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Research Paper

Comparative foliar epidermal morphology of the West African species of Amaranthaceae JUSS.

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Abstract

The species are generally amphistomatic except *Pandiaka heudelotii* that is hypostomatic. Anomocytic and paracytic stomatal types were mostly found but cyclocytic and diacytic types may accompany, especially in *Cyathula* spp. The anticlinal wall pattern varies from undulate to curved and curved-straight in the family. Other useful epidermal features are peltate scales, trichomes of different types such as candelabra type, unicellular and multicellular conical types, long acicular glandular forms that may be tuberculate or non-tuberculate and the filiform type; all can be employed at species level for delimitation purpose. Sub-familial grouping of the genera is supported while monophyly and polyphyly suggested in Gomphrenoideae and Amarantheae respectively can be inferred from the extent of character variations in the species. An indented dichotomous key is presented for species delimitation.

Introduction

The Amaranthaceae comprises about 65 genera and 900 species of herbs and shrubs with a few climbers distributed in the tropical Africa and America, and subtropical areas of warm temperate regions (HEYWOOD, 1978; TOWNSEND, 1993; WILLIS, 1966). The family is made up of several well-known species of medicinal, nutritional and horticultural importance. The classification of the family has witnessed changes in taxonomic opinions at one time or the other and this has led to grouping ang re-grouping of taxa (MARTIUS, 1825; SCHNIZ, 1934; TOWNSEND, 1993). The West African taxa are distributed within the two subfamilies and three of the four tribes recognized in the family (TOWNSEND, 1993). Tribe Amaranthoideae is better represented in the sub-region than Gomphrenoideae that is more common in the New World (MULLER & BORSCH, 2005). The West African species have been adequately described macromorphologically with features of the flowers and leaves (HUTCHINSON & DALZIEL, 1954) but there is dearth of information on micromorphology (EDEOGA & OTOIDE, 2001; OGUNDIPE, 1996). In the general anatomical accounts, METCALFE & CHALK (1950, 1979), SOLEREDER (1908) reported capitate with uniseriate stalk, unicellular or uniseriate, 2-many armed multicellular, stellate and candelabra trichomes in the family. Other characters that have been accounted for in the family are amphistomatic leaf and presence of crystal sands, raphoids and styloids as cell inclusions in many species (METCALFE & CHALK, 1979). In the present work, we document many taxo-

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nomically useful features of the leaf epidermis occurring in the West African species and also discuss the features in relation to the recent classification of the family. In the recent studies from Brazil, ROCIO & CARMO (2004) and SUZANE et al. (2010) noted that features of the epidermis are useful for distinguishing taxa in the family.

Material and methods

The specimens used for the study were obtained from field and herbarium. We visually examined all the herbarium specimens for outstanding morphological variations and 1–5 representative specimens whose exsiccate data appear in Table 1 were considered for the study and 25 microscope fields were examined per sample. Abbreviations of the herbaria follow HOLMGREN et al., 1981. For light microscopy, epidermal preparations were obtained by either macerating materials with Jeffrey's solution or soaked in concentrated nitric acid depending on sample's response to treatment, according to the approaches of KADIRI & AYODELE, 2003; KADIRI et al., 2007a, b., KHATIJAH et al., 1996; OGUNDIPE, 1996; OGUNDIPE & WUJEK, 2004, STACE, 1965). The approaches involved soaking leaf samples in the solutions for 1–6 hours. The leaf samples were removed as soon as we noticed air bubbles on the samples and the epidermal layers were separated in Petri dish containing water. The epidermal surfaces were washed in several changes of water with soft camel hair brush for removal of tissue debris, dragged onto the glass slides with outer surfaces facing up using mounting needle and few drops of 95% ethyl alcohol were added to harden the cells. The cleaned epidermises were stained using 3% aqueous solution of Safranin O, washed in 2–3 drops of 50% ethanol to remove excess stain before adding glycerine. Each mounted sample was covered with cover-slip and the edges were ringed with nail varnish to prevent dehydration. The epidermal features were assessed under Olympus light microscope and their characteristics were depicted using camera lucida drawing apparatus carried out in the postgraduate laboratory of the Department of Botany, University of Lagos, Nigeria. For electron microscopy, small pieces (7 mm^2) of the leaf material were fixed on SEM stubs with double-sided tape and sputter coated with gold. Specimens were examined and photographed in a Jeol JSM 35 scanning electron microscope in the Department of Biology, Central Michigan University, USA. Our stomatal descriptions follow STACE (1965) while SEM terminologies are as used by OGUNDIPE & WUJEK (2004), RADFORD et al. (1974), STACE, 1991, SUZANE, et al. (2010).

Results

The results are summarized in Tables 2–4; the line drawings under light microscope and SEM micrographs are presented in Figs. 1–10.

Light Microscopy: Epidermal cell

The anticlinal wall may be identical (undulate or curved) on both surfaces and either surface having different anticlinal wall pattern. Moreover, straight pattern was recorded on the adaxial surface of a few species (Figs. 1–6). The epidermal cells of most species can be irregularly shaped on both surfaces and we also observed polygonal shape on the adaxial surface and irregular shape on the abaxial surface of some species such as *A. lanata*, *A. bettzichiana*, *C. globosa*, *Cyathula* spp., *P. heudelotii* and *P. sclerenthum* (Table 2). Cell size varies among the species. *A. brasiliana* has the longest epidermal cell of 49.6(58.9 ± 5.0)74.4 µm on the adaxial surface while shortest cell of 10.6(12.0 ± 1.8)16.0 µm was recorded in *A. aspera* on the abaxial surface (Table 3). Cell wall is thicker on the adaxial epidermis than the abaxial surface except in *A. lanata, A. pungens, A. graecizans, C. argentea* and *C. trigyna*. Whereas, cell wall thickness is identical on both surfaces of *A. brasiliana, A. repens, A. pseudovirgata, A. spinosus* and *A. viridis* (Table 3). The leaf surface can be glabrous or pubescent in the species (Table 2).

Stomatal complex

The species generally have anomocytic stomata, but some species may have other accompanying types e.g. paracytic in *Alternanthera* spp., *Cyathula* spp., *P. lappacea, A. pseudovirgata A. viridis* and *G. celosiodes* (Figs. 1–6, Table 2). In addition, cyclocytic and diacytic types were observed in *Cyathula* spp. (Figs. 4G, H; 5B). All species are amphistomatic except *P. heudelotii* that is hypostomatic (Fig. 5E, F; Table 2). Stomatal size varies from 4.0(8.0 ± 1.4)10.0 µm × 2.0(3.0 ± 0.8)5.5 µm in *A. spinosus* to 20.2(21.3 ± 1.3)25.2 µm × 10.0(12.3 ± 1.0)13.4 µm in *A. pungens* (Table 4). Stomatal number is higher on the adaxial surface than abaxial surface in all species.

Table 1
Provenances of the representative specimens studied

S/N	Name of species	Name of collector	Accession number
Tribe:	**Amarantheae**		
1.	*Achyranthes bidentata* BLUME	J. LOWE	UIH 1239
2.	*A. bidentata* BLUME	LATILO & OGUNTAYO	FHI 70501
3.	*A. aspera* LINN.	OGUNDIPE & KADIRI	LUH 001
4.	*A. aspera* LINN.	M. REEKMANS	FHI 95086
5.	*A. aspera* LINN.	OGUNDIPE & KADIRI	LUH 002
6.	*Aerva lanata (*LINN.*)* JUSS. ex SCHULT.	G. W. IVENS	UIH 14770
7.	*A .lanata* (L.) JUSS. ex SCHULT.	OLORUNFEMI, BINUYO & BABAGBEMI	FHI 96748
8.	*A. lanata* (L.) JUSS. ex SCHULT.	A. B. KADIRI	LUH…..
9.	*Amaranthus dubius* MART. ex THELL	G. JACKSON	–
10.	*A. graecizans* LINN.	OGUNDIPE & KADIRI	LUH 007
11.	*A. hybridus* LINN.	OGUNDIPE & KADIRI	LUH40
12.	*A. pseudovirgata* SCHINZ	J. B. HALL	UIH 18477
13.	*A. spinosus* LINN.	OGUNDIPE & KADIRI	LUH 006
14.	*A. viridis* LINN	OGUNDIPE & KADIRI	LUH 008
15.	*Cyathula pedicellata* C. B. CLARKE	A. GLEEDHILL	UIH 10534
16.	*C. prostrata* (LINN.) BLUME	EKWUNO & others	FHI 95876
17.	*C. schimperiana* SUSSENG	S. TAMAJONG	FHI 26896
18.	*Pandiaka heudelotii* (MOQ.) HOOK. f.	I. F. LA CROIX	UIH 6756
19.	*P. heudelotii* (MOQ.) HOOK. f.	OLORUNFEMI, BINUYO & BABAGBEMI	FHI 96481
20.	*Psilotrichum scleranthum* BLUME	JEAN PAWEK	FHI 97157
21.	*Pupalia lappacea* (LINN.) JUSS.	SOLADOYE, DARAMOLA & IHE	FHI 86252
22.	*P. lappacea* (LINN.) JUSS.	OGUNDIPE & KADIRI	LUH 012
Tribe:	***Celosieae***		
23.	*Celosia argentea* LINN.	OGUNDIPE & KADIRI	LUH 009
24.	*C. argentea* LINN.	OGUNDIPE & KADIRI	LUH 010
25.	*C. argentea* LINN.	OGU	FHI 52476
26.	*C. globosa* SCHINZ	P. WIT & J. OLORUNFEMI	FHI 66925
27.	*C. laxa* SCHUM & THONN	J. LOWE	UIH 1264
28.	*C. trigyna* LINN.	I. F. LA CROIX	UIH 6757
29.	*C. trigyna* LINN.	OLORUNFEMI	FHI 91878
Tribe:	***Gomphreneae***		
30.	*Alternanthera bettzichiana* (REGEL) VOSS	NIL	UIH 20454
31.	*A. brasiliana* (LINN.) KUNTZE	J. LOWE	UIH 22235
32.	*A. ficoidea* (LINN.) P.BEAUV.	J. LOWE	UIH 19352
33.	*A. pungens* H. B. & K.	OGUNDIPE & KADIRI	LUH 005
34.	*A. repens* (LINN.) LINK. ENUM.	SHARMA	UIH 19071
35.	*A. sessilis* (LINN.) R. BR. ex BOTH-SCHINZ	OGUNDIPE & KADIRI	LUH 003
36.	*Gomphrena celosioides* MART.	OGUNDIPE & KADIRI	LUH 011

Nomenclature is according to HUTCHINSON & DALZIEL (1954).

Scanning electron microscopy

All the species studied have irregular striae and the periclinal wall is either conspicuous or inconspicuous. Stomata may be sunken or raised; usually they have thin rims and narrow to slightly wide apertures (Figs. 7–10). Epicuticular waxes may occur as flakes or evenly spread and trichomes are either restricted to the abaxial or adaxial surface or found on both surfaces of the leaf. Long trichomes which may unicellular or multicellular and smooth or tuberculed on the surface were recorded in some species of *Amaranthus* e.g. *A. pseudovirgata* and *A. spinosus, C. argentea, C. globosa, C. prostrata, C. schimperiana* and *P. lappacea*

Table 2
Comparative qualitative foliar epidermal features of the species of West African Amaranthaceae

S/N	Species	Surface	Anticlinal wall pattern	Cell shape	Stomatal types	Trichome types
Tribe:	**Amarantheae**					
1.	*Achyranthes aspera*	Adaxial	Curved	Irregular	Anomocytic/Paracytic	Absent
		Abaxial	Undulate	Irregular	Anomocytic/Paracytic	Absent
2.	*A. bidentata*	Adaxial	Curved	Irregular	Anomocytic	Absent
		Abaxial	Undulate	Irregular	Anomocytic	Unicellular/Conical
3.	*Aerva lanata*	Adaxial	Straight/Curved	Polygonal	Anomocytic	Absent
		Abaxial	Curved	Irregular	Anomocytic/Paracytic	Multicellular/Conical
4.	*Amaranthus dubius*	Adaxial	Curved	Irregular	Anomocytic/Paracytic	Multicellular
		Abaxial	Curved	Irregular	Anomocytic/Paracytic	Multicellular
5.	*A. graecizans*	Adaxial	Curved	Irregular	Anomocytic	Absent
		Abaxial	Undulate	Irregular	Anomocytic	Multicellular
6.	*A. pseudovirgata*	Adaxial	Curved	Irregular	Anomocytic/Paracytic	Absent
		Abaxial	Undulate	Irregular	Anomocytic/Paracytic	Absent
7.	*A. spinosus*	Adaxial	Curved	Irregular	Anomocytic	Absent
		Abaxial	Undulate	Irregular	Anomocytic	Absent
8.	*A. viridis*	Adaxial	Curved	Irregular	Anomocytic/Paracytic	Absent
		Abaxial	Undulate	Irregular	Anomocytic/Paracytic	Absent
9.	*Cyathula pedicillata*	Adaxial	Straight/Curved	Polygonal	Anomocytic/Cyclocytic/Paracytic	Multicellular/Conical
		Abaxial	Undulate	Irregular	Anomocytic/Diacytic	Absent
10.	*C. prostrata*	Adaxial	Straight/Curved	Polygonal	Anomocytic/Cyclocytic/Paracytic	Absent
		Abaxial	Undulate	Irregular	Anomocytic/Paracytic	Absent
11.	*C. schimperiana*	Adaxial	Curved	Polygonal	Anomocytic/Cyclocytic	Absent
		Abaxial	Undulate	Irregular	Anomocytic/Cyclocytic	Absent
12.	*Pandiaka heudelotii*	Adaxial	Curved	Polygonal	Absent	Absent
		Abaxial	Curved	Irregular	Anomocytic	Absent
13.	*Psilotrichum scleranthum*	Adaxial	Curved	Polygonal	Anomocytic	Absent
		Abaxial	Undulate	Irregular	Anomocytic	Absent
14.	*Pupalia lappacea*	Adaxial	Curved	Irregular	Anomocytic/Paracytic	Absent
		Abaxial	Undulate	Irregular	Anomocytic/Paracytic	Absent
Tribe:	***Celosieae***					
15.	*Celosia argentea*	Adaxial	Straight/Curved	Polygonal	Anomocytic	Absent
		Abaxial	Undulate	Irregular	Anomocytic	Unicellular
16.	*C. globosa*	Adaxial	Curved	Polygonal	Anomocytic	Absent
		Abaxial	Undulate	Irregular	Anomocytic	Absent
17.	*C. trigyna*	Adaxial	Undulate	Irregular	Anomocytic	Absent
		Abaxial	Undulate	Irregular	Anomocytic	Absent

Tribe:	***Gomphreneae***				
18.	*Alternanthera bettzichiana*	Adaxial	Curved	Polygonal	Anomocytic/Paracytic	Absent
		Abaxial	Curved	Irregular	Anomocytic	Absent
19.	*A. brasiliana*	Adaxial	Undulate	Irregular	Anomocytic	Absent
		Abaxial	Undulate	Irregular	Anomocytic	Absent
20.	*A. ficoidea*	Adaxial	Undulate	Irregular	Anomocytic	Absent
		Abaxial	Undulate	Irregular	Anomocytic/Paracytic	Multicellular/Conical
21.	*A. pungens*	Adaxial	Curved	Irregular	Anomocytic/Paracytic	Absent
		Abaxial	Undulate	Irregular	Anomocytic/Paracytic	Absent
22.	*A. repens*	Adaxial	Curved	Irregular	Anomocytic/Paracytic	Absent
		Abaxial	Curved	Irregular	Anomocytic/Paracytic	Absent
23.	*A. sessilis*	Adaxial	Undulate	Irregular	Anomocytic/Paracytic	Unicellular
		Abaxial	Undulate	Irregular	Anomocytic/Paracytic	Unicellular
24.	*Gomphrena celosioides*	Adaxial	Curved	Irregular	Anomocytic/Paracytic	Absent
		Abaxial	Curved	Irregular	Anomocytic/Paracytic	Multicellular/Conical

(Figs. 7E, G, H; 8G, 9A–D, G, H; 10A, H). Candelabra trichomes were encountered in *C. trigyna* and *A. viridis* (Figs. 8F, 10G). Filiform type was found in *A. graecizans* (Fig. 9E, F), and peltate scales were observed in *A. psudovirgata* and *A. spinosus* (Figs. 7F and 10E).

Discussion

The present comparative work does not only conform to the existing reports on leaf epidermal features of some species of Amaranthaceae by previous workers especially, METCALFE & CHALK, 1950, 1979; OGUNDIPE, 1996; ROCIO & CARMO, 2004 and SUZANE et al., 2010 but also shed light on features which have implications on taxa delimitation in the family. Anomocytic stomatal type, usually accompanied by paracytic type, undulate anticlinal wall pattern and irregular cell shape were observed among the species. Other stomatal types such as cyclocytic and diacytic were found only in *Cyathula pedicillata* but only diacytic was recorded in *Cyathula prostrata*. Diacytic stomata have also been reported in the Brazilian *Alternanthera brasiliana* (ROCIO & CARMO, 2004) but METCALFE & CHALK (1950, 1979) reported anomocytic stomata as being typical for the family. The taxonomic relevance of stomata has been shown in many angiosperm families (AHMAD, 1976; AKHIL & SUBHAN, 1997; BHANDARI & MUKHOPADHYAY, 1997; FORTENELLE, et al., 1994; KADIRI & AYODELE, 2003; KHATIJAH et al., 1996; KOTRESHA & SEETHARAM, 1995; 2000, OGUNDIPE, 1996; OGUNDIPE & WUJEK, 2004; ROCIO & CARMO, 2004; SOLEREDER, 1908; SUZANE et al., 2010). The variation recorded in the anticlinal wall pattern as noted by STACE (1965) may be under strict genetic control or influenced by environmental conditions. In Brazil, sinuous and straight walls were recorded in *Gomphrena* spp. while straight-curved and deeply sinuous walls were reported for *Alternanthera brasiliana* (ROCIO & CARMO, 2004; SUZANE et al., 2010). Generally, the anticlinal wall of the West African species of Amaranthaceae varies from curved to undulate or sometimes straight-curved. Trichomes were also recorded to vary among the species, this observation agrees with the reports of METCALE & CHALK (1950, 1979), OGUNDIPE

Table 3
Comparative quantitative features of the foliar epidermal cells of the species of West African Amaranthaceae

S/n	Species	Cell number	Cell length (μm)	Cell width (μm)	Cell thick (μm)
Tribe	**Amarantheae**				
1.	*Achyranthes aspera*	48(60 ± 3)75	15.6(18.5 ± 2.0)28.0	6.0(9.9 ± 2.0)12.4	1.5(1.0 ± 0.3)2.0
		35(40 ± 5)51	10.6(12.0 ± 1.8)16.0	5.0(7.5 ± 1.8)8.6	0.8(1.0 ± 0.2)1.0
2.	*A. bidentata*	58(68 ± 5)85	25.6(28.5 ± 2.2)32.0	8.0(10.9 ± 2.0)14.4	1.6(1.7 ± 0.3)2.0
		56(60 ± 5)65	25.6(32.0 ± 2.3)36.0	5.6(7.7 ± 2.0)9.6	0.8(1.1 ± 0.2)1.2
3.	*Aerva lanata*	58(63 ± 8)68	32.0(38.9 ± 3.0)44.8	12.8(14.4 ± 2.2)16.0	1.2(1.5 ± 0.2)1.6
		25(30 ± 3)50	18.0(28.8 ± 3.2)26.0	16.0(18.7 ± 2.0)22.4	2(2.3 ± 0.2)2.4
4.	*Amaranthus dubius*	75(80 ± 7)85	14.4(16.8 ± 2.4)20.0	8.0(9.9 ± 2.0)12.0	1.2(1.6 ± 0.2)2.0
		86(92 ± 5)98	16.0(22.7 ± 2.2)28.0	12.0(14.1 ± 2.0)16.0	2.0(2.1 ± 0.3)2.4
5.	*A. graecizans*	89(77 ± 7)98	16.0(23.5 ± 3.6)35.2	8.0(10.7 ± 2.0)13.6	1.2(1.5 ± 0.3)1.6
		26(29 ± 3)32	32.0(36.0 ± 2.5)40.0	14.4(16.8 ± 2.0)20.0	1.6(1.9 ± 0.2)2.4
6.	*A. pseudovirgata*	15(12 ± 3)20	22.4(26.1 ± 2.0)24.0	8.0(11.5 ± 2.0)16.0	1.2(1.3 ± 0.3)1.6
		78(84 ± 9)90	20.0(22.1 ± 2.3)24.0	6.4(8.0 ± 1.3)9.6	1.2(1.3 ± 0.2)1.6
7.	*A. spinosus*	12(15 ± 3)20	20.4(24.1 ± 2.0)20.0	7.0(10.5 ± 2.0)15.0	1.0(1.3 ± 0.3)1.5
		68(80 ± 9)90	20.0(22.1 ± 2.3)22.0	6.0(9.0 ± 1.3)10.6	1.0(1.1 ± 0.2)1.5
8.	*A. viridis*	13(15 ± 3)22	21.4(24.1 ± 2.0)28.0	8.0(10.5 ± 2.0)16.0	1.0(1.3 ± 0.3)1.5
		70(82 ± 9)94	25.0(27.1 ± 2.3)32.0	8.5(9.0 ± 1.3)10.8	1.0(1.1 ± 0.2)1.5
9.	*Cyathula pedicillata*	20(25 ± 3)30	28.0(48.0 ± 3.2)56.0	16.0(20.0 ± 2.2)26.4	1.2(1.5 ± 0.1)1.6
		38(41 ± 5)45	20.0(24.0 ± 3.0)28.0	6.4(8.8 ± 2.1)12.0	0.8(1.1 ± 0.3)1.2
10.	*C. prostrata*	58(63 ± 8)68	32.0(38.9 ± 3.0)44.8	12.8(14.4 ± 2.2)16.0	1.2(1.5 ± 0.2)1.6
		40(47 ± 5)52	32.8(36.5 ± 3.0)40.0	8.0(13.9 ± 1.6)17.6	0.8(0.9 ± 0.2)1.2
11.	*C. schimperiana*	60(68 ± 8)75	16.0(22.7 ± 2.0)28.0	11.2(13.6 ± 2.0)16.0	2.0(2.1 ± 0.2)2.4
		28(31 ± 4)35	14.4(16.8 ± 3.4)20.0	8.0(14.4 ± 2.0)24.4	1.2(1.5 ± 0.1)1.6
12.	*Pandiaka heudelotii*	30(36 ± 5)42	25.6(40 ± 5.5)51.2	20.0(23.2 ± 2.0)25.6	2(2.3 ± 0.1)2.4
		22(26 ± 3)30	20.0(25.9 ± 3.0)32.0	8.0(11.5 ± 2.1)14.4	0.8(1.2 ± 0.2)1.6
13.	*Psilotrichum scleranthum*	25(30 ± 3)50	18.0(28.8 ± 3.2)26.0	16.0(18.7 ± 2.0)22.4	2(2.3 ± 0.2)2.4
		24(30 ± 4)35	24.0(31.7 ± 3.0)36.0	8.0(19.9 ± 2.0)25.0	1.2(1.5 ± 0.3)1.6
14.	*Pupalia lappacea*	14(16 ± 3)18	32.0(44.8 ± 5.0)56.0	22.4(25.3 ± 1.8)29.6	2.4(2.7 ± 0.5)2.8
		12(16 ± 3)22	32.0(41.9 ± 3.0)58.4	12.8(14.4 ± 2.0)16.0	1.6(1.9 ± 0.5)2.4
Tribe:	***Celosieae***				
15.	*Celosia argentea*	28(31 ± 5)35	34.4(42.1 ± 4.1)48.0	17.6(20.5 ± 2.0)24.0	1.6(2.0 ± 0.1)2.4
		20(22 ± 3)25	30.4(37.6 ± 2.3)37.6	14.4(20.2 ± 1.7)24.8	1.6(1.7 ± 0.7)3.1
16.	*C. globosa*	27(31 ± 5)35	25.6(35.7 ± 5)49.6	20.8(25.1 ± 2.0)30.4	1.6(2.0 ± 0.4)2.4
		28(30 ± 5)32	38.4(48 ± 5)57.6	12.0(14.1 ± 2.0)16.0	1.2(1.5 ± 0.2)1.6
17.	*C. trigyna*	28(32 ± 3)35	30.4(36.3 ± 3.0)42.4	18.4(22.9 ± 2.0)26.4	0.8(1.2 ± 0.3)1.6
		36(46 ± 4)54	20.8(26.9 ± 2.8)36.0	12.8(14.9 ± 2.0)16.0	1.6(1.7 ± 0.3)2.0
Tribe:	***Gomphreneae***				
18.	*Alternanthera bettzichiana*	32(38 ± 3)48	32.0(35.7 ± 3.0)40.0	16(23.2 ± 2.2)29.6	1.6(1.7 ± 0.1)2.0
		35(38 ± 3)42	21.6(26.4 ± 2.3)25.6	8(10.4 ± 1.5)13.6	0.8(1.1 ± 0.1)1.2
19.	*A. brasiliana*	10(12 ± 2)15	49.6(58.9 ± 5.0)74.4	17.6(20 ± 2.0)22.4	2.0(2.4 ± 0.3)2.8
		12(15 ± 2)18	32.0(35.2 ± 3.0)40.0	14.4(16 ± 2.0)17.6	2.0(2.4 ± 0.3)2.8
20.	*A. ficoidea*	25(35 ± 3)50	18.4(21.6 ± 1.9)24.0	4.0(7.2 ± 2.0)9.6	1.2(1.3 ± 0.2)2.0
		36(40 ± 3)45	20.0(26.1 ± 2.4)32.0	9.6(12.2 ± 1.8)14.4	1.6(1.7 ± 0.2)2.0
21.	*A.pungens*	9(10 ± 2)13	46.6(54.9 ± 5.0)70.4	15.6(18 ± 2.0)20.4	2.0(2.2 ± 0.3)2.5
		12(15 ± 2)17	30.0(33.2 ± 3.0)45.0	13.4(16 ± 2.0)18.6	2.0(2.2 ± 0.3)2.6
22.	*A. repens*	22(25 ± 3)28	34.4(44.8 ± 3.1)56.0	20.0(25.9 ± 2.0)25.6	1.6(2.0 ± 0.1)2.4
		18(21 ± 2)25	25.6(38.1 ± 3.3)37.6	14.4(18.7 ± 2.0)21.6	1.6(2.0 ± 0.2)2.4
23.	*A. sessilis*	40(47 ± 2)52	19.2(22.1 ± 2.4)25.6	9.6(13.3 ± 1.5)16.0	1.2(1.6 ± 0.1)2.0
		65(75 ± 6)85	16.8(21.1 ± 2.7)24	8.0(10.7 ± 2.0)14.4	0.8(1.2 ± 0.1)1.6
24.	*Gomphrena celosioides*	28(30 ± 4)32	32.0(38.7 ± 3.2)44.0	10.4(10.9 ± 2.1)14.4	0.8(1.2 ± 0.3)1.6
		40(45)54	27.2(36.8 ± 5.0)48.0	10.4(19.4 ± 1.7)28.0	1.2(1.5 ± 0.1)1.6

Values are arranged adaxial/abaxial. Measurements: Minimum value (Mean ± Standard Deviation) Maximum value.

Table 4
Comparative quantitative features of the stomata of the species of West African Amaranthaceae

S/n	Species	Stomatal number	Stomatal length (μm)	Stomatal width (μm)
Tribe:	**Amarantheae**			
1.	*Achyranthes aspera*	10(13 ± 2)20	4.4(7.0 ± 1.3)8.6	2.0(4.0 ± 1.0)4.1
		20(18 ± 2)30	5.6(8.0 ± 1.5)9.0	3.2(4.2 ± 1.0)4.9
2.	*A. bidentata*	15(18 ± 3)20	5.4(7.0 ± 1.9)8.6	2.4(3.0 ± 1.0)4.0
		25(28 ± 2)30	6.4(8.0 ± 1.9)9.6	3.2(4.0 ± 1.0)4.8
3.	*Aerva lanata*	2(4 ± 1)6	10.4(12.3 ± 1.6)14.4	9.6(10.7 ± 1.0)12.0
		5(8 ± 2)10	6.4(7.0 ± 1.0)8.6	6.1(8.0 ± 1.0)10.0
4.	*Amaranthus dubius*	35(38 ± 3)40	4.8(6.4 ± 1.3)8.0	3.2(4.0 ± 1.2)4.8
		50(55 ± 2)60	8.0(9.9 ± 2)12.0	4.0(5.1 ± 1.2)6.4
5.	*A. graecizans*	15(18 ± 4)20	12(16.8 ± 2.4)20.8	9.6(11.7 ± 1.5)13.6
		18(21 ± 3)25	12.8(14.4 ± 1.8)16.0	8.0(11.5 ± 1.2)14.4
6.	*A. pseudovirgata*	26(33 ± 3)45	5.4(7.5 ± 1.0)15.0	2.1(3.2 ± 0.8)5.0
		30(36 ± 2)45	4.4(8.3 ± 1.4)10.0	2.0(3.0 ± 0.8)4.5
7.	*A. spinosus*	20(30 ± 3)38	5.0(7.0 ± 1.0)13.0	2.0(3.0 ± 0.8)6.0
		26(32 ± 2)43	4.0(8.0 ± 1.4)10.0	2.0(3.0 ± 0.8)5.5
8.	*A. viridis*	25(30 ± 3)38	6.0(7.0 ± 1.4)10.0	2.3(3.0 ± 0.8)6.1
		29(32 ± 2)47	5.0(8.0 ± 1.4)12.0	2.5(3.0 ± 0.8)5.6
9.	*Cyathula pedicillata*	10(17 ± 2)20	10(14.4 ± 1.2)15.6	3.50(6.1 ± 1.9)7.0
		16(19 ± 2)24	12(14.4 ± 1.3)17.6	4.0(6.1 ± 1.1)8.0
10.	*C. prostrata*	2(4 ± 1)6	10.4(12.3 ± 1.6)14.4	9.6(10.7 ± 1.0)12.0
		16(17 ± 2)19	12.0(14.1 ± 1.2)16.0	8.0(9.9 ± 1.4)12.0
11.	*C. schimperiana*	20(26 ± 2)30	6.0(8.1 ± 2.1)10.8	3.0(5.0 ± 1.0)6.4
		25(31 ± 2)32	8.0(10.1 ± 2.1)12.8	4.0(5.3 ± 1.3)7.2
12.	*Pandiaka heudelotii*	Absent	Absent	Absent
		28(31 ± 2)35	5.6(9.1 ± 2.1)10.5	3.2(5.1 ± 1.2)7.2
13.	*Psilotrichum scleranthum*	5(8 ± 2)10	4.4(6.0 ± 1.0)7.6	3.1(4.0 ± 1.0)5.0
		10(12 ± 2)15	6.4(8.0 ± 1.0)9.6	4.0(4.8 ± 1.0)5.6
14.	*Pupalia lappacea*	9(10 ± 2)12	9.6(13.3 ± 2.0)16.0	6.4(8.0 ± 1.0)9.6
		15(18 ± 2)20	14.4(16 ± 2.0)17.6	6.4(8.0 ± 1.0)9.6
Tribe	***Celosieae***			
15.	*Celosia argentea*	5(6 ± 2)7	18.4(20 ± 1.2)21.6	14.4(14.9 ± 1.1)16.0
		18(21 ± 2)25	14.4(16.0 ± 1.6)17.6	9.6(11.5 ± 1.0)12.8
16.	*C. globosa*	8(10 ± 3)15	14.0(16.1 ± 1.1)18.0	4.5(5.0 ± 1.0)7.0
		10(14 ± 3)18	12.0(14.1 ± 1.5)16.0	4.8(5.6 ± 1.0)6.4
17.	*C. trigyna*	5(8 ± 2)10	6.4(8.0 ± 1.4)9.6	2.4(3.2 ± 1.0)4.8
		18(21 ± 3)24	4.8(7.7 ± 1.6)10.4	2.4(4.0 ± 0.8)5.6
Tribe	***Gomphreneae***			
18.	*Alternanthera bettzichiana*	13(15 ± 2)18	10.0(12.1 ± 1.5)20.0	5.0(4.0 ± 1.0)8.5
		25(28 ± 2)32	20.0(22.1 ± 1.7)24.0	8.0(8.0 ± 1.0)9.6
19.	*A. brasiliana*	2(4 ± 2)5	19.2(21.3 ± 1.5)23.2	12.0(13.3 ± 1.0)14.4
		5(6 ± 2)8	12.8(14.4 ± 1.8)16.0	8.0(10.7 ± 1.0)14.4
20.	*A. ficoidea*	14(17 ± 4)20	6.4(8.0 ± 2.0)9.6	1.6(2.4 ± 1.1)3.2
		28(31 ± 3)35	6.4(8.0 ± 2.0)9.6	2.4(3.2 ± 1.3)4.0
21.	*A.pungens*	3(4 ± 2)6	20.2(21.3 ± 1.3)25.2	10.0(12.3 ± 1.0)13.4
		8(6 ± 2)10	15.8(13.4 ± 1.5)18.0	8.0(11.7 ± 1.0)15.5
22.	*A. repens*	8(9 ± 2)10	13.6(15.5 ± 1.3)18.4	6.4(7.7 ± 1.4)9.6
		13(15 ± 2)17	8.0(9.9 ± 1.5)12.0	6.4(7.2 ± 1.0)8.0
23.	*A. sessilis*	10(13 ± 2)18	9.6(11.5 ± 1.3)13.6	6.4(8.0 ± 2.0)9.6
		28(31 ± 3)35	9.6(12 ± 1.4)14.4	6.4(8.0 ± 1.0)9.6
24.	*Gomphrena celosioides*	15(16 ± 2)17	13.6(14.7 ± 2.1)16.0	10.4(11.7 ± 1.1)12.8
		4(20 ± 5)25	14.4(16.0 ± 2.2)17.6	12(13.1 ± 1.0)14.4

Values are arranged adaxial/abaxial. Measurements: Minimum value (Mean ± Standard Deviation) Maximum value.

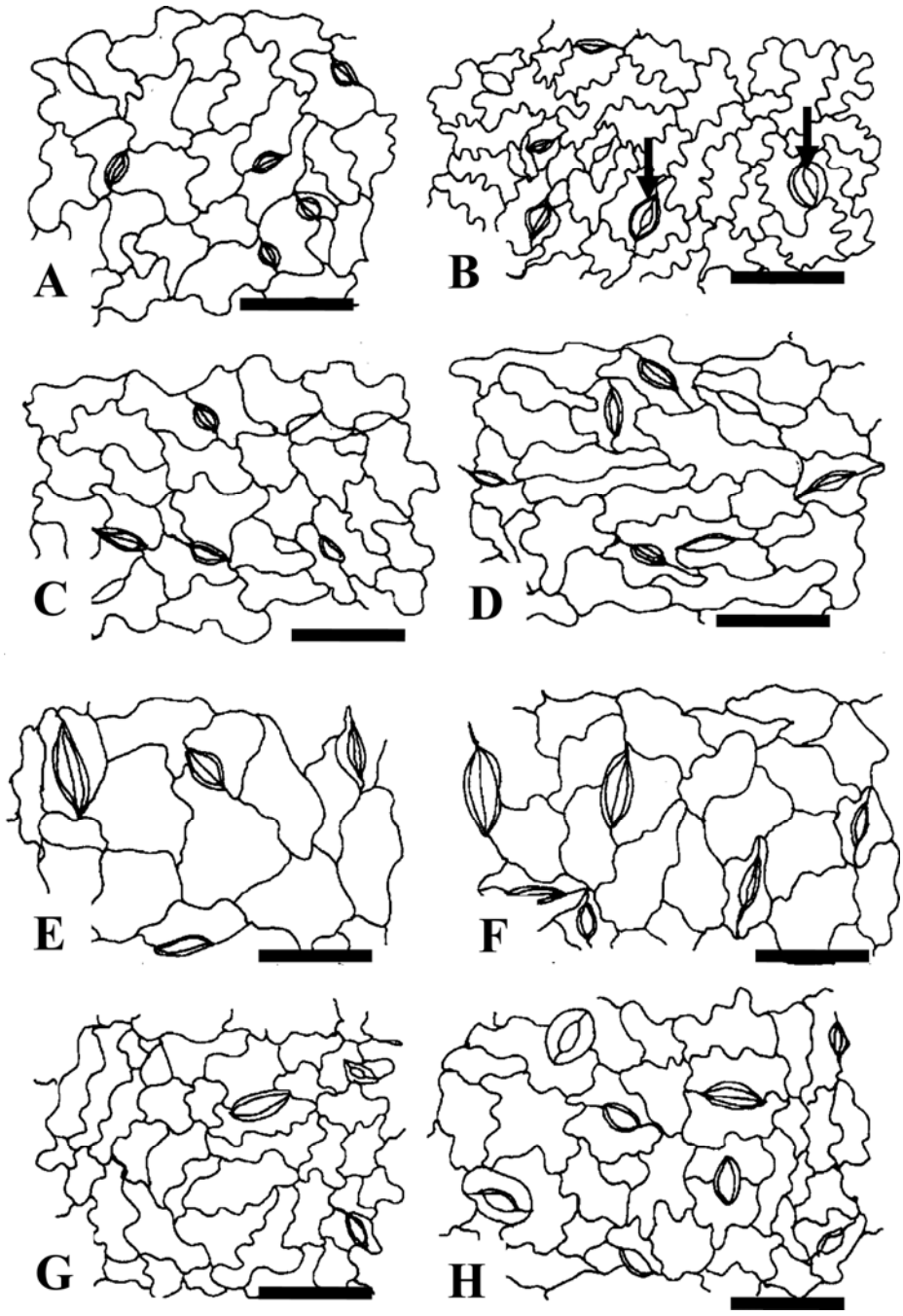

Fig. 1
Foliar epidermal characteristics of some West African species of Amaranthaceae.

A, C, E, and G: Adaxial surface. B, D, F and H: Abaxial surface. A, B: *A. bidentata*; C, D: *A. aspera*; E, F: *A. repens*; G, H: *A. ficoidea*. Anticlinal wall pattern is curved to undulate. Note the degree of curvature. From left in B, 1st arrow points at a typical paracytic stoma while right arrow shows anomocytic stoma. Scale bar is 50 µm

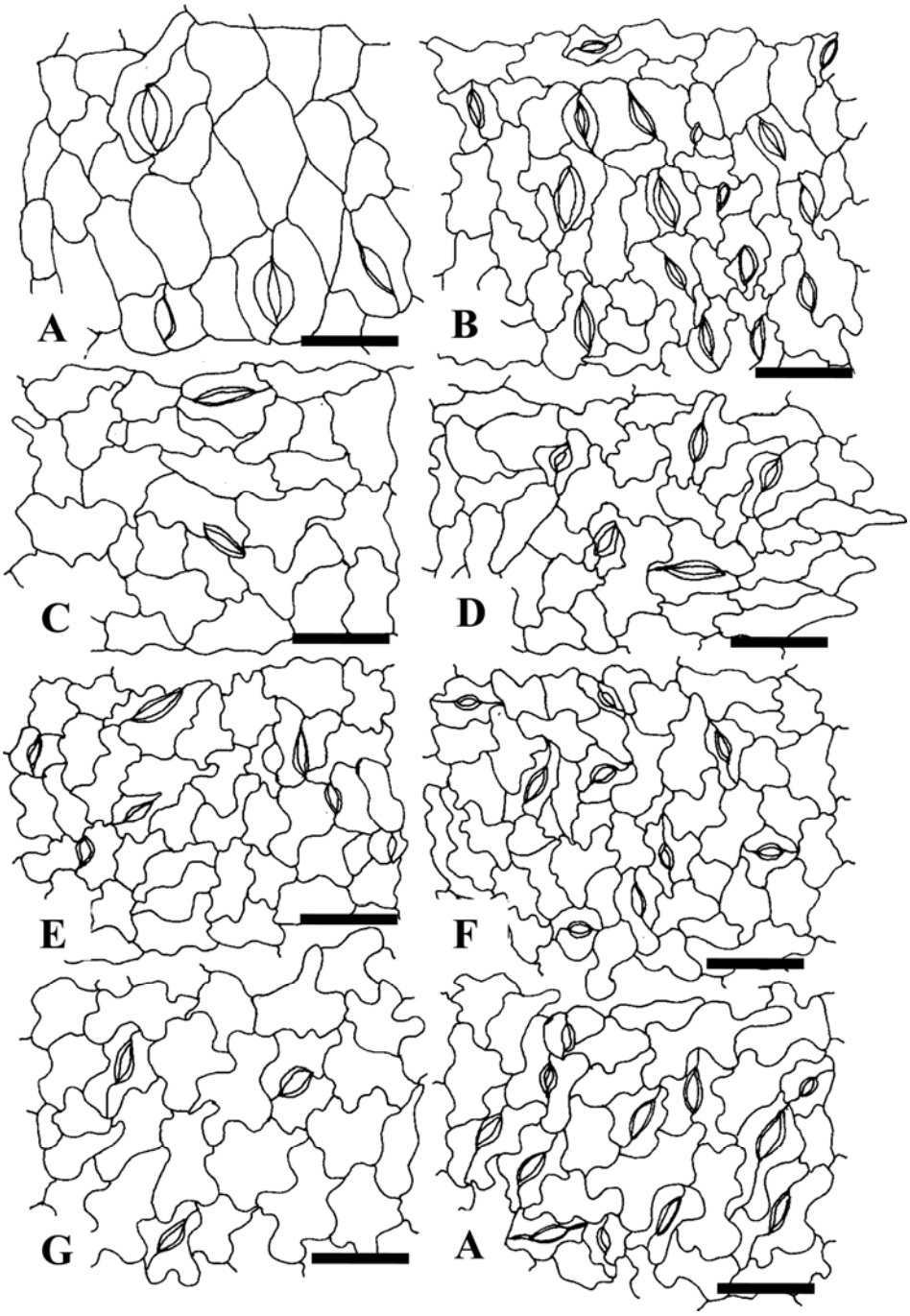

Fig. 2
Foliar epidermal characteristics of some West African species of Amaranthaceae

A, C, E, and G: Adaxial surface. B, D, F and H: Abaxial surface. A, B: *A. bettzichiana*; C, D: *A. pungens*; E, F: *A. sessilis*; G, H: *A. brasiliana*. Anticlinal wall pattern is curved to undulate. Note the degree of curvature. Scale bar is 50 μm

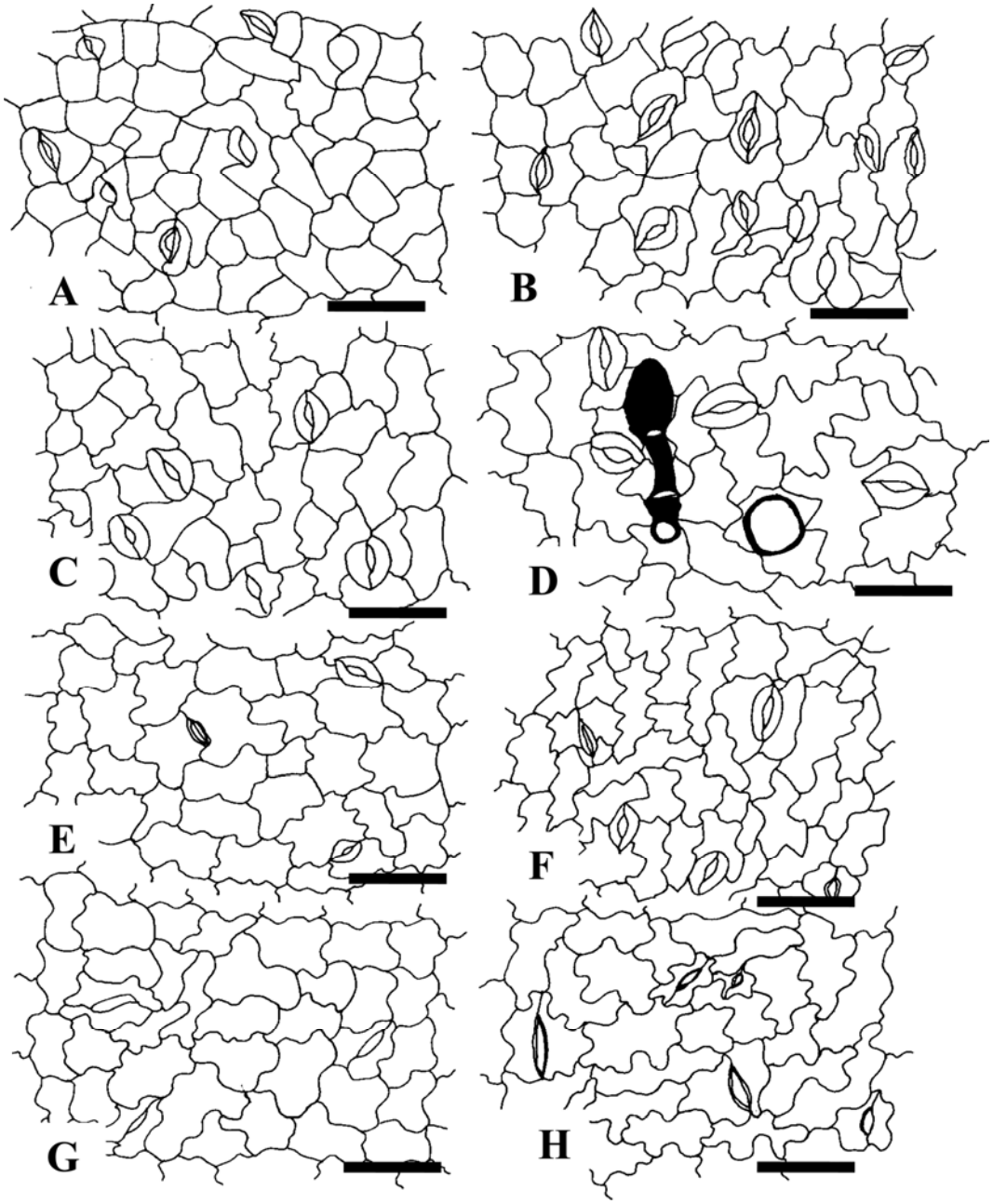

Fig. 3
Foliar epidermal characteristics of some West African species of Amaranthaceae

A, C, E, and G: Adaxial surface. B, D, F and H: Abaxial surface. A, B: *A. dubius*; C, D: *A. graecizans*; E, F: *A. pseudovirgata*; G, H: *A. spinosus*. Anticlinal wall pattern is curved to undulate. Note the degree of wall curvature in all species. Multicellular glandular trichome represented by a solid drawing and trichome scar were found in *A. graecizans*. Scale bar is 50 µm

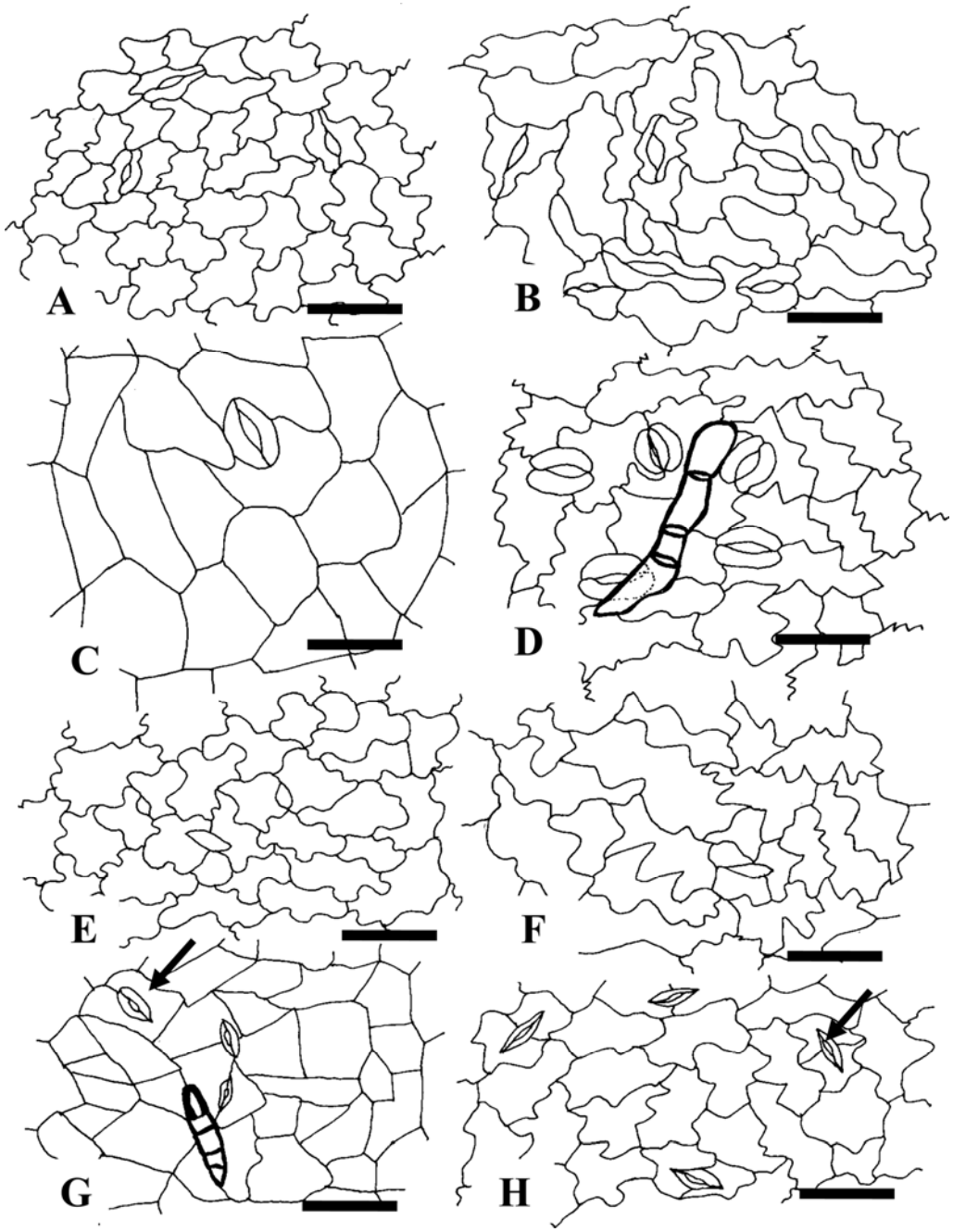

Fig. 4
Foliar epidermal characteristics of some West African species of Amaranthaceae

A, C, E, and G: Adaxial surface. B, D, F and H: Abaxial surface. A, B: *A. viridis*; C, D: *C. argentea*; E, F: *C. trigyna*; G, H: *C. pedicillata*. Anticlinal wall pattern is curved to undulate. Note the degree of wall curvature in the species. In G, the arrow points at cyclocytic stoma while 4H arrow shows diacytic stoma. Multicellular glandular trichomes with thick outline were found in D and G. Scale bar is 50 μm

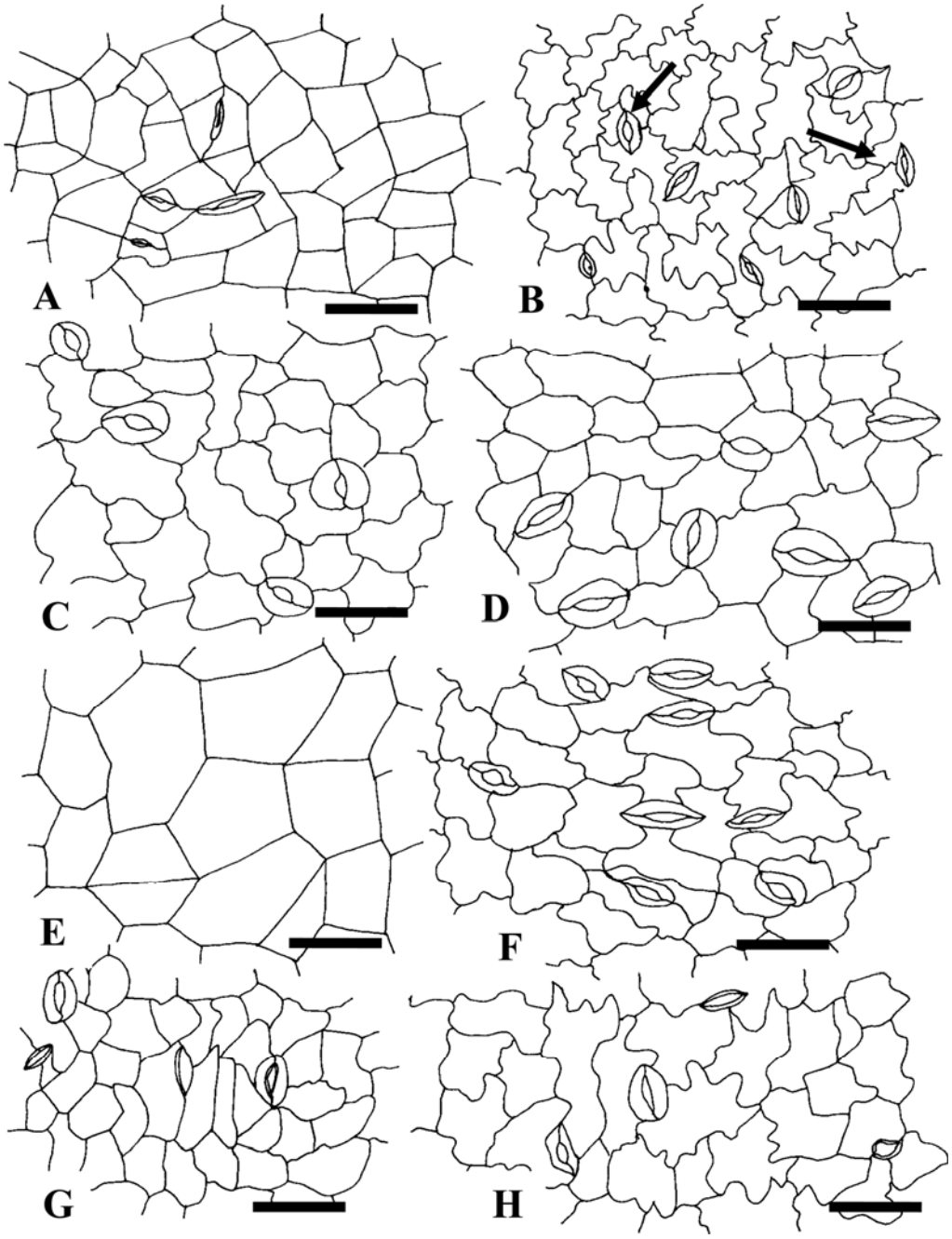

Fig. 5
Foliar epidermal characteristics of some West African species of Amaranthaceae

A, C, E, and G: Adaxial surface. B, D, F and H: Abaxial surface. A, B: *C. prostrata*; C, D: *G. celosiodes*; E, F: *P. heudelotii* G, H: *P. scleranthum*. Anticlinal wall pattern is curved to undulate and straight. Note the degree of wall curvature in the species. In B, the upper arrow points at anomocytic stoma while the lower arrow points at diacytic stoma. Stoma is absent on the adaxial surface of E. Scale bar is 50 µm

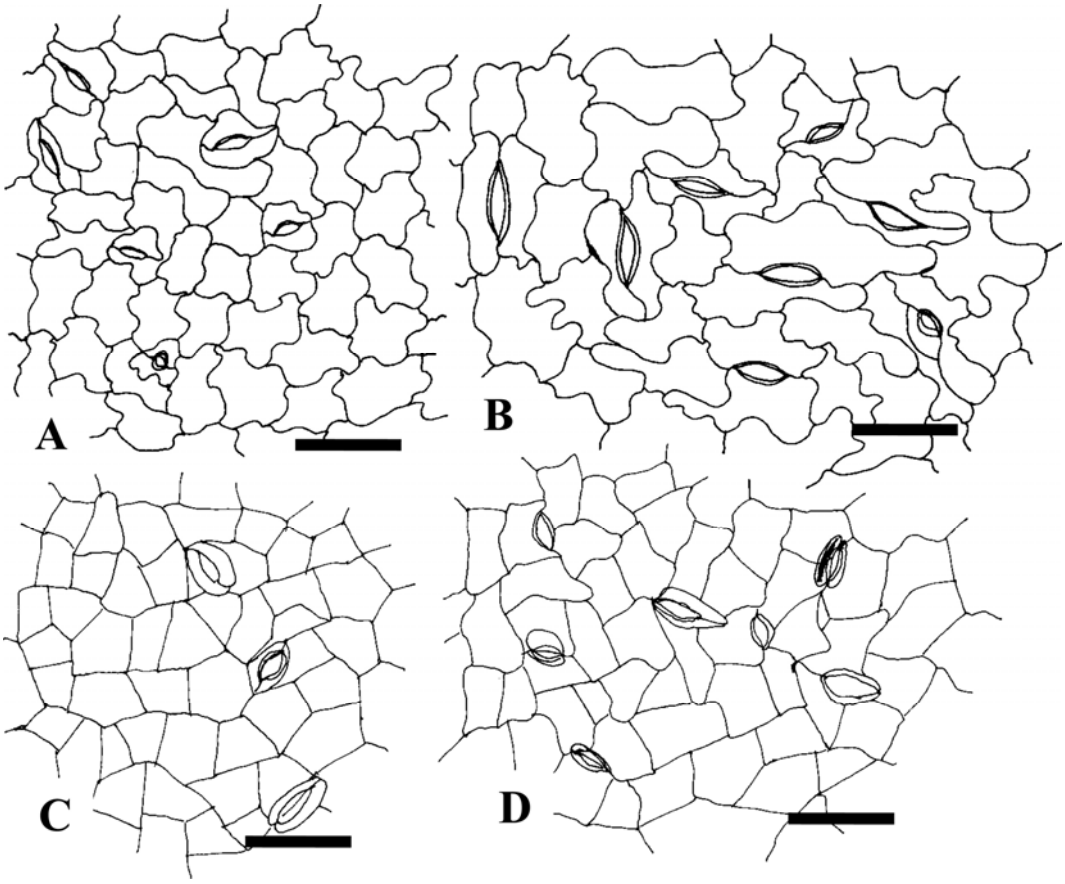

Fig. 6
Foliar epidermal characteristics of some West African species of Amaranthaceae

A and C: Adaxial surface. B and D: Abaxial surface. A, B: *P. lappacea*; C, D: *A. lanata*. Anticlinal wall pattern is curved to undulate and straight. Note the degree of wall curvature in the species. Scale bar is 50 μm

(1996), ROCIO & CARMO (2004) and SUZANE et al. (2010). We observed long acicular type which may be unicellular or multicellular, candelabra and filiform trichomes in some species belonging to a tribe in the family and a few of the types belonging to some species in other tribes. Trichomes have been shown to assist in distinguishing taxa in angiosperms (INAMDHAR & GANGDHARA, 1977; OGUNDIPE, 1996; RADFORD et al., 1978; STACE, 1965). The stomatal rims and apertures are thin and narrow respectively in the family and the stomatal orientation varies from raised to sunken both inter- and intra-generically. A combination of these features can be used to distinguish the taxa from one another as presented thus: in the tribe Amarantheae, the epidermal cells such as anticlinal wall pattern is usually curved on the adaxial surface and curved, curved-straight and undulate on the abaxial surface in all species. The curved-straight pattern was usually recorded in *Cyathula* spp. The species all have anomocytic and paracytic stomata but presence of cyclocytic and diacytic types again distinguishes *Cyathula* from other species. Celosieae is only represented in West Africa by *Celosia* (HUTCHINSON & DALZIEL, 1958) but, the three species studied reflected all the three types of anticlinal walls found in Amarantheae and they have all the typical stomatal types: anomocytic and paracytic. Also, trichomes are widely varied in the tribe. Gomphreneae represented by

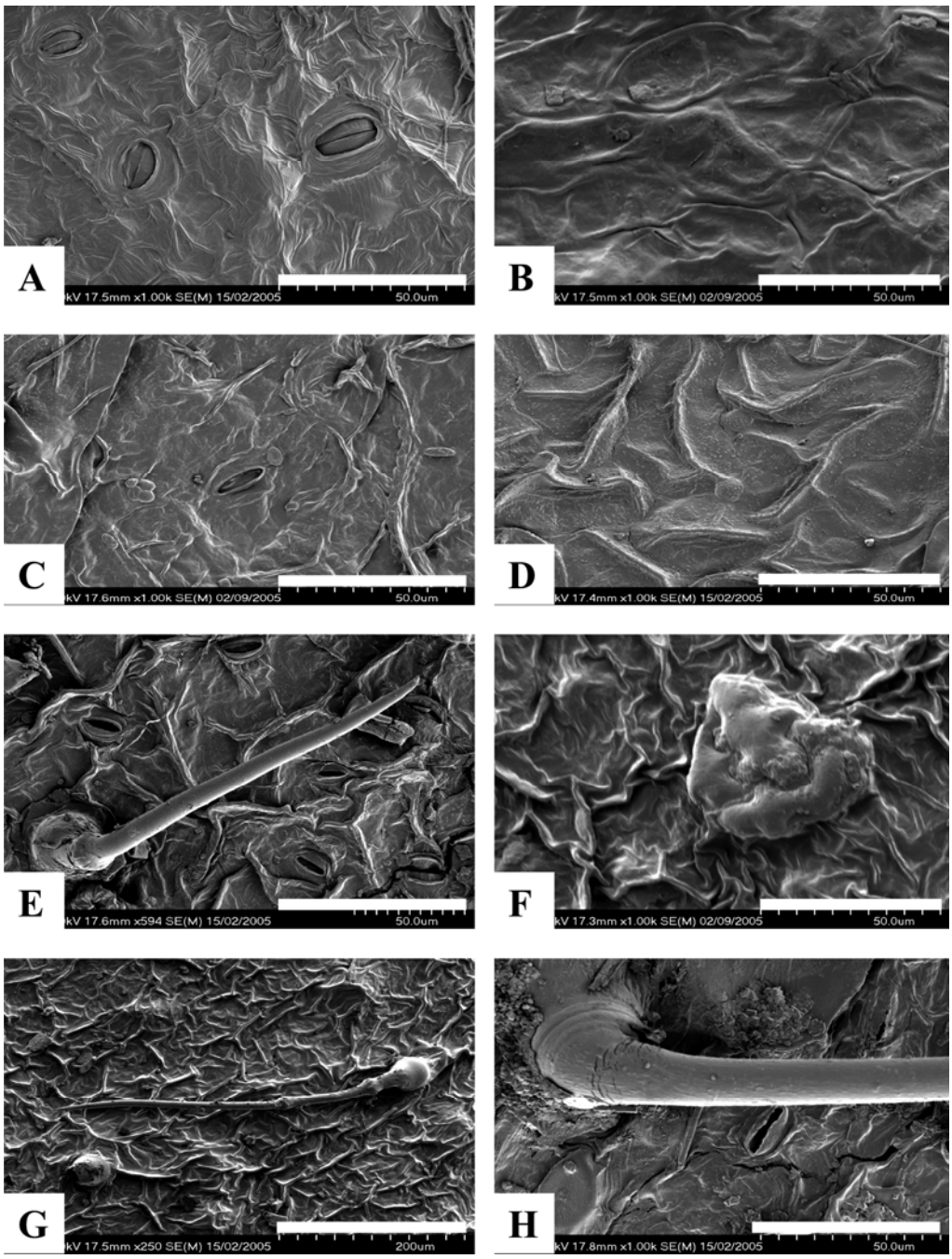

Fig. 7
Scanning electron micrographs of leaf surfaces of some species of Amaranthaceae

All abaxial surfaces. A, B: *A. aspera*; C, D: *A. lanata*; E, F: *A. pseudovirgata*; G, H: *A. spinosus*. Note irregular striae and sunken stomata in all species, long acicular trichomes in E, G and H; and multicellular peltate scale in F. Scale bar is 200 μm in G and 50 μm in others

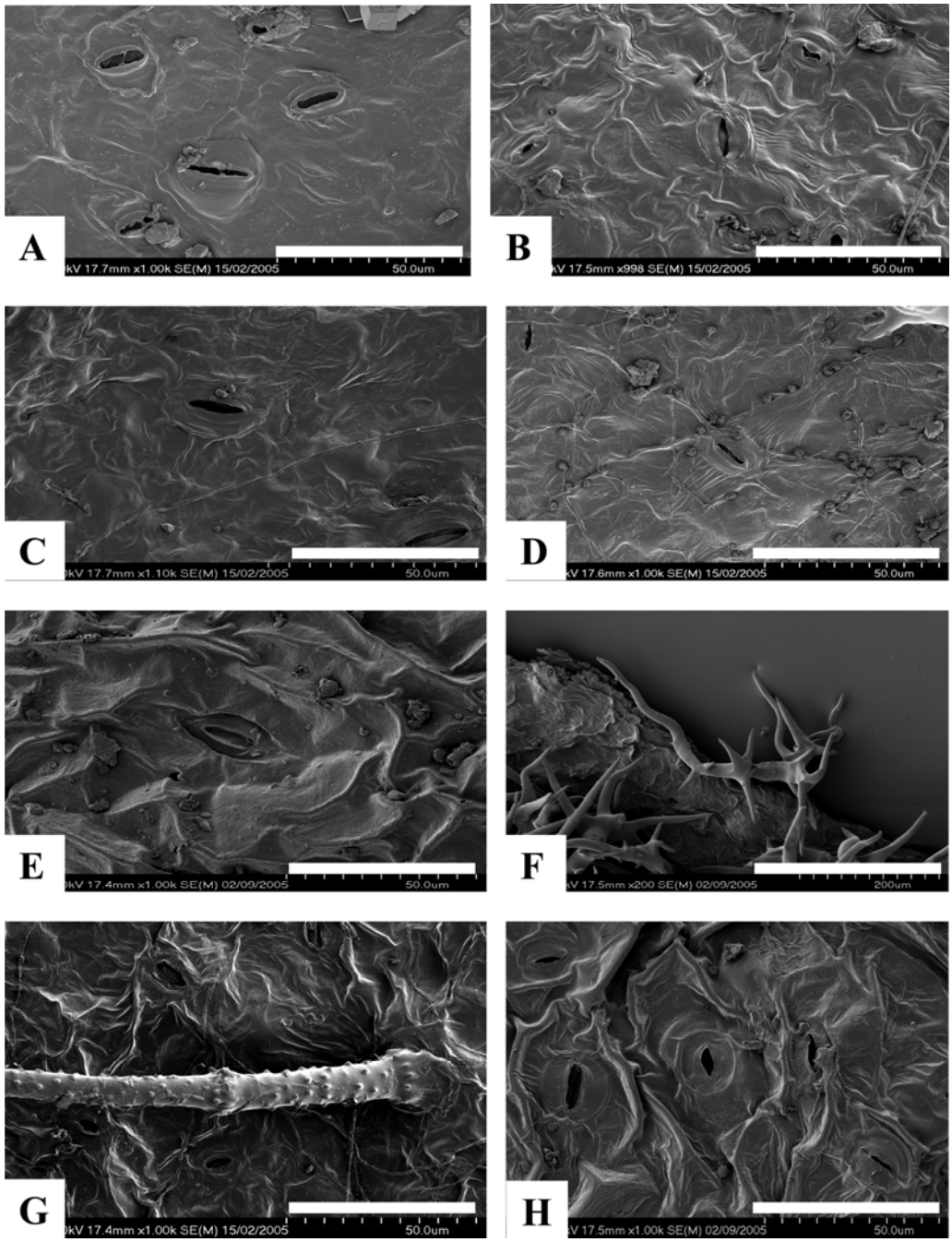

Fig. 8
Scanning electron micrographs of leaf surfaces of some species of Amaranthaceae

Adaxial surface: A, C, E and G. Abaxial surface: B, D, F and H. A, B: *A. viridis*; C, D: *C. globosa*; E, F: *C. trigyna;* G, H: *C. prostrata*. Note irregular striae in all species, raised stomata in A–D, sunken stomata in E–H, multi-branched trichome in F and long acicular type in G. Scale bar is 200 µm in F and 50 µm in others

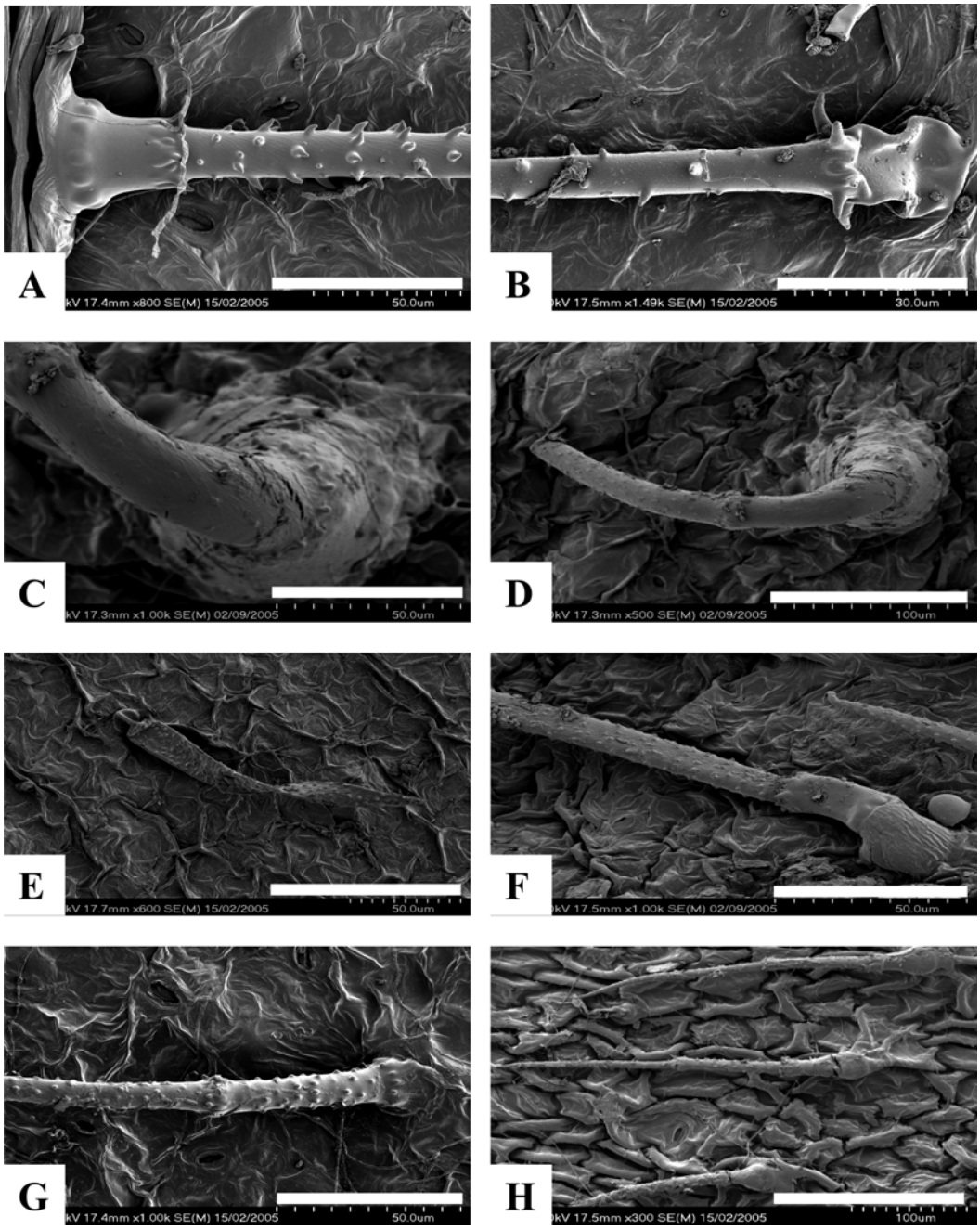

Fig. 9
Scanning electron micrographs of leaf surfaces of some species of Amaranthaceae

All abaxial surfaces except G. A, B: *C. schimperiana*; C, D: *P. lappacea*; E, F: *A. graecizans;* G, H: *A. hybridus*. Note irregular anticlinal wall in E, irregular striae, thin stomatal rim, narrow aperture and sunken stomata in all species. Scale plates in D and H, tuberculate trichomes in A and B, flat unicellular trichome in E, F and multicelluar type in G, H. Scale bar is 100 µm in D and H, 30 µm in B and 50 µm in other species

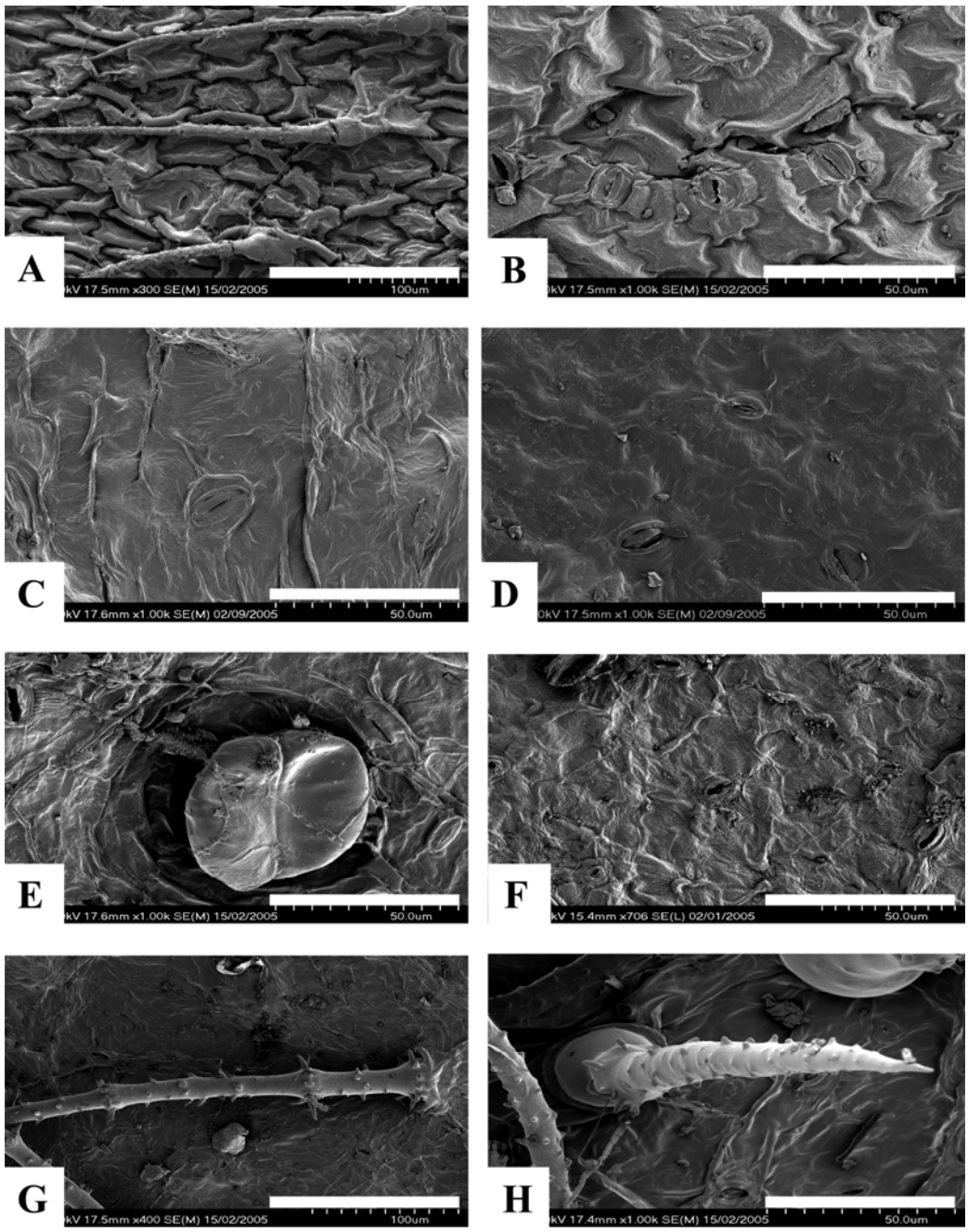

Fig. 10
Scanning electron micrographs of leaf surfaces of some species of Amaranthaceae

All adaxial surfaces except F and H. A, B: *A. hybridus* with acicular trichome; C–E: *A. spinosus*; C and D show smooth periclinal wall while E shows bicellular peltate scale. F is the lower surface of *C. pedicillata*. G, H: *A. viridis*, G shows candelabra trichome and H, tuberculate unicelluar trichome. Note the conspicuous periclinal wall and irregular striae in the species. Scale bar is 100 μm in A and G, and 50 μm in others

two genera *Alternanthera* and *Gomphrena* usually have uniform anticlinal wall patterns (curved and undulate) on both surfaces of the epidermis and they have the typical stomatal types for the family. These features are known to vary with environmental factors (METCALFE & CHALK, 1979; RADFORD et. al. 1974; STACE, 1965) but their use in solving taxonomic problems have been proved in many angiosperms families (AHMAD, 1976; AKHIL & SUBHAN, 1997; BHANDARI & MUKHOPADHYAY, 1997; FORTENELLE, et al., 1994; KADIRI & AYODELE, 2003; KHATIJAH et al., 1996; KOTRESHA & SEETHARAM, 1995, 2000; OGUNDIPE, 1996; OGUNDIPE & WUJEK, 2004; ROCIO & CARMO, 2004; SUZANE et al., 2010).

Therefore, we conclude that placement of the West African genera of Amaranthaceae into 3 tribes is supported with characters of the leaf epidermis. The species of Gomphreneae have many characters in common than any other tribe and Celosieae has more common features than Amarantheae which has the largest taxa. In our opinion, Gomphrenoideae appears to be more natural than Amaranthoideae and character affinities in Celosieae are more than in Amarantheae. Hence, our findings agree with the suggestion that Gomphrenoideae is monophyletic but our features of the epidermis support polyphyletic suggestion for Celosieae by OGUNDIPE & CHASE (2009) based on matK DNA sequence but do not support its monophyly according to BORSCH (1998) and KADEREIT et al. (2003) based on pollen characters and molecular analysis of *rbc*L. The apparent polyphyletic nature of Amarantheae observed by OGUNDIPE & CHASE (2009) is also supported because of wide variations in their epidermal characters. An indented dichotomous key for delimiting the species is presented below:

1. Leaf hypostomatic . . . . . *Pandiaka heudelotii*
1. Leaf amphistomatic
  2. Stomatal type anomocytic only
    3. Anticlinal wall curved on adaxial surface/undulate on abaxial surface
      4. Epidermal cell four times longer than wider on the abaxial surface . . . . . *Achyranthes bidentata*
      4. Epidermal cell two times longer than wider on the abaxial surface
        5. Cell shape irregular on both surfaces
          6. Peltate scale present, more cells on the adaxial surface . . *Amaranthus graecizans*
          6. Peltate scale absent, more cells on the abaxial surface . . . . *Amaranthus spinosus*
            7. Anticlinal wall undulate on both surfaces
              8. Mean stomatal size on abaxial surface 14.4 × 10.7 µm . . . . . *Alternanthera brasiliana*
              8. Mean stomatal size on abaxial surface 7.7 × 4.0 µm . . . . *Celosia trigyna*
            7. Anticlinal wall straight on adaxial surface/curved on abaxial surface
        5. Cell shape polygonal on adaxial surface/irregular on abaxial surface
          9. Stoma almost thrice as long as wide on both surfaces . . *Celosia globosa*
          9. Stoma almost twice as long as wide on both surfaces . . . . . *Psilotrichum scleranthum*
            10. Mean stomatal number not greater than 10 on abaxial surface . . . . . *Aerva lanata*
            10. Mean stomatal number greater than 10 on abaxial surface . . . . . *Celosia argentea*
    3. Anticlinal wall curved on both surfaces
      11. Cell shape polygonal on adaxial surface/irregular on abaxial surface, trichome absent . . . . . *Alternanthera bettzichiana*
      11. Cell shape irregular on both surfaces, trichome present . . . . . *Gomphrena celosiodes*
  2. Stomatal type anomocytic with some other accompanying types
    12. Cyclocytic present . . . . . *Cyathula pedicillata*
    12. Cyclocytic absent

13. Stomata number not more than 20 . *Cyathula prostrata*
13. Stomata number more than 20 . . *Cyathula schimperiana*
    14. Anticlinal wall pattern similar on both surfaces
        15. Anticlinal wall undulate
            16. Mean stomatal size on abaxial surface 8.0 × 3.2 µm . . . . *Alternanthera ficoidea*
            16. Mean stomatal size on abaxial surface 12.0 × 8.0 µm . . . . *Alternanthera sessilis*
        15. Anticlinal wall curved
            17. Epidermal cell number 18–28 . . . . . *Alternanthera repens*
            17. Epidermal cell number 75–98 . . . . . *Amaranthus dubius*
    14. Anticlinal wall pattern different on either surface
        18. Stomata number not more than 30 . . . . *Achyranthes aspera*
        18. Stomata number more than 40
            19. Trichome absent . . . . . *Amaranthus viridis*
            19. Trichome present . . *Amaranthus pseudovirgata*
                20. Stoma thrice longer than wider . . . . . *Pupalia lappacea*
                20. Stoma twice longer than wider . . *Alternanthera pungens*

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