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PHYTOMEDICINAL POTENTIALS OF SPECIES OF NEPHROLEPIS (SCHOTT.)

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ABSTRACT

Natural products have played vital roles in health care for decades. Of the different sources of natural products, plants have been a source of chemical substances which serve as drugs in their own right or key ingredients in formulations containing synthetic drugs. *Nephrolepis* is a genus of about thirty (30) species of ferns which are distributed in many parts of the world. Phytochemical screening indicated the presence of phytochemicals such as flavonoids, terpenoids, tannins, saponins and cardiac glycosides, while alkaloids and phlobatannins were completely absent in all the three species. Antinutrients such as hydrocyanide, tannin, oxalate and phytic acid were found to be present in the plants, and their concentrations were all within the permissible levels, apart from oxalates which exceeded the permissible level in *Nephrolepis exaltata* (L.) Schott. Anatomical investigations in the three species of *Nephrolepis* ((*Nephrolepis biserrata* (Sw.) Schott, *Nephrolepis exaltata* and *Nephrolepis undulata* (Afzel. ex Sw.) J. Sm.)) showed that stomatal types were, Diacytic, Anisocytic, Anomocytic and Anomalous. Epidermal cell walls were undulating in all the three species studied. Unicellular and multicellular types of trichomes were found in *N. biserrata* but trichomes were absent in *N. exaltata* and *N. undulata*. The micromeritric study of the three species of *Nephrolepis* showed that they all had fair flowing properties. The phytochemical contents and the anatomical results show similarities in the plants. It is recommended that further studies should be carried out on species of *Nephrolepis* and other ferns in general in order to determine their potential uses in phytomedicine.

KEYWORDS: Micrometry, *Nephrolepis*, Phytochemicals, Phytomedicine, Stomata, Trichomes.

INTRODUCTION

Nephrolepis is a genus of about thirty (30) species of ferns of mainly pantropical distribution but extending both north and south to as much as 40^0 latitude[1]. The genus is in the family Davalliaceae. It is commonly referred to as "Macho fern". Some other common names used to describe *Nephrolepis* include; Sword fern, and Boston fern. The genus *Nephrolepis* is morphologically quite distinct and easily defined; the stem is a short erect stock with closely tufted fronds, it has a slender lateral branch (runners) bearing numerous roots and occasional buds which develops into new plants[2]. The fronds are typically long and narrow (sword shaped). The sori is usually single and terminal on each vein on the lower surface or near the margin of the pinna, protected by a kidney-shaped or orbicular indusium. The matured plant body is sporophytic and can be differentiated into rhizome, roots, and leaves. The stem has a long slender lateral branch called runner. The runners spread for a considerable distance and bear roots. The roots are branched and adventitious and they arise from the rhizome and runners in acropetal succession. The leaves are tufted, long, narrow and simply pinnate. The fronds are 30 - 240 cm long. The pinnae are numerous, crowded, often imbricated, 3 -20 cm long, slightly falcate and articulated at the base. The margins are entire or slightly crenate. The veins are free and there is a white line of dots over the vein tips. Sori are present on the lower surface. The sori are half way between the midrib and the margin in a single row, or submarginal, or near the margin, depending on the species. The indusia are usually round – reniform with a narrow sinus. Sometimes the sinus widens to a broad curved base. The young leaves are covered with multicellular hairs and scales and show circinate venation.[2]

Scientific classification of *Nephrolepis* [3]

Kingdom	:	Plantae
Division	:	Pteridophyta
Class	:	Polypodiopsida
Order	:	Polypodiales
Family	:	Davalliaceae
Genus	:	*Nephrolepis*

The aim and objectives of this research is as follows;

1. To collect and identify common species of *Nephrolepis* in Uyo metropolis, Akwa Ibom State, Nigeria.
2. To investgate the epidermal and petiole anatomy of the plants
3. To determine the types of phytochemicals present in the plants.
4. To determine the anti-nutritional composition of the plants.
5. To determine the flow properties of powdered samples.

MATERIALS AND METHODS

COLLECTION AND IDENTIFICATION OF PLANT SAMPLES

All three plant samples; *N. biserrata, N. exaltata* and *N. undulata* were collected at Ikot Akpabio in Shelter Afrique along Oron road in Uyo, Akwa Ibom State, Nigeria, in the month of May 2018. These plant samples were identified and authenticated by Professor M. E. Bassey, a Taxonomist with the Department of Botany and Ecological Studies, University of Uyo,Uyo,Akwa Ibom State. Voucher specimens were deposited in the University of Uyo Herbarium (UUYH).

ANATOMICAL STUDIES

For the purpose of anatomical studies, the median portion of epidermal peels of matured leaflets of the plant were obtained. The specimen surface was irrigated with water holding the specimen from one end. The epidermis above the desired surface was scrapped off carefully with a sharp razor blade. Loose cells were washed away from the epidermal peel with water until the desired epidermis below was reached. The epidermal peels were bleached in sodium hypochlorite solution and rinsed in distilled water and were stained with aqueous solution of safranin for 4-8 minutes and then mounted in 10 % glycerol and was viewed with an Olympus CX21 binocular microscope.

The petiole of each plant was sectioned transversly by placing a petiole in a cork and thin sections were made using a sharp razor blade. The sections were stained with aqueous solution of safranin then rinsed carefully with water to remove excess stain and mounted in 10 % glycerol and was viewed with an Olympus CX21 binocular microscope.

Photographs of the prepared slides were taken with an Amscope.

Stomatal index was determined according to[4], Using the formula:

$$S.I = S/E+S \times 100/1$$

Where,

S = Number of stomata per unit area

E = Number of epidermal cells in same area

All measurements were made with the aid of a caliberated ocular micrometer and treated statistically.

PHYTOCHEMICAL SCREENING

The phytochemical screening was done using the methods of.[5] The analytical response was identified by the colour, ring formation, or the precipitation formed from the test.

ANTI- NUTRIENTS EVALUATION

All three plant samples were tested for the four major anti-nutrients using the methods of[6] for Hydrocyanide (HCN),[7] for Phytic acid (phytate),[8] for oxalate and[9] for Tannins.

MICROMERITRIC EVALUATION

Preparation of Plant Samples for Micromeritric Studies

The leaves of the plant samples were sliced and air-dried under room temperature. After which the dried samples were reduced to powdered form using a mortar and pistil. The powders were passed through a 350 micron mesh sieve and stored in an airtight container for further use. Various physicochemical properties were calculated, some of which include; bulk volume, tapped volume, bulk density, tapped density, Hausner's ratio, Carr's index, diamter, height of heap and the flow time. These parameters were calculated in order to determine the total flow rate of the powders.

Bulk density and Tapped density

About 10 g of the of the dried leaf powder was weighed and measured into a 100 ml measuring cylinder and the volume which is called the bulk volume (Vb) was noted. Then the cylinder was gently tapped and again the volume which is called the tapped volume (Vt) was noted. The initial volume gave the bulk density value and after tapping the volume reduced and it gave the value of the tapped density.

Bulk density (Db) =W/Vb. Where W is the weight (10 g) of the plant powder and Vb is the total volume or the bulk volume of the packing.

Tapped density (Dt) =W/Vt. Where W is the weight (10 g) of the plant powder and Vt is the final volume after tapping.

Hausner's Ratio

Hausner's ratio which is related to inter particle fraction was used to predict the powder flow properties.

Hausner's ratio (H.R) =Dt/Db. Where Dt is the tapped density and Db is the bulk density. Values less than 1.25 indicated a good flow, while values greater than 1.25 indicated a poor flow.

Carr's Index

Carr's index was another indirect nethod that was used for measuring the powder flow from the bulk density.

Carr's index (C.I) = (Dt-Db/Dt)x100. Where Dt is the tapped density and Db is the bulk density. The bulk density and tapped density were expressed in grams per mililitre (g/ml). The values below 25 % indicated good flow characteristics ,while the values above 25 % indicated a poor flow characteristics.

RESULTS

The results for the anatomical studies, phytochemical analysis, anti- nutrient composition, micromeritric evaluation and the extractive values are summarized in Tables 1 – 6 and Figures 1 – 3.

Table 1 and Figures 1 – 3 shows the result of the anatomical studies. On the adaxial (upper) surface, Results showed that the anticlinal walls of the epidermal cells were all undulate in all the three plant species. Stomata were present on the abaxial surfaces of all three plant species. The stomata were Diacytic, Anomocytic and Anicocytic types of stomata. In Table 1, the length of the stomata in the plants had a range of 10.12±0.28- 13.07±0.36 µm, while the width had a range of 6.27±0.18-8.24±0.24µm. The Stomatal Index for *N. biserrata* (Lower surface) was 10.77 %, that for *N. exaltata* (Lower surface) was 15.08 %, that for *N. undulata* (Lower surface) was 20.59% and that for *N. undulata* (upper surface) was 15.39%. In Table 2, *N. undulata* had the the longest epidermal cell (42.52±2.19 µm), followed by *N. biserrata* (29.31±1.37µm) and lastly *N. exaltata* (25.87±1.49µm), While *N. biserrata* had the widest epidermal cell (8.46±1.02µm), followed by *N. exaltata* (7.46±0.51µm) and lastly *N. undulata* (6.60±0.64µm). *N. exaltata* had the highest number of epidermal cells (64.20±0.92) next to *N. biserrata* (55.50±1.95 µm) and finally *N. undulata* (48.20±1.27 µm). In Table 1, *N. biserrata*

had a stomatal number of 6.70±0.45 µm, that for *N. exaltata* was 11.40±0.60 µm and lastly for *N. undulata* was 12.50±0.73 µm. Unicellular and multicellular trichomes were present in *N. biserrata* (fig. 1a and b). In Table 2, the trichome had a length of 66.82±6.67 µm and width of 5.38±0.24 µm.

The anticlinal walls of the epidermal cells on the adaxial surface were also undulating. Stomata were also completely absent in *N. biserrata* and *N. exaltata* but was present in *N. undulata* (Diacytic and Anomocytic) with a length of 12. 31±0.24µm (Table 1) and a width of 7.33±0.14 µm (Table 1). The stomata number was 8.90±0.64 µm (Table 1), with a stomatal index of 0.15 (Table 1). Trichomes were completely absent. The longest epidermal cell on the adaxial surface was found in *N. exaltata* (66.10±2.24 µm; Table 2) followed by *N. biserrata* (60.10±1.61µm; Table 2) and lastly *N. undulata* (48.90±1.54 µm; Table 2).

The result of the petiole anatomy is shown in figures 3A – 3C. There was presence of cuticle and pericycle was present, Sclerenchyma was found immediately after the epidermal layer. About 2-3 rows in some parts of the cortex and arranged in a semi -corde. Collenchyma were observed after sclerenchyma. Vascular bundles were broken into meristeles. Phloem and xylem vessels were present. Sclerenchyma cells were 6 – 10 rows in the cortex and parenchyma were observed in the central region in which vascular tissues were embedded.

Table 1: Results for the stomatal Information for *N. biserrata, N. exaltata* and *N. undulata.*

Plant species	Epidermal Cell Number	Surface	Stomata Distribution	Stomata Type	Stomata Length (µm)	Stomata Width (µm)	Stomata Ratio L/W	Stomata Number	Stomata Index (%)
N.biserrata	55.5±195	L	Hypostomatic	Diacytic, Anisocytic and Anomocytic	10.12±0.28	6.27±0.18	1:1	6.70±0.45	10.77
N. biserrata	60.10±61	U	Absent	Absent	Absent	Absent	-	Absent	-
N. exaltata	64.20±0.92	L	Hypostomatic	Anomalous	11.61±0.42	8.24±0.24	1:1	11.40±0.65	15.08
N. exaltata	66.10±2.24	U	Absent	Absent	Absent	Absent	-	Absent	-
N. undulata	48.20±1.27	L	Amphistomatic	Anisocytic	13.07±0.36	7.36±0.24	1:1	12.50±0.73	20.59
N. undulata	48.90±1.54	U	Amphistomatic	Anisocytic and Anomocytic	12.31±0.24	7.33±0.14	1:1	8.90±0.64	15.39
LSD 5 %	38.43 S				16.94 S	19.59 S		24.88 S	0.00 NS

Values are represented as Mean ± of ten (10) determination

Key: L = Lower surface, U = Upper surface, S= significant difference, NS= No significant difference, SEM= Standard error of the mean

Table 2: Results for the Trichome and Epidermal cell Information for *N. biserrata, N. exaltata* and *N. undulata.*

Plant species	Surface	Type of trichome	Length of Trichome (µm)	Width of Trichome (µm)	Length of Epidermal Cell (µm)	Width of Epidermal Cell (µm)	Epidermal Cell Ratio L/W	Thickness of Epidermal Cell	Epidermal Cell Type
N. biserrata	L	Unicellular and Multicellular	66.82±6.67	5.38±0.24	25.87±1.49	8.46±1.02	3:1	0.52±0.09	Undulating
N. biserrata	U	Absent	Absent	Absent	22.06±0.85	5.99±0.35	4:1	0.32±0.02	Undulating
N. exaltata	L	Absent	Absent	Absent	29.31±1.37	7.46±0.51	4:1	0.61±0.02	Undulating
N.exaltata	U	Absent	Absent	Absent	21.79±1.15	9.29±0.98	2:1	0.470.01	Undulating
N. undulata	L	Absent	Absent	Absent	42.52±2.19	6.60±0.64	7:1	0.211±0.01	Undulating
N. undulata	U	Absent	Absent	Absent	42.62±3.45	6.61±0.45	7:1	0.63±0.01	Undulating
LSD 5 %					26.04 S	1.55 NS		15.01 S	

Values are represented as Mean±SEM of ten (10) determination

Key: L= Lower surface, U= Upper surface, S = Significant Difference, N.S = No Significant difference, SEM = Standard Error of the Mean

Table 3 shows the the result of the extractive values of the plant extracts. From the table it shows that *N. biserrata* had the highest extractive value (5.779 %) meaninig it yielded more than *N. exaltata* and *N. undulata*.

Table 3: Results for the extractive value.

Sample	Weight of empty beaker (g)	Weight of sample + beaker (g)	Yield of extract (%)
N. biserrata	62.111	67.890	5.779
N. exaltata	61.702	66.879	5.177
N. undulata	60.417	62.815	2.398

Table 4 shows the results for the phytochemical screening. From the table it is seen that all the three plant species were void of alkaloid and phlobatannin. Saponnins and cardiac glycosides were strongly present in all the three plant species. Flavonoids were strongly present in *N. exaltata* and *N. undulata* but was absent in *N. biserrata.* Tannins were strongly present in *N. exaltata* and *N. undulata* but were not strongly present in *N. biserrata*. Terpenoids were absent in *N. biserrata* but were strongly present in *N. exaltata* and *N. undulata.*

Table 4: Result for Phytochemical Screening.

Phytochemical properties	*N. biserrata*	*N. exaltata*	*N. undulata*
Saponnin	+++	+++	+++
Tannin	++	+++	+++
Terpenoids	-	+++	+++
Cardiac glycosides	+++	+++	+++
Phlobatannin	-	-	-
Alkaloid	-	-	-
Flavonoid	-	+++	+++

Key: +++ = Strongly present ++ = Present + = Trace - = Absent

The result for anti-nutrient composition is shown in Table 5. From the table it is seen that *N. exaltata* had the highest concentration of hydrocyanide, tannins, phytates and oxalates while *N. undulata* had the least values in all the anti- nutrients.

Table 5: Result for Anti – Nutrient Evaluation of *N. biserrata, N. exaltata* and *N. undulata*

Anti- nutrient properties	*N. biserrata*	*N. exaltata*	*N. undulata*
Hydrocyanide (HCN)	5.89±0.01	13.66±0.04	2.71±0.05
Tannin	26.07±0.01	92.37±0.06	18.85±0.03
Phytic acid (Phytates)	8.34±0.02	10.10±0.05	5.19±0.01
Oxalic acid (oxalates)	183.68±5.09	265.62±6.35	144.07±12.74

Values are Represented as Mean ± SEM of triplicate (3) determination

Table 6 shows the result of the micromeritic evaluation. From the table it is seen that *N. biserrata* had the highest bulk and tapped volumes while it was least in *N. undulata*. *N. undulata* had the highest bulk and tapped densities while *N. biserrata* had the least. The values of the hausner's ratio, carr's index, diameter were recorded. The height of heap, the flow time and the angle of repose were also recorded.

Table 6: Results for Micromeritric Properties of *N. biserrata, N. exaltata* and *N. undulate.*

Properties	*N. biserrata*	*N. exaltata*	*N. undulata*
Bulk volume (mL)	55.0±0.58	43.33±1.20	29.17±0.44
Tapped volume (mL)	34.17±0.17	30.33±0.17	19.50 0.29
Bulk density (mL)	0.18±0.00	0.23±0.01	0.34±0.01
Tapped density (mL)	0.29±0.00	0.33±0.00	0.51±0.03
Hausner's ratio	1.58±0.03	1.43±0.00	1.51±0.03
Carr's index	36.67±1.33	31.00±1.73	33.67±1.45
Diameter of heap (cm)	7.71±0.09	7.18±0.08	7.58±0.05
Height of heap (cm)	3.00±0.05	2.60±0.00	2.80±0.06
Flow time (secs)	21.67±0.88	20.00±1.00	21.00±0.58
Angle of repose	38^0	36^0	36^0
Flow rate	0.46	0.50	0.48

Values are represented as Mean ± SEM of triplicate (3) determination

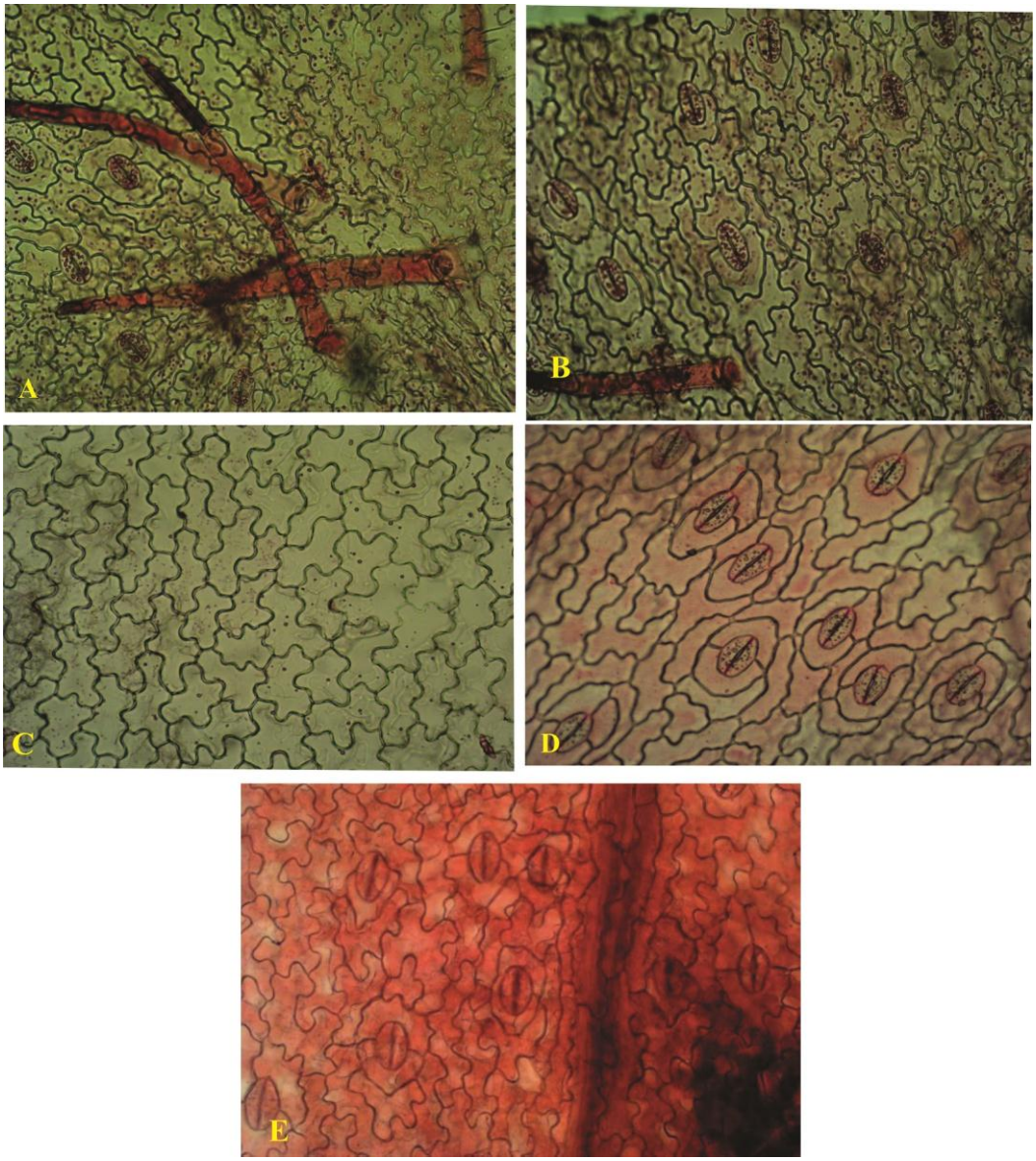

Fig. 1: (A) Multicellular trichome on *N. biserrata* abaxial epidermis,

(B) Unicellular trichome on *N. biserrata* abaxial epidermis,

(C) Anisocytic and Anomocytic stomata on *N. biserrata* abaxial epidermis,

(D) Diacytic stomata of *N. exaltata* abaxial surface

(E) Anisocytic stomata of *N undulata* abaxial surface

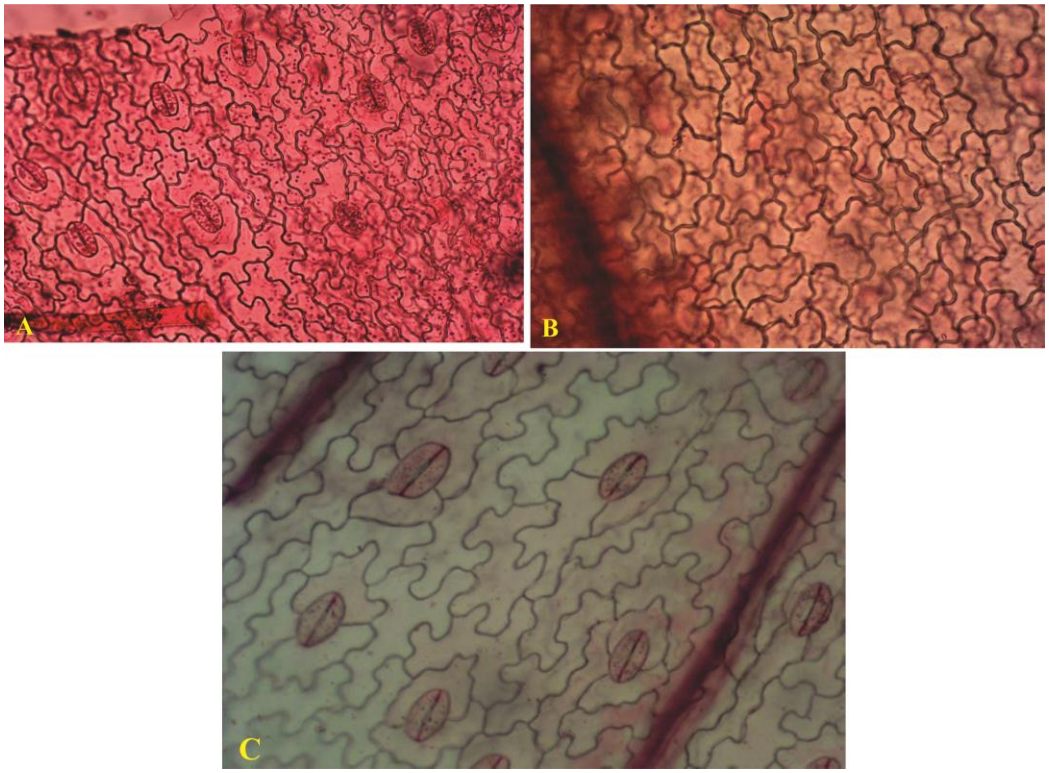

Fig. 2: (A) Undulating epidermal cellwall on *N. biserrata* adaxial surface

(B) Undulating epidermal cellwall of *N. exaltata* adaxial surface

(C) Anisocytic and Anomocytic stomata of *N. undulata* adaxial surface

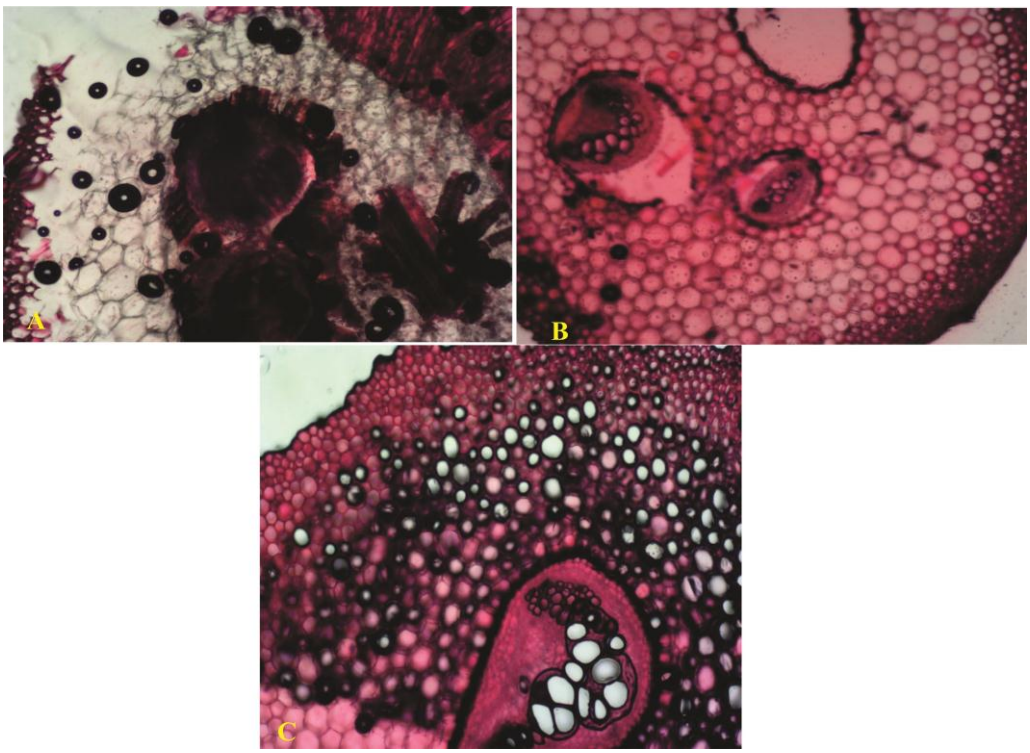

Fig. 3: (A) T. S. of petiole of *N. undulata*

(B) T. S. of petiole of *N. exaltata*

(C) T. S. of petiole of *N. biserrata*

DISCUSSION

Anatomical features are widely used systematically for identification and placing anomalous groups in satisfactory positions in classification of species. From the results (Tables 1 and 2), it has been observed that the species of *Nephrolepis* studied possessed stomata abundantly on the abaxial surfaces and they were absent on the adaxial surfaces apart from *N. undulata* that possessed stomata on both the abaxial and adaxial surfaces (Table 1) (Figures 1c - 1e and 2c). According to [4], the leaves of all the species are Hypoamphistomatic. The stomata types are diagnostic characters which may be associated with the plant habit.[10] The function of stomata is intimately and intricately associated with various physiological processes and the survival of each individual plant [10]. The undulating epidermal cell walls (Figures 1d and 2a) (Table 2) observed in the species indicates that they are forest plants occurring in shades as postulated by.[11] Shade plants tend to have more undulated walls since epidermal cell walls undulation is known to be affected by light.[12] There were similarities in measurement of length and width (Table 1) of stomata and this could be attributed to environmental factors and their degree of relatedness.

The presence of trichome in the abaxial surface of *N. biserrata* (Table 2) (Figure 1a and b) could be to reduce the rate of transpiration; This agrees with what[13] highlighted as importance of trichomes in the family *Combretaceae.*

In Figures 3A – 3C, the T.S. of petiole showed the presence of sclerenchyma followed by collenchyma. These tissues support the elongation of the petiole and its firmness in holding the frond as it provides tensile strength to the organs.[10] Vascular bundles were broken into meristeles. Phloem and xylem vessels were present. parenchyma cells were observed in the central region in which vascular tissues were embedded.

This study has revealed the presence or absence of phytochemicals in the three different plant species as summarized in Table 4. Saponnins were strongly present in all three plant species. According to[14], this implies that they all have cholesterol lowering properties and some deleterious properties (cytotoxic-permeabilizing of the intestine). Saponins have also been found to increase and accelerate the body's ability to absorb calcium and silicon.[15] Numerous studies have confirmed that saponnins possess the unique proerty of precipitating and coagulating red blood cells.[16,17] Cardiac glycosides were strongly present in the three plant samples. According to[18], Cardiac glycosides have been discovered to be effective remedies for treating heart diseases, and this is due to its ability to increase the blood preasure

which in turn increases the efficiency of the kidney. This implies that these ferns may be a panacea for individuals with heart diseases. All three plant species were void of alkaloids. Many alkaloids are known to be of medicinal value to man. Plants having alkaloids are used as medicines for reducing headache and fever and this is due to their antibacterial and analgesic properties.[19] Alkaloids are medicinal but they could be very toxic or poisinous when consumed above the required amount. Since these plant species are all lacking alkaloid, then they are safe for consumption as food with regards to alkaloid content. Flavonoids were absent in *N. biserrata*, but was strongly present in *N. exaltata* and *N. undulata*. This suggests that *N. exaltata* and *N. undulata* may have anti-allergic, anti-inflammatory, anti-microbial and anti-cancer properties and Flavonoids have been referred to as nature's response modifiers because of the strong experimental evidence of their ability to modify the body's reaction to allergies, viruses and carcinogens.[20] Tannins were strongly present in *N. exaltata* and *N. undulata* but it was not strongly present in *N. biserrata.* Tannins are attributed to analgesic and anti-inflammatory activities, they have amazing stringent properties and they are known to hasten the healing of wounds and inflammed mucous membranes.[21] Terpernoids were found to be absent in *N. biserrata*, but was strongly present in *N. exaltata* and *N. undulata*. Terpernoids have been found to be useful in the prevention and therapy of several diseases including cancer. They are also known to posses anti-microbial, anti-fungal, anti-parasitic, anti-viral, anti-allergenic, anti-pasmodic, anti-hyperglycemic, anti-inflammatory, and anti-immunomodulatory properties.[22] All three plant species were lacking phlobatannins. Phlobatannins have been reported to possess astringent properties.[23] They have also been discovered to posses wound healing properties, these are anti-inflammatory, analgesic and anti-oxidant.[24,25]

The strong presence of saponins and cardiac glycosides and the absence of alkaloids and phlobatannins in all the three ferns studied, shows that they are strongly related thus affirming their classification to the same genus. The strong presence of flavonoids, terpenoids and tannins in *N. exaltata* and *N. undulata* implies a closer relationship between them (*N. exaltata* and *N. undulata*) than with *N. biserrata*.

Four (4) different types of anti-nutrients were found in the three plant species as summarized in table 5. The hydrocyanide, phytic acid, tannin and oxalic acids concentration of the different plant species showed that *N. exaltata* has the highest concentration in all cases followed by *N. biserrata* and least in *N. undulata* respectively. The concentration of the

hydrocyanide in the plant samples were below the permissible level of 200 mg/ kg fresh weight of vegetables or forages.[26,27] The results thus suggest that the leaves of any of these plant samples are very safe for human consumption with respect to cyanide content. Phytic acid contributes to mineral deficiency in individuals whose diets rely on certain foods for their mineral intake and for people with low intake of essential minerals such as young children and those in developing countries.[28] Phytic acids also provides anti-oxidant effects and may prevent and cure colon cancer. The concentration of oxalic acid in *N. biserrata* (183.07 mg/100 g) and *N. undulata* (144.62 mg/100 g) was within the permissible level of 250 mg/100 g (Table 5). While the concentration of Oxalic acid in *N. exaltata* (265 mg/100 g) was above the permissible level of 250 mg/100 g with respect to oxalate content. According to[29], regular consumption of fresh raw samples of *N. exaltata* without proper processing could deliver toxic levels of oxalate into the body with attendant health problems of oxalate toxicosis. This can lead to hypocalcaemia, Kidney diseases (e.g, kidney stone), electrolytes imbalance and reduction of bioavailability of minerals in the body, and even death.[30,31,32,33] This finding, questions the safety of *N. exaltata* for human consumption with respect to oxalate content, though it is reduced during processing. Meanwhile, *N. biserrata* and *N. undulata* are safe for human consumption with regards to oxalate content, since they are within the acceptable level of 250 mg/100 g. The tannin content of the three plant samples were in low concentrations (*N. biserrata* = 26.07 mg/100 g, *N. exaltata* = 92.07 mg/100 g and *N. undulata* = 18.85 mg/100 g). plants that contains tannin are known to possess astringent properties. Tannins are water soluble phenolic compounds that chelate Iron (Fe) and Zinc (Zn) and thus limiting their absorption and they also interfare with digestion by exhibiting anti-trypsin and anti-amylase activity. They are not easily or completely destroyed by heat due to their high molecular weight but are preferred due to their anti-cancer activities.[34] In other words, these plants may be safe for human consumption.

The results for the Micromeritric studies as seen in Table 4 has shown that the bulk and tapped densities, Hausner's ratio, Compressibility index ie Carr's index, flow rate and the angle of repose obtained from the extracted powders all had fair flowing properties. The angle of repose is a property related to inter-particle friction of resistance to movement between particles.[35] Generally, if the value of the angle of repose exceeds 50^0, it indicates a non-satisfactory flow, while values that are near the minimum cica 25^0 flows well and easily.[36] Values that are less than or equal to 30^0 indicates a free flowing material. When it is between 20^0 and 30^0, it shows a good flowing property. 36^0 to 40^0 indicates a fair flow and

finally 41^0 to 45^0 indicates a passable flow. From the results (Table 2), it had been observed that all the three plant species (*N. biserrata*, *N. exaltata* and *N. undulata*) all had an angle of repose ranging from 36^0 to 40^0, and this suggests that they all have a fair flow. This character is indicative of the relatedness of the species.

CONCLUSION

In conclusion, Anatomical features such as stomatal types, stomatal ratio (L/W) and stomatal index and undulating epidermal cell wall justify the inclusion of the species in the same genus. Stomatal distribution, presence of trichome, thickness of epidermal cell walls and stomatal length, width and number justify their separation into different species. *N. undulata* and *N. exaltata* are more related morphologically and in terms of presence of secondary metabolites, *N. exaltata* is also different in having the higher values of anti-nutrients though they are present in all the three species. The results of the phytochemical screening show that *N. biserrata* and *N. undulata* may have potential uses as herbal products for pharmaceutical industries, however, the micromeritric studies show that all three species have fair flowing properties which enables good packaging of these potential herbal products.

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