Pollen Morphology in Ornamental Plants: Systematics and Evolutionary Significance

Daniel A. Zhigila[1,2]; Fatima B.J. Sawa[3]; Jedidah J. Akawu[2] and Cleophas B. Ajiya[2]

Abstract

Pollen grain morphology of 20 species belonging to 18 genera in 11 families of some ornamental plants was employed for palynology. Polleniferous materials were properly collected, identified and utilized with the view of ascertaining their relevance to taxonomy. Nine types of pollen apertures namely – triporate, inaperturate, monoporate, dicolpate, monocolporate, monocolpate and tricolpate were recorded. Inaperturate pollen was the most common among the species, especially in Bougainvillea glabra (Comm. ex Juss), Jatropha caucas L., Cynodondactylon (L.) Pers. and Plumeria alba L. The analysis of mean polar and equatorial measurements i.e. the pollen sizes showed that largest pollen was recorded in Thevetia peruviana (L.) Lippold (782.69 μm^2*) and the smallest was recorded in Terminalia catappa L. (27.62* μm^2*). The mean density of pollen was found to be highest in Moringa oleifera Lam. (20.60±1.21* mm^2*) and lowest in*

Daniel A. Zhigila[1,2]; Fatima B.J. Sawa[3]; Jedidah J. Akawu[2] and Cleophas B. Ajiya[2]

[1] *Department of Biological Sciences, University of Cape Town, Private Bag, Western Cape, Rondebosch, 7701, South Africa*

[2] *Botany Programme, Department of Biological Sciences, Gombe State University, Gombe, Gombe State, Nigeria*

[3] *Federal College of Horticulture Dadin Kowa, Gombe State, Nigeria*

Correspondence: zhgdan001@myuct.ac.za; dandrawus@ymail.com; +2348065588811

Ixora casei L. (1.20±0.20 mm^2). Exine thickness ranged from 2.86 μm^2 as seen in J. caucas to 0.30 μm^2 in Breynia viscosa L. Evolutionarily, the common reticulate sexine pattern would be more primitive than the vermcate one. The typical reticulate sculpture pattern (such as in Hibiscus rosa-sinensis L.) would be basic, from which transitional variants would have derived and finally led to the typical verrucae pattern in Polyanthea longifolia Sonn. Similarly, the common boat-shaped-oblong pollen (as in Euphorbia mitis Pax.) would be more primitive than the circular pollen (B. glabara), from which the latter would have derived during phylogenetic development.

Keywords: Evolution, Ornamental plants, Palynology, Pollinosis, Taxonomy.

Introduction

Plants that display aesthetic values are used in landscaping industries either for their foliage arrangements, beautiful bright flowers, scents, fruits, stem or overall form used for horticultural trade regardless of their agricultural and other botanical significance are referred to as ornamental plants. These plants are found in houses, roads, gardens, parks, offices, public institutions such as hospitals, schools, stations, etc. which are for the decoration of individuals, horticulturists, guests, and the public. Flowering and floral features of ornamentals are a key aspect, as these plants are grown for their blossoms, with many horticulturalists preferring to cultivate exotic flowers even though combination with other morphological characteristics to delimit different taxa (Hosni and Araffa, 1999; Zhigila *et al.,* 2014; Banks and Rudall, 2016). For instance, the appearance of pores on the inner surface of the acetolysed pollen has proved to have reliable taxonomic characteristics (Perveen, 2003). It is used to identify hybrid plants and to solve the taxonomic problems of for example, medicinal and other economically important plants (Bibi, 2003; Noor *et al.,* 2009). Palynology has also been very helpful in phylogenetics, evolution and allergy but there is scanty research work on such studies (Nagels *et al.*, 2009; Qin *et al.*, 2015). Luo *et al.* (2015) suggested that pollen morphology has become integral part of understanding the evolution of the whole angiosperm.

Most studies on pollen of ornamental plant focused on the morphology of the species of Asteraceae family (Waisel *et al.*, 2004) and the systematic and evolutionary significance of pollen morphology has not been explored. In this study, an attempt was made to elucidate the evolutionary and taxonomic significance of morphological differences in the pollen grains of some ornamental plants.

Materials and Methods

Plant Materials and Palynology

Florets were obtained from fresh mature flower buds (before anthesis) of the plants species and preserved in glacial acetic acid. For each taxon, ten plant accessions were used and from each plant accessions, at least five to ten anthers were investigated and prepared for light microscopy (LM), methods of Abdulrahaman *et al.* (2013) was adopted. Pollen length (long polar ends) and pollen

Statistical Analyses

The coded data matrix was subjected to statistical analysis using R for taxa cluster analysis and principal component analysis (PCA) to determine diagnostic features that could be used for delimitation of taxa was used. Mean and the corresponding standard error, simple percentage, Analysis of Variance (ANOVA) and multivariate analysis and similarity index to reflect the evolutionary tree of the taxa based on the pollen features were calculated and constructed in (R=Core= Team, 2018).

Results and Discussion

Evolution of Pollen Shapes and Apertures

The pollen structure of 21 representative taxa observed under LM is described to typify the range of pollen characters and their states in plant used for their aesthetic values (Table 2 and 3, Fig 1–6). In all the studied plants, the pollen shapes in polar view were circular to spherical (except in *Terminalia catappa* which was concave-triangular). There exists the one-furrowed grain type of *Euphorbia mitis*, the smooth single-pored globular grain of *Cynodon dactylon*, or the three-furrowed pollen grain of *Thevetia peruviana* and *Duranta repens* have undoubtedly derived each from another by evolutionary derivation of the plants that produced such comparable pollen types (Wode house, 1936).Regarding aperture (pollen type), nine types were observed namely - monoporate, monocolpate, dicolpate, tricolpate, tricolporate, tetraporate and inaperturate (Table 2 and Fig 1–4). From the evolutionary point of view, *Hibiscus rosa-sinensis, E. mitis* and *Breynia viscosa* seems to be relatively unspecialized within the

2000; Rudall and Bateman, 2007) pollen aperture have displayed common changes throughout the phylogeny of angiosperm. Observably, Luo *et al.* (2015) suggested that the range of aperture types in monocots is fewer than that observed for other angiosperms but remain key to the evolution of monocots.

Frequency of Pollen Types

Inaperturate pollen was observed in most species with the highest frequency (33.30%), followed by monocolpate pollen which recorded 24.5%. Monocolporate, tetracolpate and tricolpate had the least occurrence (1.33%) each among the taxa (Table 2 and Fig. 5). Within the species, *D. repens* L., *I. coccinea* L. and *T. peruviana* showed the highest heterogeneity and diversity of pollen type with five types each. *Cynodon dactylon, J. curcas, Luculia grandiflora* Ghose and *B. glabra* recorded 100% inaperturate pollen aperture whereas *P. alba* had 100% pericolpate (Table 2 and Fig. 6).Entomophilies (insect pollinated) plants produce relatively few pollen grains e.g. *L. grandiflora* and *T. peruviana* (Table 2).

Pollen Sculpture

The exine sculpturing types in the studied species were reticulate (Fig. 3b), psilate (Fig. 4f), baculate (Fig. 2c) or micropitted (Fig. 4d). These variations are of significance in species delimitation in that, they correlated with morphological features (Qureshi *et al.*, 2002). From evolutionary and phylogentic point of view, pollen sculpture indicates primitiveness and advancement in plants (Takhtajan, 1969). *Thevetia peruviana, Duranta repens, I.*

T. catappa and *J. coccinea* L. (0.54 ± 0.46 μm^2) were relatively advanced considering their reduced exine (Table 3).

Pollen Length, Breadth and Size

The quantitative features of the pollen morphology in the studied species. Diameter of the equatorial axis in pollen grains varied from 5.60±0.06 in *T. catappa* to 31.40±1.03 μm in *T. peruviana (*Table 3). The length of the polar axis varied from7.00±0.44 to 32.8±0.84μm in *T. catappa* and *T peruviana* respective (Table 2). For the measurement of the pollen sizes, the polar and equatorial axes of the pollen grains were measured. Based on this character, the smallest and the biggest pollen grains were found in *T. catappa* and *J. caucas* respectively. The size of the pollen grains can help to describe the evolutionary trend among the taxa; they include increase in grain size, shortening of the colpi, increase in number of apertures and increase in the process of polymorphism. Talebi *et al*. (2012) reported that primitive plants have small pollen grains in comparison with the advanced plants. According to Talebi *et al.* (2012), pollen size was also found to be an index to chromosome numerical variations (aneuploidy and polyploidy) and has proved useful in cytopalynological studies. Varis (2009) reported in *Pinus sylvestris* L. that germinated pollen grains were larger in size than non-germinated. Thus, the size of pollen grains is a sign of pollen viability and the ratio of large pollen grains has been used to assess pollen performance (Talebi *et al.*, 2012).

they are all anemorphilous pollens because EAACI (2014) reported that most airborne pollens range in size from 12 to 70 μm^2. Clinically, anemophilous species produce allergic pollens even though they form a minority of flowering plants and they produce prodigious amount of pollens important of pollinosis is primarily due to flowering plants with wind dispersed pollens (EAACI, 2014). Although these anemophilous species form a minority of flowering plants, they produce prodigious amounts important of pollinosis is primarily due to flowering plants with wind dispersed pollens.

Incongruence from Multivariate Analyses

Overall multivariate morphometric assessment of the taxa using the pollen morphological data revealed some interesting similarities among the pollen of different ornamental plants tested in this study as evident from the similarity in the PCA plot (Fig. 8) and the cladogram (Fig. 9). The phenogramdid support the taxonomic similarities among some taxa as suggested by (Clarke *et al.*, 1980; Bashir and Khan, 2003). The multivariate analysis shows intra-specific clusters, indicating significant delimitation of the taxa. Al-Ghamadi *et al.* (2013) in the study of *Astragalus* as well Akhila and Beevy (2015) in *Sesamum* had the same evidence in species delimitations. The analysis clustered the taxa into three basic nodes with clear distinction of group (Fig. 9). It is worth to note that, species of the same genus such as *T. catappa* (closely similar to *Bignona venusta* Kerr.)and *T. radii* L. (more closely similar to *Moringa oleifera*) as well as *Ixora coccinea* (on same branch with *Jacobinea coccinea*) and *Ixora casei* (closely similar to *Mangifera indica* L.) were observed to be more closely similar to

Table : Nomenclature and pollen characters and their states used in analyses of character optimization of ornamental plants

Taxa	Family	Common name	Pollen Type	Pollen Sculpture	Density (mm^2)	Pollen frequency (%)
Thevetia peruviana	Apocynaceae	Oleandar	triporate	verrucate	1.20±0.20	7.14
			monocolporate	verrucate	6.8±1.20	28.57
			inaperturate	psilate	1.2±0.20	28.57
			monoporate	reticulate	1.4±0.25	7.14
			tricolporate	verrucate	2±0.45	28.57
Duranta repens	Euphorbiaceae	Yellow bush	monoporate	psilate	1.2±0.20	3.33
			inaperturate	psilate	3.6±1.12	60
			monocolpate	verrucate	1.2±0.20	10
			dicolpate	psilate	1.6±0.25	10
			triporate	psilate	1.8±0.49	16.67
Ixora coccinea	Rubiaceae	Flame of wood	inaperturate	psilate	5.4±1.12	21.92
			monoporate	psilate	2.8±0.37	12.33
			monocolpate	psilate	9.4±0.40	57.53
			dicolpate	psilate	1.2±0.2.	4.11
			tricolporate	psilate	2.6±0.40	4.11
Bignonia venusta	Bignoniaceae	Flame vine	monocolpate	verrucate	3.2±0.20	75
			dicolpate	psilate	1.2±0.20	25
Euphorbia mitis	Euphorbiaceae	Plumeria	monocolpate	psilate	2.4±0.25	53.85
			dicolpate	psilate	1.2±0.20	30.77
			inaperturate	psilate	1.2±0.20	7.69
Breynia viscosa	Rubiaceae	Snow bush	monocolpate	psilate	5.6±0.40	60
			dicolpate	psilate	4.2±0.2.	40
Terminalia catappa	Combretaceae	Indian almond	monocolpate	verrucate	2.0±0.45	14.29
			inaperturate	psilate	3.0±1.23	28.57
			dicolpate	verrucate	1.6±0.25	57.14
Ixora casaei.	Rubiaceae	flame of wood	monocolporate	verrucate	1.2±0.20	12.5
			tricolporate	verrucate	2.8±0.20	37.5
			inaperturate	psilate	1.2±0.20	50
Nerium indicum	Apocynaceae		inaperturate	psilate	2.4±0.20	67.05
			tetracolpate	verrucate	1.2±0.20	18.18
			monocolpate	verrucate	1.2±0.20	9.09
Bouganvilia glabra	Nyctaginaceae	Bougainveilia	inaperturate	micropitted	2.8±0.58	100
Jacobinea coccinea	Rubiaceae		inaperturate	micropitted	6.4±1.69	44.44
			monocolpate	verrucate	2.0±0.32	13.89
			triporate	verrucate	6.0±2.15	41.67
Terminalia radii	Combretaceae	Satellite	monocolpate	verrucate	4.8±0.74	50
			inaperturate	verrucate	2.2±0.20	16.67
			monoporate	verrucate	1.4±0.25	8.33
			triporate	verrucate	4.2±0.20	25
Luculia grandiflora	Rubiaceae	White luculia	inaperturate	psilate	1.2±0.20	100
Jatropha caucas	Euphorbiaceae	Jatropha	inaperturate	psilate	3.0±0.32	100
Moringa oleifera	Moringaceae	Moringa	inaperturate	psilate	20.6±1.21	57.43
			triporate	verrucate	3.8±0.74	10.81
			monocolpate	baculate	8.6±1.69	24.32
			dicolpate	psilate	1.2±0.20	0.68
Phyllanthus nivosus	Euphorbiaceae	Ice plant	inaperturate	psilate	2.2±0.20	40
			monoporate	psilate	2.8±0.20	60
Hibiscus rosa-sinensis	Asteraceae	Chinese rose	dicolpate	reticulate	5.6±0.33	45
			Inaperturate	verrucate	3.2±0.76	65

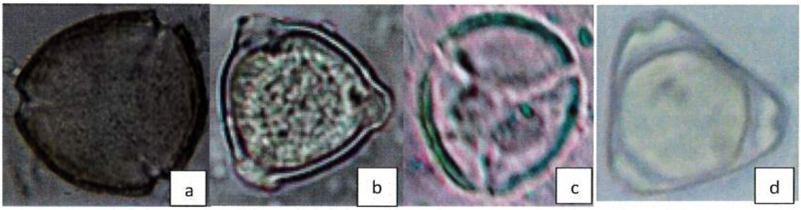

Fig. 1: Pollen morphology - triporate pollens arranged following the topology of Luo *et al*. (2015) a: polar view showing triporate pollen aperture of *Thevetia peruviana*, b: Triporate pollen in *Jacobinea coccinea*, c: tricolporate pollen in *Ixora coccinea* d: trianporate pollen in *Terminalia catappa* x1000

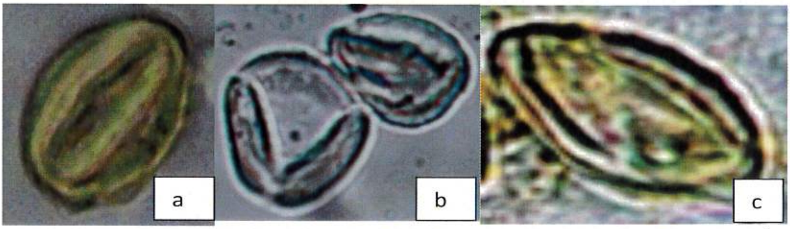

Fig. 2: Oblique equatorial view, showing two colpi located at the proximal pole – dicolpate pollens a: diparacolpate pollen of *Hibiscus rosa -sinenesis*; b: diarchcolpate pollen in *Breyniaviscosa*; c: dicircolpate pollen in *Euphorbia mitis* x1000

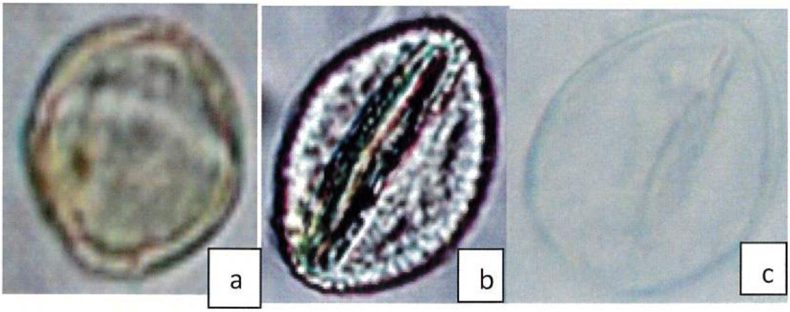

Fig. 3: Distal view of monocolpate pollens a: monoarcolpate pollen of *Jacobinia coccinea*, b: monoparacolpate pollen in *snow bush* ; c: monocircolpate pollen in *Polyanthea longifolia* x1000

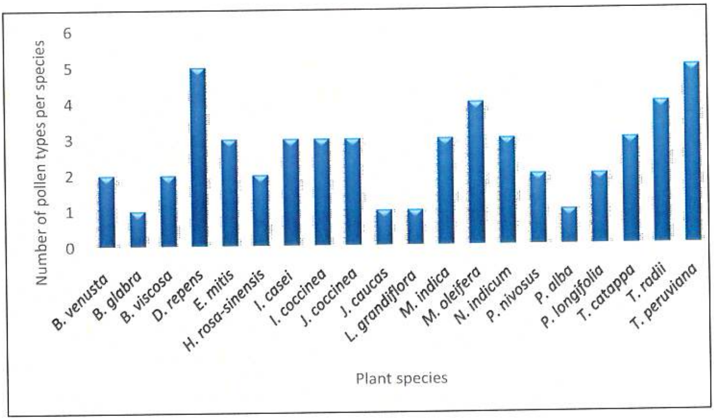

Fig. 6: Number of pollen types in each species.

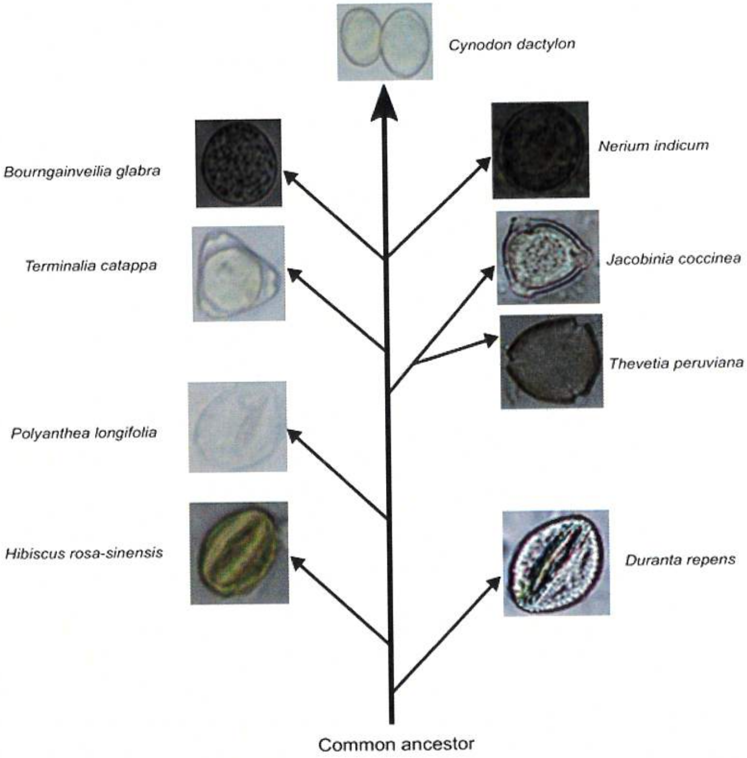

Fig. 7: Evolutionary hypothesis based on pollen features in ornamental plants

References

Akhila, H. and Beevy, S.S. (2015). Palynological characterization of species of *Sesamum* (*Pedaliaceae*) from Kerala: a systematic approach. *Plant Systematics and Evolution,* 301:2179–2188.

Al-Ghamadi, F., El-DeenOsman, A.K. and Guetat, A. (2013). Contributions to the pollen morphology of Genus Astragalus L. (Fabaceae) and its taxonomic implications. *Asian* Journal of Plant Science 12:176–189.

ASCIA (2015). The Australasian Society of Clinical Immunology and Allergy. ASCIA Information for patients, consumers and carers. Australia. Pg 1 – 4.

Banks, H., and Rudall, P.J. (2016). Pollen structure and function in *Caesalpinioid* legumes. *American Journal of Botany*103: 423 – 436.

Bashir, S. and Khan, M.A. (2003). Pollen morphology as an aid to the Identification of Medicinal plants: *Trianthema portulacastrum* L., *Boerhaavia procumbens* L., *Alternanthera pungens* Kunth. *Hamdard Medicus*, 66: 7–9

Bibi, K.(2003). Palynological Study of Selected Medicinal Plant Species of Family Malvaceae. Department of Botany University of Arid Agriculture, Rawalpindi, pp: 15-18.

Clark, W.D., Brown, G.K. and Mayes, R.A. (1980). Pollen Morphology of *Haplopappus* and related Genera (*Compositae*). *American Journal of Botany*, 67: 1391–1393.

Cronquist, A. (1968). *The Evolution and Classification of Flowering Plants*. Houghton Mifflin. Boston. Pp 34.

EAACI(2014). European Academy of Allergy and Clinical Immunology. Allergy Awareness Campaign: Food Allergy and Anaphylaxis.

Martin, J., Raymundez, M.B., Valles, J., Garnatje, T. andRaimundez, E. (2012). Palynological study of the Venezuelan species of the genus *Hymenocallis* S. alisb. (*Amaryllidaceae*). *Plants Systematics and Evolution* 298:695–701. doi:10.1007/s00606-011-0579-z

Moore, P.D., Webb, J.A. and Collinson, M.E. (1991). *An Illustrated Guide to Pollen Analysis*. 2nd Edn., Hodder and Stoughton, UK.

Nagels, A., Muasya, A.M., Huysmans, S., Vrijdaghs, A., Smets, E. andVinckier, S. (2009). Palynological diversity and major evolutionary trends in Cyperaceae. *Plants Systematics and Evolution* 277: 117–142.

NIAID(2012). National Institute of Allergy and Infectious Diseases health information. Pollen allegy. www.niaid.nih.gov.

Njokuocha R.C. and Ezenwajiaku T.E. (2010). Airborne pollen flora in Aguata Local Government Area, Anambra State, Nigeria. *Nigerian Journal ofBotany* 23(2): 211-224.

Noor, M.J., Khan., M.A. and Camphor, E.S. (2009). Palynological analysis of pollen loads from pollen sources of honeybees in Islamabad, Pakistan. *Pakistan Journal of Botany* 41(2):54

Nur, N. (1976). Studies on pollination in Musaceae. *Annals* of Botany\ 167–177.

Perveen, A. (2000). Pollen Characters and Their Evolutionary Significance with Special Reference to the Flora of Karachi. *Turkey Journal of Biology*, 24 :365–377 induced allergy. *Aerobiologia*, 20: 197–208

Willard, D.A., Cooper, S.R., Gamez, D. and Jenshen, J. (2004). Atlas of pollen and spores of the Florida everglades. *Palynology*, 28: 175 – 227.

Wodehouse, R.P.(1936). Evolution of Pollen Grains, *Botanical Review*, 2(2): 67-84

Wortley, A. H., Wang, H. Lu, L. Li, D.Z. and Blackmore, S. (2015). Evolution of angiosperm pollen: 1. Introduction. *Annals of Missouri Botanical Garden* 3: 177–226.

Xiong, Z., Chen, S., Hong, D. and Luo, Y. (1998). Pollen morphology and its evolutionary significance in Hememcallis (Liliaceae). *Nordic Journal of Botany*, 18(2):183 – 191.

Zhigila, D.A.; Sawa, F.B.J.; Abdul, S.D. and Danailu, G. (2014). Diversity of pollen morphology in accessions of *Sesamum indicum* L. *International Journal of Modern Botany*, 4(1): 22 – 28.