

## CHARACTERIZATION OF POLLEN GRAINS AND SPECIES AFFINITY OF SOME NIGERIAN SEDGES

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### ABSTRACT

Pollen grains of some common species of the genera *Kyllinga*, *Mariscus*, *Torulinium*, *Cyperus*, *Eleocharis* and *Fimbristylis* formed the main material for this study. Analysis of pollen grains in terms of size, shape, wall sculpturing and fertility was done. The results show some interspecific and intergeneric variations in terms of size and shape of the pollen grains. The wall sculpturing produced by the exine projections was the same in all the species studied. This similarity in the wall sculpturing is suggestive of a common ancestor for the group studied. The species were separated into two groups based on the pollen subtypes. The large pollen subtypes had pollen grains with the size range of  $8.30 \pm 0.61$  to  $17.72 \pm 1.31$   $\mu\text{m}$  while the small pollen subtypes had the size range of  $6.10 \pm 0.57$  to  $7.50 \pm 0.52 \mu\text{m}$ .

### INTRODUCTION

In recent years the number of workers on pollen grains have increased and reports of pollen analysis of many plants are now available.

Palynology, the study of pollen grain, is concerned both with the structure and formation of pollen grains of angiosperm and spores of cryptogams (Moore and Webb, 1978). The insight necessary to discern the potentialities of pollen analysis was until recently limited to a few Scandinavian Botanists.

Erdtman (1952), studied the pollen morphology of some Rutaceae species around the world for taxonomic and palaeo-botanical purposes. Mukhopadhyay (1971) and Mukherjee (1978) investigated the pollen grains of the genera *Campylostachys* and *Stilbe*.

Bonnefille and Riollet (1980) described in detail the pollen of eleven species of *Indigofera* with light microscopy. Ferguson and Skvaria (1981) reported differences in ornamentation and exine stratification and that these might have taxonomic value within the Family Indigofereae.

Barth (1980; 1982) has presented a scanning electron microscope general survey of the pollen morphology of Brazilian Rutaceae by examining their size, shape, wall sculpturing and genetic content for evolutionary purpose. In spite of the volume of work on pollen analysis, there has been no analysis of the Family Cyperaceae.

The Cyperaceae commonly called the sedges is an interesting group of plants represented throughout Nigeria by about 230 species. Pollen grains from the commonest species encountered in South Eastern Nigeria form the basis of the present study.

## MATERIALS AND METHODS

Pollen grains were taken directly from mature flowers in the field. The pollen grains were dusted on clean slides, mounted with FLP orcein and labelled to ascertain source and location of collection. Some were also dusted on white filter paper and packed in stacks in separate envelopes. A record of each collection was kept including the nature of the habitat. The collection was done for at least 20 plants from each of the 15 locations spread over South Eastern part of Nigeria. The collection was done twice daily for each location, that is, in the morning before 9.00am and in the evening between 5.00 and 7.00 pm.

Pollen from unopened flower buds were also collected and in this case the glumes were teased with dissecting pins to expose the anthers and release the pollen grains. The grains were also mounted in FLP orcein and studied for viability.

Each slide was examined under the light microscope at a constant magnification of  $\times 400$ . The diameter of at least 50 pollen grains of each species studied from the 15 locations were measured using the ocular micrometer. The data obtained were measured and carefully recorded

against each of the specimens used. The measurements were later converted to microns using the stage micrometer. Observations were also made on the shape of the pollen grains and the wall sculpturing were studied under oil immersion ( $\times 1000$ ), with phase contrast.

Viability of pollen grains was determined by staining with cotton-blue in lactophenol. Deeply stained grains were recorded as viable faintly and lightly stained grains were recorded as non-viable. Pollen fertility was determined as a ratio of normally germinated pollen to the total number of pollen grains after the grains were incubated for 12 hours in a sugar nutrient solution. Pollen viability results were collaborated with pollen fertility results.

## RESULTS AND DISCUSSION

A summary of the pollen features of the species of Sedges studied is given in Table 1. The different species present two shapes of pollen grains, that is spherical and oval. More of the species have spherical pollen grains while fewer possess oval pollen grains (Table 1).

A wide variation in pollen diameter within the groups studied was recorded (Table 1). The variation of pollen diameter range from  $6.10 \pm 0.55\mu\text{m}$  in *Mariscus longibracteatus* to  $9.50 \pm 0.99\mu\text{m}$  in *Cyperus dilatatus*. However, the variation within each group/species is very low. This definitely accounts for the small standard errors calculated. Intraspecific variation in pollen size was very low, but interspecific variation was very significant (Table 1).

The smallest pollen diameters were recorded in *Mariscus longibracteatus* ( $6.10 \pm 0.57\mu\text{m}$ ). *Cyperus dilatatus* has pollen diameters of  $7.50 \pm 0.99\mu\text{m}$ . Based on the diameter of the pollen grains no strong conclusion can be drawn on the affinity of the species studied. In terms of pollen shape too, there is no definite direction of affinity. Some possess oval shaped grains while others possess spherical shaped pollen. The genus *Fimbristylis* showed the highest variation in pollen diameters hence the large standard error.

The morphology of the pollen grains is also unique. The exine is more or less verrucate, that is, the surface has projecting elements whose width are as great as height and semi tectate. The grains of the genus *Eleocharis* have elements with slightly irregular shape, uniformly distributed all over the surface. The grains in all the species are monocolporate and the

sculpturing of the exine is also uniform for all the species, suggesting a common ancestor.

Pollen viability and fertility was very high in all the species and groups studied (Table 1). It was rarely below 95% (*Scirpus lateriflorus*), and showed no consistency in pattern of variation. That is that viability and fertility was not species dependent. Plate 1 showed the two types of pollen grains recorded in the species studied.

Pollen shapes is also genetically controlled and the uniformity of the grains morphology is also an indication of the common ancestry of the sedges. Among the groups studied *Kyllinga*, *Mariscus*, *Eleocharis* and *Scirpus* possess spherical pollen grains while the rest possess oval grains. The shape of the grains is generic since all the species of a particular genus in this study possess the same grain shape. The shape of the grains was uniform in all the locations for a particular genus and species.

Pollen grains have proved extremely useful in a variety of ways in the study of plants relationships. Thus pollen analysis has occupied a prominent position in Botany and paleo-botany inspite of numerous attendant problems encountered in its study. Because the distribution of pollen in the space is always changing as a result of changing climatic and ecological factors, including the activities of man, pollen analysis has been used in tracing the history of plant communities. The challenges and results of pollen analysis have given the plant scientist the inspiration to attempt a detailed reconstruction of plant communities, using the information on the distribution of pollen on the surface of the earth.

The variation in the pollen grain size of the species of sedges studied was very significant. However, the external surface of the pollen grains presented a uniform appearance of verrucate, semi tectate, monocolporate sculpturing. The exine stratification is simple and uniform all over the grain surface. Among the group of the sedges studied the appearance of pollen grains shape and size of the grains. Despite the conspicuous diversity in the pollen size and the apparent absence of a well defined separation among the group studied, there is a close affinity within the species based on the isodiametric projections on the exine (Plate 1). This type of affinity suggests a parallel evolution of members of the group after a split from a common ancestor. This conclusion is similar to that of Ray (1985) who used pollen morphology to draw and establish relationship between emembers of the Family Stilbaceae Kunth.

In terms of pollen size, the species can be rearranged in order of descending magnitude beginning with *Kyllinga erecta*. The size of pollen grains has been used to determine the position of species in evolution. It has been overtly suggested that there has been a systematic reduction in pollen grains size generally in the plant kingdom to allow for effective pollen transfer in the course of evolution. This will therefore suggest that *Kyllinga erecta* with the largest pollen grains is the oldest member of the group studied, while *Mariscus longibracteatus* with the smallest pollen grains (Table 1) is the most recent in the evolutionary hierarchy. In spite of the variation, the pollen grain size is significantly close and this fact and the uniform pattern of the exine sculpturing can be used to support suggestion that the group has a common origin or gene pool. This position had been reported by Godwin (1968).

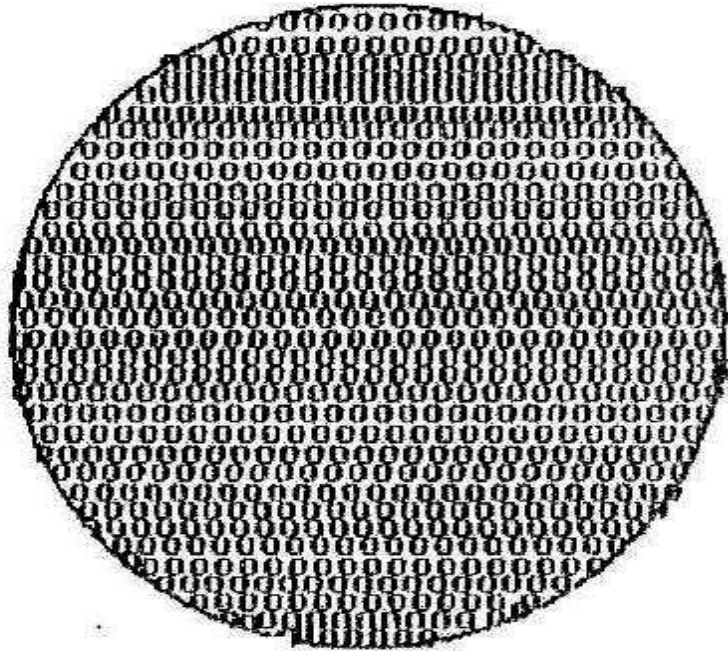
The absence of abnormal pollen grains and the high pollen fertility recorded indicates a high regularity in the meiotic events. Even though seed set was not studied, the usually large population whenever the group is encountered indicates good seed set and high seed viability.

Finally, using the data on pollen analysis, the species studied can be divided into two groups based on pollen size, that is two pollen Subtypes. Following this therefore *Kyllinga erecta*, *Kyllinga nigritana*, *Cyperus margaritaceus*, *Fimbristylis nigritana*, *Eleocharis geniculata* and *Torulium odoratum* form Subtype 1 with large pollen grains while *Scirpus cubensis*, *Mariscus alternifolius*, *Cyperus distans* and *Scirpus lateriflorus* form Subtype 2 with small pollen grains. A similar grouping was suggested by Barth (1982).

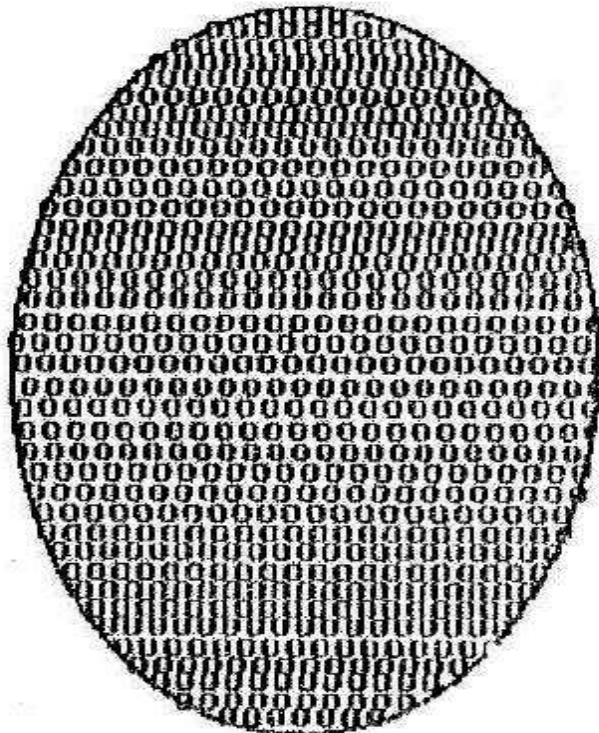
Table 1: Pollen data (source and characteristics)

Species	Source	Habitat	Diameter	Viability	Fertility	Shape
<i>Kyllinga erecta</i> Schumacher	Uyo	Sandy soil	9.30 ± 0.23	97.5	Oval	
<i>Mariscus alternifolium</i> Vahl	Eket	Swamp soil	7.50 ± 0.52	96.92	98.25	Spherical
<i>Torulium odoratum</i> Hooper	Calabar	Wet Soil	8.30 ± 0.67	89.99	96.94	Spherical
<i>Kyllinga erecta</i> Schumacher	Akamkpa	Farm land	9.12 ± 0.41	98.81	95.76	Oval
<i>Mariscus longibracteatus</i> Cherm	Uyo	Dry soil	6.10 ± 0.57	90.56	97.9	Oval
<i>Cyperus dilatatus</i> Schum/Thonn	Pt. Harcourt	Marshy soil	9.50 ± 0.99	99.15	97.54	Spherical
<i>Cyperus margaritaceus</i> Vahl	Aba	Sandy soil	8.61 ± 0.52	95.5	98.92	Spherical
<i>Cyperus dilatatus</i> Schum/Thonn	Okigwe	Rocky soil	8.98 ± 0.87	67.21	99.05	Spherical
<i>Scirpus cubensis</i> Peoppig/Kunth	Uyo	Farm land	6.80 ± 0.42	96.37	98.61	Spherical
<i>Cyperus distans</i> Linn.	Owerri	wet land	6.80 ± 0.51	98.16	96.93	Spherical
<i>Kyllinga nigritana</i> C. B. CL	Uyo	Dry land	9.21 ± 0.62	95.71	98.25	Oval
<i>Fimbristylis nigritana</i> C.B.CL	Uyo	Wet land	9.72 ± 1.31	98.11	96.36	Oval
<i>Fimbristylis nigritana</i> C. B. CL	Eket	Wet land	9.68 ± 0.89	95.07	97.05	Oval
<i>Eleocharis geniculata</i> Reomer/Sc.	Calabar	Swamp soil	8.79 ± 0.75	97.19	97.25	Spherical
<i>Scirpus lateriflorus</i> Peoppig/Kunth	Ikot Ekpene	Farm land	7.10 ± 0.37	96.45	95.97	Spherical

Plate 1 a: The pollen grain of *Mariscus longibracteatus*  
The pollen grain is spherical and small.



b: The pollen grain of *Kyllinga nigrilana*  
The pollen grain is oval and large.



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