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## Comparative Anatomy of Midrib and its Significance in the Taxonomy of the Family Asteraceae from Nigeria

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### ABSTRACT

In this study, we investigated the comparative anatomy of 17 species from 14 genera of Asteraceae occurring in Nigeria. Dried materials obtained from University of Port Harcourt Herbarium (UPH) were used for this study. The midrib of specimens were hand sectioned, stained with 1% safranin or alcian blue, mounted on slide, observed under microscope and micro-photographed using Leica WILD MPS 52 microscope camera on Leitz Diaplan microscope. The results showed that twelve species *Aspilia africana*, *Chromolaena odorata*, *Conyza sumatrensis*, *Emilia preatamissa*, *Eleutheranthera ruderalis*, *Metanthera scandens*, *Synedrella nodiflora*, *Tithonia diversifolia*, *Tridax procumbens*, *Vernonia cinaræ*, *Vernonia biafrae* and *Emilia coccinea* have secretory duct while *Adenostemma* sp., *Ageratum conyzoides*, *Bidens pilosa*, *Eclipta alba* and *Vernonia amygdalina* not have secretory ducts. The number of abaxial and adaxial parenchymatous cells, shape of the midrib, number and arrangement of the vascular bundle varied from one species or genera to other. These characters are dependable in delimitating the family.

**Key words:** Asteraceae, midrib, secretory ducts, vascular bundle, parenchymatous cells

### INTRODUCTION

The Asteraceae family consists of predominantly herbaceous but includes some woody and arboreal types (Metcalf and Chalk, 1950; Cronquist, 1981) and has approximately 25000 species and over 1100 genera (Barroso, 1986). In West Africa, the family is represented by 84 genera and 273 species (Hutchinson and Dalziel, 1954). Members of Asteraceae exhibit considerable anatomical diversity, which is attributed to their ecological specialization and could account for their occurrence in different habitats (Metcalf and Chalk, 1950). Among the members of this family, secretory structures are of great taxonomical interest and their restricted distribution has an important diagnostic value (Metcalf and Chalk, 1950; Fahn, 1979; Castro *et al.*, 1997; Solereder, 1908). Also, difference in vascular bundle types and presence of secretory cell in the phloem varied from species to species (Makbul *et al.*, 2011). In addition the differences in the midrib shape could be used to classify some members of this family (Noorbakhsh *et al.*, 2008).

Members of this family have a remarkable ecological and economical importance. These include ethno-botanical, phytochemical, antimicrobial and medicinal purposes (Kamboj and Saluja, 2008; Abii and Onuoha, 2011; Chono *et al.*, 2014; Bartolome *et al.*, 2013; Adebayo *et al.*, 2010; Ajiboye *et al.*, 2014; Teke *et al.*, 2007; Toyang and Verpoorte, 2013). In Nigeria, most of these specimens used for treatment of illnesses by the Traditional Medicine Practitioners (TMP) are sort from the local markets and are collected from the wild by the locals who may not be able to identify

the plants scientifically. Also, the traditional medicine practitioners in the urban areas rely on the dried specimens from the rural communities for the preparation of concoctions and decoctions before they administer same to their patients (unpublished). The need for an alternative way of identifying these specimens becomes inevitable. Therefore, this study seeks to explore the information from the midrib anatomy to complement the existing data in delimitating the species of this family.

## MATERIALS AND METHODS

Seventeen species from 14 genera of Asteraceae occurring in Nigeria were investigated. Dried materials were obtained from specimens in University of Port Harcourt Herbarium (UPH) and the information about the voucher specimens are listed in Table 1.

The specimens were boiled in hot water for 30 min and the cut sections of the midrib fixed in 70% ethanol. The specimen is trimmed, insert in wax block and hand-sectioned. Thereafter, thin sections selected, stained with 1% safranin or alcian blue, mounted on slide, observed under microscope and micro-photographed using Leica WILD MPS 52 microscope camera on Leitz Diaplan microscope. This method is modified by Metcalfe and Chalk (1979) and Cutler (1978).

## RESULTS AND DISCUSSION

The results of this study are presented in Table 2 and Fig. 1. Seventeen species belonging to 14 genera were studied. Among these species, twelve have secretory *Adenostemma* sp., *Ageratum conyzoides*, *Bidens pilosa*, *Eclipta alba* and *Vernonia amygdalina* do not secretory ducts. The species with secretory ducts include; *Aspilia africana* (Fig. 1a-b), *Chromolaena odorata* (Fig. 1e), *Conyza sumatrensis* (Fig. 1f), *Emilia preatamissa* (Fig. 1h), *Eleutheranthera ruderalis* (Fig. 1i), *Metanthera scandens* (Fig. 1j), *Synedrella nodiflora* (Fig. 1k-l), *Tithonia diversifolia* (Fig. 1m-n), *Tridax procumbens* (Fig. 1o), *Vernonia cinaræ* (Fig. 1q), *Vernonia biafrae* (Fig. 1r) and *Emilia coccinea* (Fig. 1s). This character has been used to delimit the members of this family (Metcalf and Chalk, 1950; Makbul *et al.*, 2011; Noorbakhsh *et al.*, 2008; Castro *et al.*, 1997; Fahn, 1979; Solereder, 1908). In their studies, Metcalfe and Chalk (1950) noted the presence of

Table 1: Description of voucher specimens studied form University of Port Harcourt Herbarium

Species	Herbarium number	Locality and date of collection
<i>Adenostemma</i> sp.	UPH 1256	Umualum Ngor Okpoala, Imo State
<i>Ageratum conyzoides</i> L.	UPH 1276	Behind College of Natural and Applied Sciences Complex, Abuja Park, University of Port Harcourt, Rivers State
<i>Aspilia africana</i> (Pers.) C.D. Adams	UPH 1356	Alakahia, Rivers State
<i>Bidens pilosa</i> L.	UPH 1256	Choba, Rivers State
<i>Chromolaena odorata</i> (L.) King and H.E. Robins.	UPH 1258	Obiga-Asa, Abia State
<i>Conyza sumatrensis</i> (Retz.) Walker	UPH 1253	Alakahia, Rivers State
<i>Eclipta alba</i> (L.) Hassk.	UPH 1346	Choba, Rivers State
<i>Eleutheranthera ruderalis</i> (Sw.) Sch. Bip.	UPH 1456	Choba, Rivers State
<i>Emilia coccinea</i> (Sims) G. Don.	UPH 1267	Eleme, Rivers State
<i>Emilia preatamissa</i> Milne-Redhead	UPH 1258	Alakahia, Rivers State
<i>Metanthera scandens</i> (Schum. and Thonn.) Roberty	UPH 1276	Beside UBA Bank, Abuja Park University of Port Harcourt, Rivers State
<i>Synedrella nodiflora</i> (L.) Gaertn.	UPH 1296	Ecological Centre, Abuja Park, University of Port Harcourt, Rivers State
<i>Tithonia diversifolia</i> (Hemsl.) A.Gray	UPH 1288	Opposite Cricket Court, Abuja Park, University of Port Harcourt, Rivers State
<i>Tridax procumbens</i> L.	UPH 2252	Junior Staff Quarters, Abuja Park, University of Port Harcourt, Rivers State
<i>Vernonia amygdalina</i> Delile	UPH 1256	Obiga-Asa, Abia State
<i>Vernonia biafrae</i> Oliv. and Hiern.	UPH 2353	Opposite NNPC Refinery, Eleme, Rivers State
<i>Vernonia cinerea</i> (Linn) Less	UPH 1250	Goelee Ltd, TDU Site, Eleme, Rivers State

Table 2: Anatomical characteristics of the midrib of the Asteraceae species studied

Species	No. of vascular bundle	Nature of vascular bundle	No. of abaxial parenchyma	No. of adaxial parenchyma	Nature of midrib	Secretory duct
<i>Adenostemma</i> sp.	3 separate traces	Arced	6-7 layers	6-7 layers	Elevated forming acute angle or curved surface	Absent
<i>Ageratum conyzoides</i>	Single trace	Oval	4-5 layers	6-7 layers	Elevated to form an obtuse angle at the apex	Absent
<i>Aspilia africana</i>	More than 13 with strand of medullary ones	Arced	6-8 layers	8-14 layers	Elevated to form more or less acute angle with the leaf blades	Present
<i>Bidens pilosa</i>	Single trace	Oval	3-5 layers	3-4 layers	Relatively flat.	Absent
<i>Chromolaena odorata</i>	5 (3 separate traces and 2 medullary ones)	Arced	4-6 layers	5-7 layers	Elevated with flattened at the apex	Present
<i>Conyza sumatrensis</i> (Retz.) Walker	Single trace	Oval	6-7 layers	4-7 layers	Relatively flat.	Present
<i>Eclipta alba</i>	3 separate traces	Arced	5-7 layers	5-7 layers	Elevated but relatively flat	Absent
<i>Eleutheranthera ruderalis</i>	3 separate traces	Arced	9-13 layers	5-8 layers	Elevated forming obtuse angle or curved surface	Present
<i>Emilia coccinea</i>	3 separate traces	Arced	4-6 layers	6-7 layers	Relatively flat to concave	Present
<i>Emilia preatamissa</i>	Single trace	Oval	5-6 layers	4-5 layers	Not elevated but flat	Present
<i>Metanthera scandens</i>	6 (4 close traces and 2 medullary ones)	Arced	7-9 layers	10-12 layers	Elevated forming obtuse angle or curved surface	Present
<i>Synedrella nodiflora</i>	3 separate traces	Arced	6-10 layers	3-4 layers	Elevated with flat apex	Present
<i>Tithonia diversifolia</i>	8 with 3 medullary ones	Arced	9-11 layers	12-14 layers	Elevated with curved apex	Present
<i>Tridax procumbens</i>	Single trace	Oval	5-6 layers	6-7 layers	Relatively flat	Present
<i>Vernonia amygdalina</i>	7 separate bundles (5 large and 2 small ones)	V or U-shaped	10-13 layers	15-20 layers	Elevated but flattened at the apex	Absent
<i>Vernonia biafrae</i>	Single trace	U-shaped	More than 25 layers	14-16 layers	Elevated forming obtuse angle or curved surface	Present
<i>Vernonia cinerea</i>	Single trace	Oval	7-10 layers	6-9 layers	Elevated forming acute angle or curved surface	Present

secretory ducts in *Chromolaena*, *Ageratum*, *Vernonia* and *Tridax*. Similarly, this character was observed in *Chromolaena*, *Ageratum*, *Tridax* and *Vernonia*. In addition, secretory ducts were found in *Aspilia*, *Eleutheranthera*, *Synedrella*, *Emilia*, *Conyza metanthera* and *Tithonia*. Makbul *et al.* (2011) used vascular bundle types, presence/absence of cavity on midrib, presence or absence of secretory cells in phloem to distinguish the members of the genus *Scorzonera* L. (Asteraceae) from North East Anatolia. They noted that some of the species have cavity in the midrib while others do not and that the occurrence of different vascular bundle types and presence of secretory cell in the phloem varied from species to species. Noorbakhsh *et al.* (2008) noted the differences in the midrib shape of *Artamisia* as a diagnostic character while the presence of secretory ducts and cavities were used by Castro *et al.* (1997), Fahn (1979) and Solereder (1908) to delimit other members of Asteraceae.

Despite the occurrence of the secretory ducts, the nature of abaxial and adaxial parenchymatous cells, the shape, number and arrangement of vascular bundles in the midrib vary among the species studied and could be used to distinguish them. For instance, the midrib of *A. africana* has arced vascular bundle with more than 13 traces including medullary ones. The parenchymatous cells in the abaxial and adaxial epidermises are 6-8 and 8-14 layers, respectively. Also, the midrib is elevated at the middle to form more or less acute angle with the leaf blades (Fig. 1a-b). *Chromolaena odorata* midrib has 5 vascular bundles (3 separate traces and 2 medullary ones) with the abaxial and adaxial parenchymatous cells 4-6 and 5-7 layers, respectively and the middle portion of the midrib elevated but flattened at the apex (Fig. 1e). Furthermore, *C. sumatrensis* has single trace of vascular bundle with oval shape. The middle portion of the midrib is relatively flat with the abaxial parenchymatous cells 6-7 layers and the adaxial cells 4-7 layers (Fig. 1f). On the other hand, the midrib of *E. ruderalis* has 3 separate traces of vascular bundles, 9-13 layers of cells at the abaxial surface and 5-8 layers on the adaxial surface. It is also elevated forming obtuse angle or convex surface (Fig. 1i). In *T. diversifolia* the midrib is elevated with curved apex and has 10 vascular bundles including 3 medullary ones with

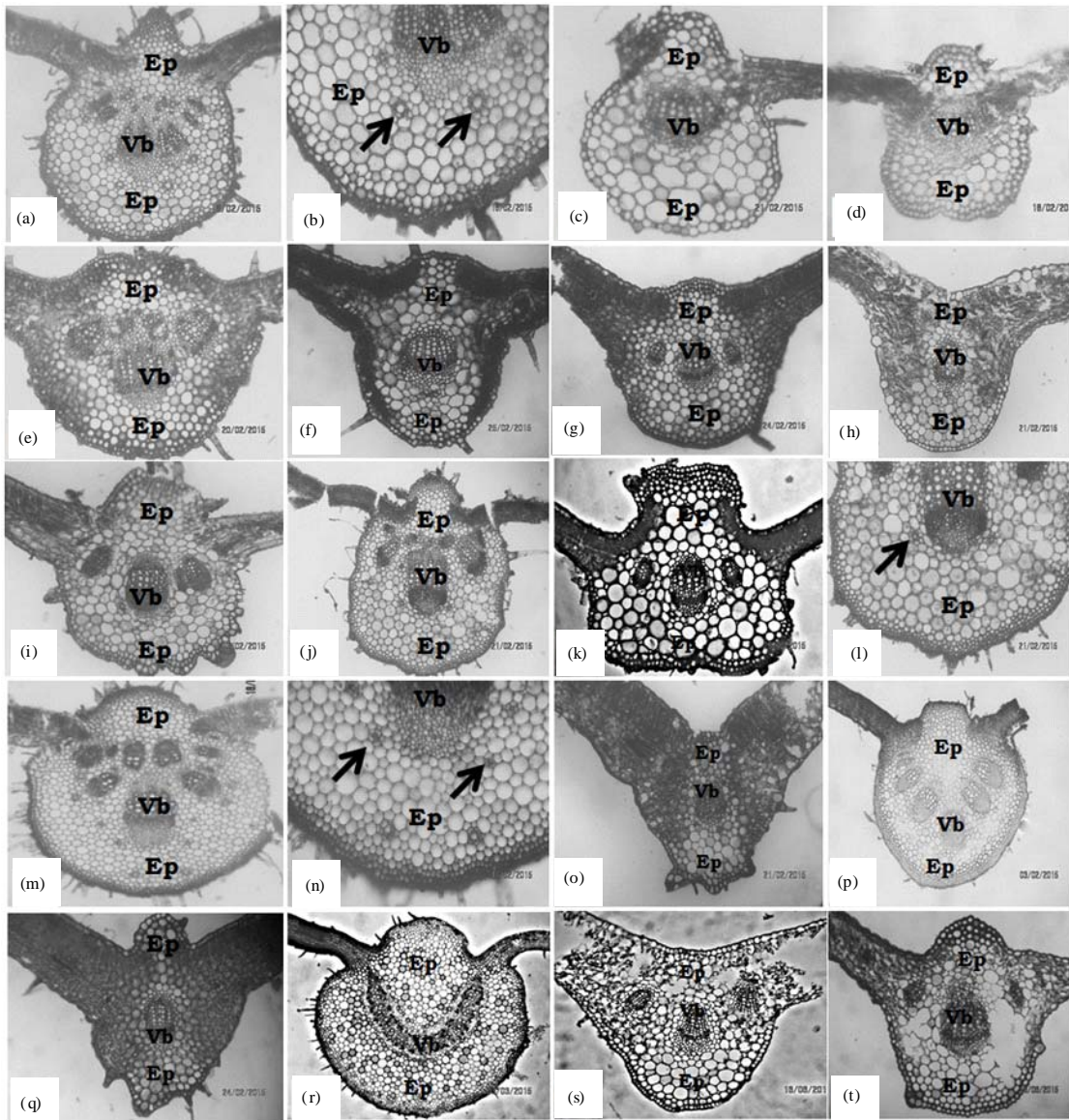


Fig. 1(a-t): Anatomical sections showing the transverse section of the midribs of (a and b) *Aspilia africana*, (c) *Ageratum conyzoides*, (d) *Bidens pilosa*, (e) *C. odorata*, (f) *Conyza sumatrensis*, (g) *Eclipta alba*, (h) *E. preatamissa*, (i) *Eleutheranthera ruderalis*, (j) *M. scandens*, (k and l) *Synedrella nodiflora*, (m and n) *T. diversifolia*, (o) *Tridax procumbense*, (p) *Vernonia amygdalina*, (q) *Vernonia cinaræ*, (r) *Vernonia biafrae*, (s) *Emilia coccinea* and (t) *Adenostemma* sp., Ep: Epidermis, Vb: Vascular bundle

arced shape. The abaxial and adaxial parenchymatous cells have 9-11 and 12-14 layers, respectively (Table 2 and Fig. 1m-n). In *T. procumbens*, the vascular bundle has single trace with oval shape. The abaxial and adaxial parenchymatous cells are 5-6 and 6-7 layers, respectively and the midrib relatively flat or concave (Fig. 1o). This same differences was also observed in *M. scandens* and *S. nodiflora*. The vascular bundles in *M. scandens* are 6 (4 close traces and 2 medullary ones) (Fig. 1j) while in *S. nodiflora* it is 3 separate traces (Fig. 1k-l).

The shapes of the midribs in the two *Emilia* species studied are relatively same however; the number of vascular bundles in the midrib could be used to distinguish them. For example, the midrib of *E. preatamissa* has one-trace of vascular bundle (Fig. 1h) while *E. coccinea* has 3-traces of vascular bundles (Fig. 1s). Among the genus *Vernonia* studied, the shape of the midrib, number of vascular bundles and parenchymatous layers in the abaxial and adaxial surfaces of the midrib varied from species to species. For instance, *V. amygdalina* has 7 separate bundles (5 large and 2 small ones) V or U-shaped, the abaxial parenchyma 10-13 layers and the adaxial parenchyma 15-20 layers while the midrib is elevated but flattened at the apex (Fig. 1p). In *V. biafrae* the vascular bundle has single trace and U-shaped, abaxial parenchymatous cell more than 25 layers while the adaxial surface is 14-16 layers and the midrib is elevated forming obtuse angle or curved surface (Fig. 1q) and in *V. cinerea* the vascular bundle has single trace, oval in shape, the abaxial and adaxial parenchymatous cells 7-10 and 6-9 layers, respectively while the midrib elevated forming acute angle or curved surface.

Considering the species without the secretory ducts, the midrib of *A. conyzoides* has single trace of vascular bundle, 4-5 layers of abaxial and 6-7 layers adaxial parenchymatous cells and the midrib elevated to form an obtuse angle at the apex (Fig. 1c). Similarly, in *B. pilosa*, the vascular bundle has one-trace, the middle of the midrib relatively flat or concave with 3-5 layers of abaxial and 3-4 layers of adaxial parenchymatous cells. In contrast, *E. alba* has 3 separate traces of vascular bundles and 5-7 layers of parenchymatous cells in both abaxial and adaxial surfaces while the midrib is elevated but relatively flat at the apex (Fig. 1g). Also *Adenostemma* sp. has arced vascular bundles with 3 separate traces, 6-7 layers of parenchymatous cells abaxial and adaxial each. The middle portion of the midrib elevated forming acute angle or convex surface (Fig. 1t).

This present findings indicates that the dried specimens from these species could be identified based on the presence or absence of secretory duct, the number of abaxial and adaxial parenchymatous cells, shape of the midrib, number and arrangement of the vascular bundle varied from one species or genera to other. These characters are dependable in delimitating the family and support the previous works by Metcalfe and Chalk (1950), Makbul *et al.* (2011), Noorbakhsh *et al.* (2008), Castro *et al.* (1997), Fahn (1979) and Solereder (1908) on Asteraceae.

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