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Pharmacognostical Evaluation, Growth Inhibitory and Antioxidant Activities of *Chasmanthera dependens* Hochst.

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ABSTRACT

The absence of pharmacognostic criteria and inadequate crude drug quality control yardsticks are the primary reasons for medicinal plant misidentification, therapeutic failure, and toxicity associated with herbal medicines. This study aims to identify the macroscopic and microscopic features, physicochemical properties, important chemical constituents, and antioxidant and growth inhibitory potential of *Chasmanthera dependens*, a species found in the rainforest of West Africa. A detailed pharmacognostic evaluation of *C. dependens* was carried out, including macroscopic and microscopic characterization, phytochemical screening, and physicochemical analysis. Antioxidant and growth-inhibitory bioassays were performed to assess its bioactivity. *Chasmanthera dependens* is characterized by a cylindrical, partly long, rough-surfaced twig with evergreen, cordate leaves. Its exudate is odorless but slightly sweet, with a bitter aftertaste. Microscopic analysis revealed covering trichomes on both adaxial and abaxial leaf surfaces, with an anomocytic type of stomata only on the abaxial surface. The chemical analysis showed the fruit is rich in phytoconstituents like alkaloids, tannins, and flavonoids, as well as important micronutrients such as zinc, copper, cadmium, magnesium, manganese, protein, lipids, and fiber. The extracts demonstrated significant antioxidant and growth-inhibitory effects. This study provides the first detailed pharmacognostic features of *C. dependens*, contributing valuable data for its identification, quality control, and potential medicinal applications.

1. Introduction

Medicinal plants have long been central to traditional medicine, and despite significant advancements in medical research, they remain highly valued. The majority of the world's population still relies on medicinal plants for primary healthcare (Sahil *et al.* 2011). Nearly 50% of the world's flora and fauna are found in tropical rainforests, which cover only 6% of the Earth's land area. The biodiversity of tropical rainforests, including those in Africa, is regarded as a global heritage (Pinho *et al.* 2020). The tropical African climate fosters a chemically rich ecosystem, evident in its abundant vegetation. Local populations primarily depend on these plants for their healthcare needs (Okaiyeto and Oguntibeju 2021).

In Nigeria alone, over 7,895 plant species, including 128 endemic species, have been reported, with the majority used in traditional medicine for millennia (Osawaru *et al.* 2013). However, only a few of these medicinal plants have undergone standardization (Folami *et al.* 2024). The Nigerian Society of Pharmacognosy is making concerted efforts to develop Nigeria's herbal pharmacopeia (Iyiola and Adegoke Wahab 2023).

The absence of standardized criteria for identifying most Nigerian medicinal plants has led to numerous reports of adulteration and misidentification (Nalawade *et al.* 2022). The lack of pharmacognostic standards and poor macroscopic properties for plant quality control are major causes of medicinal plant misidentification,

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therapeutic failure, and the toxicity often associated with herbal medicines (Das *et al.* 2021). These deficiencies contribute to the poor acceptance of herbal medicines in many developed societies (Ozioma and Chinwe 2019). Therefore, the standardization of medicinal plants with well-defined identification criteria and physicochemical parameters is crucial for scientific research on their chemical and pharmacological properties, mechanisms of action, and therapeutic potentials, ensuring reproducible results (Nafiu *et al.* 2017; Lamichhane *et al.* 2023; Wang *et al.* 2023a). Standardization of commonly used medicinal plants is crucial for ensuring their quality, safety, and efficacy in healthcare operations for various reasons, particularly because a significant number of people still depend on them for their primary healthcare needs (Ndhlovu *et al.* 2023; Wang *et al.* 2023b). Modern pharmacognostic research has improved crude drug identification and evaluation, making plant identification more reliable, accurate, and cost-effective (Jamtsho *et al.* 2024). The World Health Organization also highlights the significance of identifying macroscopic and microscopic properties of medicinal plants for quality control, standardization, and pharmacognostic investigations (Sarfraz *et al.* 2024).

Chasmanthera dependens, a climbing plant native to Africa, is widely used in traditional medicine for the treatment of malaria, to relieve pain, digestive issues, inflammatory problems and fever, respiratory conditions, skin conditions, aphrodisiac issues, infections, and hemorrhoids (Codo Toafode *et al.* 2022). The plant's roots and leaves are boiled to create a decoction, which reduces fever and fights the parasite (Olanipekun 2023). It is also used to treat digestive problems like stomach aches, diarrhea, and dysentery (Madueke *et al.* 2020). Research has shown that *C. dependens* contains bioactive compounds like alkaloids, flavonoids, saponins, and tannins (Olorunnisola *et al.* 2013). However, *C. dependens* can easily be confused with other climbing plants or species within the same family (Menispermaceae) due to their similar morphological features, such as leaf shape, flower structure, and growth habits. It is worrisome that in peer-reviewed articles, other plants have been misidentified as *C. dependens* (Enenebeaku *et al.* 2022), and several online repositories have other plants archived as *C. dependens*. These misidentifications are possible because, like many medicinal plants, proper identification yardsticks for *C. dependens* have not been determined (Madueke and Anosike 2017). *Chasmanthera dependens* is a widely utilized traditional herb in many African communities. Despite its widespread use, there are no reports of pharmacognostic standards that can be used to identify it properly. Multidisciplinary collaboration between botanists, pharmacognosists, ethnobotanists, traditional healers, and regulatory authorities is critical for addressing challenges in pharmacognostic standards for *C. dependens* and other African medicinal plants, as well as ensuring sustainable use and conservation. In Nigeria, among many other therapeutic potentials of *C. dependens*, traditional bone setters employ *C. dependence* by applying it as a balm on fracture areas to mend fractures (Kola-Mustapha *et al.* 2020). The plant is locally referred to as "ato" among the Yoruba people of Nigeria. The word "ato" means "capable of mending" in English (Olaniran *et al.* 2018).

There is a controversy about the exact number of species in the *Chasmanthera* genus. The Royal Botanical Garden recognizes two species, namely *Chasmanthera dependens* Hochst. and *Chasmanthera uniformis* Baill. (Govaerts 1999). The earlier names of *Chasmanthera dependens* include *Chasmanthera strigosa* Welw. Ex Hiern was named in 1896, and *Chasmanthera welwitschii* Troupin was named in 1951. Whereas other authors considered *Chasmanthera welwitschii* Troupin a separate genus (Afiong *et al.* 2024).

Hence, this study was designed to identify the macroscopic and microscopic features, identify its physicochemical properties, identify important chemical constituents, and determine the antioxidant and growth inhibitory potential of *C. dependens* fruit.

2. Materials and Methods

2.1. Materials

Both fresh and dried specimens of the plant of *Chasmanthera dependens*, 3.5% of 75 ml Hypochlorite, Petri dishes, ethanol; 50-100% Alcohol, glycerine, Safranin-O, Compound microscope, meter rule, digital camera, 2, 2-diphenyl-1-picrylhydrazyl (DPPH), ascorbic acid, gallic acid, Iron (II) Sulfate (Fe_2SO_4), Blender, Whatman No. 1 filter paper and Ultraviolet spectrophotometer (TitertekUniskan).

2.2. Collection and Authentication of Plant Material

Chasmanthera dependens Hochst (Figure 1Aa and Bb), which was collected from the Medicinal Garden of the Department of Pharmacognosy and Drug Development, University of Ilorin. The plant was identified and authenticated at the Herbarium

Figure 1. (Aa) Whole Plant (X 1/50 Magnification), (Ba) fruits (X 1/3 Magnification). (A) whole leaf, (B) diameter of leaf, (C) height of leaf, (D) leaf apex, (E) leaf arrangement and petiolate, (F) mature stem, (G) fracture of the stem, (H) root system (X 1/3 Magnification); (I) flower stalk (X 1/3 Magnification) and (J) complete flower (X 2Magnification) of *Chasmanthera dependens*

unit of the Department of Plant Biology, Faculty of Life Science, University of Ilorin, and a voucher number (UIL001/1042/2021) was issued. The whole fruit, roots, leaves, and stems of *C. dependents* were collected from the Medicinal Garden in the Department of Pharmacognosy and Drug Development, University of Ilorin, Ilorin, Kwara State, Nigeria.

2.3. Methods

2.3.1. Macroscopical, Organoleptic and Microscopic Evaluations

Macroscopical evaluations: Different macroscopic parameters of stem, root, fruit, and leaves were noted. Leaves evaluation includes the absence or presence of petioles and different characters of lamina, i.e., shape indentations, base, texture, venations, and apex. Root was studied for its size, shape, surface, and fracture.

Organoleptic evaluations: Organoleptic evaluations were performed according to the color, size, odor, and taste parameters. Also, the moisture content of the fruit was observed using the 5 fruits collected and was kept at a temperature of 60°C for 72 hours. The assessment of some exo-morphological features such as plant odor and texture, leaf type, arrangement, margin, base, apex, venation, and size (Hameed *et al.* 2023).

Microscopical evaluations: Qualitative microscopy of the fruits, roots, stems, and leaves of *C. dependens* was done using a compound microscope. Epidermal covering of the various parts was scraped and collected, and the transparent epidermal covering was cut in portions of 2–5 cm^2 from the standard median part of the leaf lamina near the mid-rib. Three cuttings of the epidermal covering were selected, separated, and cleaned, then rinsed with water thoroughly. About two to three thin slices of each of these parts were made and then immersed in three separate solutions of 3.5% of 75 ml of hypochlorite until each plant tissue was completely cleared. Then, they were rinsed in distilled water, and different drops of different grades of ethanol; 50-100% were added in turn to dehydrate the cells (Hikmawanti *et al.* 2024). The thin slices of tissue preparations were later stained with drops of Safranin O in 50% alcohol for about five minutes before mounting in glycerine on glass slides. The epidermal layers were mounted on glass slides with the uppermost surfaces facing up, covered with coverslips, and ringed with nail varnish to prevent dehydration. The method was modified for examination of mid-rib, fruits, stem (TS), and root (TS) via free-hand sectioning with the aid of dissecting blade and counterstaining using Safranin O. Prepared slides were covered with a cover

slip and ringed with nail varnish to prevent dehydration. They were all carefully observed under the microscope, captured, and recorded. Different cell components, i.e., cork cells, sieve tube fibers, trichomes, vascular bundles, lignified fibers, cortex cells, calcium oxalate crystals, mesocarp, endocarp, and stomatal cells were looked out for, noted, and photography was done by using a digital camera (Seong *et al.* 2023). Photomicrographs of different sections were taken at different magnifications (between x4 and x100) using a binocular compound microscope with a Digital camera placed on its Eyepiece lens, and pictures were taken (Morris 2020).

2.3.2. Preparation of Extract and Fractions

Freshly collected *C. dependens* fruit was air-dried at room temperature, and the dried fruit samples were pulverized. The powdered plant material (500 g) was extracted into 70% aqueous methanol (2 L). The extract was concentrated in vacuo at 45°C, labeled as a crude extract, and stored in a refrigerator at 4°C until it was needed. Ten grams (10 g) of crude extract obtained above was fractionated into *n*-hexane, ethyl acetate (EtoAc), and aqueous soluble fractions, and the fractions were concentrated in vacuo at 45°C. The remaining dried crude extract and fractions obtained were stored in the refrigerator at 4°C until they were needed.

Percentage yield of the extracts (% w/w) were calculated using the formula below:

$$\% \text{ yield of extract} = \frac{\text{Weight of extract}}{\text{Weight of powdered plant material}} \times 100\%$$

2.3.3. Determination of 1,1-diphenyl-2-picryl hydrazyl (DPPH) Scavenging Activity

The DPPH radical scavenging ability of the crude extracts and fractions using standard DPPH radical scavenging assay (Ajaiyeoba *et al.* 2016). A stock solution (1,000 mg/ml) of the extract and fractions was prepared by dissolving 2 g of extract/fraction in 2 ml of methanol; serial dilutions of 200, 100, 50, 25, 12.5, and 6.25 µg/ml were prepared in the 96 well-plate, and 150 µL of 0.04 µg/ml DPPH in methanol was added to each well. The microplate plate was incubated for 30 minutes in the darkness at room temperature (25°C). The UV absorbance (Absorbance sample) of the resulting solution was measured at 517 nm using the Ultraviolet spectrophotometer (TitertekUniskan). The negative control wells contained DPPH and solvent (methanol) without the extract/fraction. Ascorbic acid was used as the positive control, while methanol was used as a negative control. This experiment was carried out in triplicate. The results obtained were expressed as percentage inhibition and calculated using the formula below.

$$\text{Inhibition (\%)} = \frac{\text{Absorbance control} - \text{Absorbance sample}}{\text{Absorbance control}} \times 100\%$$

2.3.4. Growth Inhibitory Activity

The extracts and fractions were subjected to growth inhibitory assays as described below:

Sorghum bicolor Radicle Growth Inhibitory Assay: Fresh seeds of *Sorghum bicolor* were purchased from the Ipata market in Ilorin. Viable seeds were selected by introducing 100 g of the seeds in distilled water. The floating seeds and husks were decanting off while the submerged seeds were collected, aired, and washed with 95% ethanol for a minute and later rinsed with distilled water before they were dried and stored in a dark place until needed. Only viable seeds were used for the *Sorghum bicolor* radicle growth (SBRG) inhibitory assay. Ten milliliters of 39.06, 156.25, 625, 2500, and 10,000 µg/ml were prepared by a fourfold dilution of the stock solution (40 mg/ml) prepared in 5% DMSO. Cyclophosphamide was used as the positive control at a concentration similar to the extract/fractions. A volume of 10 ml of different concentrations of the extracts (39.06-10,000 µg/ml) was transferred into a pre-labeled Petri dish of 10 cm lined with filter paper underlaid with wool. Thereafter, ten (10) seeds were placed on each filter paper in the Petri dishes. The Petri dishes were incubated in a dark cupboard at ambient temperature (32°C), and the lengths of emerging radicles were measured after 48 and 96 h of incubation. Seeds of the negative control group were treated with 10 mL of 5% DMSO in distilled water. This experiment was performed in triplicates. With the aid of a scaled ruler, changes in radicle length were measured. The mean change in radicle length per concentration was calculated. The percentage of radicle growth inhibition at 48 and 96 h and IC_{50} were determined (Salawu *et al.* 2020).

Percentage inhibition of the root growth was calculated using the formula below:

$$\% \text{ inhibition} = \frac{A - B}{A} \times 100\%$$

Where:
A : length of untreated root
B : length of treated root with plant extract, fractions/ cyclophosphamide

Allium cepa Root Growth Inhibitory Assay: *Allium cepa* root growth (ACRG) inhibitory assay was conducted using the method described in our earlier work (Salawu *et al.* 2020). Moderate-size onion bulbs (50±10 g) were obtained from the Ipata market. *Allium cepa* bulbs (50±10 g) were rinsed with water (distilled) and grown with the root submerged in water in the dark at room temperature (32°C) for 24-48 h until the roots had grown up to 3 cm in length.

Twenty (20) milliliters of different concentrations (39.06, 156.25, 625, 2,500, and 10,000 µg/ml) of each extract was prepared by four-fold dilution of the stock solution (40 mg/ml) prepared in 5% DMSO. Twenty (20 ml) of various concentrations were transferred into the Petri dish of 10 cm diameter, and the roots of each onion bulb were submerged in the extract/fraction in the Petri dishes containing each extract/fraction (39.06-10,000 µg/ml). Cyclophosphamide was used as the positive control at a similar concentration with the extract/ fractions, while 5% DMSO in water was applied as the negative control. This experiment was performed in triplicates. New root growth lengths were measured at 0, 48, and 96 h, and the percentage root growth inhibition was estimated by the formula below:

Percentage inhibition was calculated as follows:

$$\% \text{ inhibition} = \frac{A - B}{A} \times 100\%$$

Where:
A : length of untreated radicle
B : length of treated radicle with plant extracts/ cyclophosphamide

2.3.5. Cytotoxicity Assay

Brine shrimp lethality (BSL) assay: The eggs of Artemia salina were incubated in seawater for 48 h to hatch the eggs. Ten milligrams of the extract were dissolved in 2 ml 5% DMSO in seawater, resulting in a 5 mg/ml stock solution. Further dilutions of 1, 10, 100, 500, and 1,000 µg/ml were made in microplates in triplicates. A 250 µL suspension of nauplii in the extract was added to each well. The plates were incubated at room temperature (RT = 25-33°C) for 24 h. 0.5% DMSO in seawater was used as the negative control, and thereafter, the number of dead nauplii in each well was counted (Salawu *et al.* 2017).

The percentage cytotoxicity was estimated by the formula below:

$$\% \text{ inhibition} = \frac{A - B}{A} \times 100\%$$

Where:
A : the mean number of shrimp in negative control wells
B : the mean number of shrimp in the wells with plant extracts/cyclophosphamide

Determination of phytochemical and physicochemical properties.

2.3.6. Phytochemical Analysis

Preliminary Phytochemical Screening: The extracts and fractions of *C. dependens* fruit were screened for the presence of secondary plant metabolites as described below using the method previously reported (Sodeinde *et al.* 2019).

Gas chromatography-mass spectrometry (GC-MS) analysis: The phytochemical investigation of the *n*-hexane fraction of *C. dependens* was performed using GC-MS (Thermo Scientific Co.) equipment. The GC-MS instrument controlled parameters are as follows: Injection source (PAL Sampler), injection volume (2µL), column (Agilent technologies-HP-5MS), column dimension (360°C: 30 m × 250 µm × 0.25 µm), initial temperature (50°C), program temperature (260°C), initial pressure (9.05 psi), average velocity (38.724 cm/ sec), holdup time (1.2912 min), run time (82.286 min). The sample dissolved in HPLC grade hexane was run full and compared by using Database- Spectrum MS-NW-1798. Suggestions with the highest confidence interval greater than 80% were selected. Compounds were identified based on a comparison of their retention and mass spectra data to those generated under identical experimental conditions by applying a two-dimensional search algorithm, considering the retention index, as well as mass spectral similarity.

2.3.7. Physicochemical Analysis

Proximate Analysis: The AOAC (Nanda *et al.* 2003) and AOCS (Agroindustriais 2013) Standard procedures were used for the proximate analysis. The freeze-dried sample of the herbal remedies proximate analyses (moisture fiber, ash, lipids, proteins, and carbs) were identified. In summary, the weight difference method was used to determine the moisture and ash. The fiber content was measured by measuring the crucible's weight loss and its contents upon lighting. The method used to determine carbohydrates was different. We

deducted 100 from the total percentages of moisture, ash, crude protein, extract, and crude fiber. The micro Kjeldahl method was used to measure the nitrogen value of the material, which is the precursor for protein. The procedure involved digesting, distilling, and then titrating the sample. Protein was obtained by increasing the nitrogen value by a factor of 6.25. All the proximate values were presented in percentage (Nanda *et al.* 2003; Agroindustriais 2013).

Elemental Analysis: According to Robert and Gustav (Nanda *et al.* 2003), trace elements found in the herbal treatment solution were identified using an atomic absorption spectrometer (Model 230ATS) provided by Light Path Optical Ltd, United Kingdom. In short, the sample was treated to nitric acid digestion before being submitted to an atomic absorption spectrophotometer (Model: BUCK Scientific ACCUSYS211 AAS) with a cathode lamp that was changed to identify various metals. Every element has a unique wavelength of absorption, and no other element absorbs this wavelength.

2.3.8. Statistical Analysis

Data obtained were expressed as means ± SEM of values obtained in triplicates from three independent experiments. Statistical differences between treated groups and standards were evaluated using one-way analysis of variance (ANOVA). A p-value <0.05 was considered to be significant. Data were analyzed using GraphPad Prism Version 6.0 Software.

3. Results

3.1. Macroscopic and Organoleptic Description of *Chasmanthera dependens*

3.1.1. Macroscopic Description of Vegetative Parts of *C. dependens*

The macroscopic and organoleptic analysis of *C. dependent* plants undertaken in this research has yielded profound insights into important yardsticks that are essential in the proper identification of the plant. The plant *C. dependens* is an evergreen climber. It was found to climb up to an average of 640.07 and 875.34 cm in height and breadth, respectively, as shown in Figure 1-Aa and Table 1. The plant used in this study was found in semi-dried land. The leaves of the plant are evergreen. Upon incision of the stem, the juvenile stem produces faint white non-viscous exudates, while the mature stem produces no exudates upon incision. Interestingly, the leaves of *C. dependens* are arranged alternately on the same axis of the stem. The twinge of the plant measured

Table 1. Evaluation of macroscopic features of *Chasmanthera dependens* whole plant

Parameter	Observation/results	
Form	Climber	
Mean height (cm)	640.07±60.97	
Mean diameter of the canopy (cm)	875.34±10.17	
Habitat	Semi-dried	
Evergreen	Yes	
Type of exudes on insertion of the stem	Juvenile stem	Faint white
	Mature stem	Nil
Leave arrangement on the branch	Alternate	
Colour of twig	Greenish yellow	
Length of twig (cm)	311.67±4.41	
Root system (fibrous/taproot)	Aerial/ brace root system	
Root length (cm)	66.00±5.00	

an average of 311.67 cm in length. The plant was observed to have a brace root system, and an average length of 66 cm was measured, as shown in Figure 1H. An emerging aerial root system was also where the stem was cut off the primary trunk.

The leaf length of the plant was observed to range from 6.83±0.17 (juvenile leaf) to 21.00±0.00 cm (mature leaf) and a diameter ranging from 7.67±0.33 (juvenile leaf) to 19.00±0.33 cm (mature leaf) as shown Table 2 and Figure 1A. The leaves are evergreen and cordate in shape. The leaf has no characteristic smell but tastes initially slightly sweet with a bitter aftertaste. The abaxial leaf surface is greyish-green and appears rougher than the adaxial green surface. The venation system in the leaf is palmate and radiates from the leaf stuck up to the leaf margin. The leaf apex is characterized by a yellowish-brown aristate apex that measured an average of 0.30±0.0 (juvenile) to 0.90±0.0 cm (mature), as shown in Table 2 and Figure 1D. The leaf base appeared to be cordate (juvenile), which measured 1.60±0.10 cm, and lobate (mature), which measured 5.00±0.29 cm, as shown in Figure 1C. The leaf margin was entire, and the leaf had a characteristic yellowish-green petiolate that measured up to 4.00±0.00 cm (juvenile) to 18.50±0.76 cm, as shown in Table 2 and Figure 1E.

Chasmanthera dependens was observed to have a stem that is cylindrical and appears smooth and green when young (diameter: 0.57±0.03 cm) or rigid, woody, and brownish-grey when mature (diameter: 7.00±0.00 cm) as shown in Table 3 and Figure 1F. There are lenticels on the young and mature stem bark that appear as white scars. The young and mature stems taste slightly sweet with a bitter aftertaste, and breaking both the young and mature displayed a bulking type of fracture, as shown in Figure 1G.

Table 2: Evaluation of macroscopic features of *Chasmanthera dependens* leaf

Parameter	Observation/results	
Leaf length (cm)	Juvenile	6.83±0.17
	Mature	21.00±0.00
Leaf diameter (cm)	Juvenile	7.67±0.33
	Mature	19.00±0.33
Shape	Cordate	
Color	Green	
Odor	Odourless	
Taste	Initially slightly sweet with a bitter aftertaste	
Adaxial surface texture	Rough	
Adaxial surface colour	Light green	
Abaxial surface texture	Rougher than Adexial Surface	
Abaxial surface color	Greyish green	
Venation system	Palmate	
Apex type	Aristate	
Apex length (cm)	Juvenile	0.30±0.0
	Mature	0.90±0.00
Base type	Juvenile	Cordate
	Mature	Lobate
Base length (cm)	Juvenile	1.60±0.10
	Mature	5.00±0.29
Leaf margin	Entire	
Petiolate length (cm)	Juvenile	4.00±0.00
	Mature	18.50±0.76

Table 3. Evaluation of macroscopic features of *Chasmanthera dependens* stem

Parameter	Observation/results	
Stem bark color	Juvenile	Green
	Mature	Brownish grey
Fracture (transverse or longitudinal)	Bulking	
Odor	odorless	
Taste	slightly sweet with a bitter aftertaste	
Texture	Juvenile	hairy and soft
	Mature	dry, rough with white patches
Diameter	Juvenile	0.57±0.03
	Mature	7.00±0.00
Shape	Cylindrical	

Table 4. Evaluation of macroscopic features of *Chasmanthera dependens* flower and whole fruit

Parameter	Observation/results	
Flower colour	Yellow	
Flower arrangement on flower stalk	Alternate	
The mean length of flower stalk (cm)	7.53±1.34	
The mean number of lower per stalk	21.00±3.33	
Mean diameter of lower (cm)	4.02±0.23	
Calyx type	Caducous, sepaloids, polysepalous	
Number of sepals	3±0.00	
Corolla type	Yellow color, regular, polypetalous	
Number of petals	3±0.00	
Carpel	three yellow apocarpous	
Stamen	three brown color	
Stem bark color	Mature:	Green
	Ripe:	Cherry
Shape	Oval	
Odor	Odorless	
Taste	Slight sweet with a bitter aftertaste	
Mean diameter (cm)	1.23±0.07	
Mean weight of fresh fruit (g)	11.99±0.35	
Mean weight of dried fruit (g)	3.60±0.15	
Moisture content (%w/w)	70.00±0.57	
Texture	Mature	Smooth and hard
	Ripe	Smooth and succulent
Mean seed diameter (cm)	0.93±0.03	
Mean seed weight (g)	0.42 ±0.02	
Seed shape	Oval	

3.2. Macroscopic Description of Floral Parts of *C. dependens*

Chasmanthera dependens is characterized by a yellowish-green floral system that consists of about 21.00±3.33 flowers per flower stalk that is 7.53±1.34 cm long. Each flower is yellowish and arranged in alternate. They are about 4.02±0.23 in diameter, as shown in Table 4 and Figure 1I. The young flower bud has three free sepaloids that weather on the opening of the flower cup. The carpel is composed of three yellow-colour polypetalous regular petals surrounding the three apocarpous carpels that are yellow-colored with three stamens that appear brownish, as shown in Table 4 and Figure 1J. In the rainy season of July to September, the plant is characterized by ripe (cherry-colored) and unripe (green-colored) berries that are oval, measure a diameter of 1.23±0.07 cm and weigh 11.99±0.35 (fresh) or 3.60±0.15 (dried). The fresh fruit has an approximate moisture content of 70.00±0.57 %w/w. The fruit has an oval-shaped woody seed that measures 0.93±0.03cm in diameter and weighs 0.42 ±0.02g, as shown in Table 4.

3.3. Microscopic Description of Various Parts of *C. dependens*

Microscopic evaluation of the leaf (adaxial and abaxial) and the transverse section of the leaf, stem, and root of *C. dependens* revealed the following details. The adaxial surface of the plant was observed to be characterized by covering epidermal trichomes consistently surrounded by eight (8) basal cells, as shown in Figure 2M. The epidermal cells are irregular in shape with straight cells, as shown in Figure 2L. Observation of the adaxial surface revealed a vascular system that is made up of tubular-shaped cells with straight cell walls, as shown in Figure 2N. No epidermal stomata were observed on the adaxial leaf surface. Interestingly, the abaxial leaf surface is rich in covering trichomes with an embedded type of base. The trichomes are observed to be embedded in the epidermal vascular tissues, as shown in Figures 2O and P. The epidermal cells are irregular in shape, similar to the adaxial epidermal cells. Unlike the adaxial epidermis, the cell walls of the abaxial epidermis are wavy. An anomocytic type of stomata characterizes the lower/abaxial leaf surface. The guard cells are surrounded by four irregular subsidiary cells, as shown in Figure 2Q.

The transverse section of the leaf showed that the mid-rib is composed of several embedded covering trichomes and conspicuous vascular tissues, as shown in Figure 2R. The mid-rib is observed to be covered by a thin cuticular layer surrounding tubular lower epidermal cells, as shown in Figure 2S. Directly adjacent to the lower epidermal cells are collenchymas cells that surround the vascular bundles. The transverse section of the *C. dependens* stem revealed a wavy pattern endodermis system. The stem of *C. dependens* is characterized by a closed collateral type of vascular bundle with the phloem adaxial to the xylem. Eighteen vascular systems were observed in a ring arrangement. The xylem is marked by metaxylem and protoxylem. Similarly, oval-shaped pericycle cells are obvious, as shown in Figure 3U. The transverse section of *C. dependens* root revived thick cuticular layer and non-wavy endodermis enclosing six (6) xylem conducting systems arranged in a circular pattern. However, the phloem fiber was observed to surround the around the xylem and a pith.

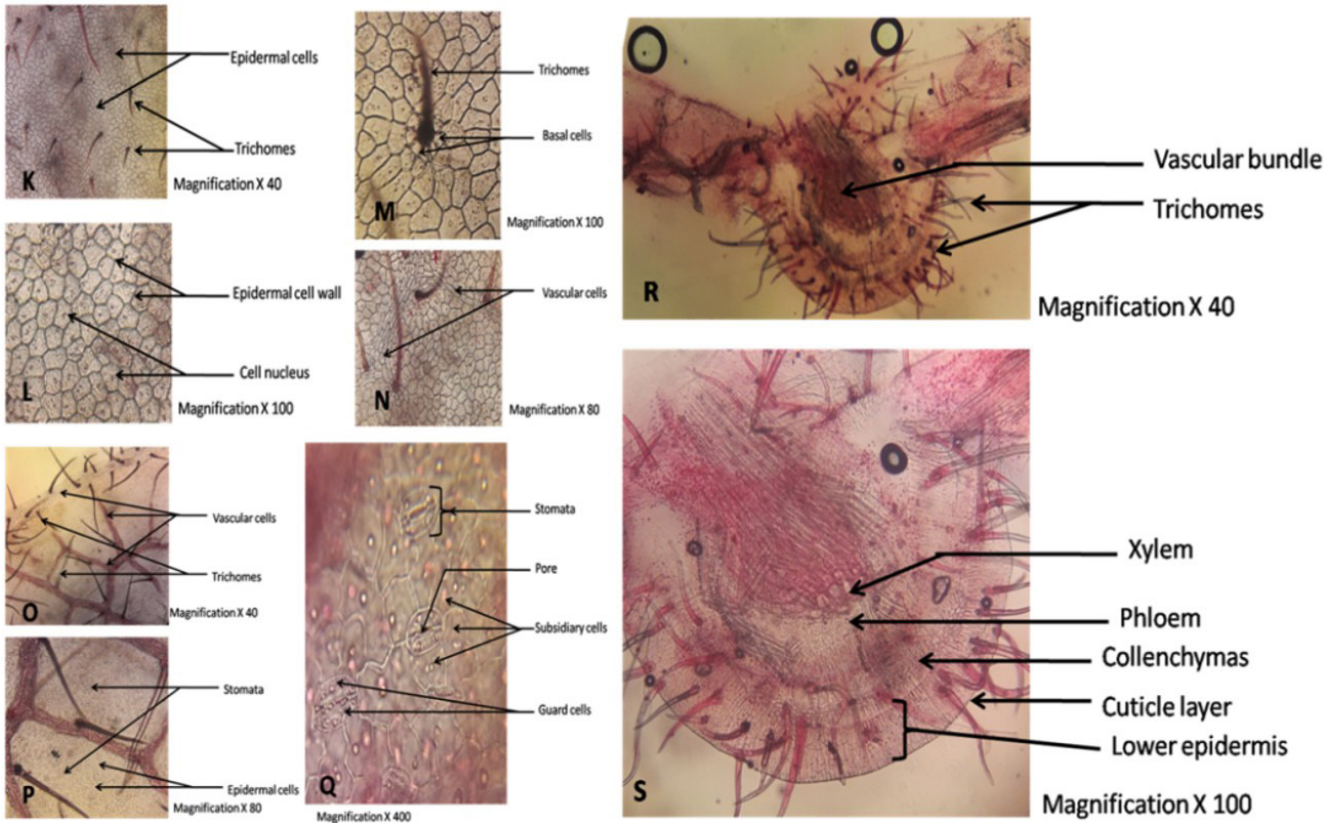

Figure 2. Microscopic features of the (K-N) adaxial leaf surface, (O-Q) abaxial leaf surface, and (R-S) leaf transverse section of *Chasmanthera dependens*

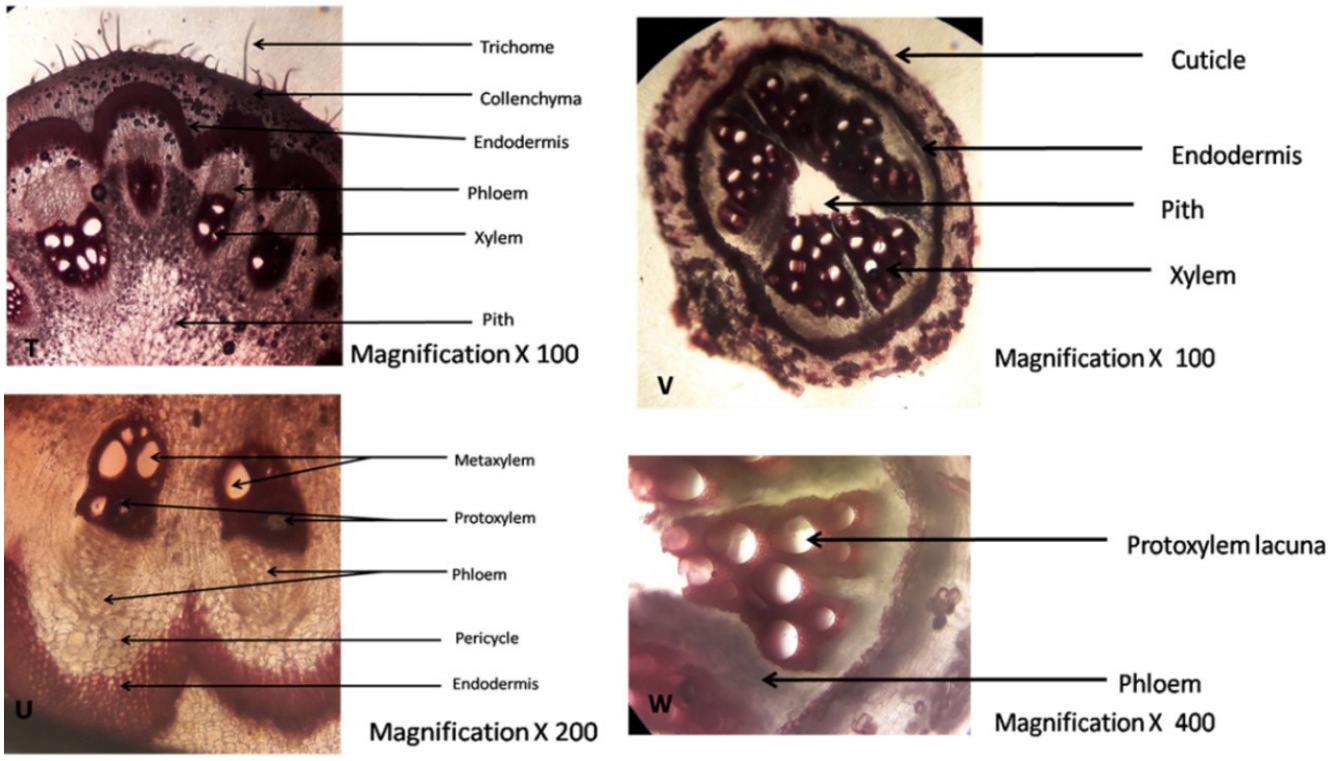

Figure 3. Microscopic features of (T-U) stem transverse and (V-W) root transverse sections of *Chasmanthera dependens*

3.4. Bioactivity of the Extract and Fractions of *Chasmanthera dependens* Whole Fruit

3.4.1. DPPH Radical Scavenging Activities

The extract, as well as the positive control (Ascorbic acid), displayed concentration-dependent DPPH radical scavenging activities. The positive control (IC_{50} = 9.67±0.50 µg/ml) displayed higher DPPH radical scavenging than the extract and fractions, as shown in Table 5. However the fractions demonstrated different degrees of DPPH radical scavenging activities with the ethyl acetate fraction (43.92±1.25 µg/ml) displaying the highest activities compared to crude extract (IC_{50} = 57.03±1.24 µg/ml), *n*-hexane (IC_{50} = 254.03±35.28 µg/ml) and aqueous fractions (IC_{50} = 284±16.89 µg/ml).

3.4.2. *Sorghum bicolor* Radicle and *Allium cepa* Root Growth Inhibition

The extract and fractions displayed concentration-dependent growth inhibition in the *Sorghum bicolor* radicle growth and *Allium cepa* root growth inhibitory assays. The aqueous fraction (43.73±1.38 µg/ml) displayed the highest growth inhibitory effects in the *Sorghum bicolor* radicle growth inhibitory assay with an IC_{50} value compared to the crude extract but better than the positive control (cyclophosphamide with IC_{50} = 690.25±13.32µg/ml) and the *n*-hexane (IC_{50} = 413.28±9.13 µg/ml) and ethyl acetate (IC_{50} = 179.59±11.47 µg/ml) fractions as shown in Table 5. Similarly, in the *Allium cepa* root growth inhibitory assay, the crude extract (IC_{50} = 22.00±4.22 µg/ml) and aqueous fraction (IC_{50} = 43.73±1.38 µg/ml) displayed a better growth inhibitory effect than cyclophosphamide (IC_{50} = 211.70±8.12µg/ml), and *n*-hexane (IC_{50} = 265.50±53.51µg/ml) and ethyl acetate (IC_{50} = 211.70±8.12µg/ml) fractions.

3.4.3. Brine Shrimps Cytotoxicity

The extract and fractions displayed concentration-dependent cytotoxicity on the brine shrimps, with the aqueous fraction (LC_{50} = 99±6.08 µg/ml) having similar cytotoxicity to cyclophosphamide (LC_{50} = 98±3.33 µg/ml) as shown in Table 5. However, crude extract (LC_{50} = 176±3.76 µg/ml), *n*-hexane (LC_{50} = 441±21.35 µg/ml), and ethyl acetate (LC_{50} = 238±5.37 µg/ml) fractions displayed weak brine shrimp lethality compared to cyclophosphamide.

3.5. Phytochemical Evaluation

3.5.1. Preliminary Phytochemical Screening

Qualitative phytochemical screenings of the extract and fractions of *Chasmanthera dependens* revealed the presence of various secondary metabolites. The

Table 5. Yield (% w/w), antioxidant, growth inhibitory, and cytotoxic activities of *Chasmanthera dependens* fruit extracts and fractions

Samples	Yield (%$^{w}/_{w}$)	DPPH radical scavenging IC_{50}±SEM (µg/ml)	Growth inhibition (µg/ml)		Cytotoxicity (µg/ml)
			SBRGI GI_{50}±SEM	*ACRGI* GI_{50}±SEM	BSL LC_{50}±SEM
Crude extract	10.78	57.03±1.34*	48.00±2.65*	48.00±2.65*	176±3.76*
n-hexane	2.96	254±35.00*	413.28±9.13	413.28±9.13	441±21.35*
Ethyl acetate	0.72	43±1.25*	179.59±11.47*	179.59±11.47*	238±5.37*
Aqueous	0.83	284±16.89*	43.73±1.38*	43.73±1.38*	99±6.08*
Standard drug		9.67±0.50^{+}	690.25±13.32^{++}	155.84±0.37^{++}	98±3.33^{++}

SBRGI: *Sorghum bicolor* radicle growth inhibition, ACRGI: *Allium cepa* root growth inhibition, *significantly different from standard drug ($P<0.05$), $^{+}IC_{50}$ of ascorbic acid, $^{++}$: IC_{50} of cyclophosphamide

crude extract possesses saponins, tannins, alkaloids, anthraquinones, flavonoids, terpenoids, carbohydrates, and reducing sugar, as shown in Table 6. The *n*-hexane fraction was rich in saponins, alkaloids, anthraquinones, and terpenoids. On the other hand, ethyl acetate fraction is rich in saponins, tannins, alkaloids, cardiac glycoside, anthraquinones, flavonoids, terpenoids, carbohydrates, and reducing sugar. At the same time, the aqueous fraction contained saponins, tannins, anthraquinones, flavonoids, carbohydrates, and reducing sugar.

3.5.2. GC-MS Analysis

However, the GC-MS analysis of the *n*-hexane fraction of *Chasmanthera dependens* led to the identification of 34 bioactive compounds (Figure 4 and Table 7). The most abundant constituents of the fraction included cis-13-octadecenoic acid 87.11% occurrence), Octadecanoic acid (5.53 % occurrence), Oleic acid, methyl ester (1.64 % occurrence), Glycerol 2-monooleate (1.63 % occurrence), and lupeol (0.82 % occurrence). Other compounds includeObtusifoliol, β-Sitosterol, Cycloeucalenol 24-methylene cycloartenol, etc., as shown in Table 7.

3.6. Elemental and Proximate Composition of *Chasmanthera dependens* Fruit

The mineral composition of sesame seed is presented in Table 8. The magnesium content (0.53±0.07 mg/100 g) was the highest. This was followed by Manganese (0.47±0.02 mg/100 g), zinc (0.42±0.07 mg/100 g), copper (0.17±0.03 mg/100 g), and Cadmium (0.01±0.00 mg/100 g).

The proximate composition of *Chasmanthera dependens* fruit is presented in Table 9. The fiber content was the highest, constituting 40.26±0.80% of the entire fruit. The fruit also contains significant amounts of carbohydrate (29.15±0.16%), ash (12.10±0.31%), moisture (9.94±0.21%), lipid (4.73±0.02%) and protein (3.83±0.46%).

Table 6. Phytochemical examination of *Chasmanthera dependens* fruit extract and fractions preparation

Constituents	Relative abundance			
	Crude extract	*n*-hexane fraction	Ethyl acetate fraction	Ethyl acetate fraction
Saponins	+	+	+	++
Tannins	+++	-	+	+
Alkaloids	+++	+	++	-
Cardiac glycosides	-	-	+	+
Free anthraquinones	+	+	+	+
Combined anthraquinones	+	-	+	+
Flavonoids	++	-	+++	+
Terpenoids	++	++	+	-
Carbohydrate	++	-	+	++
Reducing sugar	+	-	+	++

-: absence of component, +: trace presence of component, ++: moderate amount of component, +++:copious amount of component

4. Discussion

The therapeutic potential of medicinal plants continues to appreciate and evolve alongside modern medicine, and many more people depend on herbal medicine for their health care (Ghosh *et al.* 2023). The quality of herbal medicine is largely dependent on the caliber of the raw materials used in the preparation of the medicine. Often, error occurs with the selection of medicinal plants because, in many cases, plants of the same family and genus look alike and are often referred to by the same local names, resulting in the selection of the wrong plant and preparation of products with unwanted activity and toxicity (Choudhury *et al.* 2023). Hence, there is an increasing desire to standardize medicinal plants, particularly those with valuable therapeutic potential, through pharmacognostic evaluation, which involves organoleptic–microscopic evaluations and physicochemical and phytochemical assessments of the plant to provide yardsticks for proper identification of the desired medicinal plant (Wang *et al.* 2023b).

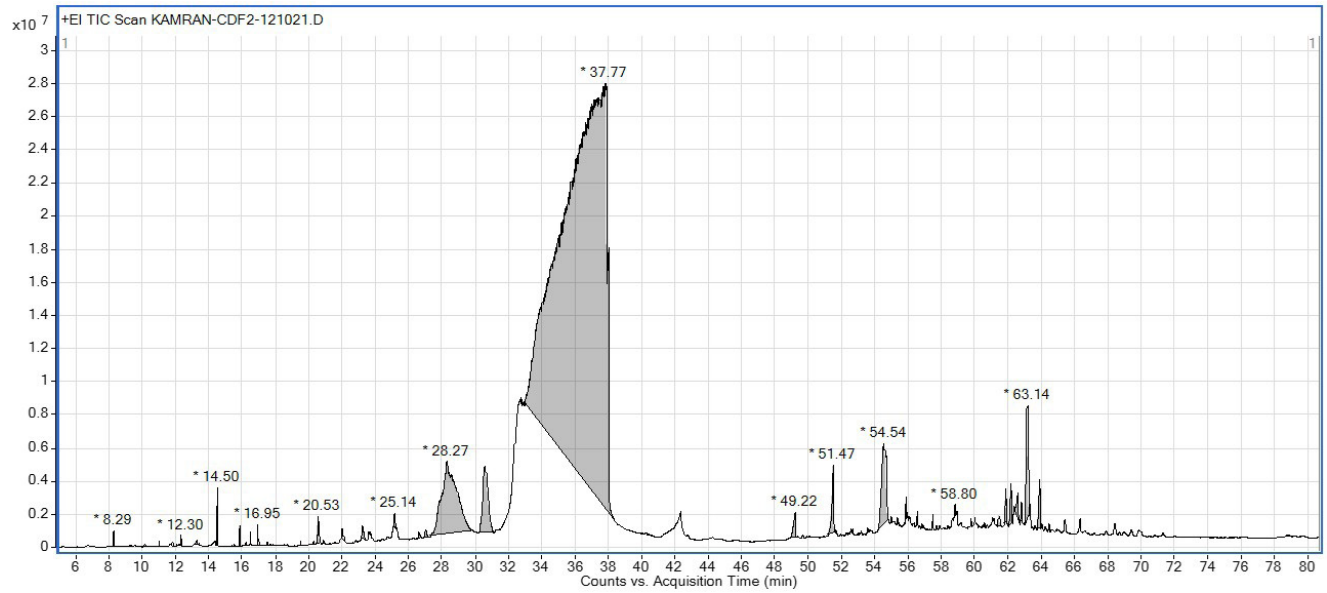

Figure 4. GC-MS Chromatograph of the n-hexane fraction of *Chasmanthera dependens* fruit

Table 7. GC-MS analysis of the *n*-hexane fraction of *Chasmanthera dependens* fruit

Compound	RT	Area sum %
4-Pentenal, 2-ethyl-	8.29	0.04
4-Propylcyclohexylamine	10.18	0.02
2,6-Octadiene, 2,6-dimethyl-	11.01	0.01
1,1-Ethanediol, diacetate	11.81	0.02
Nonanal	12.3	0.02
2-Hydroxy-4-methyl pentanoic acid	13.28	0.03
Tridecane	14.5	0.13
.(Z)-2-Decenal	15.83	0.04
2,4-Decadienal	16.49	0.03
2,4-Decadienal, (E,E)-	16.95	0.06
Tridecanal	20.53	0.19
Nonadecane	22.01	0.07
13-Tetradecenal	23.22	0.11
1-Hexadecanol, 2-methyl-	23.63	0.01
Tetradecane, 2,6,10-trimethyl-	23.71	0.01
Oleic Acid	25.06	0.02
Eicosane	25.14	0.08
7-Methyl-Z-tetradecen-1-ol	26.6	0.05
Oxiraneundecanoic acid, 3-pentyl-, methyl ester,	27.02	0.07
Octadecanoic acid	28.27	5.53
Oleic acid, methyl ester	30.58	1.64
cis-13-Octadecenoic acid	37.77	87.11
2-Hydrazino-2-imidazoline	49.22	0.28
1,2-Benzenedicarboxylic acid, diisooctyl ester	51.47	0.62
Glycerol 2-monooleate	54.54	1.63
13-Docosenamide	55.87	0.08
Glycerin 1-monooleate	58.8	0.07
Ethyl iso-allocholate	61.44	0.07
Obtusifoliol	61.83	0.18
β-Sitosterol	62.16	0.29
9,19-Cycloergost-24(28)-en-3-ol, 4,14-dimethyl-, acetate, (3β,4α,5α)-	62.58	0.2
Cycloeucalenol	62.8	0.1
Lupeol	63.14	0.82
24-Methylenecycloartanol	63.88	0.36

Table 8. Elemental analysis of *Chasmanthera dependens* fruit

Minerals	Composition (mg/100 g)
Zinc (Zn)	0.42±0.07
Copper (Cu)	0.17±0.03
Chromium (Cr)	ND
Lead (Pb)	ND
Cadmium (Cd)	0.01±0.00
Magnesium (Mg)	0.53±0.07
Nickel (Ni)	ND
Manganese (Mn)	0.47±0.02

Table 9. proximate analysis of *Chasmanthera dependens* fruit

Chemical composition	Composition (%)
Moisture content	9.94±0.21
Carbohydrates	29.15±0.16
Crude lipid	4.73±0.02
Crude protein	3.83±0.46
Crude fibre	40.26±0.80
Total ash	12.10±0.31

Up until now, there are no reports in the literature for the pharmacognostic description of *Chasmanthera dependence* or any member of the genus *Chasmanthera*; however, pharmacognostic study of some members of the Menispermaceae has been performed, including *Triclisia subcordata* Oliv (Sonibare and Adebodun 2018). Macroscopic evaluation of *C. dependence* showed that the plant is a climber with an evergreen leave arising from a green flexible twig. The leaves arising from the twig are generally cordate in shape with an aristate apex and lobate base. The climbing adventitious twig of *C. dependens* arises from a greyish-brown woody stem. During the flowing season, *C. dependens* bears yellowish flowers with three apocarpous petals, which bear a cherry color small berry fruit on fertilization.

The microscopic examination of *C. dependens* leaf, stem, and root revealed various characteristics. The adaxial surface is characterized by covering epidermal trichomes surrounded by eight basal cells, while the abaxial surface is rich in covering trichomes with an embedded base. The epidermal cells are irregular in shape and have a wavy cell wall. The stomata (anomocytic type) were found on the abaxial surface, similar to those found in *T. subcordata*. However, *T. subcordata* bears paracytic stomata (Sonibare and Adebodun 2018). The stomata are surrounded by four subsidiary cells. The transverse section of the leaf shows a mid-rib composed of embedded trichomes and conspicuous vascular tissues, covered by a thin cuticular layer surrounding tubular lower epidermal cells. The stem of *C. dependens* has a wavy pattern endodermis system, with well-developed vascular systems in a ring arrangement similar to those of *T. subcordata* (Sonibare and Adebodun 2018). The root has a thick cuticular layer and non-wavy endodermis enclosing six xylem conducting systems arranged in a circular pattern. The phloem fiber surrounds the xylem and a pit. The pith of the root is bigger than that of the stem, suggesting that it forms a canal throughout the root system.

The *C. dependens* fruit extract demonstrated positive DPPH radical scavenging activity. However, the ethyl acetate fractions possess a higher antioxidant effect compared to the crude extract and other fractions. Phytochemical screening of the fractions showed that the ethyl acetate fraction is rich in flavonoids. Flavonoids and phenols are known to possess free radicals scavenging ability (Hassanpour and Doroudi 2023). It can be extrapolated that the metabolites in the extract and fractions are responsible for the antioxidant activity. In an earlier study, the root extract of *C. dependens* displayed antioxidant activity (Enenebeaku *et al.* 2022; Nwanelo *et al.* 2023). The extract of *C. dependens* displayed growth inhibitory activity and brine shrimp cytotoxicity. However, the aqueous fraction displays consistent activities in growth inhibitory and brine shrimp cytotoxicity, which may be attributed to the high presence of saponins in the aqueous fraction of the crude extract. All saponins, both aglycone and sugar part, play an important role in cytotoxic activity and inhibit cell proliferation and growth (Podolak *et al.* 2010). Interestingly, the GS-MS analysis of the *n*-hexane fraction of *C. dependens* revealed that octadecenoic is the dominant compound. Octadecenoic acid has been reported to result in a dose-dependent reduction in the number of tumor cells and significantly inhibited cell proliferation (Yu *et al.* 2008). Plants with a high content of octadecenoic acid have been known to possess marked antioxidant and antiproliferative activities (Reza *et al.* 2021). Studies have shown that many non-polar fractions or extracts rich in terpenic compounds and fatty acids have been known to demonstrate potent antioxidant and growth-inhibitory effects (Mellado *et al.* 2019; Burkett *et al.* 2022). The other compounds identified in the *n*-hexane fraction, such as Obtusifoliol (Rattanapunya *et al.* 2021), β-Sitosterol (Baskar *et al.* 2012), and Cycloeucalenol 24-methylene cycloartenol (Pal and Raj 2023) are known to possess some significant levels of antioxidant and growth-inhibitory activities.

Elemental and proximate analysis of the *C. dependens* fruit indicated that the fruit is rich in important micronutrients such as zinc, copper, cadmium, magnesium, manganese, protein, lipids, and fiber. The result of the

element and proximate analysis of the *C. dependens* is similar to those obtained from *Sphenocentrum jollyanum*, a member of the Menispermaceae family (Akinwumi and Sonibare 2022). However, despite the rich nutritional content of the *C. dependens* fruit, the fruit is not locally consumed as food probably because of its initial bitter taste rising from the high alkaloid content (Muñoz *et al.* 2020).

In conclusion, this study has revealed for the first time the pharmacognostic features of *Chasmanthera dependens*. The macroscopic and microscopic parameters identified for the whole plant, leaf, stem, root, and flower may be useful in the preparation standards for the proper identification of *Chasmanthera dependens*. The extracts and fractions demonstrated antioxidant and growth inhibitory effects, and chemical analysis shows that the fruit is rich in phytoconstituent and micronutrients with important activities.

References

Afiong, N.N., Fils, P.B., Guekam, K.K., Muhesi, E.K., Martin, E.A., Brull, G.R., Fa, J.E., Funk, S.M., Fedoung, E.F., Betti, J.L., 2024. Traditional use of medicinal plants confirmed by the baka in Southern and Eastern cameroon. *Journal of Biosciences and Medicines*, 12, 76-106. https://doi.org/10.4236/jbm.2024.128007

Agroindustriais, P., 2013. Aoac. Official methods of analysis of the association of official analytical chemists. Caracterização, Propagação E Melhoramento Genético De Pitaya Comercial E Nativa Do Cerrado, 26, 62.

Ajaiyeoba, E., Salawu, K., Ogbole, O., Adeniji, J., 2016. Radical scavenging and cytotoxicity evaluation of eight extracts of citrus limon and citrus aurantifolia. *Nigerian Journal Of Natural Products And Medicine*. 20, 121-127. https://doi.org/10.4314/njnpm.v20i1.20

Akinwumi, I., Sonibare, M., 2022. Sphenocentrum jollyanum pierre (Menispermaceae): from traditional medicine to pharmacological activity and chemical constituents. *Trends In Phytochemical Research*. 4, 301.

Baskar, A.A., Al Numair, K.S., Gabriel Paulraj, M., Alsaif, M.A., Muamar, M.A. Ignacimuthu, S., 2012. B-Sitosterol prevents lipid peroxidation and improves antioxidant status and histoarchitecture in rats with 1, 2-dimethylhydrazine-induced colon cancer. *Journal Of Medicinal Food*. 15, 335-343. https://doi.org/10.1089/jmf.2011.1780

Burkett, W., Zhao, Z., Newton, M., Zhou, C., Bae-Jump, V., 2022. Palmitic acid, a common long-chain saturated fatty acid, demonstrates anti-proliferative and anti-tumorigenic effects in pre-clinical models of endometroid endometrial cancer (262). *Gynecologic Oncology*. 166, 140. https://doi.org/10.1016/S0090-8258(22)01483-4

Choudhury, A., Singh, P.A., Bajwa, N., Dash, S., Bisht, P., 2023. Pharmacovigilance of herbal medicines: concerns and future prospects. *Journal Of Ethnopharmacology*. 309, 116383. https://doi.org/10.1016/j.jep.2023.116383

Codo Toafode, N.M., Oppong Bekoe, E., Vissiennon, Z., Ahyi, V., Vissiennon, C., Fester, K., 2022. Ethnomedicinal information on plants used for the treatment of bone fractures, wounds, and sprains in the Northern Region of The Republic of Benin. *Evidence-Based Complementary And Alternative Medicine*. 2022, 8619330. https://doi.org/10.1155/2022/8619330

Das, M.K., Silpavathi, L., Das, D., 2021. Pharmacognosy and *in-vivo* anticancer potential of an indigenous marine macroalga, ulvafasciata delile from visakhapatnam coast, India. *Advances in Traditional Medicine*. 21, 433-442. https://doi.org/10.1007/s13596-021-00594-3

Enenebeaku, C.K., Ogukwe, C.E., Nweke, C.O., Anyado-Nwadike, S.O., Obi, M., Duru, I.A., Enenebeaku, U.E., Ogidi, O.I., 2022. Antiplasmodial and *in vitro* antioxidant potentials of crude aqueous and methanol extracts of *Chasmanthera dependens* (Hochst). *Bulletin of the National Research Centre* 46, 33. https://doi.org/10.1186/s42269-022-00721-3

Folami, S.O., Abuga, I., Balogun, M.E., Lawal, A.S., Lawal, A.Z., Adetoyi, M.G., Abubakar, M.F., 2024. Resident altitude about the usage of herbal medicine in Kwara and Lagos Metropolis, Nigeria. *Journal of Research In Complementary Medicine*. 1, 81-81. https://doi.org/10.5455/JRCM.20240624082005

Ghosh, S., Bishal, A., Ghosh, S.K., Jana, K., Gayen, B., Sahu, S., Debnath, B., 2023. Herbal medicines: a potent approach to human diseases, their chief compounds, formulations, present status, and future aspects. *International Journal of Membrane Science and Technology*. 10, 442-464. https://doi.org/10.15379/ijmst.v10i1.2608

Govaerts, R., 1999. *World Checklist of Seed Plants 3 (1, 2a & 2b): 1-1532*. Mim, Deurne.

Hameed, A., Ghani, N., Mughal, T. A., Abbas, M., Abrar, A., Javed, H., 2023. Pharmacognostical evaluation and physiochemical analysis of salsola kali as medicinal plant. *Microscopy Research and Technique*. 86, 1322-1332. https://doi.org/10.1002/jemt.24316

Hassanpour, S.H., Doroudi, A., 2023. Review of the antioxidant potential of flavonoids as a subgroup of polyphenols and partial substitute for synthetic antioxidants. *Avicenna Journal of Phytomedicine*. 13, 354.

Hikmawanti, N.P.E., Saputri, F.C., Yanuar, A., Ningrum, R.A., Mun'im, A., Hayati, H., 2024. Microscopical evaluation and TLC analysis of *Pluchea indica* (L.) less: leaf, stem, and root. *Hayati Journal of Biosciences*, 31, 71-81. https://doi.org/10.4308/hjb.31.1.71-81

Iyiola, A.O., Adegoke Wahab, M.K., 2023. Herbal medicine methods and practices in Nigeria, in: Izah, S.C., Ogwu, M.C., Akram, M. (Eds.), *Herbal Medicine Phytochemistry. Reference Series in Phytochemistry*. Springer, Cham, pp. 1-34 https://doi.org/10.1007/978-3-031-21973-3_47-1

Jamtsho, T., Yeshi, K., Perry, M.J., Loukas, A., Wangchuk, P., 2024. Approaches, strategies and procedures for identifying anti-inflammatory drug lead molecules from natural products. *Pharmaceuticals*. 17, 283. https://doi.org/10.3390/ph17030283

Kola-Mustapha, A., Yohanna, K., Ghazali, Y., Ayotunde, H., 2020. Design, formulation and evaluation of *Chasmanthera dependens* Hochst and *Chenopodium ambrosioides* Linn based gel for its analgesic and anti-inflammatory activities. *Heliyon.* 6, 1-8. https://doi.org/10.1016/j.heliyon.2020.e04894

Lamichhane, S., Sahariah, B.J., Das, B., Khatiwara, D., Sola, P., Adhikari, R.P., Gogoi, B., Alam, T., 2023. Herbal drug standardization: a systemic review. *Journal of Drug Delivery and Therapeutics*. 13, 149-153. https://doi.org/10.22270/jddt.v13i4.6012

Madueke, A., Nwanelo, V., Tabansi, E., Onoh, P., Anichebe, R., Okosisi, A., Anosike, A., 2020. Evaluation of antioxidant properties of choloroform extract of *Chasmanthera dependens* roots. *Med. Sci. Forum.* 2, 21. https://doi.org/10.3390/CAHD2020-08606

Madueke, C.A., Anosike, C.A., 2017. Anti-ulcerogenic effect of the chloroform extract of *Chasmanthera dependens* on indomethacin and aspirin-induced ulcer in rats. *Journal of Nutritional Ecology and Food Research*. 4, 131-137. https://doi.org/10.1166/jnef.2017.1164

Mellado, M., Soto, M., Madrid, A., Montenegro, I., Jara-Gutiérrez, C., Villena, J., Werner, E., Godoy, P., Aguilar, L.F., 2019. *In vitro* antioxidant and antiproliferative effect of the extracts of ephedra chilensis K presl aerial parts. *Bmc Complementary And Alternative Medicine*. 19, 1-10. https://doi.org/10.1186/s12906-019-2462-3

Morris, K., 2020. *Modern Microscopic Methods of Bioaerosol Analysis*, first ed. Bioaerosols Handbook. Crc Press. https://doi.org/10.1201/9781003070023-11

Muñoz, I.J., Schilman, P.E., Barrozo, R.B., 2020. Impact of alkaloids in food consumption, metabolism and survival in a blood-sucking insect. *Scientific Reports*, 10, 9443. https://doi.org/10.1038/s41598-020-65932-y

Nafiu, M., Hamid, A., Muritala, H., Adeyemi, S., 2017. Preparation, standardization, and quality control of medicinal plants in Africa. *Medicinal Spices and Vegetables from Africa*, 171-204. https://doi.org/10.1016/B978-0-12-809286-6.00007-8

Nalawade, A.S., Gurav, R.V., Patil, A.R., Patwekar, M., Patwekar, F., 2022. A comprehensive review on morphological, genetic and phytochemical diversity, breeding and bioprospecting studies of genus *Chlorophytum* Ker Gawl. from India. *Trends In Phytochemical Research*. 6, 19-45.

Nanda, V., Sarkar, B., Sharma, H., Bawa, A., 2003. Physico-chemical properties and estimation of mineral content in honey produced from different plants in Northern India. *Journal of Food Composition and Analysis*. 16, 613-619. https://doi.org/10.1016/S0889-1575(03)00062-0

Ndhlovu, P.T., Asong, J.A., Omotayo, A.O., Otang-Mbeng, W., Aremu, A.O., 2023. Ethnobotanical survey of medicinal plants used by indigenous knowledge holders to manage healthcare needs in children. *Plos One*, 18, E0282113. https://doi.org/10.1371/journal.pone.0282113

Nwanelo, V., Agwu, L., Ugwuoke, S., Osuji, D., Odo, C., Aguchem, R., Nwangene, K., Ngwu, L., Ekemezie, C., Chikelue, S., 2023. An assessment of the antinociceptive effect of methanolic root extract of *Chasmanthera dependens*. *Letters in Applied NanoBioScience.* 12, 176.

Okaiyeto, K., Oguntibeju, O.O., 2021. African herbal medicines: adverse effects and cytotoxic potentials with different therapeutic applications. *International Journal of Environmental Research and Public Health*. 18, 5988. https://doi.org/10.3390/ijerph18115988

Olanipekun, M.K., 2023. Ethnobotanical relevance and conservation of medicinal plants used to treat human diseases in ifedore, ondo-state, Nigeria. *Asian Journal of Ethnobiology*. 6, 7-19. https://doi.org/10.13057/asianjethnobiol/y060102

Olaniran, A.D., Morenikeji, E.S., Adeyemi, O.A., Anuoluwapo, T.O., Omoniyi, O.S., 2018. The doctrine of signatures in herbal prescriptions in ikale and ilaje communities of ondo state, Southwestern Nigeria. *J. Med. Plants Res*. 12, 222-227. https://doi.org/10.5897/JMPR2018.6576

Olorunnisola, O., Adetutu, A., Balogun, E., Afolayan, A., 2013. Ethnobotanical survey of medicinal plants used in the treatment of malarial in ogbomoso, Southwest Nigeria. *Journal of Ethnopharmacology*. 150, 71-78. https://doi.org/10.1016/j.jep.2013.07.038

Osawaru, M., Ogwu, M., Ahana, C., 2013. Current status of plant diversity and conservation in Nigeria. *Nigerian J Life Sci*. 3, 168-178. https://doi.org/10.52417/njls.v3i1.120

Ozioma, E.O.J., Chinwe, O.A.N., 2019. Herbal medicines in African traditional medicine. *Herbal Medicine*. 10, 191-214.

Pal, D., Raj, K., 2023. *Role of Lipid-Soluble Bioactive Substance from Avocado for Inhibition of Prostate and Other Cancers*, first ed. Therapeutic Platform Of Bioactive Lipids. Apple Academic Press. https://doi.org/10.1201/9781003301608-13

Pinho, B.X., Peres, C.A., Leal, I.R., Tabarelli, M., 2020. Critical role and collapse of tropical mega-trees: a key global resource. *Advances in Ecological Research*. 62, 253-294. https://doi.org/10.1016/bs.aecr.2020.01.009

Podolak, I., Galanty, A., Sobolewska, D., 2010. Saponins as cytotoxic agents: a review. *Phytochemistry Reviews*. 9, 425-474. https://doi.org/10.1007/s11101-010-9183-z

Rattanapunya, S., Sumsakul, W., Bunsongthae, A., Jaitia, S., 2021. *In vitro* antioxidants and anticancer activity of crude extract isolates from euphorbiaceae in Northern Thailand. *The Thai Journal of Pharmaceutical Sciences*. 45, 394-399. https://doi.org/10.56808/3027-7922.2518

Reza, A.A., Haque, M.A., Sarker, J., Nasrin, M.S., Rahman, M.M., Tareq, A.M., Khan, Z., Rashid, M., Sadik, M.G., Tsukahara, T., 2021. Antiproliferative and antioxidant potentials of bioactive edible vegetable fraction of *Achyranthes ferruginea* Roxb. in cancer cell line. *Food Science & Nutrition*. 9, 3777-3805. https://doi.org/10.1002/fsn3.2343

Sahil, K., Sudeep, B., Akanksha, M., 2011. Standardization of medicinal plant materials. *International Journal of Research in Ayurveda & Pharmacy*. 2, 1100-1109.

Salawu, K.M., Ajaiyeoba, E.O., Ogbole, O.O., Adeniji, J.A., Faleye, T.C., Agunu, A., 2017. Antioxidant, brine shrimp lethality, and antiproliferative properties of gel and leaf extracts of *Aloe schweinfurthii* and *Aloe vera*. *Journal of Herbs, Spices & Medicinal Plants*. 23, 263-271. https://doi.org/10.1080/10496475.2017.1318328

Salawu, K.M., Oyerinde, A.A., Aliyu, A., Obajemihi, O.I., 2020. Growth inhibitory properties and antimicrobial evaluation of *Aloe schweinfurthii* (Baker) leaf rind extract. *Journal of Pharmacy & Bioresources*. 17, 174-179. https://doi.org/10.4314/jpb.v17i2.11

Sarfraz, M., Alam, F., Din, K.M., Malik, S., Quddoos, A., 2024. Pharmacognostic characterization of dicleptera chinensis by scanning electron microscopy, light microscopy, and analytical techniques. *Microscopy Research and Technique*. 87, 279-290. https://doi.org/10.1002/jemt.24429

Seong, D., Lee, E., Kim, Y., Han, S., Lee, J., Jeon, M., Kim, J., 2023. Three-dimensional reconstructing undersampled photoacoustic microscopy images using deep learning. *Photoacoustics*. 29, 100429. https://doi.org/10.1016/j.pacs.2022.100429

Sodeinde, O., Salawu, K., Ogbole, O., Ajaiyeoba, E., 2019. Phytochemical, antioxidant, brine shrimp lethality and antiproliferative analyses of *Solanum dasyphyllum* Schum. & Thonn. leaf and fruit extracts [Solanaceae]. *Savannah Veterinary Journal*. 2, 13-17. https://doi.org/10.36759/svj.2019.043

Sonibare, M., Adebodun, S., 2018. Macroscopic and microscopic evaluation of *Triclisia subcordata* Oliv. (Menispermaceae) towards Its Standardization. *Nigerian Journal of Pharmaceutical Research*. 14, 189-196.

Wang, H., Chen, Y., Wang, C., 2023a. Advancing herbal medicine: enhancing product quality and safety through robust quality control practices. *Frontiers in Pharmacology*. 14, 1265178. https://doi.org/10.3389/fphar.2023.1265178

Wang, H., Chen, Y., Wang, L., Liu, Q., Yang, S., Wang, C., 2023b. Advancing herbal medicine: enhancing product quality and safety through robust quality control practices. *Frontiers In Pharmacology*. 14, 1265178. https://doi.org/10.3389/fphar.2023.1265178

Yu, F., Lu, S., Yu, F., Shi, J., Mcguire, P.M., Wang, R., 2008. Cytotoxic activity of an octadecenoic acid extract from euphorbia kansui (Euphorbiaceae) on human tumour cell strains. *Journal of Pharmacy and Pharmacology*. 60, 253-259. https://doi.org/10.1211/jpp.60.2.0015