Journal of Biological Studies
ISSN 2209-2560

PKP PUBLIC KNOWLEDGE PROJECT

JBS An Open Access Peer Reviewed Journal

Research Full-Text Paper
DOI: 10.22034/jbs.2022.153279

Morphology and phytochemical diversity among some species in the family cyperaceae

Sekinat Okikiola Azeez[1]*, Stella Abimbola Ojo[1], Oluwatumininu Grace Dunmade[1], Titilope Olamide Ogundele[1], Bolajoko A. Akinpelu[2]

[1] *Department of Botany, Obafemi Awolowo University, Ile – Ife, Nigeria.*
[2] *Department of Biochemistry and Molecular Biology, Obafemi Awolowo University, Ile – Ife, Nigeria.*

Received November 25, 2021; Accepted May 20, 2022; Published online June 30, 2022

Abstract

Classification of members of Cyperaceae is complicated basically due to the size and complexity of their inflorescences. This study employed morphological attributes to delimit some species belonging to the *Cyperus*, *Pycreus*, *Mariscus*, *Kyllinga*, *Fimbristylis*, and *Rhynchospora*; and further explored and analyzed the secondary metabolites from these species if they could be helpful in their characterization. Eighteen morphological characters were considered and analyzed through cluster analysis. Additionally, thin layer chromatography (TLC) was employed to screen phytochemicals such as flavonoid, steroid, tannin, saponin, triterpene, alkaloid, and cardiac glycosides in some of the sedges. The morphological study showed that the cluster analysis grouped all the species studied into two main clusters. The first main cluster contained all the *Cyperus* species studied except *C. nipponus* and *C. rotundus*. However, a few members of other genera studied were embedded within the *Cyperus* cluster. On the other hand, the second main cluster was further divided into two sub-clusters; the first sub-cluster had only *Fimbristylis* species while the second sub-cluster comprised *Kyllinga*, *Pycreus*, *Mariscus* species and two *Cyperus* species. In this present study, the inflorescence type and achene morphology played a significant role in delimiting the accessions studied. The TLC profiling of the species of Cyperaceae studied revealed their phytochemical diversity which can be harnessed in pharmaceutical industries. Moreover, the phytochemicals detected (flavonoid, steroid, tannin, saponin, triterpene, alkaloid, and cardiac glycosides) have been demonstrated to be useful in the delimitation of the species within each genus studied and elucidated the relationship that exists among them. However, they have little or no taxonomic value in delimiting the genera studied within the family Cyperaceae. Further research is required with respect to characterization of the diverse secondary metabolites detected in some members of Cyperaceae studied

Keywords: Chemosystematics, Genetic relationship, Secondary metabolites, Sedges

*Corresponding author email: azeezs@oauife.edu.ng / sekinatokiki@gmail.com

1 Introduction

The sedge family (Cyperaceae) is the third-largest monocotyledon family, with approximately 5500 species distributed among 109 genera (Pal and Choudhury, 2017; Xu and Zhou, 2017). Twenty-four of these genera were recorded in West Africa (Hutchinson and Dalziel, 1972). In Nigeria, 230 species belonging to 23 genera have been identified (Lowe and Stanfield, 1974). The members of the family are distributed throughout the world excluding Antarctica. They occur in diverse habitats with over 80% growing in damp or wet habitats (Lowe and Stanfield, 1974; Goetghebeur, 1998; Pal and Choudhury, 2017). The genus *Carex* L. (with 2000 species) is the largest genus in the family followed by *Cyperus* L. (Bruhl, 1995; Goetghebeur, 1998; Muasya et al., 2002; Tantwai, 2017). *Bulbostylis* Kunth., *Cyperus* and *Pycreus* P. Beauv. are common in tropical Africa (Goetghebeur, 1998). Sedges may be annual or perennial and they are morphologically diverse (Goetghebeur, 1998; Carter, 2005). The family is characterized by a large amount of variation in chromosome numbers and was opined to have a basic chromosome number of X=5. Aneuploids, as well as polyploids, have been documented in Cyperaceae (Goetghebeur, 1998; Roalson, 2008). The majority of the family members are adapted to wind pollination. The genus *Rhynchospora* Vahl. has been identified as one of a few genera in which pollination strategy transits from wind to insect pollination (Goetghebeur, 1998; Lucero et al., 2014).

The members of Cyperaceae are important in ecosystem management; some members of the family (such as *Cyperus difformis* L., *C. iria* L., *C. rotundus* L. *C. esculentus* L. and *Fimbristylis* Vahl. species) invade the agricultural fields, lawns and gardens and they are difficult to control or eradicate (Carter, 2005; Barrett, 2013). *Cyperus papyrus* L. has been used in paper production and it also has horticultural value. Moreover, it has been identified as a possible source of bioenergy (Goetghebeur, 1998; Carter, 2005). A few members of the family are edible e.g. *C. esculentus var. sativus* Boeck. (Tigernut) while many have been identified to have medicinal value such as *C. rotundus* which were used in the treatment of various ailments (Cheema et al., 2017). Fibres from some sedges including *Scirpus* L. species have served as weaving materials. In addition, *Schoenoplectus lacustris* L. was documented to have played a good role in water purification and certain *Fimbristylis* species were reported as indicators of copper deposit (Carter, 2005).

Cyperaceae was reported to be more closely related to Juncaceae and Thurniaceae than Poaceae considering the cladistics analysis based on molecular and morphological studies (Carter, 2005). This assertion is further supported by the report on the presence of diffused centromeres and meiosis of post-reductional type in these families (Goetghebeur, 1998). The classification of members of Cyperaceae is challenging mainly due to complexity and the small inflorescence size (Carter, 2005). The family had been previously divided into two or four subfamilies (Bruhl, 1995; Goetghebeur, 1998; Muasya et al., 2009). Delimitation of genera in Cyperaceae using morphological data alone is still a difficult task to achieve (Muasya et al., 2002; Meshack, 2007). Furthermore, circumscription in the genus *Cyperus* has been a matter of considerable disagreement (Lye and Masterhazy, 2012) and Reid et al., (2017) pointed out that generic delimitation in *Cyperus* and allied genera have not been completely resolved by

molecular evidence. More so, Desai and Raole (2014) stated that morphological study alone cannot resolve the taxonomic issues in the *Cyperus* clade. Ankanna et al., (2012) opined that a combination of botanical and phytochemical studies would provide a better understanding of a plant group. According to Goetghebeur (1998), phenolic and quinonoid constituents in the members of the family may likely play a good role in its classification (Goetghebeur, 1998). Additionally, secondary constituents in this family may be new sources of new pharmaceutical products (Babu and Savithramma, 2014). Moreover, phytochemicals have been reported to be of pharmaceutical importance (Oladipo et al., 2017). In recent years, attention has been given to secondary metabolites in plants and their importance in taxonomy cannot be overemphasized (Misra and Srivastava, 2016). Chemical variation in plants is of considerable taxonomic value in confirmation or support of putative classifications derived from other sources of taxonomic characters such as morphology (Mannheimer, 1999). Phytochemical constituents in plants have been employed to solve taxonomic problems and have been used to reveal the pattern of chemical diversity, as well as the phylogenetic relationships previously (Hegnauer, 1986; Ankanna et al., 2012; Oladipo et al., 2017; Olatunji and Afolayan, 2019). In addition, the classification of plants based on chemical constituents has an advantage over traditional methods considering the ease of working methodology (Singh, 2016), coupled with the fact that the traditional classification of plants is not enough to arrive at a satisfactory classification. Other methods are needed and chemical characters are helpful (Hegnauer, 1986). However, it was argued that chemosystematics is better in establishing consanguinity rather than elucidating phylogenetic relationships which involve tracing plant origin (Olatunji and Afolayan, 2019). However, it is noteworthy that the diversity of secondary metabolites in Ranunculaceae has been used to study the evolutionary relationships among the members of the family in addition to other taxonomic tools. Moreover, flavonoids have been used in the delimitation of plant species up to the family level (Hegnauer, 1986).

This study, therefore, employed morphological attributes to delimit the species studied in the genera *Cyperus*, *Pycreus*, *Mariscus*, *Kyllinga*, *Fimbristylis*, and *Rhynchospora* and thin layer chromatography (TLC) was explored to analyze the secondary metabolites from these species if they could be helpful in their characterization, elucidate their chemical diversity and the relationship that exists among them, in addition to their likelihood of being the new sources of new pharmaceutical products.

Materials and Methods

2. 1. Sample collections

Thirty-four (34) accessions belonging to six genera were investigated in this study (Table 1). The genera that are represented include *Cyperus* (fifteen accessions), *Fimbristylis* (four accessions), *Kyllinga* (six accessions), *Mariscus* (four accessions), *Pycreus* (four accessions) and *Rhynchospora* (one accession). The majority of the species were collected from various locations within Obafemi Awolowo University, Ile-Ife, Nigeria and Ile-Ife town while others were collected along Ogbomoso-Igbeti road and Old Oyo National Park, Sepeteri, Oyo State, Nigeria while on a field trip (Table 1). The plants were identified and authenticated at IFE and FHI herbaria. In addition, Flora of Nigerian Sedges (Lowe and Stanfield, 1974), Flora of West Tropical Africa (Hutchinson and Dalziel, 1972), a handbook of West African Weeds (Akobundu

and Agyakwa, 1998) and other literatures were consulted.

2. 2. Morphological study

The species studied (Figure1) were characterized based on 18 morphological characters (Table 2). These characters were coded numerically and subjected to Cluster Analysis using Ward's methods. The achenes' length and breadth were measured and mean and standard deviation were calculated respectively. The achenes were photographed at x4 under a dissecting microscope equipped with a camera.

2. 3. Thin layer chromatographic analysis

Leaf samples were air-dried for three to four weeks and milled into powder. The powdered substances were separately extracted with absolute methanol for 48 hours. The methanol extracts of the plant species studied were manually spotted by using capillary tubes on precoated TLC Silica gel plates 60 F254 (MERCK, Germany) (20 x 20 cm with 0.2 mm thickness (Wagner et al., 1996). The plates were activated at 120 ^{o}C for 3 min. and then transferred into a chromatography tank already saturated with appropriate solvent systems. After the separation of phytochemical constituents, specific spraying reagents (Table 3) were used to identify alkaloids, flavonoids, tannins, saponins, cardiac glycosides, steroids, and triterpenes. The retardation factor (Rf) was calculated from the following:

$$\text{Retardation factor (Rf)} = \frac{\text{Distance moved by solute}}{\text{Distance moved by solvent}}$$

Table 1. The collection of species used in the study

S/N	Species	Code	Locations
1	*Cyperus compressus* L.	CC	N7^{0}29′54.003848 E4^{0}34′10.38144
2	*C. cuspidatus* Kunth	CCU	N8^{0}24.720 E4^{0}15.483
3	*C. difformis* L.	CD	N7^{0}30′57.83868 E4^{0}31′34.18104
4	*C. dilatatus* Schum &Thonn.	CDI	N7^{0}31′19.42212 E4^{0}31′45.97212
5	*C. distans* L.	CDS	N7^{0}29′54.00348 E4^{0}34′31.3814
6	*C. esculentus var. esculentus* L.	CEE	N8^{0}24.720 E4^{0}15.483
7	*C. esculentus* L. *var sativus* Boeck	CES	N7^{0}30′2.33856 E4^{0}34′30.01764
8	*C. haspan* L.	CH	N7^{0}30′38.13913 E4^{0}31′24.7209
9	*C. imbricatus* Retz.	CIM	N7^{0}29′38.55588 E4^{0}31′37.85412
10	*C. iria* L.	CI	N7^{0}30′50.22972 E4^{0}31′21.3042
11	*C. nipponicus* Franch. & Sav.	CN	N8^{0}24.720 E4^{0}15.483
12	*C. pseudovegetus* Steud.	CP	N7^{0}30′38.13912 E4^{0}31′24.72096
13	*C. rotundus* L.	CR	N7^{0}27′39.64524 E4^{0}34′12.36324
14	*C. sphacelatus* Rottb.	CS	N7^{0}31′19.42212 E4^{0}31′45.97212
15	*C. strigosus* L.	CST	N7^{0}30′50.94612 E4^{0}34′21.45612
16	*C. tuberosus* Rottb.	CTU	N7^{0} 30′38.13912 E4^{0} 31′24.72096
17	*Fimbristylis dichotoma* (Linn.) Vahl *var. dichotoma*	FDD	N7^{0} 29′38.55588 E4^{0} 34.31′.38144
18	*F. dichotoma var pluristriata* (C.B.Cl.) Napper	FDP	N7^{0} 30′5.94612 E4^{0} 31′21.3024

19	*F. ferruginea* (Linn.) Vahl (Unspotted infl.)	FFa	N8⁰ 24.720′ E4⁰ 15.466′
20	*F. ferruginea* (Spotted infl.)	FFb	N8⁰ 24.720′ E4⁰ 15.466′
21	*F. littoralis* Gaudet	FL	N7⁰ 30′38.13912 E4⁰ 1′24.7209
22	*Kyllinga bulbosa* Beauv.	KB	N7⁰31′18.50664″E4⁰31′37.4628′
23	*K. erecta* Schumach.	KE	N7⁰31′18.50664″E4⁰31′37.4628′
24	*K. nemoralis* (Forst.) Dandy ex Hutch.	KNE	N7⁰ 31′9.95448 E4⁰31′33.83832
25	*K. nigritana* C.B.Cl	KN	N8⁰25′8.41667 E3⁰50′3.83333
26	*K. pumila* Michx.	KP	N7⁰31′7.95396′′ E4⁰ 31′48.774′
27	*K. odorata* Vahl	KO	N8⁰25′8.41667 E3⁰50′3.83333
28	*Mariscus alternifolius* (small inflorescence (infl.)* Vahl	MA	N7⁰31′7.95396 E4⁰31′48.774
29	*M. alternifolius* (big infl.)	MA	N7⁰29′47.17896 E4⁰31′15.852
30	*M. flabelliformis* Kunth	MF	N7⁰29′38.35588 E4⁰31′37.85412
31	*M. longibracteatus* Cherm.	ML	N7029′19.34412 E4032′18.43188
32	*Pycreus acuticarinatus* (Kuek.) Cherm.	PA	N8⁰ 24.720′ E4⁰15.466′
33	*P. flavescens* (Linn.) P. Beauv. ex Rchb.	PF	N7⁰30′38.13912E4⁰31′24.72096
34	*P. polystachyos* (Rottb.) Beauv.	PP	N7⁰30′38.13912E4⁰31′24.72096
35	*P. pustulatus* Vahl	PPU	N7⁰31′24.67532 E4⁰ 28.7441
36	*Rhynchospora corymbosa* (Linn.) Britton	RC	N7⁰29′38.55588 E4⁰31′37.85412

Table 2. Morphological characters and character states studied

S/N	Characters	Character states
1	Habit	Herbs
2	Longevity	Annual/ perennial
3	Plant Colour	Green/ light green/ lemon green
4	Root type	Fibrous
5	Culm	Tufted triquetrous glabrous/tufted trigonous/tufted tall hairy/tufted stout/stout tall triquetrous/triquetrous glabrous/tufted tall triquetrous glabrous/tall stout triquetrous glabrous/tufted tall stout triquetrous glabrous/tufted trigonous smooth/tall stout coarse/tufted triquetrous/tufted 4-angled/slender triquetrous glabrous
6	Leaf	Basal/ basal and hairy/ Shorter than the culm/ equal or shorter/ longer than the culm
7	Bracts	2-5/ 3-7/ 5-8

8	Inflorescence type	Simple umbel/ simple to compound umbel/compound umbel/ capitate/ 1 head/ 1 head with one or more buds/ 3 heads or more heads/simple anthela/simple anthela with spo
9	Inflorescence colour	Greenish/ yellowish green/ brownish green/ golden yellow/ white/ whitish green/ reddish green/reddish brown, cream or golden/creamy green
10	Rhizome	Present/ absent
11	Tuber/nut	Present/ absent
12	Achene shape	Triquetrous/trigonous/digonous/obovate/ovate/oval/oblong/oblong to ovate/oblong to fusiform/ellipsoid/oblong to oval/concave/convex/biconvex/ellipsoid to obovate/lineotate/tuberculate/stipitate/Flattened
13	Achene colour	Glossy/black/ dark brown/ light brown/ reddish brown/ cream to golden/ yellowish golden/dark brown to black/ golden to reddish/ brown
14	Achene pigmentation	Reddish brown stipe, not pigmented stipe achene/stipe heavily pigmented and mostly part of the achene/light brown stipe/dark brown apex/reddish achene/stipe and peripheral of achene
15	Achene ornamentation	Rugulose/reticulate/granulate/cracked/smooth
16	Achene Apex	Apiculate/blunt acute/subacute to blunt obtuse/cuspidate/subacute to cuspidate/blunt subacute/blunt obtuse/attenuate/mucronate/subacute/obtuse/acuminate/acuminate to attenuate
17	Achene length (mm)	0.5-1.5/1.6-2.5/>2.5
18	Achene breadth (mm)	0.25-0.75/ 0.76-1.25/ >1.25

Table 3**.** Solvent system/spraying reagent used in chromatographic procedure of the species studied

S/N	Secondary metabolites	Solvent system	Solvent ratio	Spraying reagent
1	Alkaloids	EtOAc:MeOH:H_2O	10:1.4:1.0	Dragendroff
2	Flavonoids	Toluene:Acetone:Formic acid	4.5:4.5:1.0	1% ethanolic $AlCl_3$
3	Tannins	EtOAc: Formic acid:MeOH	3.3:0.8:0.2	5% $FeCl_3$
4	Saponins	Chloroform:MeOH	1.2:0.2	50% Vanillin H_2SO_4
5	Cardiac glycosides	Chloroform:Acetone:Acetic acid	8.5:10:0.5	Antimony in chloroform
6	Steriods	Hexane: EtOAc	7.2:2.9	70% H_2SO_4 Acetic acid in ethanol
7	Triterpenes	100% DCM (Dichloromethane)	100%	Pentachloride

All Rf values obtained were subjected to single linkage cluster analysis (SLCA).

2 Results and Discussions

3. 1. Morphological characterization

The dendrogram obtained from cluster analysis of the 34 accessions (30 species) showed two main clusters (Figure 2). The first main cluster grouped all the *Cyperus* species studied except *C. nipponicus* and *C. rotundus* with *F. littoralis*, *R. corymbosa* and *M. longbracteatus* being embedded within this cluster. Two groups were observed within the second main cluster. The first group comprises all other *Fimbristylis* accessions studied while the second group includes *Kyllinga*, *Mariscus*, *Pycreus* and two *Cyperus* species. Furthermore, *Kyllinga* and *Pycreus* species studied were grouped together with the exception of *K. odorata* which was clustered along *M. flabelliformis, M. alternifolius*, *C. nipponicus* and *C. rotundus.* It should be noted that no seeds were recovered from *C. rotundus, M. flabelliformis, K. bulbosa* and *K. odorata* in this study. The FFa and FFb accessions are the same species (*F. ferruginea*). The only character that separated them was the presence or absence of reddish spots on their inflorescence. In addition, FFD and FDP are varieties of *F. dichotoma*, it was not suprising that they were grouped togrther. The cluster analysis also revealed that *K. nemoralis, K. bulbosa* and *K. nigritana* were more closely related than the other *Kyllinga* studied which is similar for *P. flavescens* and *P. acuticarinatus* (Figure 2). The achene morphology of *Fimbristylis* species studied is unique and may be diagnostic for them. This study showed that inflorescence type and achene morphology especially the shape and ornamentation played a significant role in the delimitation of the species studied in this family (Figure 3; Table 4).

3. 2. Phytochemical screening and analysis

Twenty-nine species from the collection (Table 1) were screened for seven secondary metabolites. These include flavonoids, steroids, tannins, saponins, triterpenes, alkaloids and cardiac glycosides. Different spots at different retardation factor (Rf) values for each phytochemical screened represent different types of the phytochemical of interest (Table 5). The phytochemical profiles showed that saponin was the most abundant with the highest deposit in *Cyperus* followed by cardiac glycoside (Figures 4 and 5).

In the chemical profile for flavonoid, seventeen different spots with different Rf values were detected in all the species of Cyperaceae screened. Flavonoid was absent *in C. distans* and *K. odorata.* The highest number of spots (seven) was observed in *M. flabelliformis* indicating seven different types of flavonoids while the least (one) was observed in *C. iria, C. haspan* and *C. imbricatus.* Flavonoid with Rf value 0.98 (in 24 species) is the commonest followed by 0.58 (in 20 species) While the flavonoid class with Rf 0.12 was found only in *C. haspan*. The percentage positive response for flavonoid is 93.94% (Figures 4 and 5; Table 5). Steroids were present in all the specimens screened. Single spot was detected in all species except in *C. esculentus var. sativus* where two spots were identified and the second spot with Rf 0.26 is peculiar only to this species. Steroid class with 0.98 Rf value was predominant and accounted for approximately 60% of the species studied. (Figure 5, Table 5).

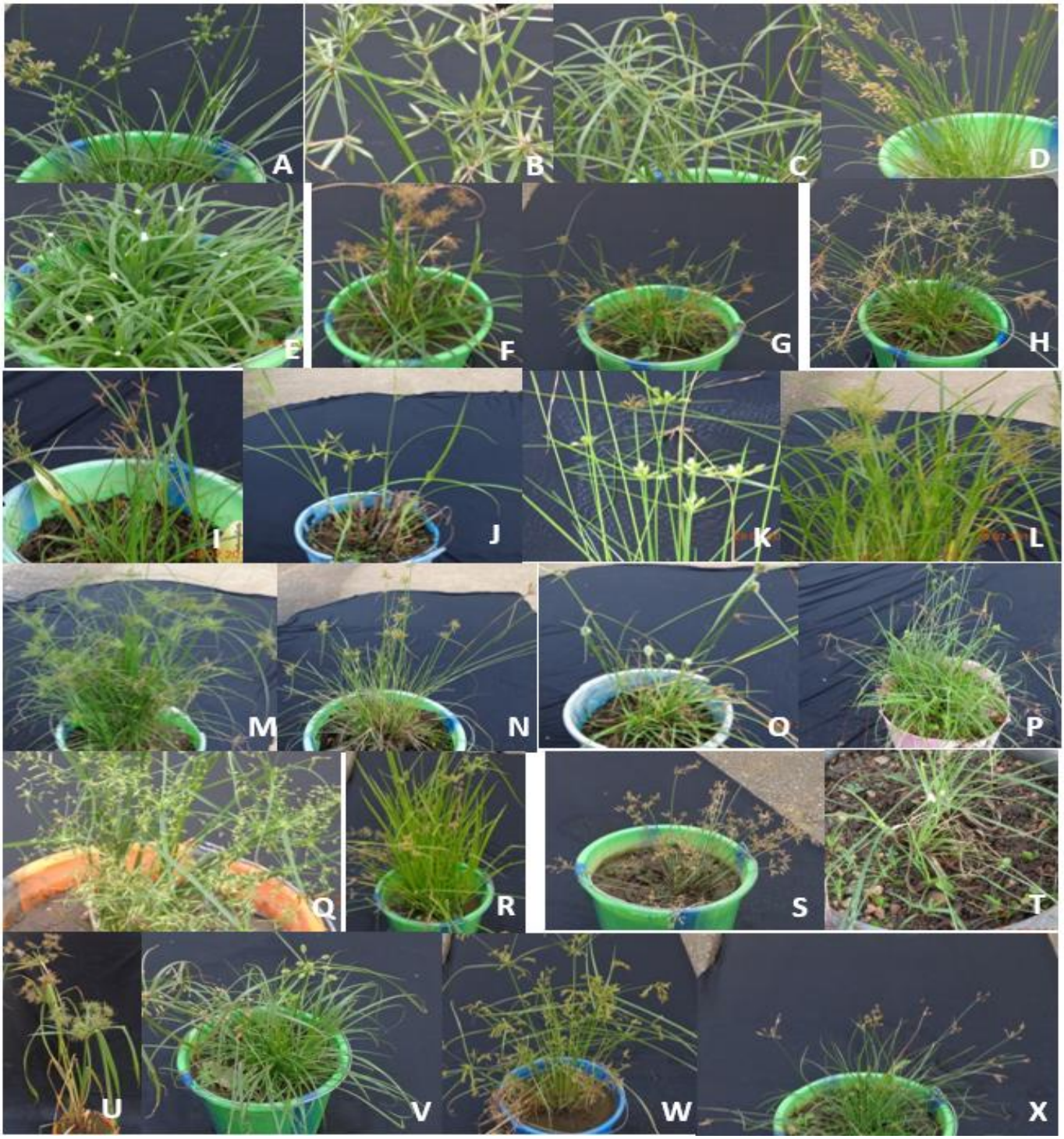

Figure 1. Habit of some of the selected members of Cyperaceae studied.

A. *C. pseudovegetus* B. *C. compressus* C. *K. erecta* D. *F. dichotoma var. dichotoma* E. *K. nemoralis* F. *C. distans* G. *P. flavescens* H. *C. sphacelatus* I. *C. rotundus* J. *C. esculentus var. esculentus* K. *P. polystayous* L. *M. longbracteatus* M. *M. flabelliformis* N. *P. acuticarinatus* O. *K. nigritana* P. *K. pumila* Q. *C. haspan* R. *C. difformis* S. *F. littoralis* T. *K. odorata* U. *C. strigosus* V. *M. alternifolius* W. *C. iria* X. *F. dichotoma var. pluristriata*

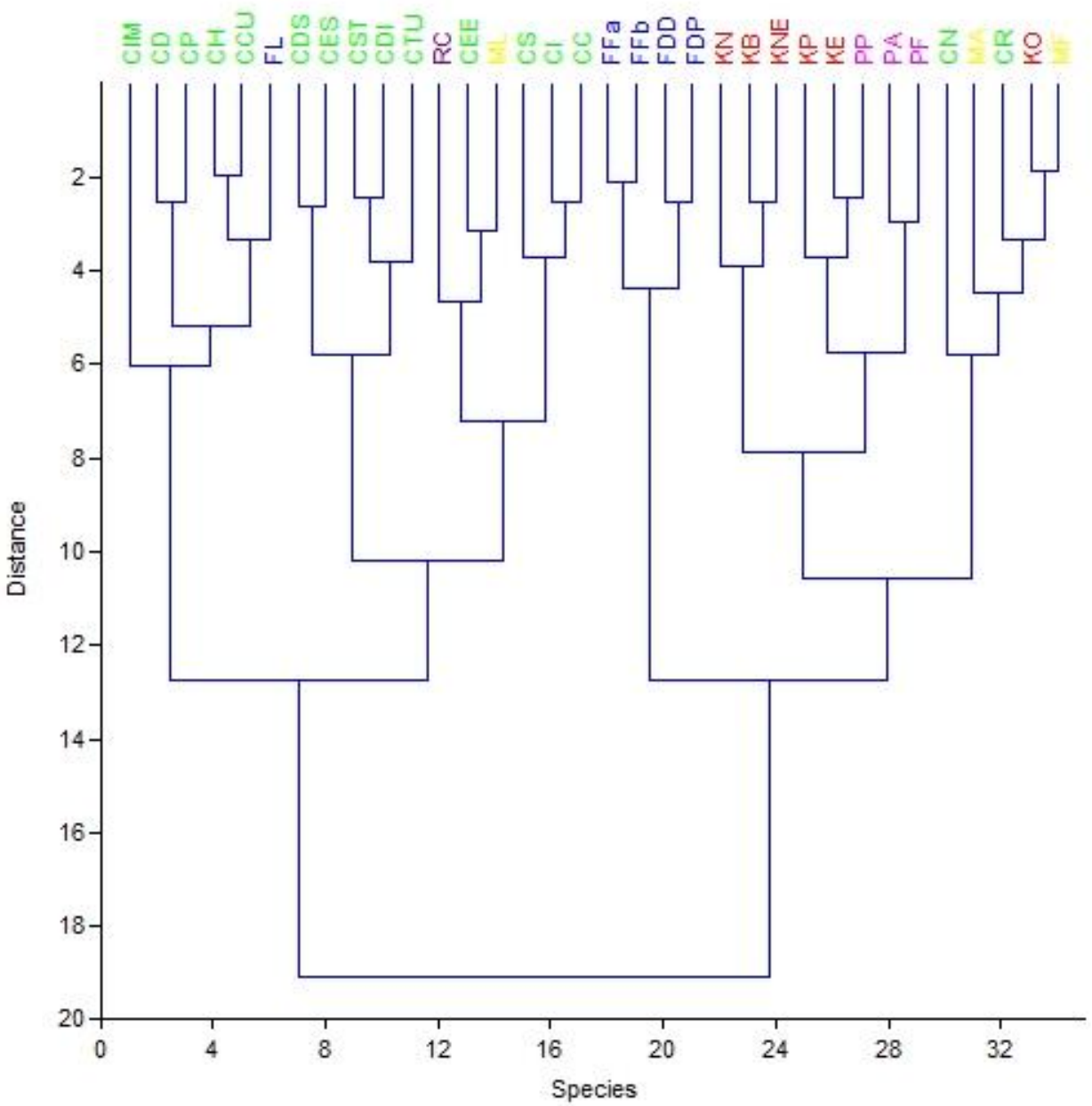

Figure 2. Dendrogram of relationship among species studied in Cypearceae based on morphology

Figure 3. Achene morphology of some selected members of Cyperaceae studied.
A. *C. compressus* B. *C. difformis* C. *C. dilatatus* D. *C. distans* E. *C. esculentus var. esculentus* F. *C. haspan* G. *C. imbricatus* H. *C. nipponicus* I. *C. sphacelatus* J. *C. strigosus* K. *C. tuberosum* L. *C.esculentus var. sativus* M. *C. iria* N. *C. pseudovegetus* O. *F. dichotoma var. dichotoma* P. *F. ferruginea* (unspotted infl.) Q. *F. ferruginea* (spotted infl.) R. *F. littoralis* S. *F. dichotoma var. pluristriata* T. *K. pumila* U. *K. erecta* V. *K. nigritana* W. *P. acuticarinatus* X. *K. nemoralis* Y. *M. alternifolius* Z. *M. longbracteatus* AA. *R. corymbosa* AB. *P. polystachyos* AC. *P. flavescens*

Table 4. Achene attributes of some of the selected members of Cyperaceae studied

S/N	Species	Shape	Colour	Ornamentation	Pigmentation	Apex shape	Length(mm) (mean±SD)	Breadth(mm) (mean±SD)
1	*Cyperus compressus*	Triquetrous, obovate, concave	Dark brown to black	Smooth	absent	mucronate	1.42±2.08	0.99±1.75
2	*C. cuspidatus*	Trigonous, obovate	Glossy, yellowish	Granulate	absent	mucronate	0.55±1.21	0.32±0.95
3	*C. difformis*	Trigonous, convex, ellipsoid	Glossy, yellowish	Granulate	absent	apiculate	0.61±2.25	0.29±1.23
4	*C. dilatatus*	Trigonous, oblong, concave	Dark brown to black	Reticulate	absent	apiculate	1.55±2.25	0.57±1.42
5	*C. distans*	Trigonous, oblong to fusiform	Brown black	Rugulose	absent	apiculate	0.99±7.13	0.42±3.58
6	*C. esculentus var. esculentus*	Triquetrous, obovate, concave	Dark brown to black	Smooth	absent	apiculate	1.77±5.60	1.04±3.38
7	*C. esculentus var. sativus*	Trigonous, concave, ellipsoid	Glossy, dark brown	Rugulose	absent	attenuate	1.43±1.31	0.58±0.93
8	*C. haspan*	Trigonous, concave, oblong to oval, stipitate	Glossy, yellowish	Granulate	absent	mucronate	0.51±2.73	0.31±1.04
9	*C. imbricatus*	Biconvex, oval	Glossy, yellowish	Reticulate	absent	Blunt acute	0.58±2.73	0.35±1.93
10	*C. iria*	Triquetrous, concave, obovate	Dark brown	Reticulate	absent	mucronate	1.17±3.74	0.58±2.73
11	*C. nipponicus*	Trigonous, lineotate	Glossy, cream	Rugulose	reddish	Sub-acute	1.73±2.46	0.35±1.55
12	*C. pseudovegetus*	Trigonous, oblong	Glossy, brown	Granulate	absent	Acuminate to attenuate	0.76±1.95	0.25±0.67
13	*C. sphacelatus*	Triquetrous, concave, ellipsoid to obovate	Glossy, brown	Smooth	absent	mucronate	1.23±3.28	0.64±2.85
14	*C. strigosus*	Trigonous, oblong	brown	Rugulose	absent	acuminate	1.76±4.79	0.48±2.18
15	*C. tuberosus*	Trigonous, oblong, concave	Dark brown	Reticulate	absent	mucronate	1.51±3.19	0.53±1.83
16	*Fimbristylis dichotoma var. dichotoma*	Biconvex, obovate, stipitate	Glossy, cream to golden	Cracked	Pigmented stipe	Sub-acute	0.86±1.58	0.66±4.85

17	*F. dichotoma var pluristriata*	Biconvex, digonous, obovate, stipitate	Glossy, golden to reddish	Cracked	Stipe heavily pigmented and most part of the achene	cuspidate	0.92±3.04	0.56±2.91
18	*F. ferruginea* (Unspotted infl.)	Biconvex, obovate, stipitate	Glossy, cream to golden	Cracked	Stipe and peripheral of the achene	cuspidate	0.94±2.64	0.92±1.94
19	*F. ferruginea* (Spotted infl.)	Biconvex, obovate, stipitate	Glossy, cream to golden	Cracked	Pigmented stipe	Sub-acute to cuspidate	0.92±2.70	0.66±2.83
20	*F. littoralis*	Trigonous, obovate, stipitate	Glossy, cream to golden	Reticulate	absent	obtuse	0.48±1.61	0.28±1.27
21	*Kyllinga erecta*	Ellipsoid, stipitate	black	Rugulose	Dark brown pigmentation at the apex	Blunt obtuse	1.37±4.73	0.57±3.38
22	*K. nemoralis*	Ellipsoid, stipitate	brown	Rugulose	Light brown stipe	Blunt obtuse	1.85±13.490.	0.87±8.34
23	*K. nigritana*	Oblong to ovate, stipitate	Dark brown to black	Rugulose	absent	Blunt obtuse	1.31±3.39	0.45±2.75
24	*K. pumila*	Oblong to ovate	Glossy, brown	Reticulate	Light brown stipe	Blunt obtuse	1.71±8.92	0.53±3.44
25	*Mariscus alternifolius*	Trigonous, ellipsoid	Reddish brown	Rugulose	absent	apiculate	1.49±5.00	0.46±2.07
26	*M. longibracteatus*	Triquetrous, ellipsoid to obovate, stipitate	Dark brown to black	Reticulate	absent	Sub-acute to blunt obtuse	0.91±8.51	0.35±2.61
27	*Pycreus acuticarinatus*	Biconvex, obovate	Light brown to reddish brown to brown	Rugulose	absent	cuspidate	0.77±2.85	0.52±3.55
28	*P. flavescens*	Biconvex, ovate, stipitate	Glossy, black	Reticulate	absent	Blunt sub-acute	0.80±1.59	0.58±2.72
29	*P. polystachyos*	Oblong	Dark brown	Rugulose	absent	Blunt obtuse	0.83±1.47	0.34±3.99
30	*Rhynchospora corymbosa*	Obovate, flattened, long tubercle	Glossy, light	Reticulate	absent	attenuate	6.83±0.51	1.97±0.13

Saponin was detected in all the plants screened except *M. alternifolius* (Big infl.) Thirty-three different spots (Figure 5; Table 5) were identified in the chemical profile of this secondary metabolite with the highest spots (eight) observed in *M. flabelliformis* while *C. pseudovegetus*, *C. sphacelatus* and *C. imbricatus* showed single spot with the same Rf value (0.98). Tannins were present in all the members of Cyperaceae screened with 9 different spots. (Figure 5; Table 5). The spot at Rf value 0.98 is common to all the species studied except *F. dichotoma var*. *pluristata* (0.88, 0.94) and *R. corymbosa* (0.94). The highest spots for tannins (seven) were recorded in *K. pumila*.

Nineteen different spots were identified in triterpene plate. It was absent in about 21% of the species studied (*C. difformis*, *C. distans*, *C. haspan*, *C. iria*, *C. imbricatus*, *C. sphacelatus*, and *M. longibracteatus*). The spot with Rf value 0.52 is unique to *P. polystachyos* which may be diagnostic for this species. The maximum number of spots (six) was recorded in *C. pseudovegetus* (Figure 5; Table 5). All the species screened possess cardiac glycosides. A total of 18 different spots were identified in all the species studied (Figure 5; Table 5). The chemical profile revealed seven different spots in *K. nemoralis*, *P. polystachyos* and *M. flabelliformis* while the least recorded in all the other species studied was three. A total of fifteen different alkaloids was detected in all the species screened with a minimum number of single spot and maximum of four spots. Four different spots were visible in *C. esculentus var*. *sativus* and *C. rotundus*. The alkaloid at Rf value of 0.98 was common to all the species studied except *M. alternifolius* (Small infl.) and *K. odorata* (Figure 5; Table 5).

The single linkage cluster analysis (SLCA) for all the species studied showed that the presence of the screened phytochemicals in some members of Cyperaceae studied has little or no taxonomic value at the family level (Figure 6). However, at the generic level, they seemed to help in the delimitation of the species within each of the genera studied. The SLCA for *Cyperus* species studied showed two main clusters. The first cluster separated *C. cuspidatus* and *C. esculentus var*. *sativus* out. The second main cluster branched into four sub-clusters in which one of them contained only one species, *C. compressus*. The third sub-cluster included *C. sphacelatus*, *C. dilatatus* and *C. tuberosus*. *Cyperus rotundus* was included in the fourth sub-cluster, though separated from *C. sphacelatus*, *C. dilatatus* and *C. tuberosus* but it was still closer to them than *C. esculentus* varieties (Figure 7). In *Kyllinga* species studied, SLCA showed that *K. pumila* and *K. bulbosa* are more closely related while *K. odorata* was separated, however, it was shown to be closer to *K. nemoralis* (Figure 8). *Fimbristylis littoralis* was showed to be distantly related to the other two species studied. The two accessions of *F. ferruginea* studied were grouped together (Figure 9). Similarly, *M. flabelliformis* was separated from the other two species studied while *P. pustulatus* was shown to be distantly related to *P. acuticarinatus* and *P. polystachyos* (Figures 10, 11).

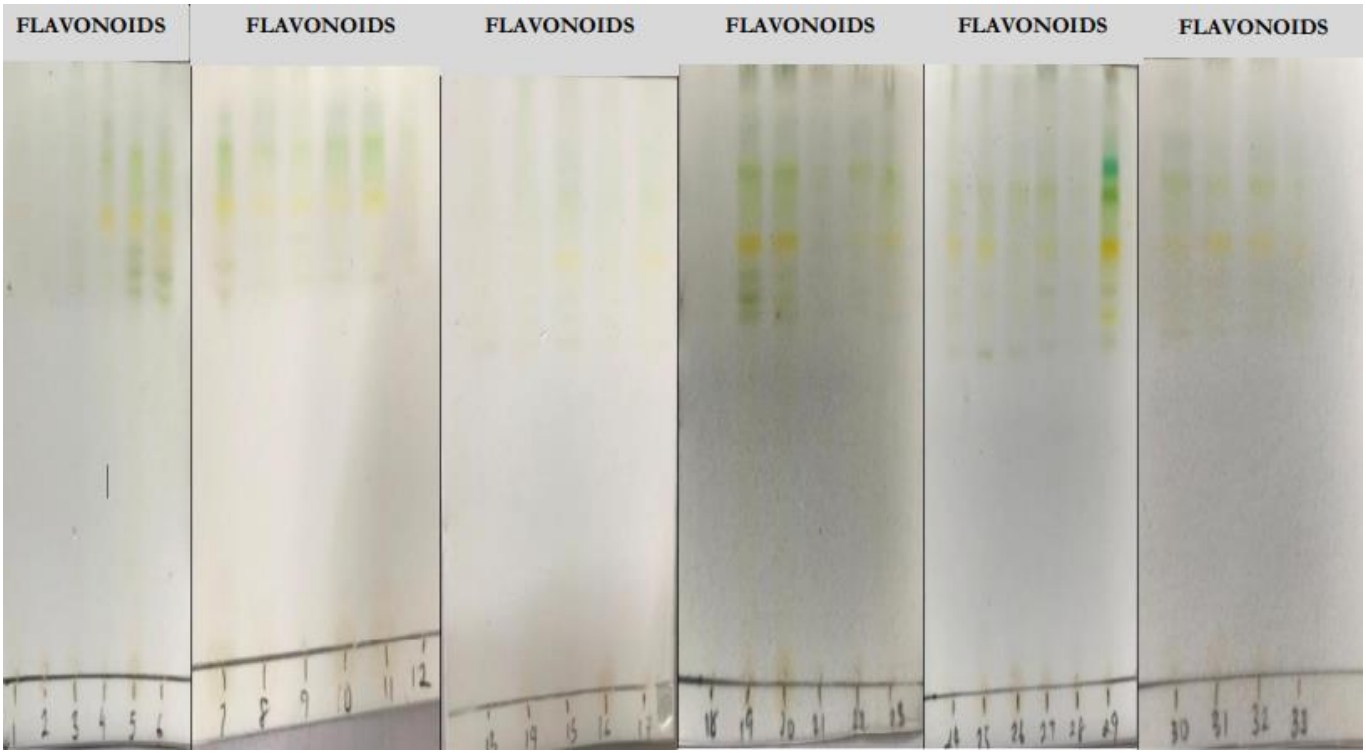

Figure 4. A representative of TLC chromatogram of the methanolic crude leaf extract of the species studied. The chromatogram indicates the presence of flavonoids. Each spot indicates presence of a unique phytoconstituent and the distance travelled was represented by the use of Rf value

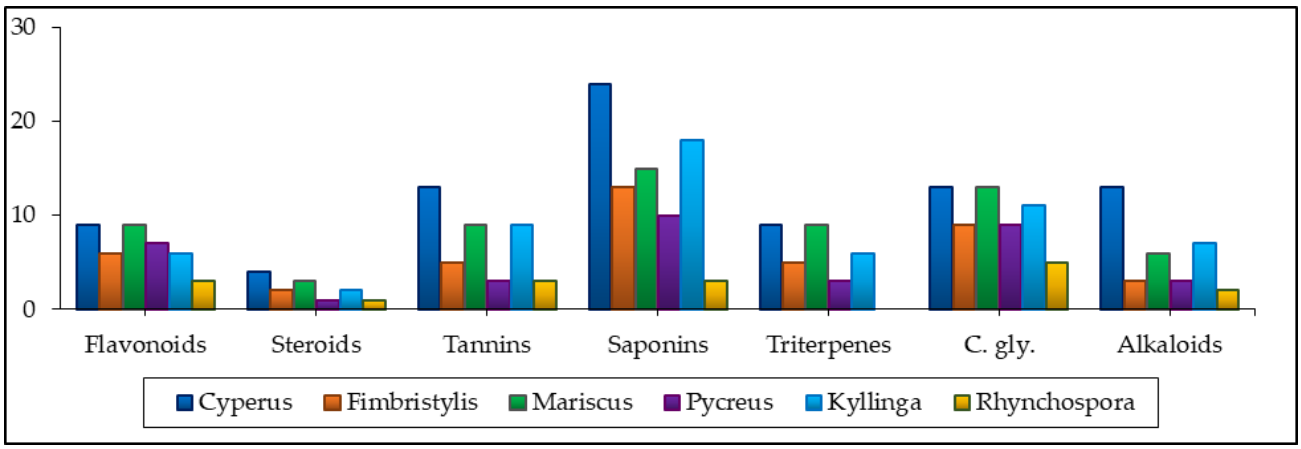

Figure 5. The profiles of all the phytochemicals screened in the members of Cyperaceae studied

Table 5. The Rf values of different phytochemicals in the Cyperaceae species studied

S/N	Plant Species	Flavonoids	Tannins	Saponins	Steriods	Triterpenes	C. glycosides	Alkaloids
1	*Cyperus compressus*	0.58,0.8,0.98	0.56,0.62,0.98	0.88,0.97	0.96	0.50	0.60,0.84,0.88, 0.90,0.94	0.04,0.40,0.98
2	*C. cuspidatus*	0.54,0.58,0.6 0, 0.98	0.50,0.88,0.94,0.98	0.50,0.52,0.86, 0.90, 0.93,0.94	0.98	0.54	0.60,0.82,0.90, 0.96	0.06,0.30,0.98
3	*C. difformis*	0.58,0.8,0.98	0.70,0.90,0.98	0.50,0.51,0.90	0.97	-	0.58,0.84,0.89, 0.92	0.98
4	*C. dilatatus*	0.52,0.78,0.9 8	0.98	0.50,0.88,0.92	0.98	0.50	0.62,0.88,0.90, 0.96	0.04,0.98
5	*C. distans*	-	0.98	0.40,0.90,0.97	0.98	-	0.62,0.92.0.96	0.98
6	*C. esculentus var. esculentus*	0.58,0.8,0.98	0.58,0.08,0.80,0.90, 0.94,0.98	0.50,0.51,0.91,0.94	0.98	0.50	0.54,0.84,0.88, 0.92,0.96	0.10,0.98
7	*C. esculentus var. sativus*	0.58,0.6,0.62, 0.78, 0.82, 0.98	0.88,0.92,0.98	0.44,0.46,0.80, 0.86, 0.96	0.26, 0.98	0.20,0.50	0.60,0.80,0.88, 0.92,0.96	0.08,0.60,0.70 , 0.98
8	*C. haspan*	0.12	0.52, 0.98	0.20, 0.96	0.98	-	0.62,0.90,0.98	0.34,0.98
9	*C. imbricatus*	0.98	0.94.0.98	0.98	0.98	-	0.84,0.88,0.92, 0.96	0.04,0.98
10	*C. iria*	0.98	0.98	0.51,0.85,0.93	0.96	-	0.62,0.92,0.96	0.98
11	*C. pseudovegetus*	0.6,0.64,0.80	0.94,0.98	0.98	0.98	0.04,0.20,0.28, 0.30, 0.32, 0.42	0.60,0.66,0.86, 0.94	0.72,0.76,0.98
12	*C. rotundus*	0.62,0.64,0.6 6, 0.80	0.94,0.98	0.42,0.46,0.80 ,0.84, 0.90, 0.98	0.98	0.10	0.62,0.70,0.82, 0.90, 0.94, 0.96	0.08,0.68,0.76 , 0.98
13	*C. sphacelatus*	0.58,0.8,0.98	0.88,0.90,0.98	0.98	0.98	-	0.60,0.66,0.86,	0.08.0.98

							0.94	
14	*C. strignosus*	0.58,0.8,0.98	0.90,0.94,0.98	0.50,0.51,0.91,0.94	0.98	0.5	0.58,0.84,0.88, 0.92, 0.96	0.08,0.98
15	*C. tuberosus*	0.58,0.62,0.8 0	0.50,0.88,0.92,0.98	0.44,0.46,0.78,0.87	0.97	0.20,0.50	0.60,0.84,0.88, 0.90,0.94	0.08,0.98
16	*Fimbristylis dichotoma var. dichotoma*	0.54,0.98	0.98	0.50,0.52,0.89,0.96	0.98	0.60,0.84,0.88, 0.92,0.96	-	0.98
17	*F. dichotoma var.pluristriata*	0.54,0.58,0.8 8, 0.98	0.88,0.94	0.49,0.52,0.53,0.86 ,0.90,0.94	0.97	0.60,0.84,0.88, 0.92,0.96	0.54	0.98
18	*F. ferruginea (spot)*	0.58,0.8,0.98	0.98	0.90,0.96	0.97	0.60,0.84,0.88, 0.90,0.94	0.14,0.20,0.50	0.98
19	*F. ferruginea(un)*	0.58,0.8,0.98	0.98	0.89,0.94	0.98	0.60,0.84,0.88,0.90 ,0.94	0.32	0.98
20	*F. littoralis*	0.58,0.62,0.8	0.52,0.88,0.90,0.94, 0.98	0.50,0.58,0.70, 0.80, 0.89,0.98	0.97	0.62,0.68,0.84,0.88 ,0.92,0.96	-	0.10,0.22,0.98
21	*Kylllinga bulbosa*	0.58,0.8,0.98	0.90,0.94,0.98	0.64,0.70,1.00,0.94	0.98	0.62,0.88,0.92,0.94	-	0.98
22	*K. nemoralis*	0.58,0.8,0.98	0.90,0.94,0.98	0.52,0.84,0.86,0.92 ,0.95,0.98	0.96	0.62,0.70,0.76,0.78 ,0.88,0.92,0.94	0.08,1.00,0.30 ,0.32	0.98
23	*K. nigritana*	0.2,0.52,0.98	0.90,0.98	0.52,0.89,0.94	0.96	0.62,0.88,0.92,0.96	0.5	0.06,0.42,0.98
24	*K. pumila*	0.58,0.8,1.00	0.52,0.62,0.70,0.88, 0.90,0.94,0.98	0.50,0.70,0.80, 0.88,0.92,0.97	0.98	0.62,0.84,0.90,0.94 ,0.96	0.5	0.08,0.40,0.98
25	*K.odorata*	-	0.18,0.52,0.60,0.98	0.10,0.42,0.88,0.94	0.98	0.42	0.60,0.92,0.94, 0.96	0.10,0.32
26	*Mariscus alternifolious* (Big infl.)	0.54,0.58,0.6, 0.8,0.98	0.62,0.94,0.98	-	0.97	0.28,0.04,0.60	0.58,0.80,0.92, 0.96	0.10,0.36,0.98

27	*M. alternifolious* (Small infl.)	0.82,0.88	0.60,0.94,0.98	0.29,0.38,0.9,0.93	0.98	0.10,0.44	0.62,0.82, 0.88 0.94,0.96	0.4
28	*M. flabelliformis*	0.54,0.58,0.6, 0.76,0.82,0.8 6,0.98	0.98	0.50,0.52,0.66, 0.80,0.86,0.92,0.94 , 0.98	0.96	0.60,0.68,0.72,0.82 ,0.86,0.94,0.96	0.20,0.24,0.26, 0.40,0.6	0.68,0.74,0.98
29	*M. longibracteatus*	0.58,0.8,0.98	0.98	0.5,0.51,0.7,0.88, 0.9	0.98	-	0.58,0.84,0.88,	0.10,0.98
30	*Pycreus acuticarinatus*	0.54,0.58,0.8, 0.98	0.94,0.98	0.50,0.52,0.86, 0.90,0.93,0.94	0.98	0.60,0.84,0.88,0.92 ,0.96	-	0.06,0.98
31	*P. polystachyous*	0.58,0.8,1.00	0.90,0.94,0.98	0.50,0.52,0.54, 0.86,0.94,0.98	0.98	0.62,0.79,0.76,0.78 ,0.88,0.92,0.94	0.52	0.06,0.98
32	*P. pustulatus*	0.52,0.76,0.9 8	0.94,0.98	0.50,0.88,0.95	0.98	0.62,0.88,0.94	0.28,0.50	0.04,0.98
33	*Rhyconspora corymbosa*	0.58,0.80,0.9 8	0.94	0.50,0.86,0.97	0.98	0.58,0.84,0.88,0.92 ,0.96	-	0.10,0.98
-	Total number of spots	17.00	9.00	33.00	4.00	19.00	18.00	15.00
-	Percentage of the spots (%)	14.78	7.82	28.69	3.47	16.52	15.65	13.04

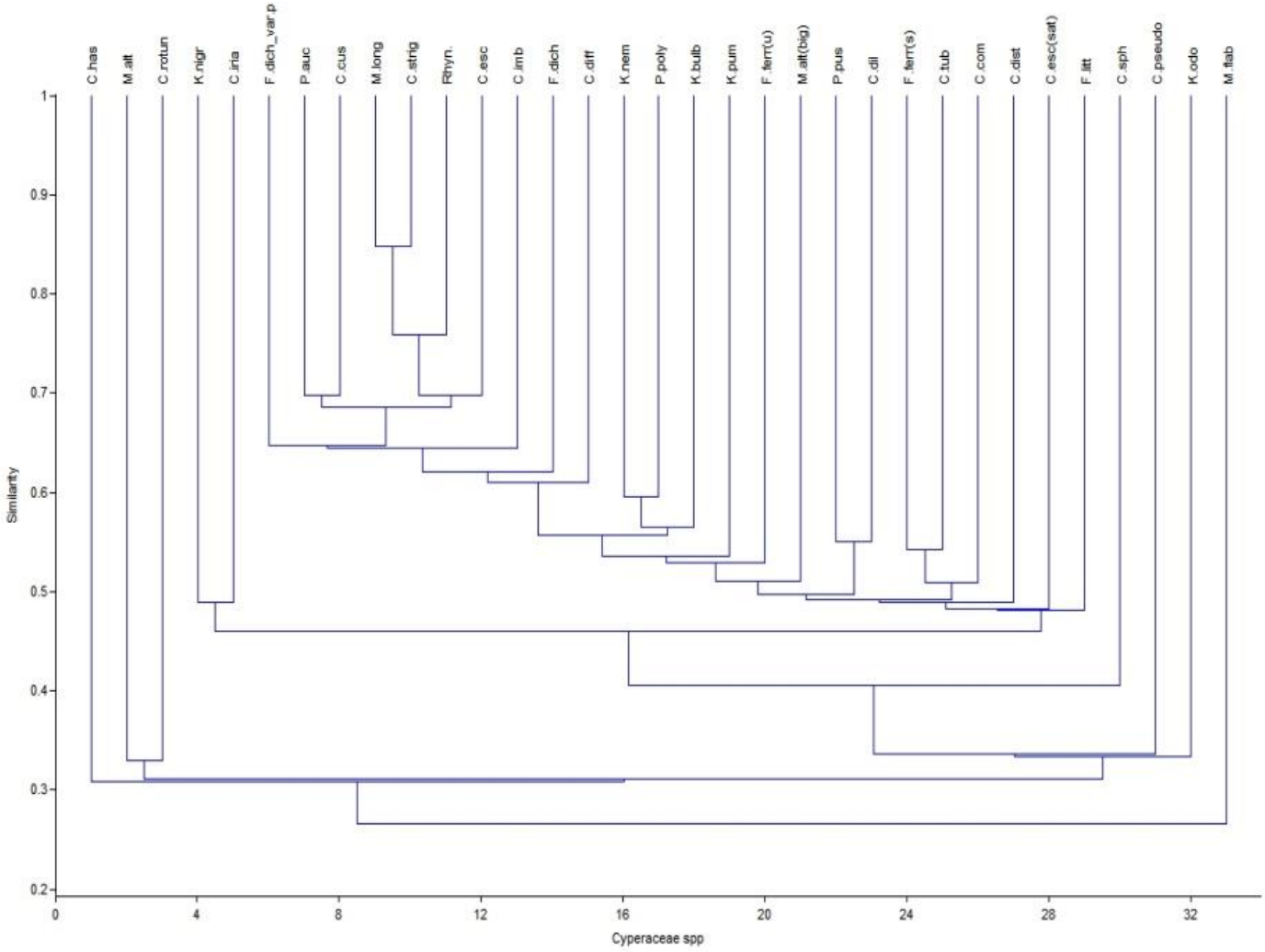

Figure 6. Dendrogram showing relationship among the members of Cyperaceae studied based on phytochemical constituents

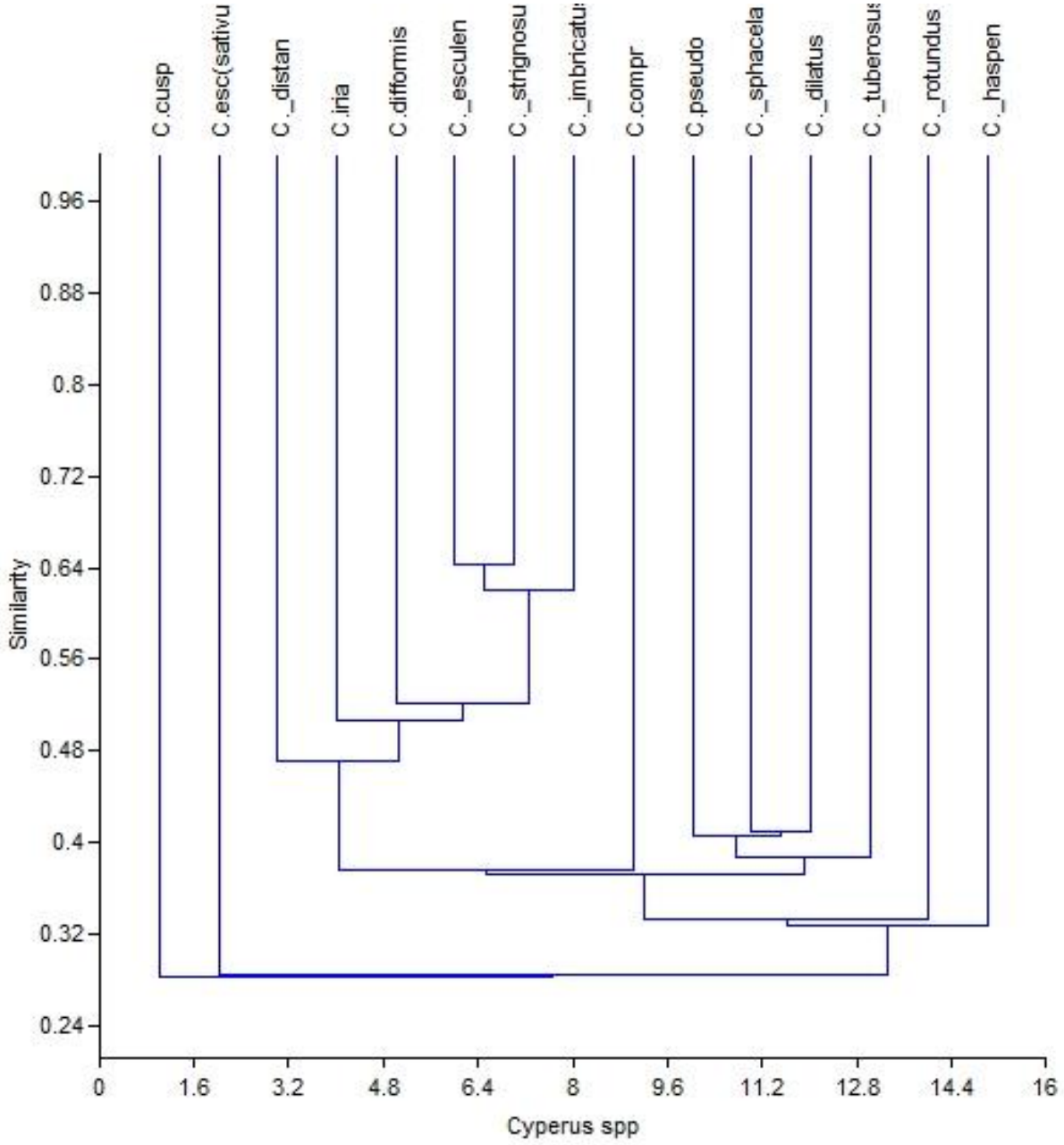

Figure 7. Dendrogram showing relationship among *Cyperus* species studied based on phytochemical constituents

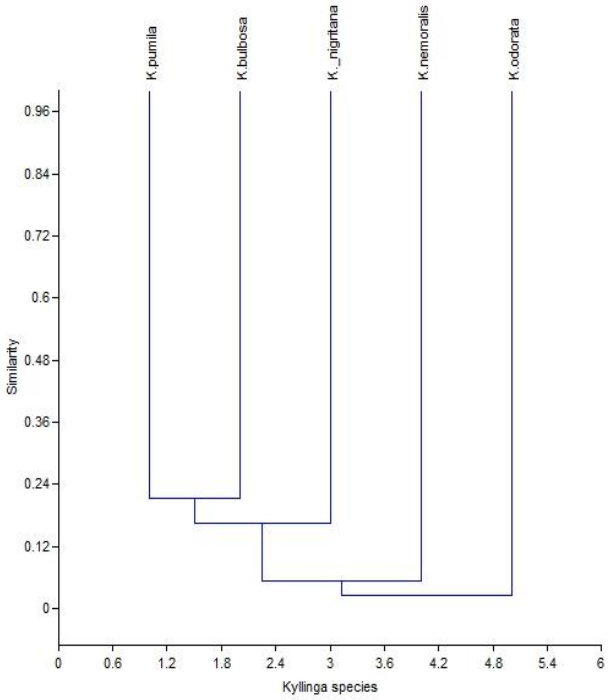

Figure 8. Dendrogram showing relationship among *Kyllinga* species studied based on phytochemical constituents

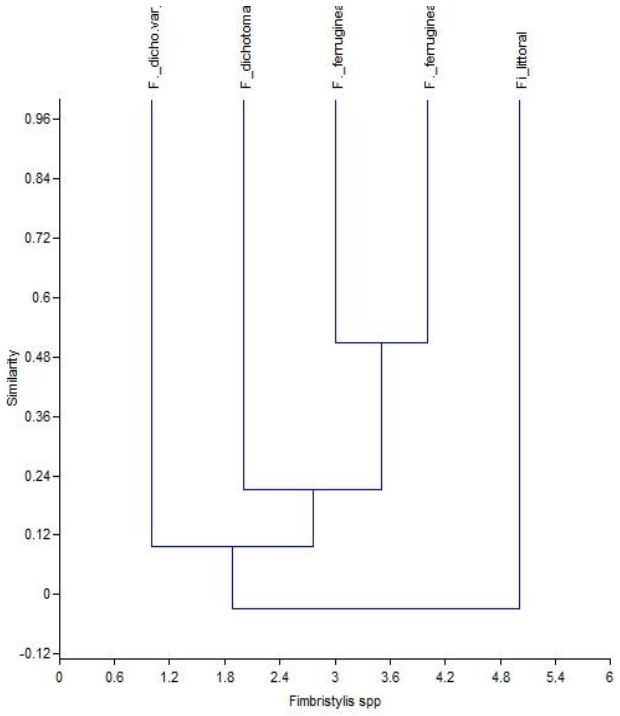

Figure 9. Dendrogram showing relationship among *Fimbristylis* species studied based on phytochemical constituents

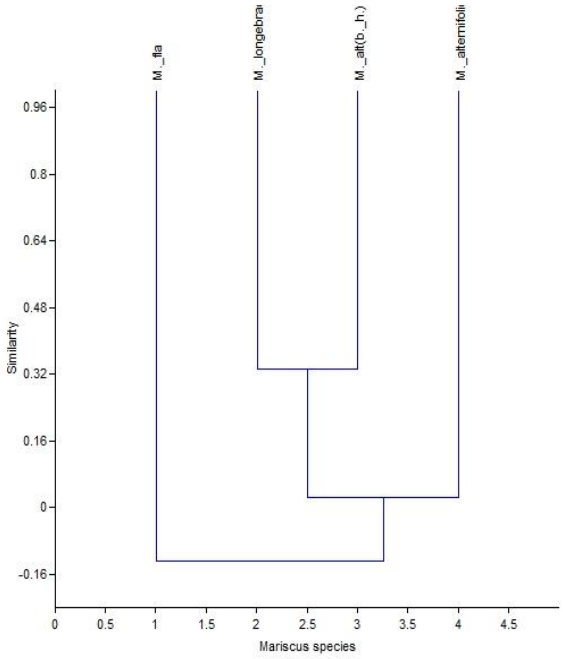

Figure 10. Dendrogram showing relationship among *Mariscus* species studied based on phytochemical constituents

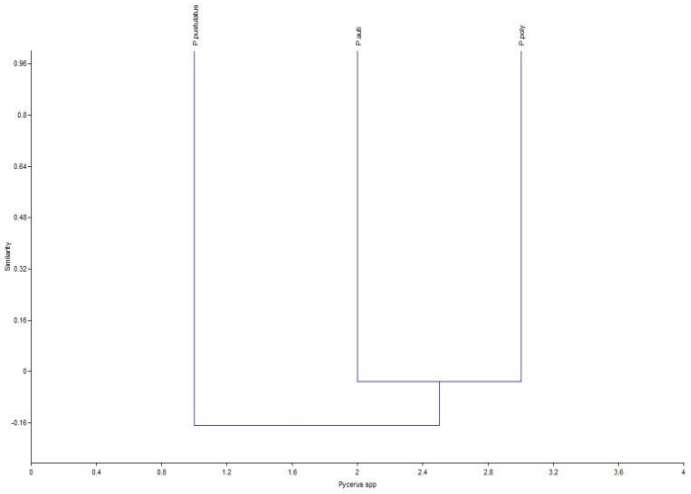

Figure 11. Dendrogram showing relationship among *Pycreus* species studied based on phytochemical constituents

The relationship elucidated among the species studied through the morphological analysis in the present study was not unexpected because they all belong to the subfamily Cyperoideae

except *R. corymbosa* which belongs to the subfamily Caricoideae, tribe Rhychosporeae (Bruhl, 1995). *Kyllinga, Cyperus, Mariscus* and *Pycreus* are in the tribe Cypereae while *Fimbristylis* was placed in the tribe Abildgaaridieae (Bruhl, 1995). Longevity and achene morphology separated *Fimbristylis littoralis* from the other *Fimbristylis* species studied. *Mariscus longbracteatus* was embedded within the cluster of most of the *Cyperus* species studied while *M. alternifolius* and *M. flabelliformis* was grouped along with members of other genera in the tribe Cypereae. There was controversy concerning the status of *Marisus* as a distinct genus /Subgenera (Goetghebeur, 1986; Bruhl, 1995; Goetghebeur, 1998; Vrijdaghs *et al*., 2011; Desai and Raole, 2014). Moreover, it was stated that *Cyperus* is not monophyletic due to the fact that other cyperoid genera are embedded within *Cyperus* s. s. More so, there are different opinions on whether *Cyperus* and allied genera should be merged as a genus or should be regarded as *Cyperus* s. l. subgenera or each taxon as a distinct genus (Goetghebeur, 1989; Bruhl, 1995; Goetghebeur, 1998; Meshack, 2007; Muasya et al., 2001; Muasya et al., 2002; Vrijdaghs et al., 2011; Desai and Raole, 2014). Though *Rhynchospora* is a special genus in the family Cyperaceae being one of a few genera in which pollination strategy transit from wind to insect pollination (Lucero et al., 2014). However, the only species of *Rhynchospora* studied in this current study was embedded in the *Cyperus* cluster.

Considering the number of spots identified in the samples screened for saponin, it can be said that it is the most diverse among the secondary metabolites studied. Conversely, Steroids showed only four different spots; making steroid the least diverse among all the secondary metabolites screened. Alkaloid with Rf 0.98 was common to all the species studied except *M. alternifolius* (small infl.) and *K. odorata.* However, alkaloid at spotted at Rf 0.22 was unique to *F. littoralis*; this can be diagnostic for this species. Similarly, tannins at Rf 0.98 was detected across all the species studied except *F. dichotoma* var. *dichotoma* and *F. littoralis.* Among the *Kyllinga* species from this study, saponin at Rf 0.7 was detected both in *K. bulbosa* and *K. pumila* which were grouped together. *Kyllinga odorata* that was distant from other species studied did not share any secondary metabolite at the same Rf with them. Moreover, flavonoid was not detected in *K. odorata* likewise, *C. glycoside* in *K. bulbosa*. *Pycreus acuticarinatus* and *P. polystachyos* were grouped together; they shared more secondary metabolites at different Rf values in alkaloids, flavonoids, tannins, saponin and triterpenes than *P. pustulatus.* Also, phytochemicals at Rf 0.9, 0.98 and 0.88 in saponin, steroid and triterpene respectively are common to *M. longibracteatus* and *M. alternifolius* (small infl.)

The presence of alkaloids, tannins and steroids in the entire selected species screened coupled with flavonoids and saponins nearly in all the Cyperaceae species studied inferred that they may be pharmaceutically useful (Oladipo et al., 2017). Alkaloids are valuable in pharmacological applications of stimulants and anesthetics in the central nervous system (Madziga et al., 2010). Flavonoids comprise one of the most important groups of polyphenols; making up over 60% and serve to bring down the danger of coronary heart disease. Additionally, they are used as cancer prevention agents and employed as a natural antioxidant due to their capacity to scavenge free radicals (Kim et al., 1990; Rice-Evans et al., 1996; Olatunji and Afolayan, 2019). Tannins have antimicrobial activity (Scalbert, 1991) and this helps in the treatment of gastrointestinal disorders (Burkill, 1985). Studies have reported anti-inflammatory, antioxidant, antiviral, antibacterial, antiparasitic, anticancer, antiseptic, and antidiuretic properties of tannins (Souza et al., 2006). Steroids possess the ability to display activities such as

antifungal, antiviral, antileukemic and muscle-relaxant activities (Kokpol et al., 1984). Some steroids have been documented to be used in the treatment of sexual dysfunction (Oyedemi et al., 2012). Saponins help to reduce cholesterol levels which in turn lower the risk of cardiovascular diseases such as hypertension (Francis et al., 2002). Moreover, certain saponins are being used in the food and beverage industry as well as in cosmetics (Price et al., 1987; Petite et al., 1995; Uematsu et al., 2000). Cardiac glycosides help in the treatment of heart-related ailments and they have potential anti-cancer activity (Prassas and Diamandis, 2008). Antifungal and antibacterial properties are present in terpenoids (Amaral et al., 1998). *Cyperus rotundus* and *K. nemoralis* are among the monocotyledons screened for the presence of secondary metabolites by Ankanna et al., (2012). They reported the presence of alkaloids and triterpenoids in these plants while flavonoids, glycosides, saponins and tannins were not detected in them. However, *C. rotundus* tested positive for steroid, the latter tested negative. In this present study, all the phytochemicals tested were present in both *C. rotundus* and *K. nemoralis.*

Cyperus sphacelatus, C. dilatatus and *C. tuberosus* were grouped together by SLCA based on their phytochemical composition in this study. These three species are very similar morphologically with the formal two more closely related than the latter. The latter two are the most confused ones especially when they are still young. *Cyperus rotundus* is more related to them than *C. esculentus* according to this present study. Interestingly, both the morphological and phytochemical characterization separated *F. littorilis* from other the two *Fimbristylis* species studied. Kang et al., (2018) stated that the phylogenetic and phytochemical relationship does not coincide with each other, though partial consistency could be found among them. The two accessions of *F. ferruginea* studied were grouped together; even though they are slightly differed in their inflorescence; one is spotted while the other is unspotted, the two accessions are the same species. They possess the same set of flavonoids, tannins, alkaloids, and cardiac glycosides while they have different spots with different Rf values for steroids, saponins and triterpenes.

The presence of a set of secondary metabolites in different species belonging to a genus indicated a generic phenomenon. Genes are involved in the biosynthesis of metabolites and some are implicated in the production of a specific class of secondary metabolites. In addition, the corresponding gene products of individual members may use the same or similar substrate to produce similar metabolic products. A number of genes are responsible for the biosynthesis of phytochemicals and there are many metabolic genes in plants that are responsible for the production of diverse phytochemicals in a tissue-specific manner (Matsuda et al., 2010; Iu et al., 2017). Flavonoids have revealed a wide range of chemical structures that were shown to have a genetic basis for their variation (Saito, 2013; Zhao et al., 2013). Mostly, the chemical structure of secondary metabolites is specific and unique to taxonomically related species which may not be unconnected to their restricted occurrence (Singh, 2016).

3 Conclusion

The cluster analysis based on morphological character showed that *Cyperus* is not monophyletic as earlier reported as other members of other genera studied were embedded in the *Cyperus* cluster. The TLC profiling of the species of Cyperaceae studied revealed their phytochemical diversity. Flavonoids, alkaloids, tannins, saponins, triterpenes, cardiac glycosides and steroids were detected in the leaf extract of the species studied and they possess certain medicinal

attributes, which can be utilized in the treatment of certain ailments, food preparation and also explored in pharmaceutical industries. Moreover, the phytochemicals detected in this study have been demonstrated to be useful in delimiting the species within each genus studied. However, they have little or no taxonomic value in delimiting the genera studied within the family Cyperaceae. It should be pointed out that the morphological grouping did not correspond with the phytochemical grouping in this study, even though, partial consistency was observed as shown among the *Fimbristylis* grouping in this study. Further research is required in the characterization of the diverse secondary metabolites detected in some members of Cyperaceae studied.

4 Acknowledgments

Special thank goes to Doctor Oladipo T. O. and Doctor Bello, A. O. for their contributions to the preparation of this manuscript.

5 Conflict of interests

The authors declared that there is no conflict of interests regarding publication of this paper.

References

Akobundu, I. O., & Agyakwa, C. W. (1998). *A handbook of West African Weeds*. International Institute of Tropical Agriculture, Ibadan, Nigeria.

Amaral, J. A., Ekins, A., Richards, S. R., & Knowles, R. (1998). Effect of selected monoterpenes on methane oxidation, denitrification, and aerobic metabolism by bacteria in pure culture. *Applied and Environmental Microbiology*, *64*(2), 520-525. https://doi.org/10.1128/AEM.64.2.520-525.1998.

Ankanna, S., Suhrulatha, D., & Savithramma, N. (2012). Chemotaxonomical studies of some important monocotyledons. *Botany Research International*, *5*(4), 90-96. doi: 10.5829/idosi.bri.2012.5.4.501.

Babu, H. R., & Savithramma, N. (2014). Screening of secondary metabolites of underutilized species of Cyperaceae. *Int J Pharm Sci Rev Res*, *24*, 182-187. www.globalresearchonline.net.

Barrett, R. L. (2013). Ecological importance of sedges: a survey of the Australasian Cyperaceae genus *Lepdosperma*. *Annals of Botany*, 111, 499-529. doi: 10.1093/903/mct008.

Bruhl, J. J. (1995). Sedge genera of the world: Relationships and a new classification of the Cyperaceae. Australian Systematic Botany, 8, 125–305. https://doi.org/10.1070/SB9950125.

Burkill, H. M. (1985). *The Useful Plants of West Tropical Africa*. 2nd ed. Royal Botanic Gardens.

Carter, R. (2005). An introduction to the sedges of Georgia. *Tipularia*, 20, 15-44.

Cheema, P., Saggoo, M. I. S., & Kumar, N. (2017). Cytomorphology of some medicina sedges from North West India. *International Journal of Pharmacy and Pharmaceutical Research (Human),* 10(2), 231-243. www.ijppr.humanjournal.com.

Desai, R. J., & Raole, V. M. (2014). Synflorescence architecture studies in some Indian Cypereae (Cypereaceae). *Rheedea*, 24(2), 89-98.

Francis, G., Kerem, Z., Makkar, H. P., & Becker, K. (2002). The biological action of saponins in animal systems. *British Journal of Nutrition,* 88(6), 587-605. https://doi.org/10.1079/BJN2002725.

Goetghebeur, P. (1989). Studies in Cyperaceae 9. Problems in Leptotypification and infrageneric taxonomy of Cyperus L. *Bulletin de la Societe Royale de Botanique de Belgique,* 122, 103-114. https://doi.org/10.2307/20794159.

Goetghebeur, P. (1998). Cyperaceae. In K. Kubitzki (Ed.), *The families and genera of vascular plants, 4: flowering plants, monocotyledons: Alismatanae and Commelinanae (except Gramineae)* (pp. 141–190). Berlin, Germany: Springer. http://hdl.handle.net/1894/LU-278975.

Hegnauer, R. (1986). Comparative phytochemistry and plant taxonomy. *Giornale Botanico Italiano,* 120 (1-6), 15-26. https://doi.org/10.1080/11263508609428018.

Hutchinson J., & Dalziel J. M. (1972). *Flora of West Tropical Africa.* (2nd edn.) Crown Agents: London.

Iu, F., Zhang, W., Li, Y., & Wang, H. L. (2017). Establishment of model system between phytochemicals and gene expression profiles in Macrosclereid cells of *Medicago trucatula. Scientific Reports,* 7, 2580. https://doi.org/10.1038/s41598-017-02827-5.

Kang, K. B., Kang, S., Kim, M. S., Lee, D. Y., Han, S. I., Kim, T. B.,Park, J. Y., Kim, J., Yang, T., & Sung, S. H. (2018). *Phytochemistry*, 155, 126-135. https://doi.org/10.1006/j.phytochem.2018.07.012.

Kim, J. Y., Germolec, D. R., & Luster, M. I. (1990). Panax ginseng as a potential Immunodulator: studies in mice. *Immunopharmacology and Immunotoxicology,* 12, 257.

Kokpol, U., Chittawong, V., & Mills, H. D. (1984). Chemical consitituents of the roots of *Acanthus illicifolius. Journal of Natural Products* 49, 355-356. https://doi.org/10.1021/np50044a033.

Lowe, J., & Stanfield, D. P. (1974). *The Flora of Nigerian Sedges.* Ibadan University Press: Ibadan. ISBN 978 121 001.

Lucero, L. E., Vegetti, A. C., & Reinheimer, R. (2014). Evolution and development of the spikelet and flower of *Rhynchospora* (Cyperaceae). *International Journal of Plant Science,* 175(2), 186-201. https://doi.org/10.1086/674317.

Lye, K. A., & Masterhazy, A. (2012). Studies in African Cyperaceae 35. *Kyllinga carinalaevis* sp. nov. from west tropical Africa. *Nordic Journal of Botany,* 30, 385-388. https://doi.org/10.1111/j.1755-1051.2012.01289.x.

Madziga, H. A., Sanni, S., & Sandabe, U. K. (2010). Phytochemical and elemental analysis of *Acalypha wilkesiana* leaf. *Journal of American Science*, 6(11), 510-514. http://www.americanscience.org.

Mannheimer, C. A. (1999). An overview of chemotaxonomy and its role in creating a phylogenetic classification system. *AGRICOLA,* 1998/1999: 87-90.

Matsuda, F., Hirai, M. Y., Sasaki, E., Akiyama, K., Yonekura-Sasakibara, K., Provart, N. J., Sakurai, T., Shimada, Y., & Saito, K. (2010). AtMet Express Development: a phytochemical atlas of *Arabidopsis* development. *Plant Physiology*, 152, 566-578. https/doi.org/10.1104/pp.109.148031.

Meshack, N. D. (2007). *Cyperaceae tribe Cypereae: Phylogenetic relationships and evolutionary patterns of diagnostic characters*. A dissertation submitted to the University of Cape Town, in partial fulfillment of requirements for the award of Honours degree in Botany.

Misra, A., & Srivastava, S. (2016). Chemotaxonomy: An approach for conservation and exploration of industrially potential medicinal plants. *Journal of Pharmacognosy and Natural Products,* 2, 4. https://doi.org/10.4172/2472-0992./000e108.

Muasya, A. M., Simpson, D. A., & Chase, M. W. (2001). Generic relationship and character evolution in *Cyperus* s.l. (Cyperaceae). *Systematics and Geography of Plants*, 71(2), 539-544. https/doi.org/10.2307/3668698.

Muasya, A. M., Simpson, D. A., & Chase, M. W. (2002). Phylogenetic relationships in *Cyperus* L. s.l. (Cyperaceae) inferred from plastid DNA sequence data. *Botanical Journal of Linnean Society* 138, 145-153. http://dx.doi.org/10.1046/j.1095-8339.2002.138002145.

Musaya, A. M., Simpson, D. A., Verboom, G. A., Goetghebeur, P., Naczi, R. F. C., Chase, M. W., & Smets, E. (2009). Phylogeny of Cyperaceae based on DNA sequence data: Current progress and future prospects. *Botanical Review* 75, 2-21. https://doi.org/10.1007/s12229-008-9019-3.

Oladipo, O. T., Akinpelu, B. A., Folorunso, A. E., Godwin, A., Omotoso, S. E., Dosunmu, O. A., & Joseph., W. A. (2017). Chemotaxonomic Study of Six Nigerian Ficus Species (Moraceae). *Notulae Scientia Biologicae* 9(2): 250-255. https://doi.org/10.15835/nsb9210024.

Olatunji, T. L., & Afolayan, A. J. (2019). Comparative Quantitative study on phytochemical contents and antioxidant activities of Capsicum annum L. *The Scientific World Journal*, Vol 2019. Article ID 4705140 13pp. https://doi.org/10.1155/2019/4705140.

Oyedemi, S. O., Oyedemi, B. O., Arowosegbe, S., & Afolayan, A. J. (2012). Phytochemical Analysis and Medicinal Potentials of Hydroalcholic Extract from *Curtisia dentata* (Burm.f) C.A. Sm Stem Bark. *International journal of Molecular Sciences* 13, 6189-6203. https://doi.org/10.3390/ijms13056189.

Pal, H., & Choudhury, M. D. (2017). Cyperaceae: A Review of its Taxonomic Treatments. *Pleione* 11(2), 195-207. https://doi.org/10.26679/Pleione.11.2.2017.195-207.

Petite, P. R., Sauvire, Y. D., Hillaire-Buys, D. M., Leconte, O. M., Baissac, Y. G., Ponsin, G. R., & Ribes, G. R. (1995). Steriods, saponins from fenugreek seeds: extraction, purification and pharmalogical investigation on feeding behavior and plasma cholesterol. *Steriods* 60(10), 674-680. https://doi.org/10.1016/0039-128x(95)00090-d.

Prassas, I., & Diamandis, E. P. (2008). Novel theraupeutic applications of cardiac glycosides. *Nature Review (Drug Discovery*), 7, 926-935. https://doi.org/10.1038/nrd2682.

Price, K. K., Johnson, L. I., & Feriwick, D. (1987). The chemical and biological significance of saponins in food and feeding stuffs. *Food Science and Nutrition* 26, 27-135. https://doi.org/10.1080/10408398709527461.

Reid, C. S., Doyle, V. P., Carter, J. R., Vargas-Rodriguez, Y., & Urbatsch, L. E. (2017). Molecular systematics of targeted flat sedges (*Cyperus,* Cyperaceae) of the Americas. *Plant Ecology and Evolution*, 150(3), 343-357. https://doi.org/10.5091/placevo.2017.1202.

Rice-Evans, C. A., Miller, N. J., & Paganga, G. (1996). Structure–antioxidant activity relationships of flavonoids and phenolic acids. *Free Radical Biology and Medicine* 20(7), 933-938. https://doi.org/10.1016/0891-5849(95)02227-9.

Roalson, E. H. (2008). A synopsis of chromosome number variation in Cyperaceae. *Botanical Review*, 74, 209-393. https://doi.org/10.1007/S12229-008-9011-4.

Saito, K. (2013). Phytochemical genomics- a new trend. *Current Opinion in Plant Biology*, 16, 373-380. https://doi.org/10.1080/10408398709527461.

Scalbert, A. (1991). Antimicrobial properties of tannin. *Phytochemistry* 30(12), 3875-3883.

Singh, R. (2016). Chemotaxonomy: A tool for plant classification. *Journal of Medicinal Plant Studies,* 4(2), 90-93. https://doi.org/10.1016/0031-9422(91)83426-L.

Souza, S. M., Aquino, L. C., Milach, A. C., Bandeira, M. A., Nobre, M. E., &Viana, G. S. (2006). Antiinflammatory and anti-ulcer properties of tannins from *Mycracrodon urundeuva* Allemao (Anacardiaceae) in Rodents. *Phytotherapy Research* 21(3), 220-225. https://doi.org/ 10.1002/ptr.2011.

Tantwai, K. (2017). *Studies for identification and conservation of Cyperus species using DNA barcoding*. A thesis submitted in the partial fulfillment of the requirement for the degree of Doctor of Philosophy in Molecular Biology and Biotechnology of Jawaharlal Nehru Krishi Vishwa Vidyalaya, Jabalpur, Madhya Pradesh.

Uematsu, Y., Hirata, K., & Saito, K. (2000). Spectrophotometric determination of saponins in Yucca extract used as food additive. *Journal of AOAC International,* 83, 1451-1454. https://doi.org/10.1093/jaoac/83.6.1451.

Vrijdaghs, A., Reynders, M., Muasya, A. M., Larridon, I., Goetghebeur, P., & Smets, E. F. (2011). Morphological and development of spikelets and flowers in *Cyperus* and *Pycreus* (Cyperaceae). *Plant Ecology and Evolution*, 144(1), 44-63. https://doi.org/10.5091/placevo. 2011.436.

Wagner, H., Baldr, S., & Zgainski, E. M. (1996). *Plant drug analysis*. New York, Berlin, Springer.

Xu, Z., & Zhou, G. (2017). *Identification and Control of Common Weeds*: Volume 1. Hangzhou and Springer Science + Business media Dordrecht, Zhejiang University Press. https://doi.org/10. 1007/978-94-024-0954-3_2.

Zhao, N., Wang, G., Norris, A., Chen, X., & Chen, T. (2013). Studying plant secondary metabolism in age of genomics. *Critical Reviews in Plant Sciences*, 32(6), 369-382. https://doi.org/10.1080/07352689.2013.789648.