.

Thomas V, Dave Y. 1991. Comparative and phylogenetic significance of colleters in Apocynaceae. Feddes Repertorium 102: 23-28.

Thomas V, Dave Y. 1994. Significance of follicle anatomy of Apocynaceae. Acta Societatis Botanicorum Poloniae 63: 9-20.
Thomas V, Dave Y, Menon ARS. 1989. Anatomy and histochemistry of colleters in *Roupelia grata* Wall. (Apocynaceae). Nordic Journal of Botany 8: 493-496.
Thurston EL. 1974. Morphology, fine structure, and ontogeny of the stinging emergence of *Urtica dioica*. American Journal of Botany 61: 809-817.
Thurston EL. 1976. Morphology, fine structure, and ontogeny of the stinging emergence of *Tragia ramosa* and *T. saxicola* (Euphorbiaceae). American Journal of Botany 63: 710-718.
Thurston EL, Lersten NR. 1969. The morphology and toxicology of plant stinging hairs. The Botanical Review 35: 393-412.
Tiagi B, Dixit G. 1965. Studies in the floral anatomy of some Asclepiadaceae. Bulletin of the Botanical Society of Bengal 19: 111-123.
Valente MC. 1977. A flor de *Oxypetalum banksii* Roem. et Schult. subsp. *banksii*. Estudo da anatomia e vascularização (Asclepiadaceae). Rodriguésia 29: 161-283.
Valente MC. 1980. A flor de *Oxypetalum banksii* Roem. et Schult. subsp. *corymbiferum* (Fourn.) Font. et Val., comb. nov. - vascularização floral. Rodriguésia 32: 81-98.
Valente MC. 1983. Vascularização floral em *Peplonia nitida* Decaisne (Asclepiadaceae). Atas da Sociedade Botânica do Brasil 1: 55-62.
Valente MC. 1984. *Ditassa eximia* Decne (Asclepiadaceae). Anatomia vegetal. Atas da Sociedade Botânica do Brasil 2: 53-59.
Valente MC. 1994. Germinação dos polínios em *Matelea maritima* subsp. *ganglinosa* (Vell.) Font. (Asclepiadaceae). Atas da Sociedade Botânica do Brasil 3: 129-135.
Valente MC. 1995. *Matelea maritima* subsp. *ganglinosa* (Vell.) Font. - Anatomia e vascularização floral (Asclepiadaceae). Arquivos do Jardim Botânico do Rio de Janeiro 33: 75-98.
Valente MC. 1996. *Matelea maritima* subsp. *ganglinosa* (Vell.) Font. – Anatomia vegetal (Asclepiadaceae). Arquivos do Jardim Botânico do Rio de Janeiro 34: 145-176.
Valente MC, Costa CG. 2005. Estudo anatômico da flor de *Marsdenia loniceroides* E. Fournier (Asclepiadoideae - Apocynaceae). Rodriguésia 56: 51-66.
Valente MC, Pereira JF, Alencastro FMMR. 1973. Contribuição ao estudo das Asclepiadaceae brasileiras. IX – Estudos taxonômico e anatômico de: *Oxypetalum appendiculatum* Mart., *Oxypetalum pilosum* Gardn. e *Oxypetalum sublanatum* Malme. Anais da Academia Brasileira de Ciências 45: 121-149.
Valente MC, Silva NMF. 1984. Anatomia floral de *Barjonia erecta* (Vell.) Schum. (Asclepiadaceae). Rodriguésia 36: 95-106.
Vieira MF, Shepherd GJ. 2002. *Oxypetalum banksii* subsp. *banksii*: a taxon of Asclepiadaceae with an extragynoecial compitum. Plant Systematics and Evolution 233: 199-206.
Vijayaraghavan MR, Cheema K. 1977. Ontogenetical and histochemical studies on the translator apparatus in *Calotropis procera* R.Br. I. The retinaculum. Acta Histochemica 59: 15-20
Vijayaraghavan MR, Shukla AK. 1976. The nature of covering around the aggregate of microspores in *Pergularia daemia* (Forsk.) McC. & Blat. Annals of Botany 40: 417-421.
Vogel S. 1990 The role of scent glands in pollination. On the structure and function of osmophores. New Delhi, Amerind Publishing.
Walker DB. 1975. Postgenital carpel fusion in *Catharanthus roseus* (Apocynaceae). I. Light and scanning electron microscopic study of gynoecial ontogeny. American Journal of Botany 64: 457-467.
Warnaar F. 1982. Investigation on *Hoya* species. V. Determination of the amount of latex present in *Hoya australis* R.Br. ex Traill. and *Hoya bella* Hook. and its relation with shoot development. Zeitschrift fur Pflanzenphysiologie 105: 307-314.
Wiemer AP, Sérsic AN, Marino S, Simões AO, Cocucci AA. 2012. Functional morphology and wasp pollination of two South American asclepiads (Asclepiadoideae-Apocynaceae). Annals of Botany 109: 77-93.
Wilson KJ, Nessler CL, Mahlberg PG. 1976. Pectinase in *Asclepias* latex and its possible role in laticifer growth and development. American Journal of Botany 63: 1140-1144.
Wolff D, Meve U, Liede-Schumann S. 2008. Pollination ecology of Ecuadorian Asclepiadoideae (Apocynaceae): how generalized are morphologicallyspecialized flowers? Basic and Applied Ecology 9: 24-34.
Woodson RE Jr. 1941. The North American Asclepiadaceae. I. Perspective of the genera. Annals of the Missouri Botanical Garden 28: 193-244.
Woodson RE Jr. 1954. The North American species of *Asclepias* L. Annals of the Missouri Botanical Garden 41: 1-211.
Woodson RE Jr, Moore JA. 1938. The vascular anatomy and comparative morphology of Apocynaceae flowers. Bulletin of the Torrey Club 65: 135-166.
Wyatt R, Broyles SB. 1994. Ecology and evolution of reproduction in milkweeds. Annual Review of Ecology and Systematics 25: 423-441.
Wyatt R, Lipow SR. 2007. A new explanation for the evolution of pollinia and loss of carpel fusion in *Asclepias* and the Apocynaceae *s.l.* Annals of the Missouri Botanical Garden 94: 474-484.
Yang L-L, Li H-L, Wei L, *et al*. 2016 A supermatrix approach provides a comprehensive genus-level phylogeny for Gentianales. Journal of Systematics and Evolution 54: 400-415.
Yoder LR, Mahlberg PG. 1976. Reactions of alkaloid and histochemical indicators in laticifers and specialized parenchyma cells of *Catharanthus roseus* (Apocynaceae). American Journal of Botany 63: 1167-1173.