

## Full Length Research Paper

# Pharmacognostic evaluation of the leaf of *Microtrichia perotitii* DC. (Asteraceae)

Abdullahi M. N.<sup>1\*</sup>, Ilyas, N.<sup>2</sup>, Hajara I.<sup>2</sup> and Kabir Y. M.<sup>2</sup><sup>1</sup>Department of Applied Science, College of Science and Technology, Kaduna Polytechnic, Kaduna, Nigeria.<sup>2</sup>Department of Pharmacognosy and Drug Development, Ahmadu Bello University, Zaria, Nigeria.

Received 25 January, 2018; Accepted 28 February, 2018

*Microtrichia perotitii* DC. belongs to the family Asteraceae (Compositae) and it is a herb found in the West African countries. The aim of this research was to establish pharmacognostic standards for *M. perotitii* through macroscopic, microscopic, chemo-microscopic and physico-chemical investigations. The macroscopic studies revealed the shape of the leaf as small with acute apex, asymmetric base, pubescent surface with long petiole and a serrated margin. Organoleptically, the leaf is green, slightly bitter and pepperish with an unpleasant odour. The microscopic studies of the leaf showed epidermal cells with irregularly thickened walls, numerous anomocytic stomata, and multicellular covering trichomes on both upper and lower epidermis. The transverse section of the leaf showed that it was dorsiventral with separated elements of vascular bundle. The powdered leaf revealed calcium oxalate crystals (prism and rosette), starch (oval) and xylem (spiral). Quantitative-leaf microscopy revealed the leaf constants as palisade ratio (3.2-3.4); stomatal number (258-285); stomatal index (19.5-24.7); vein-islet number (6.0-8.0) and vein-let termination number (8.0-11.0). The physico-chemical constants of the leaf showed moisture content (12.67%), total ash (20.33%), acid-insoluble ash (27.3%), water-soluble ash (81.18%), water extractive values (40.9%) and alcohol extractive values (25.45%). This is the first time the pharmacognostic parameters of the leaf of *M. perotitii* were studied and it will be quite useful for its identification, standardization and inclusion in various pharmacopoeias.

**Key words:** *Microtrichia perotitii*, macroscopical, microscopical, physico-chemical, pharmacognostic.

## INTRODUCTION

Standardization of herbal medicines (drug) is a process of establishing or prescribing a set of peculiar identities or specific characteristics which are generally unique and of unshared qualities. However, amongst various techniques used in identification of plant drugs, pharmacognostic study is always the most reliable. Basic component are standardization and authentication of natural drugs

(Chandaz, 2014). Plant used in traditional medicines are authenticated through morphological, physico-chemical and phytochemical analyses (Chandaz, 2014). One of the major challenges of identification of plant is adulteration which simply means replacing the original plant with another one with the intention of increasing either the weight or potency of the product

\*Corresponding author. E-mail: amnuhu2006@yahoo.com.

(Rokad et al., 2018). The therapeutic efficacy of any medicinal plant depends generally on the quality and quantity of the chemical constituents it contains (Majumder, 2011; Kumar and Rathinam, 2013). Information about the efficacy of medicinal plants that were passed from generations to generations after sometime could be incorrect, distorted, or even lost and under such circumstances proper scientific evidence must be established through pharmacognostic evaluation (Sumitra, 2014; Evans, 2009). Medicinal plants are the greatest assets to human health and a treasure for discovering new potential compounds with various therapeutic effects. The use of plant drugs locally is becoming increasingly popular especially in modern society because of incidences of adverse effects of synthetic drugs after they have been used. This is a common practice amongst developing countries most especially in Africa where it is their greatest hope (Gireesh et al., 2015; Elujoba et al., 2005).

*M. perotitii* DC. belongs to the family Asteraceae and is widely distributed in West African countries of Nigeria, Senegal, Mali, Port of Guinea, Sierra Leone, Ivory Coast, Ghana and Dahomey (Hutchinson and Dalziel, 1963). In Nigeria it is found in northern part of the country where it is known as *Maijankai* or *Sawun keke* in Hausa, *Osete* in Igbira and *Shaware pepe* in Yoruba. The herb is an annual plant and is a diffused much branched pubescent varying from 1ft in height. The leaves are obovate, obtuse, cuneately narrowed into petiole, coarsely toothed above  $\frac{1}{2}$  -  $1\frac{1}{2}$  inch long,  $\frac{1}{3}$  - 1 inch broad. The petiole of lower leaves is 1inch or more at the upper. Other features are in accordance to some members of the family (Daniel, 1877; Andrews, 1954; Watson and Dallwitz, 1992). Traditionally, *M. perotitii* is used for treating pain related diseases such as toothaches, cuts and burns and rashes in children. Others include, skin disease, rheumatism, diarrhea and jaundice. Earlier phytochemical studies of the leaves revealed the presence of tannins, flavonoids, alkaloids, carbohydrates, cardiac glycosides, saponins, phenolics and terpenoids (Abdullahi et al., 2011). In spite of these medicinal uses of this herb, there are no pharmacognostic reports on the leaves' constants and other parameters as such this study is aimed at providing pharmacognostic standards of the leaves for the first time.

## MATERIALS AND METHODS

### Collection and preparation of the herb

The herb *M. perotitii* was collected from Rigasa village, Kaduna State, Nigeria in the month of April 2016. The fresh herb was compared with specimen authenticated by Mal. Musa M. of the herbarium unit, Department of Biological sciences, Ahmadu Bello University, Zaria and was given a voucher number 998 for future references. The fresh herb was allowed to dry under the shade for three weeks until all the leaves have fallen-off the stalk. They were

reduced to coarse powder with traditional pestle and mortar. Both fresh and the coarse powder were used for the studies.

### Macroscopic study

The fresh leaves of *M. perotitii* were subjected to morphological studies in order to evaluate appearance such as surface, dimensions, point of attachment, lamina including composition, inclusions, shape, base, venation, margin and apex as well sensory profile that is colour, odour, texture and taste. The methods described by Brain and Turner (1975), WHO (2007 and Evans (2009) were adopted.

### Microscopic study

Both fresh and powdered leaves were studied:

**Qualitative examination:** For this study, the surface of the leaf of *M. perotitii* was prepared by peeling –up the upper and lower epidermis with sterilized forceps while the transverse section was prepared with a sharp razor blade. Each of the preparations was cleared of opaque materials by boiling them in 70% chloral hydrate solution. Small quantity of the powdered leaves was also prepared by clearing them in 70% chloral hydrate solution after being boiled in a test-tube on a Bunsen flame. Epidermal and other anatomical features were observed from the surfaces and transverse section of the leaf. The standards methods of Brain and Turner (1975), WHO (1984, 2007) and Evans (2009) were adopted.

**Chemo-microscopic examination:** For this study, the powdered leaves of *M. perotitii* were first cleared of obscured materials by boiling them in 70% chloral hydrate solution. Fragments of the leaves were transferred onto clean microscopic slide with dilute glycerol to prevent dehydration or hardening. Detecting reagents for metabolites such as starch, lignins, cellulose, calcium oxalate crystals, fixed oils and fat, calcium carbonates, mucilages and tannins were applied for their presence in cells inclusions or cell wall. These examinations were carried out according to the methods described by Brain and Turner (1975), WHO (2007) and Evans (2009).

**Quantitative leaf examination:** For this study, fresh leaf of *M. perotitii* was prepared by peeling–off both the upper and lower epidermis and clearing them by in boiling 70% chloral hydrate solution. The cleared leaf was mounted on clean microscopic slide and covered with cover slip. A set-up camera Lucida at x10 objective (1 small stage micrometer division = 10  $\mu$ m; calibration factor = 2.7) was set for the determination of stomatal number, stomatal index, vein-islet number, veinlet termination number and palisade ratio. The length and width of stomata and trichomes were measured by calibrating the eyepiece micrometer using stage micrometer as described by Evans (2009).

**Physical constant examination:** For this study, various physico-chemical parameters were determined for the powdered leaves of *M. perotitii* and these included; moisture content, total ash, acid-insoluble ash, water soluble ash, water soluble extractive values and alcohol extractive values. The evaluation was carried out according to the methods described (WHO, 1998; Evans, 2009; Pratima and Mathad, 2011).

### Statistical analysis

The data obtained were expressed as mean  $\pm$  standard error of

**Table 1.** Macroscopic and organoleptic characters of the leaf of *Microtrichia perotitii*.

Features	Descriptions
Shape	Small
Dimension (length x breadth)	(3.6- 3.7 mm) × (1.6 -2.1 mm)
Arrangement	Alternate
Petiole	Long
<b>Lamina</b>	
I. Composition	Simple
li .venation	Pinnate
lii. Margin	Serrated
Iv. Apex	Acute
V. Base	Unequal
Vi. Texture	Brittle
Vii. Surface	Pubescent
<b>Organoleptic properties:</b>	
I. Colour	Greenish
li. Odour	Unpleasant
lii. Tastes	Slightly bitter and pepperish

mean (SEM), and n represents the number of replicates in a particular experiment.

## RESULTS

### Macroscopic examination

Macroscopically, the leaf of *M. perotitii* was observed to be small with pubescent surface and alternately arranged along the smooth and soft stalk. Its apex is cute, the base is unequal and the margin is serrated while the venation is pinnate. The dimension of the leaf was 3.6 to 3.7 mm in length and 1.6 to 2.1 in breadth. The leaf when it is fresh is always greenish with unpleasant odour while its taste is slightly bitter and pepperish (stringent) (Figure 1). The summary of the macroscopic and organoleptic observations are given in Table 1.

### Microscopic examination

The result of microscopic evaluation of the leaf of *M. perotitii* showed typical characteristics of leaves in terms of taxonomic importance that is the presence of lower and upper epidermis, xylem, phloem, vascular bundles, mesophyll, collenchyma and trichomes. The observed shapes of the cells showed that they appeared elongated at the lower epidermis while they are diagonal at the upper epidermis. The anticlinal walls are wavy at the lower epidermis while they are straight at upper epidermis.

The stomata were anomocytic type and were more

frequent at the lower epidermis. Uniseriate multicellular trichomes were identified at both the upper and lower surfaces but were rare at the upper surface (Figures 2 to 6). A summary of the features are summarized in Table 2. From the powdered leaves, fragments of spiral types of xylem vessels, calcium oxalate crystals and starch grains were identified (Figure 6).

### Chemo-microscopic evaluation

The chemo-microscopic evaluation of the powdered leaf of *M. perotitii* revealed the presence of cellulose cell wall, lignified cell wall, calcium oxalate crystals, tannins and fat and oils (Table 3).

### Quantitative evaluation

The result of quantitative evaluation of the leaf of *M. perotitii* is presented in Table 4.

### Physico-chemical examination

The result of the physical constant evaluation is presented in Table 5. The value of moisture content that is loss on drying has indicated the drug cannot easily be deteriorated by fungal or microbial attacks.

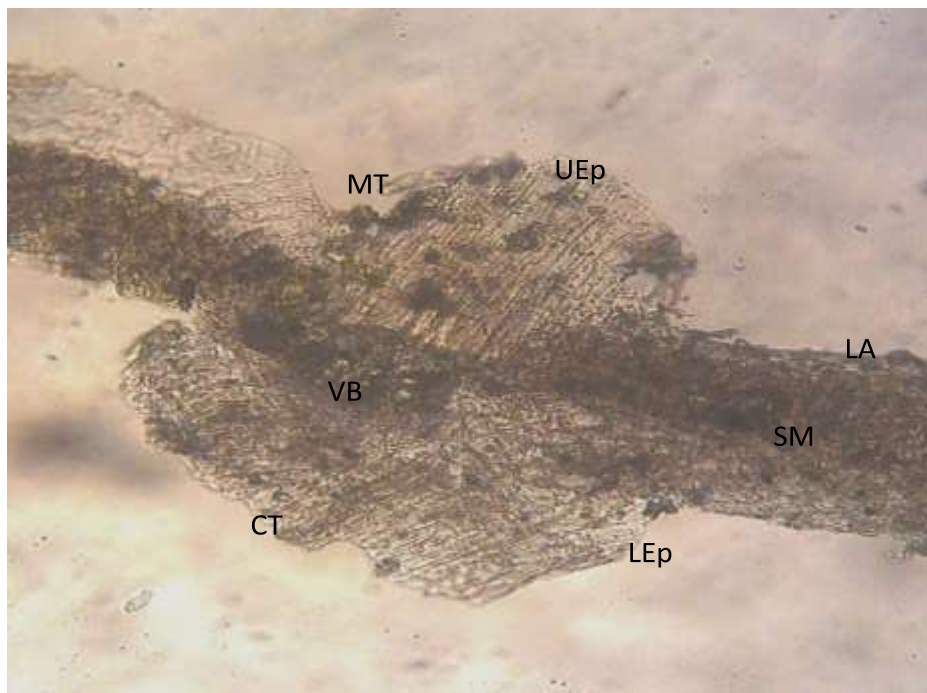
## DISCUSSION

The pharmacognostic analysis of *M. perotitii* carried-out will help in establishing its botanical identity. The standardization of the herbal medicines is necessary to assure the quality of the drug and will also help in checking and preventing substitution and adulteration of foreign material, that is by mixing or substituting the original drug material with other spurious, substandard, defective, spoiled, useless other parts of the same or different plants (Rokad et al., 2018). It is a common knowledge that plant drugs are used locally for treatment of various disease conditions without recourse to standardization in order to establish the correct identity of the drug (Periyanayagam and Karthikeyi, 2013).

The macroscopic evaluation is a technique of qualitative evaluation based on the study of morphological and sensory profiles of drugs. Therefore, the macroscopic characters of *M. perotitii* studied can serve as diagnostic parameters especially its organoleptic characteristics (Singh et al., 2010; Sathis et al., 2011). The organoleptic characteristics of the leaf were unique being bitter and pepperish with unpleasant odour. Microscopic evaluation is one of the simplest and cheapest methods to establish the correct and accurate identity for a plant drug (Patel and Zaveri, 2011). The microscopic evaluation of the leaf



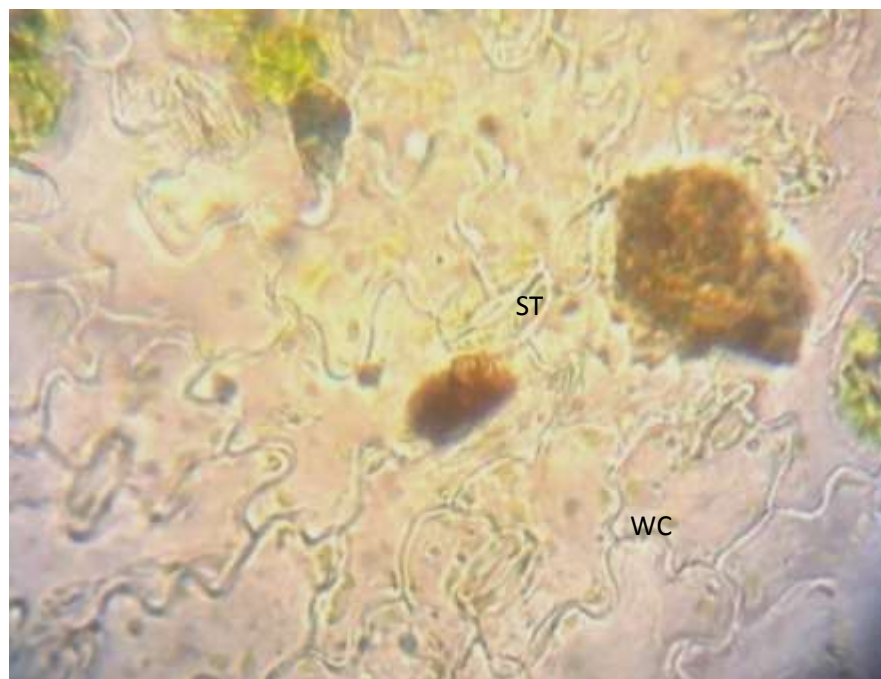
**Figure 1.** Leaf morphology of *Microtrichia perotitii* ×100.



**Figure 2.** Photomicrograph of the transverse section of the midrib of the leaf of *Microtrichia perotitii* showing multicellular trichomes (MT), upper epidermi (UEp), covering trichomes (CT), spongy mesophyll (SM), lower epidermis (LEp), lamina (LA) and vascular bundles (VB)×100.



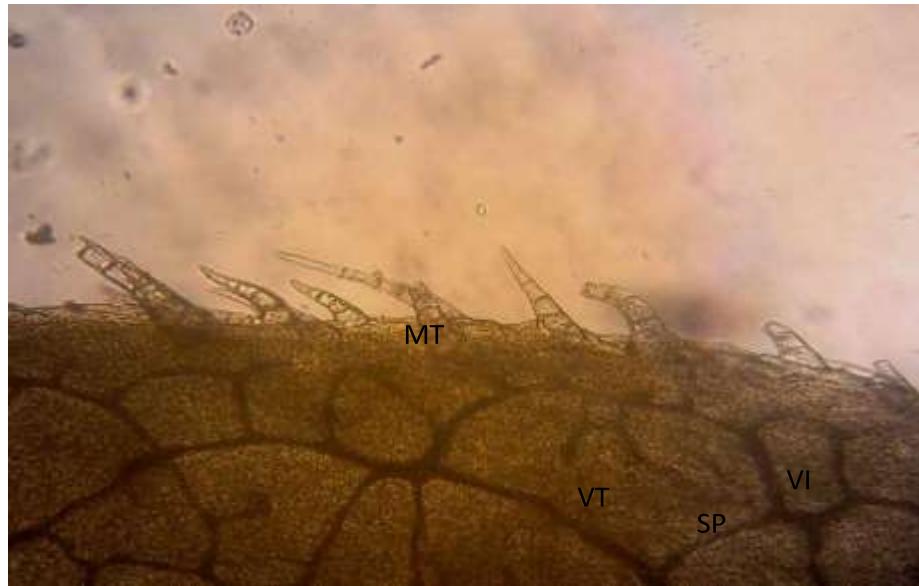
**Figure 3.** Photomicrograph of the surface preparation of upper epidermis of the leaf of *Microtrichia perotitii* showing anomocytic stomata (ST)  $\times 100$ .



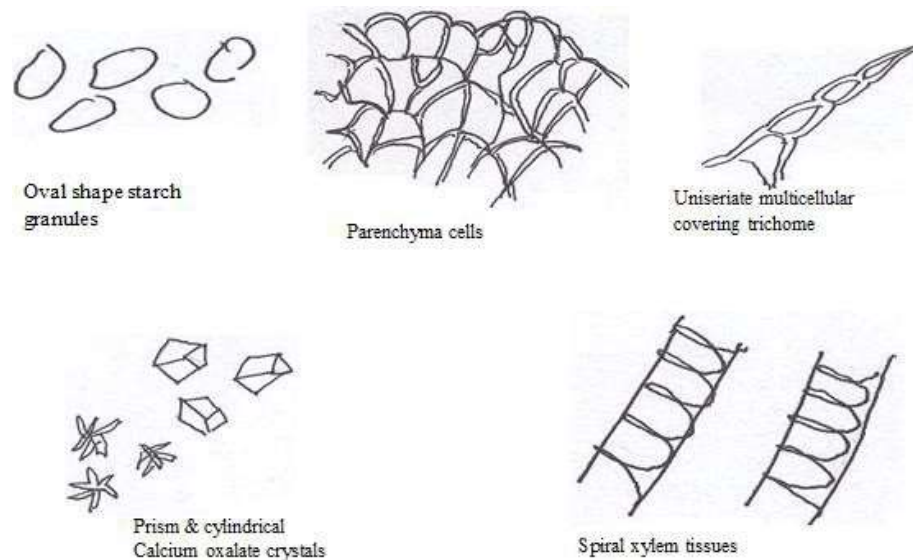
**Figure 4.** Photomicrograph of the surface preparation of lower epidermis of the leaf of *Microtrichia perotitii* showing anomocytic stomata (ST) and wavy anticlinal cell wall (WC)  $\times 100$ .

of *M. perotitii* showed that the leaf has anomocytic stomata occurring at both adaxial and abaxial epidermis thus indicating efficient gaseous exchange for photosynthesis and loss of water (Shaukat et al., 2010).

Multicellular trichomes also appeared on both surfaces of the leaves although not so frequent at the upper surface. The presence and types of trichomes are useful diagnostic features and they contribute to



**Figure 5.** Photomicrograph of lower epidermis of the leaves of *Microtrichia perottii* showing multicellular trichomes (MT), veinlet (VI), vein termination (VT) and spongy mesophyll (SP) x 100.



**Figure 6.** Some identified microscopic diagnostic features from the powdered leaves of *Microtrichia perottii* x 400.

plant resistance against herbivory and reduction of the rate of transpiration in the plants (Ahlam and Bouran, 2011; Priyanka et al., 2011a, b). The transverse section of the leaf indicated that it is dorsiventral and showed the appearance of a typical leaf structure. Calcium oxalate crystals were observed and they may be involved in the dispersing of light to the chloroplasts in the photosynthetic parenchyma cells of the leaves. The crystals were thought to have a physiological role

sequestering excess calcium within plant cells (Popescu et al., 2010). Starch granules were observed and appeared in oval shape. Although the starch is the main form in which plants store carbon, they are sometimes converted into sugar by amyloplasts when the plant needs energy (Alison, 2010). Chemo- microscopic evaluation of the leaf showed proteins, starch, cellulose and tannins including calcium oxalate crystals and lignins which are indications of the presence of

**Table 2.** Epidermal characters and their descriptions.

Features	Characters	
	Lower epidermis	Upper epidermis
i. <b>Cells:</b> Shapes	Elongated	Polygonal
Anticlinal wall	Wavy	Straight
Thickening	Smooth and cellulose	Smooth and cellulose
Papillae	Absent	Absent
Cuticle	Present	Present
ii. <b>Stomata:</b> Type	Anomocytic	Anomocytic
Frequency	Numerous	Frequent
Size (l,b,µm)	27×16.2	27×13.5
iii. <b>Trichomes:</b> Type	Uniseriate multicellular	Uniseriate multicellular
Frequency	Frequent	Rare
Size(h,bµm)	135×21.6	132×21.6

l = length; base= base; h = height.

**Table 3.** Chemo-microscopy result of the leaf of *Microtrichia perotitii*.

Constituents	Mounting reagent	Result/observation	Inference
Starch	Iodine solution	Blue-black colouration appeared on some grains within the chloroplasts and powdered leaves	Present
Lignin	Phloroglucinol and conc. HCl	No red colouration observed on the walls of some lignified collenchymas.	Present
Cellulose	Chloro-zinc iodine solution	Blue colouration observed on walls of epidermal cells	Present
Calciumoxalate	Chloralhydrate solution	brightly coloured crystals observed which later dissolved in conc. HCl and disappeared in the collenchymas cells	Present
Oils and fats /warming	Sudan iv solution	Reddish colouration was observed in some parenchyma cells and was distinct	Present
Mucilages	Ruthenium red solution	Dark solution observed at epidermis and vascular tissues	Absent
Tannins	5% ferric chloride solution	Greenish black colouration was observed in some parenchyma cells	Present

**Table 4.** Quantitative leaf microscopic features of *Microtrichia perotitii*.

S/N	Features	Characteristics
i.	Palisade ratio	3.2 -3.3* - 3.4
ii.	Stomata number	258-271.5* -285(adaxial epidermis) 289-299.5*-310( abaxial epidermis)
iii.	Stomatal index (%)	19.5- 22.1*-24.7 (adaxial epidermis) 29.3 -31.6*- 34.5 (abaxial epidermis)
iv.	Vein-islet number	6.0 - 7.2* - 8.0
v.	Veinlet termination number	8.0- 9.6* -11.0

\*n = 5.

alkaloids, flavonoids and glycosides in the leaves (Prabhu et al., 2009).

Quantitative microscopic evaluation of the leaf of *M. perotitii* has provided values for palisade ratio, stomatal number, epidermal layer, stomatal indices, vein islet number and veinlet termination number. These

information set genuity and standard for the herb as well as distinguishing it from co-generic species that may be closely related and cannot easily be characterized by general microscopy (Veeranjaneyulu and Rama, 1984).

The physico-chemical evaluation of the leaves of *M.*

**Table 5.** Physicochemical constants from the leaves powder of *Microtrichia perotitii*.

Parameter	Mean values (%)
Moisture content	12.67±0.62
Total Ash	20.33±0.20
Acid-insoluble Ash	2.73±0.13
Water-soluble Ash	8.18±0.30
Water-soluble extractives	40.9±1.73
Alcohol- soluble extractives	25.45±1.24

n = 3.

*perotitii* could serve as a significant role in standardization and quality control by means of purity, stability and phytochemical composition of the herb (Bharat and Parabia, 2010). The moisture content for *M. perotitii* calculated by loss was 12.67% which is less than 14% standard requirement for crude drugs. This is an indication that the powder of this herb can be stored for a longer period of time without spoilage (Ahmad et al., 2012; Kadam et al., 2012; Mubo et al., 2014). The total ash value for *M. perotitii* lies within fair limits and thus signified its quality and purity and gives idea about the total inorganic content in it (Ugur and Selima, 2011; Woratouch et al., 2011). The acid- insoluble ash value of 2.73±0.13% obtained for the leaves is an indication that the herb was in good physiological condition thus contained less extraneous matter in the form of contamination with silicious materials (earth and sand) and provides information about non-physiological ash produced due to adherence of inorganic dirt and dust to the crude drug. The water-soluble ash was 8.18±0.30% and this parameter is used to detect the presence of materials exhausted by water (Schoffstall, 2000; Singh and Sharma, 2010; Adedapo et al., 2011; Pratima and Pratima, 2011; Ahamad et al., 2012; Kamalakannan et al., 2012 ; Kunle et al., 2012; Veena and Pracheta, 2013; Sangram et al., 2015).

Extractive values of plants give useful estimation of chemical constituents in the drug as well as a measure of the stability of phytocompounds in the plant drug in a given solvent. The value for the water soluble extractive for the leaves of *M. perotitii* were much higher than that of the alcohol thus indicating higher polarity and more penetration of the water molecules into the cellular membrane in order to extract all inter-cellular components of the plant material. The leaves therefore, exhibited more amount of water soluble components compared to alcohol extract and may provide estimation of specific constituents soluble in a particular solvent (Periyanyagam et al., 2013; Vipin et al., 2015).

All the results obtained for the pharmacognostic evaluation of the leaves of *M. perotitii* were compared to those of some members of Asteraceae in order to maintain a standard since this is the first report about the leaves in the literature.

## Conclusion

The pharmacognostic study on the leaves of *M. perotitii* is being reported for the first time in literature. The findings will be useful in the identification of the herb, detection of adulterants and its monograph. It will also provide avenue for further studies on microscopic evaluation of the leaves.

## CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

## ACKNOWLEDGEMENT

The authors wish to acknowledge the contributions and support of our various institutions for providing the enabling environment to carry out this research. Need to be mentioned are all the technical staff of our research laboratories.

## REFERENCES

- Abdullahi M N, Ibrahim H, Ilyas N (2013). Evaluation of Phytochemical Screening and Analgesic Activity of Aqueous Extract of The Leaves of *Microtrichia perotitii* DC (Asteraceae) in Mice using Hotplate Method. *Med. Plant Res.* 3:537-543.
- Abdullahi MN, Ibrahim H, Ilyas N (2011). Phytochemical Screening and Biological Studies of the Leaves of *Microtrichia perotitii* DC (Asteraceae). *Br. J. Med. Plants* 1(3):88-97.
- Adedapo A, Jimoh F, Afolayan A (2011). Comparison of the nutritive value and Biological activities of the Acetone, Methanol and water extracts of the Leaves of *Bidens pilosa* and *chenopodium album*. *Acta polon Pharmaceu. Drug Res.* 68(1):83-92.
- Ahlam SE, Bouran IA (2011). Microscopical studies on the Leaf and Petiole of *Vernonia amygdalina* Del. *Adv. Appl. Sci. Res.* 2(2):398-406.
- Ahmad J, Showkat M, Naquri KJ (2012). Preliminary Pharmacognostical Standardisation of Aerial parts of *Artemisia absinthium* L. *Int. Res. J. Pharm.* 3(1):217-220.
- Alison MS (2010). Starch and Starch granules. John Innes Centre, Norwich, UK. eLS. Wiley online Library. John Wiley and Sons.
- Andrews FW (1954). The Flowering Plants of the Sudan, Vol.3 Compositae-Graminae. Published for the Sudan Govt. by T. Bande and co Ltd. Arbroath. Scotland. pp. 1-63.
- Bharat G, Parabia MH (2010). Pharmacognostic evaluation of the bark



- and seeds of *Mimusop selengi* L. Int. J. Pharm. Pharmaceut. Sci. 2:110-113.
- Chandaz S (2014). Importance of pharmacognostic study of medicinal plants. An overview. J. Pharmcog. Phytochem. 2(5):69-73.
- Daniel O (1877). Flora of Tropical Africa. Vol III Umbelliferae to Ebenaceae. Published under the Authority of the first commissioner of Her Majesty's works. By L. Reeve and Co., 5 Henrietta Street, Covent. Garden London. pp. 302-303.
- Elujoba AA, Odeleye OM, Ogunyemi CM (2005). Traditional medicinal development for medical and dental healthcare delivery system in Africa. Afr. J. Trad. Med. 2(1):46-61.
- Evans WC (2009). Trease and Evans. Pharmacognosy. 16<sup>th</sup> edition. W. B. Saunders. Toronto Harcourt Pub. Ltd <https://www.elsevier.com/books/trease-and-evans-pharmacognosy/evans/978-0-7020-2933-2>
- Gireesh MA, Sandeep RP, Vinayak U, Pramod JH, Harsha VH (2015). Pharmacognostic evaluation of *Achyranthes coynei*: Leaf. Egyp. J. Basic Apld. Sci. 2:25-31.
- Hutchinson J, Dalzie JM (1963). Flora of West Tropical Africa, 2<sup>nd</sup> edn Vol 12. Crown Agent for overseas Government and Administration Millbank, London SW 1:325-397.
- Kadam PV, Vadav KN, Navasare VS, Bhilwade SK, Patil MJ (2012). *Eclipta alba*: A Phytopharmacognostic study. Int. J. Pharm. Phytopharmacol. Res. 1(6):350-353.
- Kamalakkannan P, Kavitha R, Elamathi R, Deepa T (2012). Sridhar, S. Study of Phytochemical and Antimicrobial potential of Methanol and Aqueous extracts of Aerial Parts of *Elephantopus scaber* L. Int. J. Cur. Pharmaceut. Res. 4(1):18-21.
- Kunle OA, Olujemisi F, Egharevba HO, Ahmadu PO (2012). Standardization of Herbal Medicines: A review. Int. J. Biodiv. Cons. 4(3):101-112.
- Majumder P (2011). Evaluation of pharmacognostic, phytochemical profile along with quantitative physicochemical standards on the root of medicinal herb *Peporemia pellucid.* (L) HBK. J. Pharmaceut. Biomed. Sci. 10(10):1-3.
- Mubo AS, Tolulope AO, Mike OS (2014). A Pharmacobotanical Study of two Medicinal species of Fabaceae. Asian Pac. J. Trop. Biomed. 4(2):131-136.
- Patel S, Zaveri M (2011). Pharmacognostic study of the Roots of *Justica gendarussa* Burm. J. Trad. Med. 6(2):61-72.
- Periyanyagam K, Gopalakrishnan S, Karthikeyan V (2013). Evaluation of Pharmacognostical and Phytochemical properties of the Leaves of *Psidium guajava* Linn- Chittidhar variety. Innov. J. Health Sci. 1(3):10-13.
- Periyanyagam K, Karthikeyan V (2013). Pharmacognostical, SEM and XRF profile of the leaves of *Artocarpus heterophyllus* L. (Moraceae) - A contribution to combat the NTD. Innov. J. Life Sci. 1(1):23-28.
- Popescu ML, Mihaela D, Diana DU (2010). Contributions to the Pharmacognostical and Phytobiological study on *Taxacum officinale* (L) weber. Pharmacia. 58(5):646-653.
- Prabhu K, Karar PK, Ponudural K, Hemalatha S (2009). Pharmacognostic investigation of the leaves and stem of *viburnum erubescens* wall D.C. Trop. J. Pharmaceut. Res. 6:557-566.
- Pratima H, Pratima M (2011). Pharmacognostic Evaluation and Phytochemical Analysis of Leaves of *Cajanus cajan* L. J. Adv. Dev. Res. 2(2):181-185.
- Priyanka Y, Deepak S, Nayaka S (2011). Microscopic studies of *Tridex procumbens* L. Int. J. Biomed. Res. 2:508-517.
- Kumar AR, Rathinam KS (2013). Pharmacognostical Studies on *Artemisia annual.* Int. J. Res. Pharm. Biotechnol. 1(1):64-67.
- Rokad N, Pande J, Chanda S (2018). Pharmacognostic and phytochemical studies of *Ipomoea pes-caprae*, An halophyte from Gujarat. J. Pharmcog. Phytochem. 7(1):11-18.
- Sangram KP, Debajyoti D, Chandan D (2015). Phytochemical Screening, Pharmacognostic study and Physicochemical Evaluation of Leaf of *Crotalaria pallida* Aiton Der Pharmacia Let. 7(10):239-247.
- Sathis KD, David B, Prashanthi G, Harani A (2011). Pharmacognostical Evaluation Study on *Crotalaria jumcea* Linn. Amer.-Euras. J. Sci. Res. 6(3):139-145.
- Schoffstall AM, Gaddis BA, Druelinger ML (2000). Microscale and Miniscale Organic Chemistry Laboratory Experiments. McGraw Hill companies USA pp. 86-103.
- Shaukat M, Huma S, Manyam A, Shahnaz G, Ghazala HR (2010). Parmacognostic Studies on Fresh Mature Leaves of *Holoptetea integrifolia* (ROXB) Plarich. Pax. J. Bot. 42(6):3705-3708.
- Singh A, Saharan AV, Kumawat IC, Khatri A, Bhandari A (2014). A Pharmacognostical Study of *Vernonia cinerea* Less (Asteraceae) and Evaluation of Anti-inflammatory and Antibacterial activities of Stem. Egyp. Pharmaceut. J. 13(2):104-112.
- Singh S, Machawal L, Chauhan MG (2010). Pharmacognostic study of male leaves of *Trichosanthes dioica*.Roxb with special emphasis on microscopic technique. J. Pharmacog. Phytothera. 2(5):71-75.
- Sumitra C (2014). Importance of pharmacognostic study of medicinal plants: An overview. J. Pharmcog. Phytochem. 2(5):69-73.
- Ugur C, Selima K (2013). Nitrate, Moisture and Ash Contents of Edible wild plants. J. Cell. Plant Sci. 2011; 2(1):1-5.
- Veena S, Pracheta S (2013). Microscopic Studies and Preliminary Pharmacognostical Evaluation of *Euphorbia neriiifolia* L. Leaves. Ind. J. Nat. Prod. Res. 4(4):348-357.
- Vipin CP, Bhuwanendra S, Arshad A (2015). Pharmacognostic Evaluation of Leaves and Stem of *Murraya koenigii*. UK. J. Pharmaceut. Biosci. 3(1):37-41.
- Veeranjaneyulu K, Rama VSD (1984). Stomatal frequency and Resistance of some Tropical members of Asteraceae. Proc. Indian Natl. Sci. Acad. 50(3):317-320.
- Watson L, Dallwitz M (1992). The Families of Flowering Plants: Description, Illustration, Identification and Information 1992; Accessed 18<sup>th</sup> August, 2008. Available <http://www.biodiversity.uno.edu/delta/>
- Woratouch T, Thatree P, Suchada S (2011). Pharmacognostic Evaluation of Lagerstroemia Species leaves. J. Med. Plant Res. 5(8):1330-1337.
- World Health Organisation (1998). Quality control Methods for Medicinal Plants Materials. England. pp. 10-21, 22-45.
- World Health Organisation (2007). Guidelines for assessing quality of herbal medicines with reference to contaminants and residues. England. pp. 210-215.