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Pharmacognostic Studies on *Pulicaria crispa* (Asteraceae)

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ABSTRACT
Pulicaria crispa (Forssk.) Oliv. is a member of the family Asteraceae, an aromatic herb native to arid and semi-arid regions of Africa and Asia, traditionally used in the management of Inflammation and infectious conditions. However, despite its relevance, there is limited pharmacognostic information available to support its proper identification and quality control. This lack of standardized data may contribute to misidentification and unsafe use of the plant in Nigeria. Establishing baseline pharmacognostic characteristics is therefore important for quality assurance and proper plant authentication. The procedure involved the determination of physicochemical parameters, microscopic and micromorphometric examination of the leaf, stem, and root using standard microscopic techniques, as well as preliminary phytochemical screening of the plant extracts employing established analytical methods. Macroscopic examination showed leaves that are lanceolate to oblong in shape, with reticulate venation, serrated margins, a pubescent lamina surface, and an apex ranging from obtuse to acute. Microscopic analysis of the leaf epidermis revealed undulating polygonal epidermal cells, amphistomatic leaves bearing anomocytic stomata, and the presence of trichomes on both the abaxial and adaxial surfaces. Anatomical transverse sections demonstrated an isobilateral leaf with a collateral vascular system, a stem differentiated into epidermis, cortex, and stele, and a root composed of an epiblema, cortex, and vascular cylinder. The alcohol- and water-soluble extractive values were 6% and 16%, respectively, while the moisture content, total ash, acid-insoluble ash, and water-soluble ash values were 8.7%, 20%, 15%, and 1.5%, respectively. Thin-layer chromatographic analysis indicated the presence of phenolic compounds and terpenoids**.** The findings establish diagnostic pharmacognostic parameters for *Pulicaria crispa*, supporting its proper identification and quality control.

Keywords: Pharmacognostic, Phytochemicals, *Pulicaria crispa,* Histoanatomy, leaves, roots, and stems

INTRODUCTION
Pulicaria crispa is a member of the family Asteraceae, comprising approximately 80–100 species widely distributed across Europe, North Africa, and large parts of Asia, with notable diversity in Mediterranean and arid regions. In West Africa, *P. crispa* extends into Nigeria, where it has been reported in the savannah and semi-arid zones,

particularly in parts of Kaduna, Katsina, Kano, Jigawa, and Borno States, environments that support xerophytic and aromatic herbs (POWO, 2023). It is a is a perennial herb characterized by decumbent branches and a strong ecological association with Sahelian landscapes. Its natural distribution extends from Mauritania through northern Nigeria and eastwards across Africa to the Arabian Peninsula, where it is commonly encountered in savannah and semi-desert zones.

In Nigerian traditional medicine, *Pulicaria crispa* is used for the management of infections, fever, inflammatory conditions, gastrointestinal disorders, and respiratory complaints. Its ethnomedicinal relevance is well recognized among local communities that rely on herbal remedies for primary healthcare. Species of the genus are predominantly herbs adapted to dry and semi-arid environments and are valued for their ethnomedicinal relevance (POWO, 2023).

Pulicaria crispa; It is an aromatic herb, usually annual to perennial, characterized by erect stems, pubescent lanceolate leaves, and yellow capitulate inflorescences, features that favour survival in dry and semi-arid environments.

P. crispa is used in Nigeria in folk medicine as an antimalarial, galactagogue, and also rubbed in the temple for headache, among other uses (Kankara *et al*., 2022). Despite scientific research into the phytoconstituents and pharmacological activities of this plant, no literature is available on the pharmacognostic characters of the plant, which are essential for identification, authentication, and the setting of standards for quality control, being the first step in medicinal plant studies. As a result, this work was carried out to study the diagnostic characters of the plant by applying microscopic techniques as well as thin layer chromatographic profile to present the chemical entity of the plant. The information obtained can be used in the preparation of a monograph for the identification of the plant.

Scientific studies have demonstrated that *Pulicaria crispa* is a rich source of phenolic compounds and terpenoids. Phytochemical investigations have reported the occurrence of several sesquiterpene lactones, including xantholide, pseudo-guainolide, and asecosesquiterpene derivatives, in addition to flavonoids such as quercetin, quercetin-3-O-glucoside, quercetin-7-O-glucoside, and quercetin-3-methyl ether. Further analyses of *P. crispa* specimens collected from Saudi Arabia revealed the presence of β-sitosterol, β-amyrin, choline, quercetin, and an unidentified triterpene, highlighting the chemical diversity of the species (Stavri *et al*., 2008; El-Kamali *et al*., 2010). Assaf and Ali (1996) reported myrcene, limonene, terpinene, elemene, carvone, sabinol, and d- cardinene from the oil of *P. crispa*. Scientific investigations into the biological activities of extracts of *P. crispa* proved numerous activities, such as anti-inflammatory and antileukemic activities (Al Yahya *et al.,* 1984). The plant also possessed cancer chemopreventive and cytotoxic agents (AlMotwaa and Al-Otaibi, 2022) and immunomodulatory properties (Maghraby *et al*., 2010; Abo-Elghiet *et al*., 2023).

MATERIALS AND METHODS

Plant Collection, Identification, and Preparation

The whole plant of *P. crispa* (including leaves, fruits, stems, and roots) was collected in March 2024. Preliminary field identification was carried out using standard botanical

and taxonomic criteria in line with contemporary plant classification systems. The plant identity was subsequently authenticated by comparison with a reference herbarium specimen (Voucher No. ABU094) at the Herbarium of the Department of Biological Sciences, Ahmadu Bello University, Zaria, and further confirmed by a qualified plant taxonomist, Dr Namadi Sunusi. A voucher specimen was prepared and deposited in the Department of Pharmacognosy and Drug Development for future reference.

Fresh samples of the leaves, stems, and roots collected from the same source were immediately fixed in formalin–acetic acid–alcohol (FAA) solution to preserve cellular integrity and prevent enzymatic degradation. This fixation method ensures optimal preservation of tissue architecture, including epidermal layers, mesophyll, vascular tissues, and supporting structures, thereby facilitating accurate anatomical studies (Suvarna, 2019).

The remaining plant material was air-dried under shade at ambient temperature to minimize the degradation of thermolabile and photosensitive constituents. The dried material was pulverized into a fine, uniform powder using a mechanical grinder. The powdered sample was stored in clean, dry, airtight containers, properly labeled, and kept under appropriate storage conditions until required for further pharmacognostic and phytochemical analyses, in accordance with World Health Organization guidelines (WHO, 2018).

Macroscopic Evaluation of *Pulicaria crispa*

Macroscopic and organoleptic characteristics of the whole plant parts were evaluated based on parameters such as colour, odour, taste, texture, and morphological features. The assessment was conducted in accordance with established WHO guidelines for the identification and quality control of medicinal plant materials (WHO, 2025).

Microscopic Evaluation *Pulicaria crispa*

Powder Microscopy

Microscopic examination of the powdered whole plant material was performed to identify diagnostic cellular features for quality control and authentication. A small quantity of the powdered sample was cleared using chloral hydrate, mounted in glycerin on a clean glass slide, and examined under a light microscope at varying magnifications. Diagnostic features such as cell types, tissues, and inclusions were observed and documented following WHO guidelines (WHO, 2018).

Leaf Epidermal Studies on *Pulicaria crispa*

Epidermal strips were obtained from both the adaxial and abaxial surfaces of fresh leaves. The peels were stained with methylene blue, mounted in glycerin, and examined microscopically to evaluate epidermal cell characteristics, stomatal type, distribution, and trichome morphology. The procedure followed standard methods described by Brain and Turner (1975) and WHO (2018).

Quantitative Microscopy on the leaves of *Pulicaria crispa*

Quantitative microscopic parameters of the leaves were determined, including stomatal number, stomatal index, stomatal size, veinlet termination number, and palisade ratio.

These parameters were evaluated using standard pharmacognostic procedures as described by Evans (2009) and WHO (2018).

Histoanatomical Studies on the leaf, stem, and root *of Pulicaria crispa*

Transverse sections of the leaf, stem, and root were prepared using standard histological techniques (Carleton, 1980; Suvarna, 2019). Fresh tissues were fixed in FAA, dehydrated through a graded alcohol series, and embedded in paraffin wax. Thin sections were obtained using a rotary microtome, mounted on glass slides, deparaffinised, and stained with appropriate stains. The prepared sections were examined under a light microscope, and photomicrographs were captured using a digital camera (Sony®) attached to the microscope.

Physicochemical Analysis of the powdered plant of *Pulicaria crispa*

Physicochemical parameters of the powdered plant material, including moisture content, total ash, acid-insoluble ash, water-soluble ash, and extractive values, were determined according to standard procedures outlined in the British Pharmacopoeia (BP, 2012).

Extraction and Preliminary Phytochemical Screening

The powdered plant material was successively extracted using n-hexane and 70% aqueous ethanol. For each solvent, a measured quantity of the powder was macerated with intermittent shaking to enhance extraction efficiency. The mixtures were filtered, and the filtrates were concentrated under reduced pressure using a rotary evaporator to obtain the respective crude extracts.

The dried extracts were subjected to preliminary phytochemical screening using standard qualitative methods to detect major classes of secondary metabolites, including alkaloids, flavonoids, tannins, saponins, glycosides, and terpenoids. The procedures followed established protocols described by Brain and Turner (1975), Sofowora (2008), Harborne (2007), Evans (2009), and WHO (2011).

Thin Layer Chromatography (TLC) Profiling

Thin-layer chromatography (TLC) was employed for qualitative phytochemical profiling of the extracts. The n-hexane and aqueous ethanol extracts were applied onto silica gel-coated TLC plates and developed using solvent systems of hexane: ethyl acetate (9:1) and chloroform: ethyl acetate (9:1), respectively.

The developed plates were visualized under ultraviolet (UV) light (254 and 365 nm), followed by spraying with p-anisaldehyde–sulphuric acid reagent and heating at 105 °C to enhance spot development. The colour, number, and Rf values of the resolved spots were recorded for phytochemical characterization (Stahl, 2005).

RESULTS

Macroscopic Characteristics of *P. crispa*

Pulicaria crispa was collected from a semi-arid region in Northern Nigeria (Zaria–Kano axis; approximately 11.1° N, 7.7° E). The plant was observed to be a small, aromatic herb exhibiting an annual to perennial growth habit, with erect to decumbent stems. The detailed morphological features of the plant are presented in **Table I,** while the organoleptic characteristics of the powdered material are summarized in **Table II**. Representative image of the plant in its natural habitat are shown in **Plate 1**. The plant is characterized by lanceolate to oblong leaves with serrate margins, pubescent surfaces,

and alternate phyllotaxy. The leaves are whitish-green in colour, with a crispy texture and decurrent base. The stem shares a similar whitish-green coloration, while the plant bears yellow flowers and a primary root system. The powdered drug exhibited a green colour, aromatic odour, characteristic taste, and a woolly appearance.

Plate 1: Picture of the plant in its natural habitat.

Table I. Macroscopic Characteristics of *P. crispa*

Evaluative Features	Characteristics
Leaf shape	Lanceolate/ Oblong
Leaf venation	Reticulate
Leaf margin	Serrate
Leaf surface	Pubescent
Leaf apex	Obtuse/ Acute
Leaf Texture	Crispy
Leaf base	Decurrent
Phyllotaxy	Alternate
Flower color	Yellow
Leaf color	Whitish-green
Stem color	Whitish green
Root	Primary

Table II. Organoleptic Characteristics of *P. crispa* powder (Whole plant).

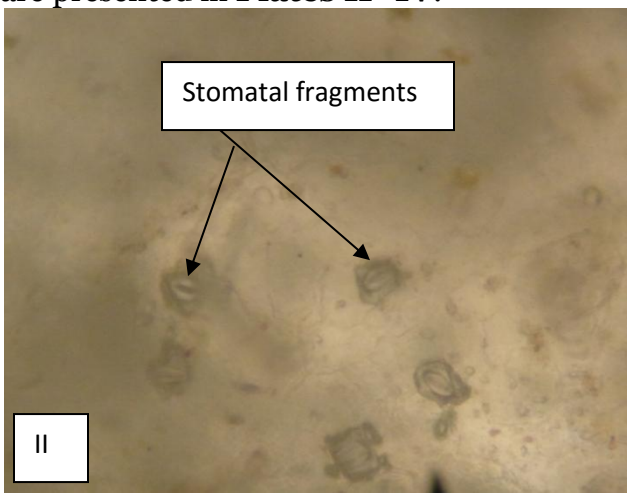
Evaluative features	Characteristics
Colour	Whitish-Green
Odour	Aromatic
Taste	Characteristic taste
Appearance	Wooly

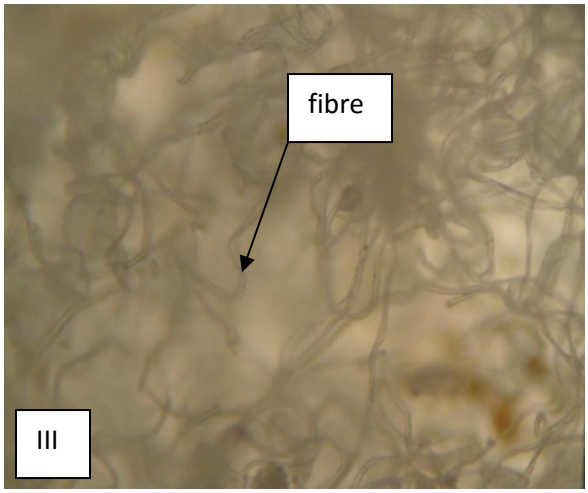
Microscopic Characteristics of *P. crispa*

Powder Microscopy

Microscopic examination of the powdered whole plant revealed several diagnostic features useful for identification and quality control. These included fragments of lignified fibres, xylem elements, and crystal sheaths containing prismatic calcium oxalate crystals. In addition, stomatal fragments, portions of leaf lamina with visible venation, and abundant trichome fragments (both intact and broken) were observed.

These microscopic characteristics correspond with those observed in the fresh plant material, supporting the integrity of the powdered sample. Representative micrographs are presented in **Plates II–IV**.





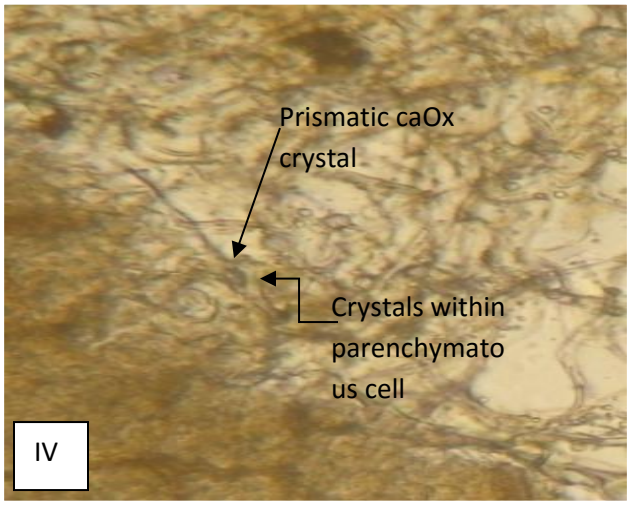


Plate II: Stomatal Fragments of powdered *P. crispa Plant [X100]*

Plate III: Fibres in powdered *P. crispa Plant [X100]*

Plate IV: Prismatic Calcium Oxalate crystals in powdered *P. crispa Plant [X100]*

Leaf Epidermal Features

Surface preparations of the leaves showed that *P. crispa* is **amphistomatic,** with stomata present on both adaxial and abaxial surfaces. The stomatal type was identified as **anomocytic**.

Adaxial Epidermis

The adaxial (upper) epidermis consisted of irregularly shaped epidermal cells with thick, undulating anticlinal walls extending throughout the cell depth. Stomata were present but less abundant compared to the abaxial surface. Both glandular (capitate) and non-

glandular trichomes were observed, including long, coiled, unicellular, unbranched covering hairs.

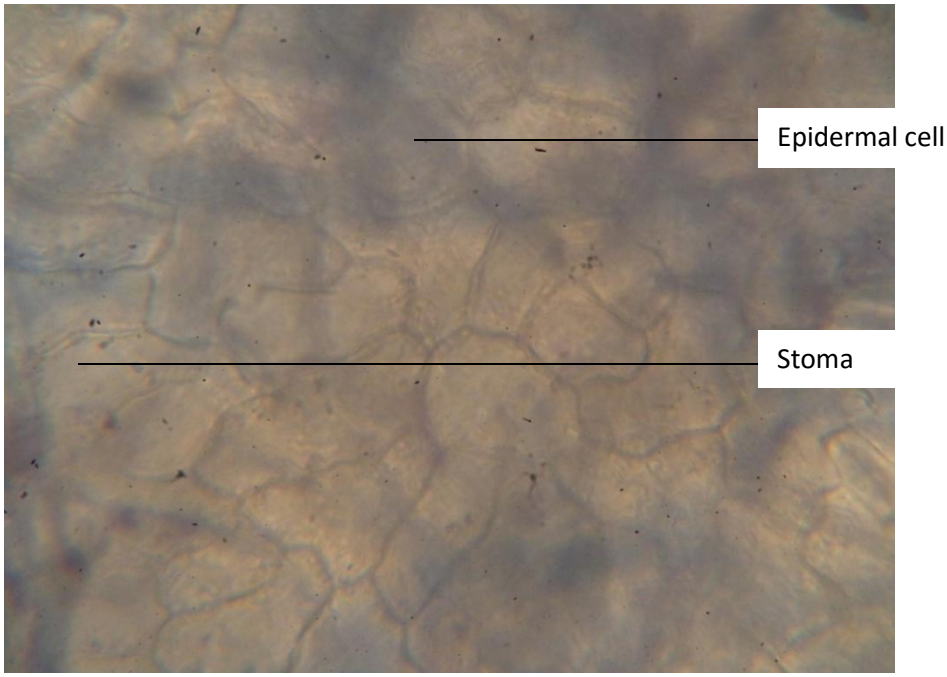


Plate V: Micrograph of the Upper Epidermal Layer of *P. crispa* leaf (Mag. ×400):

Abaxial Epidermis

The abaxial (lower) epidermis displayed polygonal cells with thick, undulating anticlinal walls, generally smaller than those of the adaxial surface. Stomata were more numerous on this surface. Both capitate and peltate glandular trichomes were present, along with abundant non-glandular trichomes similar in morphology to those observed on the upper surface.

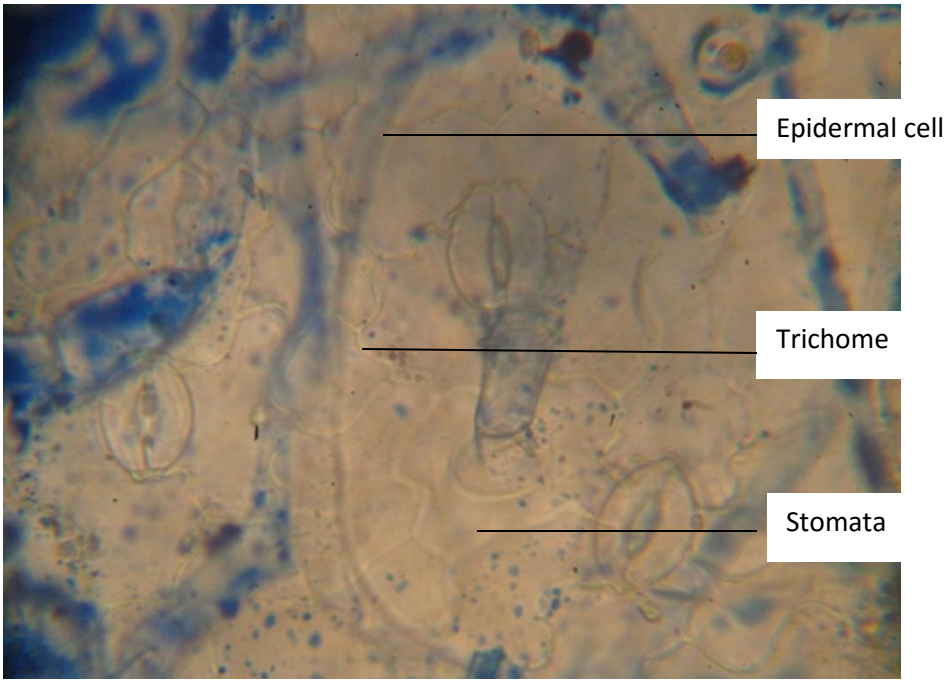


Plate VI: Micrograph of the Lower Epidermal Layer of *P. crispa* leaf stained with methylene blue solution. (Mag. ×400)

Histoanatomical Features of *P. crispa*

Leaf Anatomy

Transverse sections of the leaf revealed an **isolateral leaf structure**, a notable diagnostic feature distinguishing it from typical dorsiventral dicot leaves. Both adaxial and abaxial epidermal layers were present and interrupted by stomata and trichomes. Palisade mesophyll tissues were observed beneath both epidermal layers, separated by a central spongy mesophyll region. Vascular bundles were embedded within the mesophyll matrix (Plate V).

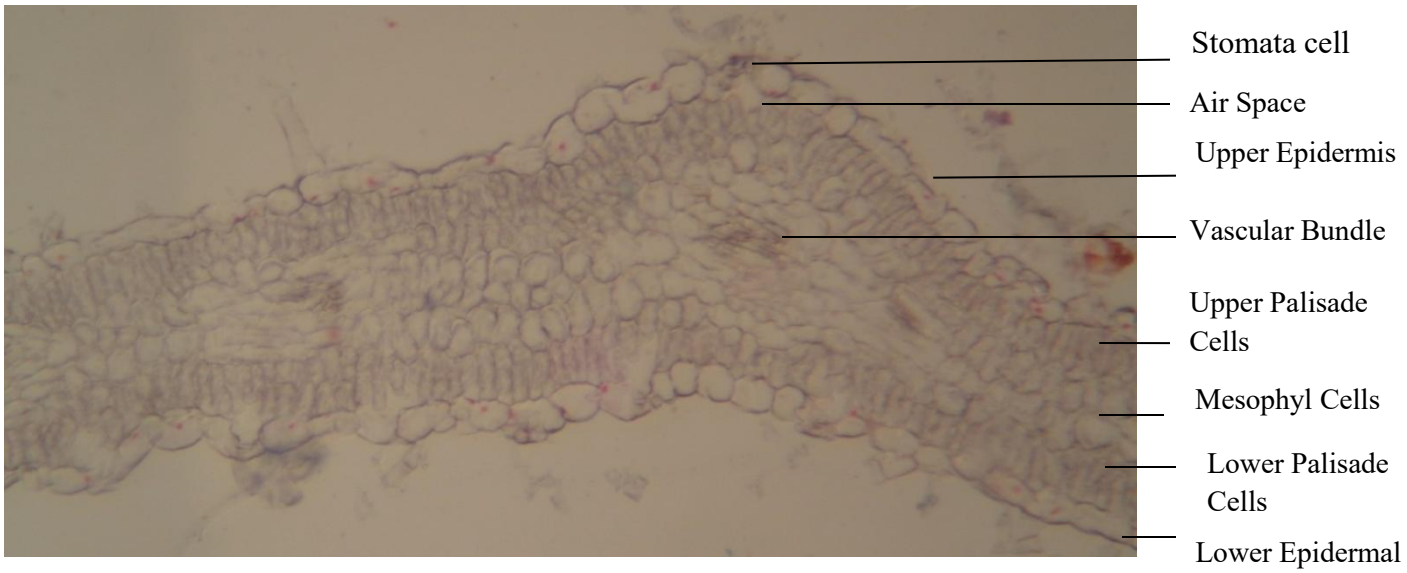


Plate III: Micrograph of the Leaf Tranverse Section of *P. crispa* stained with Saffi and Fast green solution. (Mag. ×100)

The midrib region exhibited a uniseriate epidermis with underlying collenchymatous support tissue. The central region consisted of parenchymatous cells containing three vascular bundles, with the median bundle being the largest. The vascular bundles were collateral and reinforced by sclerenchymatous tissues surrounding both xylem and phloem elements.

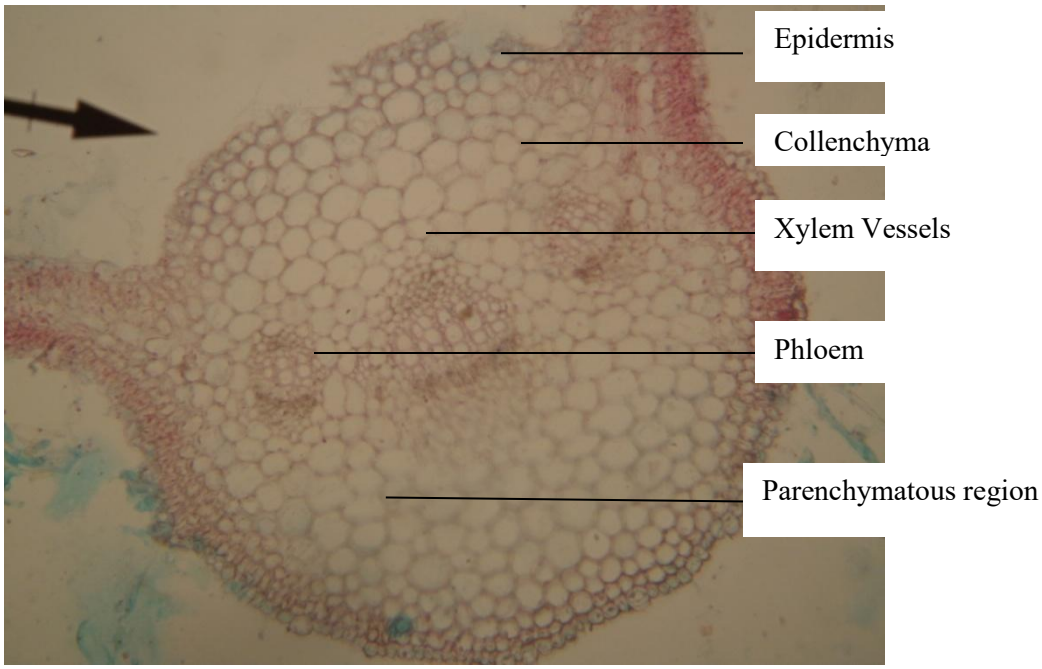


Plate IV: Micrograph of the Transverse section through the Midrib of *P. crispa* Leaf stained with Saffranin and Fast green solution. (Mag. ×100)

Stem Anatomy

The transverse section of the stem revealed three distinct tissue regions: the epidermis, cortex, and vascular cylinder (stele). The epidermis was single-layered, followed by a cortex that contained a leaf trace. The vascular cylinder exhibited a **eustele arrangement**, with collateral vascular bundles. The phloem was externally protected by periphloemic sclerenchymatous tissues, while the cambium was located between the phloem and xylem, and the xylem occupied the inner region.

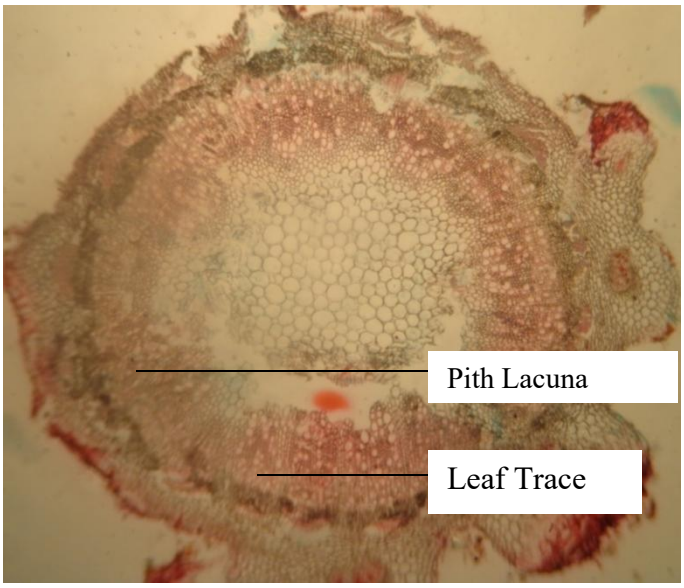


Plate VI a: Micrograph of the Transverse Section Passing through the Stem of *P. crispa* stained with Saffranin and Fast green solution. Mag. ×40.

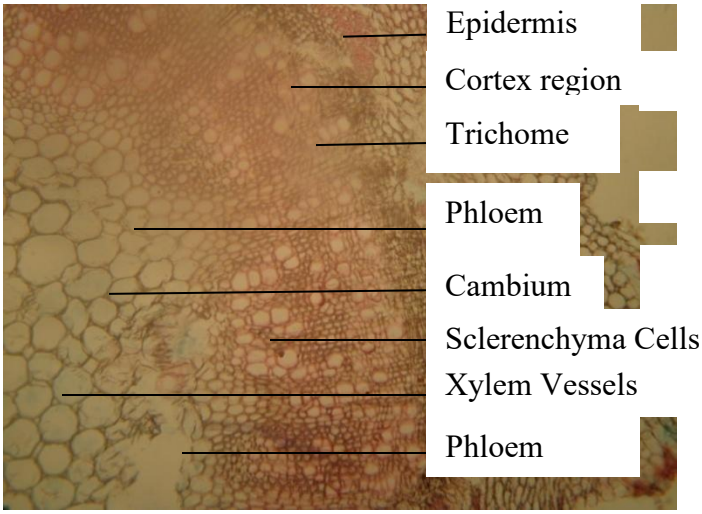


Plate VI: Micrograph of the Transverse Section Passing through the Stem of *P. crispa* stained with Saffranin and Fast green solution. (Mag ×100)

Root Anatomy

The transverse section of the root revealed an outer epiblema, followed by a parenchymatous cortex and a central stele. The vascular cambium produced secondary phloem outward and secondary xylem inward.

The xylem was more prominently developed than the phloem, which appeared relatively reduced and protected by sclerenchymatous tissues. Pith rays were also observed within the vascular.

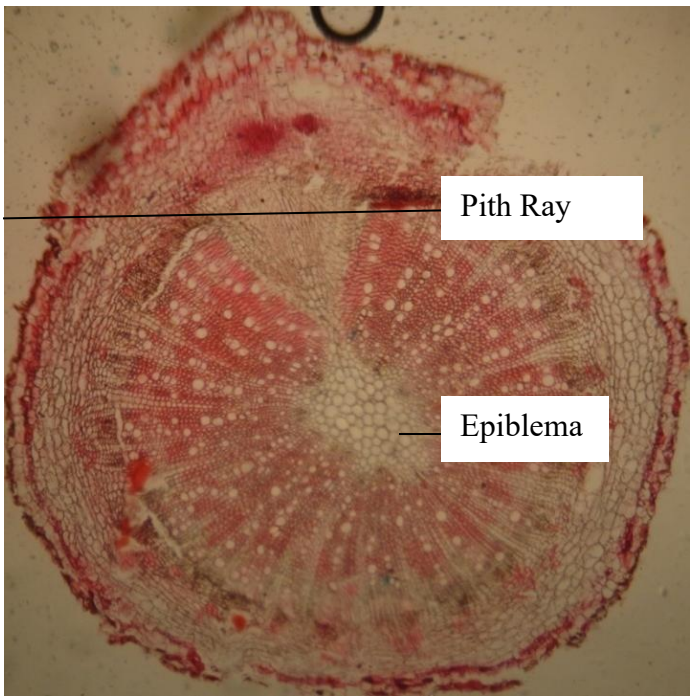


Plate VII: Micrograph of the Transection Passing through the Root of *P. crispa* stained with Saffranin and Fast green. (Mag. ×40)

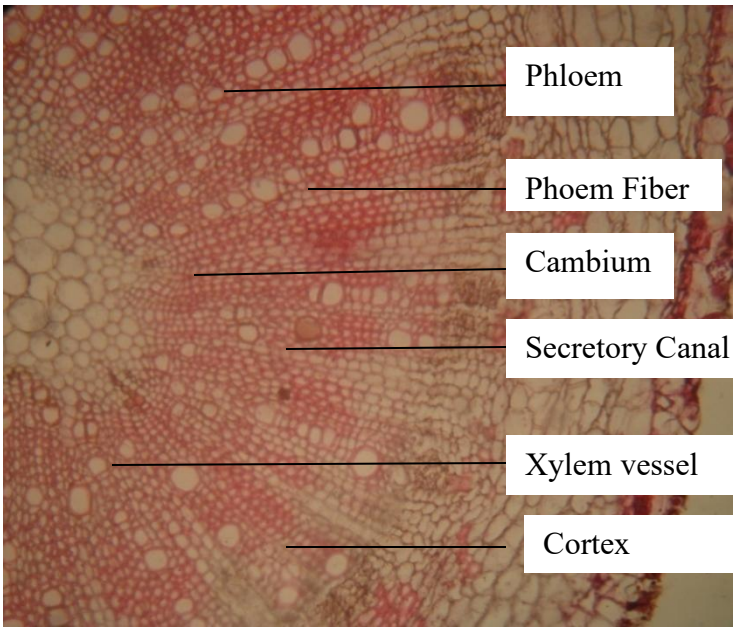


Plate VIII: Micrograph of the Transection Passing through the Root of *P. crispa* stained with Saffranin and Fast green. Mag. 100:

Quantitative Microscopy

Quantitative microscopic evaluation of the leaf provided average measurable diagnostic parameters both on the adaxial and abaxial surfaces respectively. The stomatal number (15 and 33), stomatal index (12.4 and 26.8), palisade ratio (17.1 and 12.8), and veinlet termination number of 3.3. Stomatal dimensions were approximately 2.45 µm in length and 1.75 µm in width. The abaxial surface showed a higher stomatal number and index compared to the adaxial surface, consistent with amphistomatic leaf characteristics. The detailed values are presented in **Table III**.

Table III: Quantitative Microscopical Values for *P. crispa* leaf.

Evaluative Parameter	**Values***	
	Adaxial Surface	**Abaxial surface**
Stomatal number	11.0 – 15.0 - 19.0	25.0 – 33.0 – 40.0
Stomatal index	14.0 – 12.4 – 10.7	27.7 – 26.8 – 25.9
Palisade ratio	17.8 – 17.1 – 16.3	13.5 – 12.8 – 12.0
Veinlet termination number	4.0 – 3.3 – 2.5	

*average is underlined.

Physicochemical Parameters

The physicochemical constants of the powdered plant material are summarized in **Table IV**. The results showed that the water-soluble extractive value (16.0 % w/w) was higher than the alcohol-soluble extractive value (6.0 % w/w), indicating a predominance of polar constituents. The total ash value (20.0 % w/w) was relatively high, suggesting the presence of significant inorganic content, while the acid-insoluble ash (15.0 % w/w) indicated possible siliceous matter. The moisture content (8.7 % w/w) was within acceptable limits, suggesting good stability and reduced susceptibility to microbial degradation.

Table IV: **Results of Some Physical Standards of Powdered *P. crispa***

Evaluative Parameter	**Values (% w/w)***
Alcohol soluble extractive value	6.0
Water-soluble extractive value	16.0
Moisture content	8.7
Ash value	20.0
Acid Insoluble Ash	15.0
Water soluble Ash	1.5

*Average of three determinations.

Phytochemical constituents

Preliminary phytochemical analysis of the ethanol extract revealed the presence of several classes of secondary metabolites, including cardiac glycosides, anthraquinones, saponins, steroids/triterpenes, flavonoids, tannins, and alkaloids. The results of the qualitative tests, along with their corresponding observations and inferences, are presented in **Table V.**

Table IV: Qualitative Phytochemical constituents present in the Ethanol extract of *P.crispa*

S/N	Constituent	Test	Observation	Inference
1	Cardiac glycosides	Keller-killiani Test	Reddish-brown color	Present
		Kedde Test	Purple-blue colour	Present
	Anthraquinones	Borntrager Test	Pink colour	Present
		Modified Borntrager Test	Pink or red colouration	Present
3	Saponins	Frothing Test	Frothing column	Present
		Haemolysis Test	Haemolysis	Present
4	Steriod/Triterpenes	Salkowski Test	Reddish-brown colour	Present
		Liberman Burchard Test	Reddish- brown ring	Present
5	Flavonoids	Shinoda Test	Pink or red colour	Present
		Sodium Hydroxide Test	Yellow to colourless	Present
6	Tannins	Ferric chloride Test	Blue-black precipitate	Present
		Bromine Water test		
			Yellow precipitate	Present
		Lead sub acetate Test	Whitish-yellow precipitate	Present
7	Alkaloids	Dragendorff Test	Red-Brownish Precipitate	Present
		Mayer Test	Cream Precipitate	Present
		Wagner Test	Reddish-brown Precipitate	Present

Thin Layer Chromatography (TLC) Profile of n-hexane and ethanol extracts of *P. crispa*

Thin-layer chromatographic analysis of both n-hexane and ethanol extracts revealed multiple resolved spots, indicating the presence of diverse phytoconstituents. The

hexane extract showed several distinct spots with varying Rf values and colour reactions under daylight, UV light, and after derivatization with anisaldehyde–sulphuric acid reagent. Similarly, the ethanol extract exhibited multiple components with characteristic chromatographic profiles. The detailed Rf values and colour characteristics are presented in **Tables VI and VII,** while the chromatograms are shown in **Figure 9.**

Table V: Thin Layer Chromatographic Analysis of the Hexane Extract of *Pulicaria crispa*

Solvent System: Hexane: Ethyl acetate (19:1)

Spot	R_f value	colour in Day Light	Colour in UV	Colour with Anisaldehyde/H_2SO_4
Origin	0.00	Yellow-brown	yellow-brown	dark color
A	0.04	Colourless	yellow-brown	dark green
B	0.09		orange-brown	blue
C	0.15			grey
D	0.19			green
E	0.28		white	pink
F	0.35		orange	grey
G	0.37			grey
H	0.45		orange	purple
I	0.51			purple
J	0.58			grey
K	0.73			purple
L	0.78			blue
M	0.82			blue
N	0.89	Green	yellow-brown	purple

Table VI: Thin TLC Characteristics of the Ethanol Extract of *P. crispa*

Solvent system: Chloroform: Ethyl acetate (9:1)

Spots	R_f value	Colour in Day Light	Colour in UV	Colour with Anisaldehyde/H_2SO_4
Origin	0.00	Yellowish brown	Yellowish brown	Brownish black
A	0.04	Colourless	White	Grey
B	0.08	Colourless	Orange	Greyish green
C	0.12	Colourless		Greyish green
D	0.15	Colourless		Purple
E	0.21	Colourless		Pink
F	0.25	Colourless	White	Purple
G	0.56	Colourless	Orange	Grey
H	0.68	Colourless		Purple
I	0.74	Colourless	Orange	Green

DISCUSSION

Pulicaria crispa is widely utilized in traditional medicine across West and North Africa for the management of malaria, headache, inflammatory conditions, gastrointestinal disorders, and febrile illnesses (Kankara *et al*., 2022). Despite its extensive ethnomedicinal relevance, standardized pharmacognostic data required for its proper identification and quality control remain limited (WHO, 2018). The present study therefore established comprehensive pharmacognostic parameters to support its authentication, standardization, and safe utilization.

Pharmacognostic evaluation remains the cornerstone of crude drug standardization, encompassing macroscopic, microscopic, physicochemical, and phytochemical analyses (Evans, 2009; WHO, 2011). The integration of these approaches in this study provides a robust framework for the development of a monograph for *P. crispa*, thereby addressing a critical gap in its scientific documentation.

The macroscopic characteristics observed, including lanceolate to oblong leaves with serrate margins, pubescent surfaces, and whitish-green coloration, are consistent with diagnostic features reported for members of the Asteraceae family. These morphological traits are often associated with xerophytic adaptations, enabling survival in semi-arid environments such as the Zaria–Kano axis (Liu *et al*., 2010; El-Kamali *et al*., 2010). The aromatic odour of the powdered material further suggests the presence of volatile constituents, particularly essential oils rich in terpenoids, which have been previously

reported in *Pulicaria* species and are known contributors to their therapeutic properties (Maghraby *et al*., 2010; Bakkali *et al*., 2008).

Microscopic evaluation revealed amphistomatic leaves with predominantly anomocytic stomata, undulating epidermal cells, and abundant glandular and non-glandular trichomes. These features are characteristic of Asteraceae and serve as reliable taxonomic markers. Trichomes, particularly glandular types, are known sites for the biosynthesis and storage of secondary metabolites such as essential oils, flavonoids, and terpenoids, thereby contributing to the pharmacological potential of the plant (Metcalfe & Chalk, 2012; Werker, 2000).

The isolateral leaf anatomy observed in the transverse section, characterized by palisade mesophyll on both adaxial and abaxial surfaces, represents an adaptive feature for efficient photosynthesis under high light intensity conditions typical of open and arid habitats. This anatomical arrangement enhances light utilization and reduces photoinhibition (Metcalfe & Chalk, 2012; Fahn, 1990). The presence of well-developed vascular bundles reinforced by sclerenchymatous tissues further supports structural integrity and efficient translocation of nutrients.

The stem anatomy exhibited a typical dicotyledonous organization with a uniseriate epidermis, parenchymatous cortex, and a eustele arrangement of vascular bundles. The presence of periphloemic sclerenchyma indicates mechanical reinforcement, while the cambial layer suggests potential for secondary growth. Similarly, the root anatomy, with a well-developed xylem relative to phloem, reflects adaptation for efficient water conduction in semi-arid environments. The occurrence of secretory structures may further indicate sites of accumulation of bioactive compounds (Metcalfe & Chalk, 2012; Fahn, 1990).

Powder microscopy revealed diagnostic features such as lignified fibres, trichomes, xylem elements, and calcium oxalate crystals, which are critical for the identification of powdered crude drugs. These features serve as reliable markers to detect adulteration and ensure quality control, especially in the absence of intact plant material (WHO, 2018; Wallis, 2005). Furthermore, the quantitative microscopy parameters obtained, including stomatal index, stomatal number, and palisade ratio, provide reproducible numerical standards essential for pharmacopoeial identification and standardization.

The physicochemical parameters obtained in this study further support the quality profile of *P. crispa*. The higher water-soluble extractive value compared to alcohol-soluble extractive value suggests a predominance of polar constituents, which aligns with the traditional use of aqueous preparations such as decoctions and infusions. The moisture content falls within acceptable pharmacopoeial limits, indicating reduced susceptibility to microbial spoilage. The relatively high ash values reflect inherent mineral content typical of plants growing in savannah soils rather than contamination, although this parameter also underscores the need for proper purification and processing (Evans, 2009; WHO, 2011).

Preliminary phytochemical screening confirmed the presence of multiple classes of bioactive compounds, including alkaloids, flavonoids, tannins, saponins, cardiac glycosides, anthraquinones, and triterpenes. These metabolites are well documented for

their diverse pharmacological activities such as antioxidant, anti-inflammatory, antimicrobial, and antimalarial effects, thereby providing scientific justification for the ethnomedicinal uses of *P. crispa* (Sofowora, 2008; Harborne, 2007; Cowan, 1999). In particular, flavonoids and tannins are known for their free radical scavenging and anti-inflammatory properties, while saponins and alkaloids contribute to antimicrobial and antiparasitic activities.

The TLC profiling of both hexane and ethanol extracts yielded distinct and reproducible chromatographic fingerprints characterized by multiple spots with varying Rf values and colour reactions. These profiles indicate the presence of diverse phytoconstituents, including terpenoids and phenolic compounds. TLC fingerprinting is widely recognized as a rapid, reliable, and cost-effective method for herbal drug standardization and quality control, particularly in resource-limited settings (Stahl, 2005; Reich and Schibli, 2007; WHO, 2011).

Overall, the findings from this study provide a comprehensive pharmacognostic profile of *Pulicaria crispa*, establishing baseline standards for its identification, quality control, and future pharmacological investigations.

CONCLUSION

The present study established a reproducible pharmacognostic standards for Pulicaria crispa through integrated macroscopic, microscopic, histoanatomical, physicochemical, phytochemical, and chromatographic evaluations. The diagnostic features generated provide a reliable scientific basis for the identification, authentication, and quality control of the plant. The presence of diverse bioactive secondary metabolites, supported by characteristic TLC fingerprints, further validates its ethnomedicinal applications and underscores its potential as a source of therapeutic agents. These findings contribute meaningfully to the development of a pharmacognostic monograph and support the safe and standardized use of P. crispa in research and herbal medicine. Based on these findings, the established pharmacognostic parameters are recommended as reference standards for routine quality assessment. Further studies should focus on advanced chromatographic profiling (e.g., HPLC, GC-MS, and LC-MS), quantitative phytochemical analysis, and detailed pharmacological investigations to substantiate its therapeutic efficacy. In addition, comprehensive toxicological evaluations are necessary to establish safety profiles, while formulation and stability studies will facilitate the development of standardized herbal products. Finally, the inclusion of *P. crispa* in regional and international pharmacopoeias is advocated to enhance its recognition, regulation, and wider utilization.

CONFLICTS OF INTEREST

The authors declare no conflicts of interest

REFERENCES

Abo-elghiet, F., Rushdi, A., Ibrahim, M. H., Mahmoud, S. H., Rabeh, M. A., Alshehri, S. A., & El Menofy, N. G. (2023). *Chemical profile, antibacterial, antibiofilm, and antiviral activities of Pulicaria crispa most potent fraction: An in vitro and in silico study*. Molecules, 28(10), 4184. https://doi.org/10.3390/molecules28104184

Al-Fatimi, M. (2019). Ethnobotanical survey of medicinal plants in central Yemen. *Journal of Ethnopharmacology, 241*, 111973. https://doi.org/10.1016/j.jep.2019.111973

Alhazimi, H.M.G. and Al-Khathlan H. Z. (1992). Chemistry of Various Pulicaria species (Asteraceae). *Journal of Chem. Soc. Pakistan*. 14(3), 233 – 240.

Almotwaa, S. M., & Al-Otaibi, W. A. (2022). Determination of the chemical composition and antioxidant, anticancer, and antibacterial properties of essential oil of Pulicaria crispa from Saudi Arabia. Journal of the Indian Chemical Society, 99(2), Article 100341. https://doi.org/10.1016/j.jics.2022.100341

Al-Yahya M. A., El – Sayed A. M., Mossa, J. S., Kozlowski J. F., Antoun, M. D., Ferin, M., Baird, W. M., and Cassady, J. M. (1988). Potential Cancer Chemopreventive and Cytotoxic Agents from *Pulicaria crispa*. *Journal of Natural Product*. 51(3):621 – 4. PMID:3404160

Al-Yahya, M. A., El-Sayed, A. M., Mahmood, M. A., Hassan and E-Meshal, I. (1989). The volatile oil of Saudi *Pulicaria crispa*. *Arab Gulf J. Scient Res*; 7(2), 1-6

Al-Yahya, M. A., Khafagy, S., and Shihata A. (1984). Phytochemical and Biological Screening of Saudi Medicinal Plants, Part 6: Isolation of 2 α-Hydroxyalantolactone, The Antileukemic principle of *Francoeuria crispa*. *J. Nat. Prod*. 47(6), 1013-1017

Angiosperm Phylogeny Group (APG IV). (2016). *An update of the Angiosperm Phylogeny Group classification for the orders and families of flowering plants*. Botanical Journal of the Linnean Society, 181(1), 1–20. https://doi.org/10.1111/boj.12385

Assaf, M. H and Ali, A. A (1996). *Study of the Constituents of the Essential Oil of Pulicaria crispa. (Asteraceae) Growing wild in Egypt*. Pharmacognosy Dept. Fac. Of Pharmacy, Assiut Uni. Pp 9-12.

Bakkali Fouad., Averbeck, S., Averbeck, D., & Idaomar, M. (2008). Biological effects of essential oils – A review. *Food and Chemical Toxicology*, 46(2), 446–475.

Bohlmann,F., Heinz, K. K., and El-Amary, N. A. (1979). Neuartige Sesquiterpenelacetone aus *Pulicaria crispa* Phytochemistry, 18(7), 1231.

Boulos, L. (2021). *Flora of Egypt* (Vol. 4). Al Hadara Publishing.

Brain, K. R. and Tuner, T. D (1975). The Practical Evaluation of Phytopharmaceuticals. WRIGHT-SCIENTECHNICA, BRISTOL. ISBN 0 85608 012 8 Pp 5, 25, 26.

British Pharmacopoeia (2012). General Medical Council. Version 16.0

Burkill, H. M. (1985). *The Useful plants of West Tropical Africa*. Royal Botanic Garden, Kew. Pp. 489 – 490.

Carleton, H. M. (1980). *Carleton's Histological Technique*, Fourth Edition. Oxford Medical Publication. (Ed.). Drury R. A. B., Wallington E. A. Fourth Edition. Oxford Medical Publication. Pp 49-51.

Cowan Marjorie M.. (1999). Plant products as antimicrobial agents. *Clinical Microbiology Reviews*, 12(4), 564–582.

El – Kamali, H. H., Habeballa, R., Abdalla, I., Muhammad, A. Y., Abdelkarim, N. D., Abbas, I. M., Ali, M. S. (2010). Genetic Relationships of Two Pulicaria Species and Identification of their Putative Hybrids Using Rapd Markers. *World Applied Sciences Journal* 8(6):687-693. ISSN 1818 – 4952. IDOSI Publications

El-Kamali, H. H., Habeballa, R. S., Abdalla, I. A., Mohammed, A. Y., Abdelkarim, N. D., Abbas, I. M., & Ali, M. S. (2010). Genetic relationships of two *Pulicaria* species and identification of their putative hybrids using RAPD markers. *World Applied Sciences Journal, 8*(6), 687–693.

Evans, W. C. (2009): *Trease and Evans pharmocognosy*. Elseviers 16th edition Pp. 10 – 11.

Fahn Abraham. (1990). *Plant Anatomy* (4th ed.). Pergamon Press.

Gibbs, S. (2006). An Introduction to Planar Chromatography. In: Methods in Biotechnology; Natural Products Isolation (ed) Sarker, S.D., Latif, Z. and Gray, A. I. 2nd edition. Humana Press, Totowa, New Jersey.

Harborne, J. B. (2007). *Phytochemical methods: A guide to modern techniques of plant analysis* (3rd ed.). Chapman & Hall.

Hutchinson, J and Dalziel, J. M (1963). *Flora of West Tropical Africa*. Vol. II Ed. 2: 258.

Kankara, S. S., Nuhu, A. I., Abdullahi, M. S., Bello, A., & Lawal, U. (2022). Indigenous traditional knowledge of medicinal plants used for the management of HIV/AIDS opportunistic infections in Katsina State, north-western Nigeria. *Ethnobotany Research and Applications, 23*, Article 35, 1–17. https://doi.org/10.32859/era.23.35.1-17

Liu, L. L., Yang, J. L., & Shi, Y. P. (2010). Phytochemicals and biological activities of *Pulicaria* species. *Chemistry & Biodiversity, 7*(2), 327–349.

Maghraby, A. S., Shalaby, N., Abd-All, H.I., Ahmed, S. A., Khalid, H. M. and Bahgat, M. M. (2010). Immuno modulatory Effects of Extract of P. crispa before and after Schistosoma mansoni Infection. *Acta poloniae pharmaceutica – Drug Research,* vol. 67. No. 1 Pp. 75 – 79.

Metcalfe, C. R., & Chalk, L. (2012). *Anatomy of the dicotyledons* (Vol. 2, 2nd ed.). Oxford University Press.

Nguyen, T. K. T. Stevenson, G. Obatomi, D. Bach, P (2000). Determination of Lipids in Animal Tissues by High-Performance Thin Layer Chromatography with Densitometry. *Journal of Planar Chromatography*.

Plants of the World Online. (2023). *Pulicaria* Gaertn. Royal Botanic Gardens, Kew. https://powo.science.kew.org

Reich Ernst. & Schibli Andreas. (2007). *High-Performance Thin-Layer Chromatography for the Analysis of Medicinal Plants*. Thieme Medical Publishers.

Ruzin, S. E. (1999). *Plant microtechnique and microscopy*. Oxford University Press. https://global.oup.com/academic/product/plant-microtechnique-and-microscopy-9780195089561

Santos, R. M., Oliveira, A. B., Ferreira, J. L. P., & Costa, F. N. (2018). Leaf anatomical characters as taxonomic tools in Asteraceae. *Brazilian Journal of Botany, 41*(2), 423–435. https://doi.org/10.1007/s40415-018-0456-9

Sofowora, A. (2008).*Medicinal plants and traditional medicine in Africa* (3rd ed.). Spectrum Books.

Stavri, M., Mathew, K. T., Gordon, A., Shnyder, S. D., Falconer, R. A., Gabbons, S. (2008). Guaianolide sesquiterperes from *P. crispa* (forssk.) Oliv. *Journal of Phytochemistry,* 69(9) 1915 – 1918. 10. 1016.

Suvarna, S. K., Layton, C., & Bancroft, J. D. (2019). *Bancroft's theory and practice of histological techniques* (8th ed.). Elsevier.

Ullah, R., Alqahtani, A. S., Noman, O. M. A., Alqahtani, A. M., Ibenmoussa, S., & Bourhia, M. (2020a). A review on ethno-medicinal plants used in traditional medicine in the Kingdom of Saudi Arabia. *Saudi Journal of Biological Sciences*, *27*(10), 2706–2718. https://doi.org/10.1016/j.sjbs.2020.06.020

Wallis Thomas Edward. (2005). *Textbook of Pharmacognosy* (5th ed.). CBS Publishers.

Werker Eva. (2000). Trichome diversity and development. *Advances in Botanical Research*, 31, 1–35.

World Health Organization (WHO). (2018). *Quality control methods for herbal materials*. WHO Press. https://www.who.int/publications/i/item/WHO-TRS-1010