



Comparative Morphological Features of Pollen Grains and Their Significance in The Taxonomy of Seventeen Cola Species in Nigeria

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Abstract

Cola Shott and Endl belongs to the family Sterculiaceae and the name was coined by Schott. and Endl. in 1832. Most Cola species grow in moist environments and they are indigenous to Africa. Fresh flower buds from the field which were later dried and dried specimens from herbaria were used for the study. The methods of acetolysis by Erdtman (1952) and Ayodele (2000) were used to prepare the pollen grains. Observation and study of the pollen grains were done using light microscope. Photomicrographs were taken using Olympus CX31 microscope with a camera attached. The pollen features studied include the surface sculptures, furrows, pores, shape/classes, polar axis and the equatorial diameters. The descriptions were according to Moore et. al., (1991), Erdtman (1952) and Ayodele (2000). Most of the pollen grains were sub-prolate but some were prolate and prolate-spheroidal in shape. Polar axis ranges from 20 μ to 50 μ . The pollen sizes ranged from small to large. The taxa showed three types of pores which were aporate, diporate and triporate. On the other hand, four types of furrows were observed namely; tricolpate, dicolpate, monocolpate and acolpate. The surface sculptures were either scabrate or psilate. The shapes of the pollen grains were either elliptic, circular, oblong or oval. The polar views showed spherical shapes except in Cola marsupium where they were either triangular or spherical. The morphological features of the pollen grains are not sufficient enough to classify or distinguish the Cola species as some features are overlapping. The pollen morphologies when combined with some other features can be very useful in the delimitation of the cola species.

Key Words: Cola, Pollen, Morphology, Taxonomy, Nigeria

Introduction

Cola Schott and Endl belongs to the family Sterculiaceae. It is a genus of about 125 species restricted to the forest regions of Africa (Keay et al., 1960; Nielson, 1965; Burkill, 2000). About 50 species are recognized in west Africa, 31 of which are found in Nigeria (Hutchinson and Dalziel, 1954 and Keay et al., 1960). Most of the Nigerian

species are found in the low-land forest and forest outliers.

The are predominantly trees with few shrubs. The genus Cola was separated from Sterculia Linn on the bases of flower and fruit characters (Russel, 1955). Before 1832 when the name Cola was coined by Schott and Endl, a few Cola species were known under the generic name sterculia (Opeke,

1982). In 1805, Palisot de Beauvois collected some specimens of *Cola* during his visit in 1786 to parts of what is now Nigeria (Tachie-Obeng and Brown, 2001). *Cola* is indigenous to Africa (Airy-Shaw, 1985).

Taxonomists have overlooked the importance of pollen morphological characters although they were fully appreciated by others such as Lindly, Vonmohi and Fritsche in 1830-1840.

Rarely as Faegri (1956) noted do taxonomists take the pollen grains into account when describing or revising a group of plant and when descriptions are given of the pollen grains they tend to be superficial (Davis and Heywood, 1963). Pollen sizes vary greatly among different species as well as among different pollens of the same species (Shaheen et al., 2009). According to Teksen et al., (2010), the sculpturing of the exine provided valuable characters for separating the species of the genus *Fritillaria* L. in Turkey. Bibi et al., (2008) claimed that pollen size is a reliable taxonomic tool to separate species. El Nagggar (2003) described pollen size as being useful at tribal level. According to Davis and Heywood (1963), the useful characters of the pollen grains in taxonomy are: number and position of furrows, number, position and complexity of the apertures and the sculpturing of the exine.

The shapes and the sizes of the pollen grains have less diagnostic values. The uniqueness of the pollen grain features rest in the fact that the influence which determine their morphology are both hereditary and environmental.

It is therefore fully accentuated that pollen grains are used for solving taxonomic problems (Arti, 2009).

Materials and Methods

The materials used were fresh specimens from the field which were later shade-dried in the laboratory and dried Herbarium specimens. The method of acetolysis by Erdman (1952) and Ayodele (2000) were employed. The dried flower buds were crushed in plastic test tubes using glass rod.

3ml of freshly prepared acetolysis mixture (nine-part acetic anhydride to one-part concentrated tetraoxosulphate IV acid) was added to the crushed flower buds in a centrifuge tube. This was heated in

water bath at 70°C to boiling point, stirring the content occasionally. The centrifuge tubes were left in the water bath for three minutes.

The supernatant was decanted into a specially labeled bottle called acetolysis waste bottle leaving the sediment in the centrifuge tubes. Water was added to the sediments and shaken vigorously resulting in the formation of foam. Drops of methylated spirit were added to remove the foam and the suspension was centrifuged. The supernatant was decanted.

The washing with water, centrifuging and decanting the supernatant was repeated four times. Water was added and the content sieved using a 120µm sieve. This was also centrifuged and decanted. Fifty percent aqueous glycerin was added to the sediment and left for 2 hours. The tubes were shaken vigorously and centrifuged for 10 minutes at 4000 rpm, decanted and inverted over filter paper to drain for three hours.

100% glycerin was added to the content, shaken vigorously and transferred to some labeled storage vials. Slides were prepared by shaking the content in the vials and put a drop on a clean glass slide, covered with cover slip and seal the edges with nail varnish. Observation and study of the pollen grains were done using light microscope with a Camera attached. Pollen features studied include the surface sculptures, furrows, pores, shapes/classes, polar axis and the equatorial diameters. Descriptive terminologies were based on Erdtman (1952), Moore et al., (1991) and Ayodele (2000).

Results

The result of the features or characters of the pollen grains were recorded as in tables 1 and 2 and plates 1-3. The polar axis of the pollen grains ranges from 20µm in *C. lateritia* and *C. nigerica* to 50µm in *C. lepidota* and *C. marsupium* table 1. Equatorial diameter lies between 15µm in *C. gigantea* and *C. lateritia* to 47.5µm in *C. lepidotatable* 1. Triplicate pollen grains were found in *C. acuminata*, *C. anomala*, *C. hispida*, *C. laurifolia*, *C. millennii*, *C. marsupium* and *C. verticillata*, table 2, plates 1A, 1B, 2J, 2M, 2T, 2V and 3AF while *C. heterophilla* has diporate pore table2, plate 1H and the other taxa are aporate. The types of furrows observed were tricolpate (three furrows) as in *C.*

megalophylla table2, plate 2S, dicolpate (two furrows) in *C. heterophylla*, plate 1G, monocolpate (one furrow) as in *C. anomala*, *C. laurifolia* and *C. millenii*, table2, plates 1C, 2L and 2u. Other species are acolpate (without furrow).

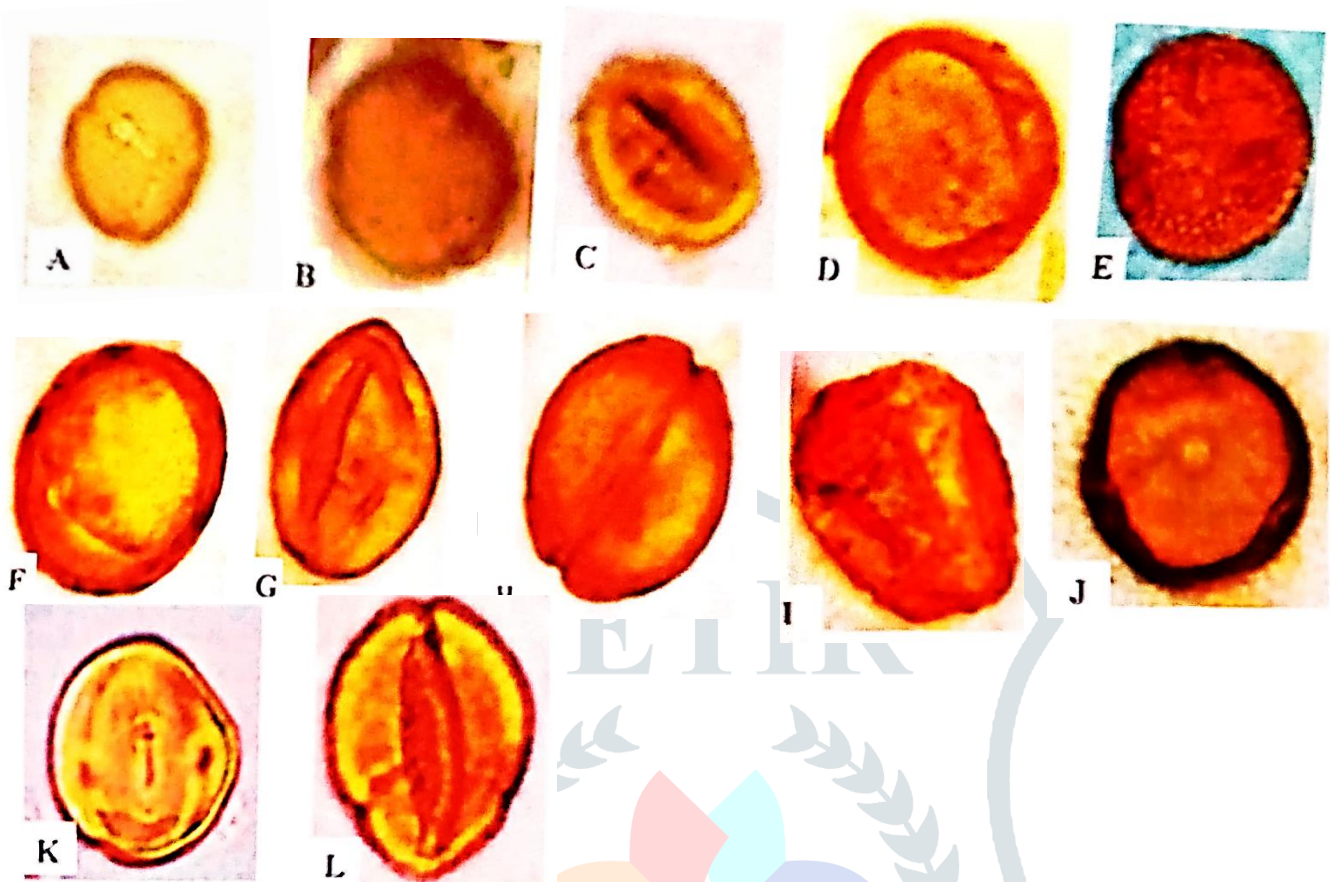
The surface sculptures were scabrate in *C. gigantea*, *C. hispida*, *C. laurifolia*, *C. lepidota*, *C. marsupium*

and *C. pachycarpata*, plates 1E, 1I, 2L, 2O, 2V and 3AB respectively.

In equatorial view, the taxa were elliptic in shape but oblong in *C. hispida*, and *C. megalophylla*, plates 1I and 2Q. The polar view showed spherical shapes except in *C. marsupium* where they were either spherical or triangular table1, plates 2V and 2W.

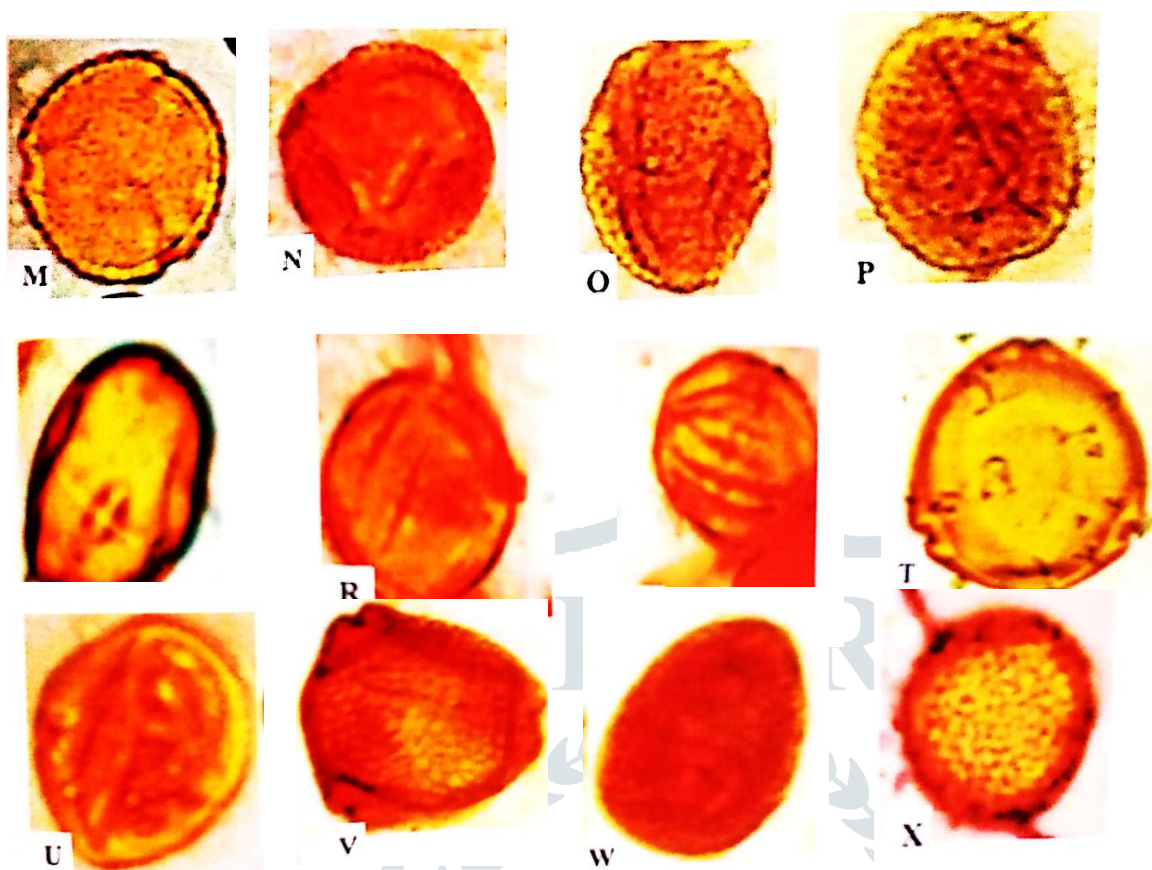


Plate 1. Photomicrographs of the pollen grains of *Cola* species in Nigeria



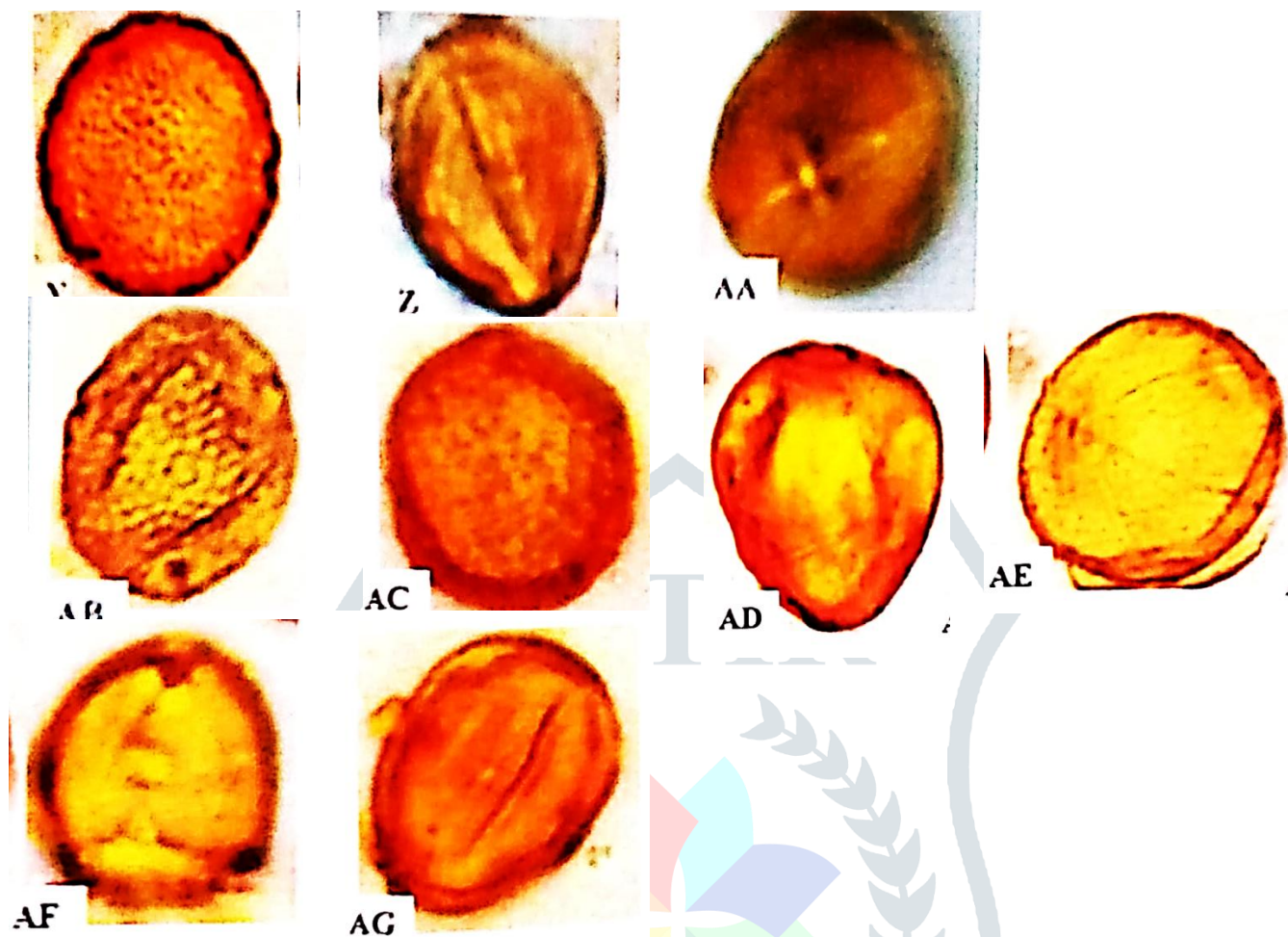
- A. *C. acuminata* showing triporate and psilate pollen grains.
 B. *C. anomala* showing Polar view, triporate and psilate pollen grains.
 C. *C. anomala* showing equatorial view, elliptic, monocolpate and psilate pollen grains.
 D. *C. flaviflora* showing inaperturate, elliptic, psilate pollen grains.
 E. *C. gigantea* showing inaperturate, scabrate and triporate and psilate pollen grains.
 F. *C. heterophylla* showing psilate and elliptic pollen grains.
 G. *C. heterophylla* showing dicolpate and psilate pollen grains.
 H. *C. Heterophylla* showing elliptic, diporate and psilate grains.
 I. *C. hispida* showing oval shape and scabrate pollen grains.
 J. *C. hispida* showing elliptic, psilate and triporate pollen grains.
 K. *C. lateritia* showing inaperturate psilate, elliptic pollen grains.
 L. *C. laurifolia* showing monocolpate, scabrate triporate pollen grains.

Plate 2. Photomicrographs of the pollen grains of *Cola* species in Nigeria



- M. *C. laurifolia* showing scabrate, circular and triporate pollen grains.
 N. *C. lepidota* showing scabrate and elliptic pollen grains.
 O. *C. lepidota* showing scabrate, dicolpate and circular pollen grains.
 P. *C. lepidota* showing scabrate, acolpate pollen grains.
 Q. *C. megalophylla* showing circular and psilate pollen grains.
 R. *C. megalophylla* showing elliptic and inaperturate pollen grains.
 S. *C. megalophylla* showing elliptic, psilate and tricolpate pollen grains.
 T. *C. millenii* showing psilate, triporate and circular pollen grains.
 U. *C. millenii* showing elliptic and monocolpate pollen grains.
 V. *C. marsupium* showing triporate, scabrate and elliptic pollen grains.
 W. *C. marsupium* showing elliptic and scabrate pollen grains.
 X. *C. nigerica* showing scabrate oval and acolpate pollen grains.

Plate 3. Photomicrographs of the pollen grains of *Cola* species in Nigeria



Y. *C. nigerica* showing triporate, scabrate and circular pollen grains.

Z. *C. nitida* showing psilate, and inaperturate pollen grains.

AA. *C. nitida* showing elliptic, acolpate and psilate pollen grains.

AB. *C. pachycarpa* showing dicolpate, scabrate and oval pollen grains.

AC. *C. pachycarpa* showing oval, scabrate and inaperturate pollen grains.

AD. *C. rostrata* showing psilate, inaperturate and heart-shaped pollen grains.

AE. *C. rostrata* showing psilate, elliptic and acolpate pollen grains.

AF. *C. verticillata* showing circular triporate and psilate pollen grains.

AG. *C. verticillata* showing elliptic psilate and acolpate pollen grains.

Pollen Morphological Features of *Cola Species* in Nigeria. T1

Taxa	Type of pore	Type of furrow	Surface sculpture	Equatorial view	Polar view
<i>C. acuminata</i>	Triporate	Dicolpate	Psilate	Elliptic	Spherical
<i>C. anomala</i>	Triporate	Monocolpate	Psilate	Elliptic	Spherical
<i>C. flaviflora</i>	Inaperturate	Acolpate	Psilate	Elliptic	Spherical
<i>C. gigantea</i>	Inaperturate	Acolpate	Scabrate	Circular	Spherical
<i>C. heterophylla</i>	Triporate or Diporate	Dicolpate	Psilate	Elliptic	Spherical/triangular
<i>C. hispida</i>	Triporate	Acolpate	Psilate	Oblong	Spherical
<i>C. lateritia</i>	Inaperturate	Acolpate	Scabrate	Elliptic	Spherical
<i>C. laurifolia</i>	Triporate or inaperturate	Monocolpate	Psilate	Oval	Spherical
<i>C. Lepidota</i>	Inaperturate	Dicolpate	Scabrate	Oval	Spherical
<i>C. megalophylla</i>	Inaperturate	Tricolpate	Psilate	Oblong	Spherical
<i>C. millenii</i>	Triporate	Monocilpate	Psilate	Elliptic	Spherical
<i>C. marsupium</i>	Triporate	Acolpate	Scabrate	Triangular/oval	Oval
<i>C. nigerica</i>	Triporate	Acolpate	Psilate	Elliptic	Spherical
<i>C. nitida</i>	Inaperturate	Acolpate	Psilate	Oval/elliptic	Spherical
<i>C. pachycarpa</i>	Inaperturate	Dicolpate	Scabrate	Elliptic	Spherical
<i>C. rostrata</i>	Inaperturate	Acolpate	Psilate	Cone-shaped	Spherical
<i>C. verticillata</i>	Inaperturate	Acolpate	Psilate	Elliptic	Spherical

Pollen Morphological Features of *Cola Species* in Nigeria.T2

Taxa	P/E (%)	Shape/Class	Polar Axis (P)	Equatorial diameter (E)	Polen size
<i>C. acuminata</i>	119	Sub – prolate	22.5 (25.3±1.4) 27.5	17.5(21.3±1.8)22.5	Small/rather small
<i>C. anomala</i>	123	Sub – prolate	25(25.4±1.4)27.5	17.5(20.6±1.8)22.5	Small
<i>C. flaviflora</i>	117	Sub – prolate	30(32.3±1.1)36	30(32±1.2)35	Small
<i>C. gigantea</i>	129	Sub – prolate	20(21.3±1.2)22.5	15(16.0±0.5)17.5	Small
<i>C. heterophylla</i>	153	Prolate	22.5(36±2.9)32.5	20(23.5±1.2)27.5	Rather small/medium
<i>C. hispida</i>	108	Spheroidal	32.5(35.8±2.6)37.5	27.5(33.3±5.3)35	Medium
<i>C. lateritia</i>	119	Sub – prolate	20(23.3±2.6)27.5	15(19.6±3.0)25.0	Small
<i>C. laurifolia</i>	118	Sub – prolate	22.5(23.3±1.2)25	17.5(19.8±0.8)20	Small
<i>C. lepidota</i>	120	Sub – prolate	37.5(45±3.8)50	30(37.5±6.9)47.5	Medium/rather large
<i>C. megalophylla</i>	124	Sub – prolate	25(29.2±3.0)35	20(23.7±2.0)27.5	Small/rather small
<i>C. milleni</i>	116	Sub – prolate	30(35.8±4.7)40	22.5(30.8±5.4)37.5	Medium
<i>C. marsupium</i>	124	Sub – prolate	25(34.2±9.0)50.0	20(27.5±7.4)37.5	Rather small/medium
<i>C. nigerica</i>	117	Sub – prolate	20(22.8±3.2)30	17.5(19.5±2.6)25	Small
<i>C. nitida</i>	125	Sub – prolate	27.5(31±3.5)37.5	20(24.7±4.3)32.5	Medium/rather small
<i>C. pachycarpa</i>	112	Spheroidal	37.5(41.7±2.8)45	32.5(37.2±4.1)40	Medium/rather large
<i>C. rostrata</i>	127	Sub – prolate	35(43±3.7)47.5	30(33.9±2.8)40	Medium/rather large
<i>C. verticillate</i>	126	Sub – prolate	25(28.3±2.5)32.5	17.5(22.5±2.2)25	Small/rather small

Discussion

The study analyzed critically the morphological features of pollen grains of seventeen *Cola* species. The features that were carefully observed include the types of pores, furrows, surface sculptures, sizes, equatorial and polar views. The *Cola* species can be classified into three based on the types of pores. These are those without pores (aporate), those with two pores (diporate) and those that have three pores (triporate). Infact, some species even exhibit two types of pores. In *Cola heterophylla*, both diporate and triporate grains were present while *Cola laurifolia* on the other hand showed both inaperturate and triporate grains. This makes it difficult to separate the species based on the pores present.

The sizes of the pollen grains are other features which are overlapping and makes it also difficult to distinguish the *Cola* species from one another. This is inline with the statement made by Davis

and Heywood (1963) that the pollen size is not a good taxonomic feature.

On the contrary, Bibi et al., (2008) were of the opinion that pollen size is a reliable taxonomic tool to separate species.

The number and position of the furrows, number, position and complexity of the pores and sculptures of the exine are the features of the pollen grains which are of taxonomic importance (Davis and Heywood, 1963; Guinet, 1986; Trytone, 1986; and Teksen et al., 2010).

The presence of both diporate and triporate grains in *Cola heterophylla* distinguishes it from other taxa. Most of the pollen grains of the *Cola* species are acolpate (without furrow). *Cola pachycarpa* is the only species that has two furrows (dicolpate). The study of the pollen grains in this taxa revealed that pollen alone cannot be used as a criteria for the identification of the taxa or species.

Conclusion

This study which involved pollen grains from seventeen Cola species revealed that the morphological features of the Cola species cannot be used to identify each taxon or even to group them in the same genus. Although some of the Cola species have the same or similar morphological features but they have some blending character more especially in terms of size. Most of the pollen grains have features which are common among some of the taxa. This work has therefore provided some features or characters which are very useful in the identification of the Cola species if combined with other characters like chemical and anatomical.

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