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Palynological and Carpological Features in Four *Jatropha* Species (Euphorbiaceae) as Taxonomic Characters

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ABSTRACT: *Taxonomic relationships of four Jatropha species namely: J. curcas L., J. gossypifoliaL., J. multifidaL. and J. podagrica Hook. were studied using their pollens, fruits and seeds. Four types of pollens were seen in the four species namely: panporate, syncolpate, monovesiculate and bivesiculate pollens. Panporate type occurs in all the species with 100% frequency in J. curcas and J. multifida. The pollen sizes ranges from 822 µm in syncolpate pollen type in J. gossypifolia to 2013.73 µm in the panporate pollen type in J. multifida. Syncolpate, and monovesculate and bivesculate pollens occurred only in J. gossypifolia and J. podagrica respctively. The lowest mean fruit and seed length (12.10 mm and 6.17 mm respectively) and width (13.44 mm and 4.14 mm respectively) values were in J. gossypifolia. The highest mean fruit and seed length (27.52 mm) and width (12.44 mm) values were recorded in the J. multifida respectively. A combination of palynological and carpological features is shown to be important in the taxonomy and systematics of the four Jatropha species.*

Key words: palynomorphs, stomata, epidermal cells, taxonomy, *Jatropha* species

Introduction

The genus *Jatropha*, which belongs to the family Euphorbiaceae and consists of 175 species (Olowokudejo, 1993) which are mostly trees, rhizomatous sub shrubs and herbs (Dehgan, 1984). The genus *Jatropha*is characterized by leaves which may be simple to palmately 3, 5 or 7 lobed or divided into a maximum of 11 segments. Earlier taxonomic treatments of the genus were evaluated on the basis of morphological (Dehgan and Webster, 1979; Dehgan, 1980), wood anatomy (Oladipo and Illoh, 2012a) and leaf and seed electrophorensis (Oladipo and Illoh, 2012b; Oladipo*et al*., 2008), leaf epidermal features (AbdulRahaman and Oladele, 2010). Leaves in various species were reported to be heterogenous with regard to size and architecture (McVaugh, 1945; Dehgan and Webster, 1979; Dehgan, 1982). Leaf sizes varies from 2-3 mm in extreme xeric habitats to 20 cm or more under mesic conditions.

The four *Jatropha* species are important for their potentials. The seeds of *J. curcas* contain 27-40% oil (Achten*et al*., 2007) that can be processed to produce a high-quality biodiesel fuel, usable in a standard diesel engine. *Jatrophamultifida* i.e. coral plant is grown for its distinctive large leaves and its flashy red flowers. This is a perfect container plant for a sunny patio or at poolside. The leaves have a strange and unusual tropical look, and coral plant is often grown as a novelty specimen or accent. It is also a welcome shrub in mixed shrub borders and often used in cactus and succulent gardens. *Jatrophapodagrica* is also known for its incredible ability to attract a variety of butterflies wherever it is grown. It is an attention grabber and a must in every tropical garden. It has a large bottle-like caudex and huge leaves up to 10-12 inches in diameter. The more shade, the bigger the leaves. The bark of *J. gossypifolia*is considered an emmenagogue and had been used to procure abortions. The seed oil is used to treat constipation, leprosy and in paralytic affections. The leaves in a decoction are used to treat fever in the form of a bath, while the juice is given to treat sores on the tongue of infants (Nadkami*et al*., 1976; Odugbemi, 2008).

Meanwhile, flowers, especially colours of flower, are used to identify the *Jatropha* species (Hutchinson and Dalziel, 1958; Iwu, 1993). The flower colour alone might not be enough; other features such as leaf epidermis, pollens, electrophoresis etc should be incorporated. The ability to identify plants from their pollen has enabled botanists and ecologists to reconstruct past assemblages of plants and identify periods of environmental change (Fægri and Iversen 1989; Moore *et al*. 1991). Morphological characteristics of pollen grains also can be useful characters in studies of plant taxonomy because many pollen traits are influenced by the strong selective forces involved in various reproductive processes, including pollination, dispersal, and germination (e.g. Erdtman, 1952; Moore, *et al*. 1991; Nowicke and Skvarla, 1979; Stuessy, 1990). At the same time, characters subject to strong selection can be misleading if they reflect convergent evolution (similar evolutionary responses by unrelated taxa to similar environmental conditions). Thus, the use of pollen morphology as a taxonomic character is challenging, and pollen characteristics must be considered in concert with other characteristics in evolutionary reconstructions.

In this study, it is the intention of the authors to document pollen, fruit and seed characteristics of four members of the genus *Jatropha* (Euphorbiaceae) and examine how these characters relate to our current understanding of the systematics of these plants.

Materials and Methods

Study materials

Flowers and fruits with seeds of four species of *Jatropha* namely *J. curcas, J. gossypifolia, J. multifida* and *J. podagrica* were collected from living plants in their natural habitats and were studied palynologically and carpologically respectively. The plants were identified at the Herbarium Unit of the Department of Plant Biology, University of Ilorin, Ilorin, Nigeria.

Microscopic studies and isolation of pollens

Pollens were collected from the anthers of the flowers of plant studied. The pollens were smeared on the glass slide and a few drops of isopropyl alcohol (IPA) were added to remove waxy surface from the pollen. This was left for 10 mins. The specimen on the slide was mounted with glycerine for microscopic observations. The cover slip was ringed with nail varnish to create a semi-permanent pollen slide. Observations were recorded with photographs of pollens and tables. This method followed that of Horrocks*et al*., (1999).

Pollen identification

Pollens observed were identified using some pollen atlas (Bambara and Leidy, 1991; Jones *et al*., 1995).

Frequency of pollen types

Using 35 fields of view of OLYMPUS microscope at x40 objective as quadrat, the number of pores on pollens was noted to determine the frequency of the different pollen types present. Frequency of each pollen type was expressed as percentage occurrence of such pollen type based on all occurrences using this formula:

p/y x 100

Where

p = the occurrence of each pollen type in the field of view

y = total occurrences of all pollen types.

Density of pollen types

The density of pollen type was determined as the number of pollen types per square millimeter (mm^2). Fields of view at x40 objective in a square millimeter was used for the counting.

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Pollen size

Pollen size was measured as the product of length multiplied by breadth and also multiplied with 0.79 using eye piece micrometer. A sample of 30 pollens was used.

Fruit and seed size measurement

Using an electronic digital calliper (Titan 23175 model) the length and width of fruits and seeds of the four *Jatropha* species were measured. Minimum and maximum length and width with mean values recorded.

Photographs of pollens, fruits and seeds

Photographs of pollens from prepared microscopic slides were taken with Amscope microscopic camera (MU1000, FMA050) and Kodak digital camera (Kodak Easy Share C913). The camera is attached to the computer system (laptop) where specimen viewing on the microscope is observing on the computer system. Photographs of the fruits and seeds were also taken using Kodak digital camera (Kodak Easy Share C913).

Statistical Analysis

All data were reported and analyzed using analysis of variance [ANOVA] and Duncan's multiple range test [DMRT]. A computer software SPSS version 16.0 (2007) was used. A probability value 0.05 was used as bench mark for significance difference between parameters.

Results

The four *Jatropha* species studied in this work possessed four types of pollens namely panporate, syncolpate, monovesculate and bivesculate (Table 1; Figs. 1A – H). Panporate pollen is the most common type occuring in the four species with 100% frequency in *Jatropha curcas, J. multifida*,while *J. gossypifolia*and *J. podagrica* possessed one and two other types of pollens respectively. Along with panporate pollen, *J. gossypifolia* also has syncolpate; *J. podagrica* has monovesculate and bivesculate pollens.

The mean values of the friuts and seeds of the four *Jatropha* species is presented in Table 2, while the plates showing the pictures of fruits and seeds is in Fig. 2. The mean fruit and seed sizes are significantly different (P<0.05) in all the studied species. The lowest mean fruit and seed length (12.10 mm and 6.17 mm respectively) and width (13.44 mm and 4.14 mm respectively) values were in *J. gossypifolia*. The highest mean fruit and seed length (29.69 mm and 16.32 mm) were in *J. curcas* and fruit and seed width (27.52 mm and 12.44 mm) values were recorded in the *J. multifida* respectively.

Table 1: Pollen features of four species of *Jatropha*

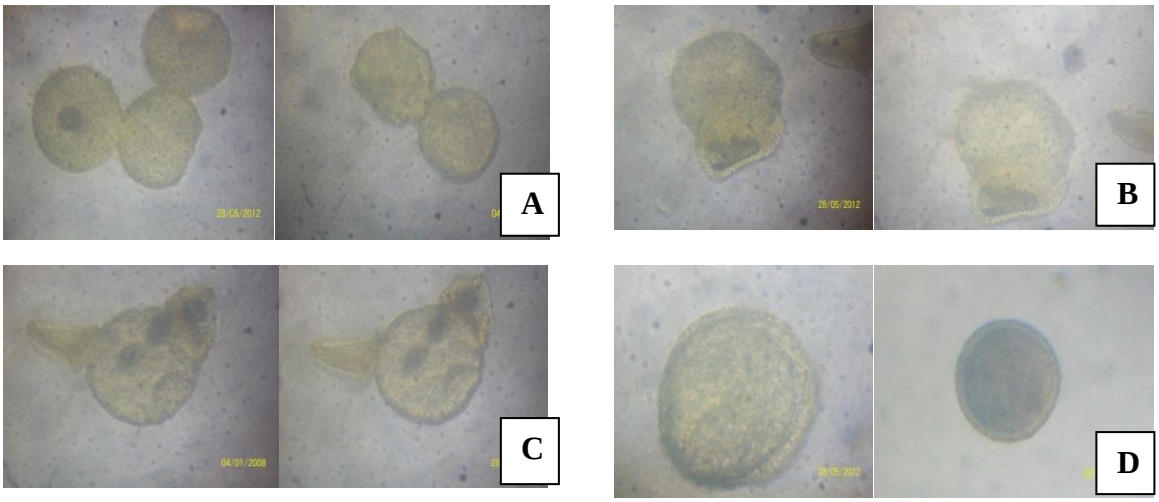
Species	Pollen types	Pollen frequency (%)	Pollen size (µm)
Jatrophacurcas	Panporate	100	1,553.67 [c]
Jatrophagossypifolia	Panporate	78	831.34 [a]
	Syncolpate	22	822.06 [a]
Jatrophamultifida	Panporate	100	2,013.75 [e]
Jatrophapodagrica	Panporate	85	1,036.20 [b]
	Monovesculate	5	960.32 [b]
	Bivesculate	10	1,749.58 [d]

Means with same letters along columns are not significantly different

Table 2: Fruit and seed size of four *Jatropha* species

Species	Fruit (mm)						Seed (mm)					
	Length			Width			Length			Width		
	Min.	Max.	Mean	Min.	Max.	Mean	Min.	Max.	Mean	Min.	Max.	Mean
Jatrophacurcas	28.54	31.27	29.69 [d]	22.16	29.48	26.78 [b]	14.48	17.17	16.32 [d]	9.68	16.67	11.16 [c]
Jatrophagossypifolia	10.71	13.45	12.10 [a]	11.01	27.47	13.44 [a]	5.24	6.63	6.17 [a]	3.89	4.41	4.14 [a]
Jatrophamultifida	25.56	27.02	26.12 [c]	26.24	29.21	27.52 [b]	15.71	17.04	16.23 [c]	11.69	13.63	12.44 [d]
Jatrophapodagrica	14.04	17.81	15.70 [b]	11.89	14.28	13.20 [a]	11.25	12.46	11.96 [b]	5.91	6.80	6.40 [b]

Means with same letters along columns are not significantly different



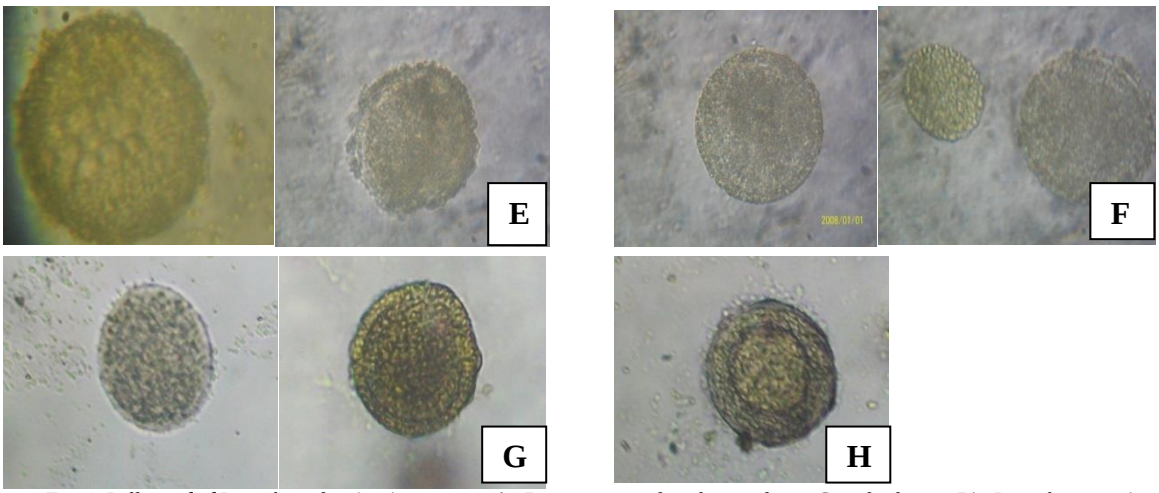


Fig. 1: Pollens of of*Jatrophapodagrica* (panporate –A, B – monovesculate, bivesculate – C, and salcate – D), *Jatrophacurcas*(panporate – E), *Jatropha multifida* (panporate – F), and *Jatropha gossypifolia* (panporate – G, syncolpate – H) x600

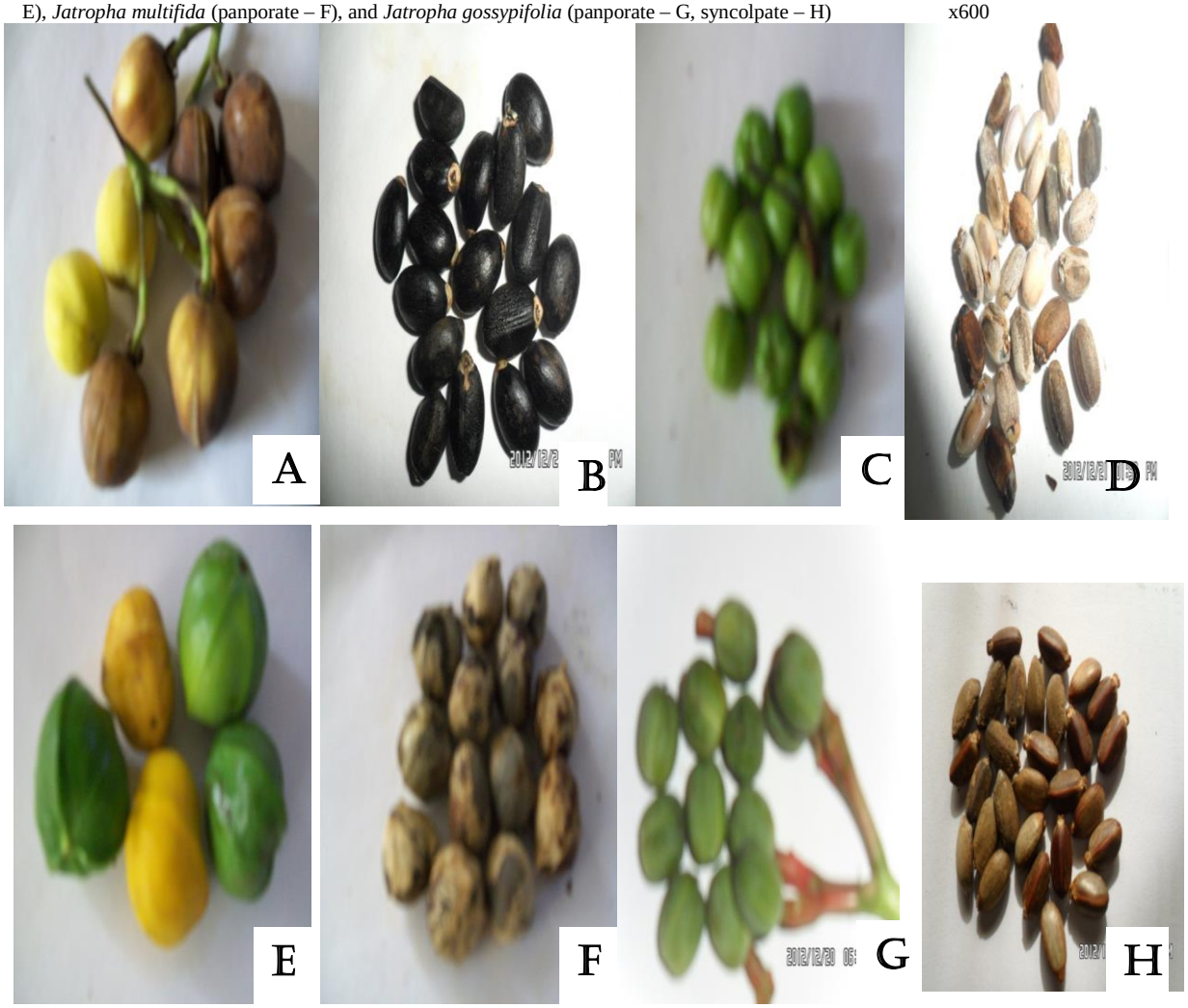


Fig. 2:Fruits and seeds of*Jatropha curcas* (A &B), *Jatropha gossypifolia* (C & D), *Jatropha multifida*(E & F) and *Jatropha podagrica* (G & H)

DISCUSSION

Palynological (i.e. study of Palynomorphs - organic-walled microfossils between 5 and 500 micrometres in size e.g. pollens, spores etc) and carpological (i.e. study of fruit and seed) features have been used for classification in taxonomy and systematics of many plants (Sukhorukov, 2007; Oswald *et al*., 2001; Nwokocha, *et al*., 2012). The utilization of morphological characters such as vegetative and floral for delimitation of closely and distantly related taxa is age long. Recently, Akyalcin, *et al.* (2006), Mbagwu and Edeoga (2006), Agbagwa (2007), Jafari, *et al*. (2009) and Silva *et al*. (2011) employed these important taxonomic characters e.g. vegetative and floral in the elucidation of different plant genera. Similarly AbdulRahaman and Oladele (2010); AbdulRahaman, *et al*. (2009); AbdulRahaman and Oladele (2005); Abubakar and Yunusa (1998) and Olowokudejo and Pereira-Sheteolu (1988) have stressed the usefulness of leaf epidermal features such as stomalal types, density index and size, epidermal cell types and anticlinal cell wall patterns as good taxonomic tools in delimiting species of the same genus or genera within a family.

Four types of pollens observed in the four *Jatropha* species and occurrence of panporate type in all of the; is an indication that pollen is a good taxonomic character. This confirm their relationships i.e. similarity and difference. Although, pollen has been used to delimit many species in many plant genera (Oswald *et al*., 2001); the authors are not aware of any previous or earlier work on the pollen structures of the four studied *Jatropha* species as taxonomic and systematic character. This work, therefore, may be a pioneer study in this regard.

The fruits and seeds of the *Jatropha* species showed variations of taxonomic relevance (Fig. and, Table 2). *J. gossypifolia* and *J. podagrica* fruits are 3-seeded globose capsule, sparingly pubescent to glabrous, usually green in colour, turning brown and dehiscing into 2-valved cocci when mature; comparatively, *J. multifida* are 3-seeded ellipsoid capsule, glabrous, green to yellow in colour turning dark brown when mature and dehisces. *J. curcas* was observed to have both ellipsoid and tear-drop-shaped fruits implying that there may be morphological and ecotypes in this species. This fact is of significant important since only the ellipsoid shaped fruit has been reported (Fairless, 2007; Nwokocha, *et al*., 2012). This pattern was earlier identified by Nwokocha, *et al*., 2012 in their studies on four species of *Jatropha* in Niger Delta, Nigeria. Also noteworthy is significance differences in the length and width in fruits and seeds of the four species. Fruit and indeed seed of *J. curcas* are the largest while that of *J. gossypifolia* are the smallest.

In conclusion, the similarities and dissimilarities in pollens, fruits and seeds of the four *Jatropha* species is evidence that these features can be used to delimit the species within the genus.

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