



RESEARCH ARTICLE

Crude drug analysis and elemental content of the leaves and stem bark of *Adansonia digitata* L. (Malvaceae), an indigenous Ghanaian medicinal plant

Cindy Kitcher^{1*}, Nana Ama Mireku-Gyimah¹, Emelia Oppong Bekoe¹, Joseph Adusei Sarkodie¹, Samuel Frimpong-Manso², Gloria Tattah¹, Memuna Maiga¹ & Tonny Asafo-Agyei³

¹Department of Pharmacognosy and Herbal Medicine, University of Ghana, Accra, P. O. Box LG43, Ghana

²Department of Pharmaceutical Chemistry, University of Ghana, Accra, P. O. Box LG43, Ghana

³Plant Development Department, Center for Plant Medicine Research, Mampong-Akuapem, Ghana

*Email: cikitcher@ug.edu.gh

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ABSTRACT

Adansonia digitata L. is a tree indigenous to Ghana and West Africa. It is traditionally used for medicinal, religious and nutritional purposes. Different parts of the plant are used traditionally for the treatment of diseases such as anaemia, malaria, asthma and diarrhoea among others. It is therefore necessary to provide standard parameters for identification and for the purpose of quality control. This study thus sought to investigate the pharmacognostic characteristics and elemental properties of the leaves and stem bark of *A. digitata* grown and used in Ghana. The macroscopic and microscopic characteristics, phytochemical, physicochemical, fluorescence and elemental properties of the leaf and stem bark were determined using standard protocols. The results of the study showed that the leaves of *A. digitata* were palmate compound and alternately arranged with stipules at each node. The outer bark was observed to be grey in color while the inner bark was pink to brown and laticiferous. Anomocytic stomata and stellate trichomes were also observed microscopically on the leaf surface. The powdered stem bark contained brachysclereids and prismatic calcium oxalate crystals. Saponins, tannins, flavonoids and alkaloids were detected in both leaf and stem bark. They additionally exhibited different fluorescence characters in various solvents. The plant contained major and minor nutritional elements in varying quantities. The results of this study can serve as reliable parameters for accurate identification and authentication of *A. digitata* L. hence ensuring quality.

Introduction

Medicinal plants play a major role in fulfilling the health care needs of various nations such as China, India, Ghana and other West African nations (1–3). The use of herbal remedies as an adjunct or alternative to conventional medicine in the management of diseases is also becoming increasingly popular all over the world (4). In the West African nation of Ghana, the use of indigenous herbal medicine is widespread and traditional medicine has huge impacts on the local economy (5). *Adansonia digitata*, the baobab, (Malvaceae), is the most widespread tree species of the genus *Adansonia*, and it is native to the African continent, mostly occurring in

Southern African countries such as Namibia, Malawi and Zimbabwe among others (6–8). The trees are deciduous, usually growing as solitary individuals and are large and distinctive elements of savannah or scrubland and deciduous vegetation (7, 9).

The baobab has other common names which include ‘monkey bread tree’ and ‘upside down tree’ (10). In some African countries, it is referred to as ‘shadjo-tso’, in Ga (Southern Ghana) ‘anderabai’ in Temne (Congo) and ‘kukulu’ in Grusi-Lyela (Burkina Faso) (11).

In several cultures, the baobab is valued as a source of food, water, and medicine and is also steeped in legendary myths and superstitions (12).

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Nutrition

The seeds can be eaten fresh or dried, and ground into flour and incorporated into soups and stews (13). In some parts of Senegal, the oil extract of the seeds is used in the preparation of some festive dishes (11). The fruit pulp, which is high in Vitamin C is used as an ingredient in the preparation of juices and jams (10) and at times as a seasoning in local diets (13). The young leaves of *A. digitata* are also cooked as spinach, or dried and powdered and used in the preparation of sauces over staples (14). The hollow trunks are used for storing water in some parts of West Africa (15).

Traditional medicinal uses

The use of *A. digitata* for the traditional treatment of various ailments and spiritual purposes has been reported. In parts of Africa and India, the leaves, seeds and fruit pulp are used in folk medicine as an antipyretic (16). The stem bark also serves the same purpose in Ghana and Nigeria where it is used in treating malaria (13, 17) and its aqueous extract is used to resolve anemia (18). Decoctions of the stem bark are also taken for wound healing purposes in Mali (7). Decoctions and infusions of the leaves are also used for the treatment of diarrhea in South Africa, kidney and bladder diseases in Tanzania and asthma in Kenya among other ailments (16). The raw seed powder is administered to stop hiccups in infants and children (19). Extracts of the stem and root bark are also used by some traditional Congolese practitioners for the management of sickle cell anemia (20). In some parts of Southern Ghana, *A. digitata* is believed to be a god to which prayers are said to, and sacrifices made for the fertility of women, prosperity, protection and guidance (11).

Pharmacological activities

Studies are there showing the ability of *A. digitata* methanol leaf extract to exert antidiabetic and antihyperlipidemic properties in experimental diabetic rats (21). Activity of the aqueous bark extract against sickle cell disease has also been established (22). Polysaccharides extracted from the fruit of *A. digitata* have as well been shown to possess anti-inflammatory, antiviral and antioxidant activities (23, 24). In addition, the fruit fiber has been shown to have analgesic activity (15). The stem and root bark extracts were shown to be antimicrobially active against clinical isolates of bacteria and fungi (25, 26). The aqueous extract of the fruit pulp is also reported to have hepatoprotective activity (27). This extract also has cardio protective effects against ISP-induced oxidative stress in rats (28).

Phytoconstituents

Various secondary metabolites have been identified and or isolated from *A. digitata*. The leaf and stem bark have been reported to contain β -sitosterol, scopoletin, betulinic acid and taraxerone (29). The alkaloid adansonin, which is reported to be responsible for the antimalarial activity occurs in the stem bark (30). Also, the fruit pulp and leaves have been found to be rich in procyanidins,

and flavonol glycosides such as tiliroside (31). The fruit pulp and seed kernels contain substantial amounts of calcium, potassium and magnesium (32).

Despite the various benefits derived from herbal medicines, including ones from baobab in Ghana, their uses are associated with challenges including limited knowledge and lack of quality assessment of the crude medicinal plants (33). It is therefore essential to document pharmacognostic specifications of various medicinal plant materials (34). This ensures the plant's identity and lays down standardization parameters which will help prevent adulteration and misidentification. Such parameters include macroscopic and microscopic studies, phytochemical analysis, physicochemical analysis, fluorescence analysis and mineral content investigation (34–36). Some of these parameters have been known to differ among same plant species situated at different locations due to environmental factors and human practices (37).

Owing to widespread use of the baobab and the challenges associated with the quality assessment of medicinal plants, the study sought to provide basic crude drug specifications and mineral content of the leaves and stem bark of *A. digitata* used in Ghana.

Materials and Methods

Plant collection and preparation

The fresh leaves and stem bark of *Adansonia digitata* were collected from the University of Ghana campus (N 05° 39'06.1, W 00° 10'55.2) in July, 2019. They were authenticated at the herbarium of the Center for Plants Medicine Research (CPMR) in the Eastern region of Ghana by Mr. Tonny Asafo-Agyei, a taxonomist, and a voucher specimen with number CPMR 4900 was prepared and deposited there.

The collected leaves, and stem bark were cleared of foreign matter and air dried at room temperature for fourteen days. They were then pulverized into fine powder and stored in air tight containers. Fresh leaves to be used for microscopic observation were sectioned and cleared of all pigment by boiling in chloral hydrate solution for 4 hours. The cleared sections were then stored in glycerin awaiting further investigation.

Macroscopic evaluation

The leaves and stem bark of *A. digitata* were visually observed for their morphological characteristics. The leaf was examined and described using parameters such as type of leaf, size, color, shape, apex, margin, venation and base. Descriptive features such as color and texture of outer bark and inner bark, slash and fracture were noted and employed in establishing its macromorphology (38, 39).

Microscopic evaluation

Microscopic observations were made under low (x10) and high power (x40) magnifications using

the Leica compound light microscope (Wetzlar, Germany).

Portions of the leaf lamina, excluding the margin and midrib were cut into 4 mm x 4 mm sections. The sections were then placed in a test tube containing chloral hydrate solution and boiled until they became clear of all pigment. The cleared leaf sections were then mounted in chloral hydrate solution and observed for features such as stomata, trichomes and venation. Quantitative surface data determinations including vein islet and veinlet termination numbers, stomata number and stomata index were all determined. The powdered leaf and stem bark were also mounted in chloral hydrate solution and observed for the presence of ergastic substances. Phloroglucinol in concentrated hydrochloric acid was also used to detect lignified content in the stem bark powder of *A. digitata* (40, 41).

Physico-chemical analysis

Physicochemical properties of the powdered plant parts were established by determining the moisture content, total ash, acid insoluble ash, water soluble ash, as well as petroleum ether, ethanol and water soluble extractive values according to already published protocols (40, 41).

The loss on drying method was employed in determining the moisture content. Five gms of each powdered material was initially weighed and oven-heated at 105 °C. The samples were dried and weighed intermittently until there was no difference between two consecutive weights. The loss in weight of each material was calculated as a percentage of the air-dried material.

The total ash was determined by igniting four grams of each powdered material at a maximum temperature of 600 °C until it turned white. Total ash was then calculated as a percentage of the air dried material. To determine the acid-insoluble ash, hydrochloric acid was added to the total ash and boiled briefly. This was then filtered using an ashless filter paper and neutralised with hot water. The ash residue together with the filter paper were then dried and ignited until a constant weight was attained. The water-soluble ash was also determined by first boiling the total ash in water. The insoluble residue was then collected on an ashless filter paper and dried. The dried residue, together with the ashless filter paper were ignited to constant weight. Both acid-insoluble ash and water-soluble ash were calculated as percentage weight of the air-dried plant samples.

The extractive values of the leaf and stem bark of *A. digitata* were determined using the cold maceration method. Four gms of each powdered material was weighed and macerated with a hundred millilitres of the given solvent in a conical flask. This was allowed to stand for twenty four hrs with intermittent shaking. The extract was then filtered into a crucible and dried on a water bath until the solvent was evaporated. The remaining extract was then allowed to cool and weighed. The

extractive value was calculated as a percentage of the air-dried plant sample.

Preliminary Phytochemical screening

Preliminary phytochemical tests for saponins, tannins, flavonoids, alkaloids and reducing sugars were carried out using standard methods (38, 42) detailed below.

Saponins (froth test)

Each powdered plant material was suspended in distilled water and shaken vigorously for one minute. The presence of a persistent froth indicated the presence of saponins.

Tannins (ferric chloride test)

An aqueous extract of each powdered plant material was initially prepared by boiling in distilled water and filtering. To each filtrate, a few drops of 0.1% ferric chloride solution was added. The appearance of a blue-black or brownish-green color suggested the presence of hydrolysable or condensed tannins respectively.

Flavonoids (alkaline reagent test)

The aqueous filtrate of each powdered material was treated with a few drops of sodium hydroxide solution. The formation of an intense yellow color, which turned colorless upon the addition of dilute acid, indicated the presence of flavonoids.

Alkaloids (Dragendorff's test)

Chloroform extracts of the powdered plant materials were prepared, filtered and evaporated off. To the residue, dilute hydrochloric acid was added, shaken and filtered. The filtrate was treated with two drops of Dragendorff's reagent. An orange-red precipitate indicated the presence of alkaloids.

Reducing sugars (Fehling's test)

Aqueous extracts of the powdered plant samples were prepared and heated in a water bath. To each filtrate, an equal volume of Fehling's A and B solutions were added and heated. The appearance of a brick-red precipitate indicated the presence of reducing sugars.

Fluorescence analysis

Each powdered stem and leaves were treated with distilled water then observed under natural day light and ultra violet (UV) light (254 nm and 365 nm). Color radiations emitted were observed and noted. The same was repeated for various reagents namely 1N H₂SO₄, 1N HCl, glacial acetic acid, 1N acetic acid, 1N NaOH, ethanol, ethyl acetate and chloroform (42, 43).

Elemental analysis

The elemental analyses of the leaf and stem bark of *A. digitata* was performed using Energy Dispersive X-ray Fluorescence (ED XRF). In preparing the plant sample, 4 gms each of the finely powdered leaves and stem bark was weighed, mixed with the binder Fluxana, and homogenized for 3 minutes. This was then pressed to obtain pellets of 32 mm

diameter. For each plant part, the pellets were prepared in triplicates. The ED XRF was then used to determine the elements present in the plant samples via the radiations emitted from the X-ray tube (44).



Fig. 1. *Adansonia digitata* tree in natural habitat.

Results

Macroscopic description

Adansonia digitata is a large single-stemmed tree with spreading branches (Fig. 1). It consists of multifoliate palmate compound leaves, and are

Table 1. Macroscopic characteristics of *Adansonia digitata* leaves

Macroscopic parameter	Description
Type	Palmate compound
Shape	Elliptic to Oblanceolate
Apex	Acuminate
Margin	Entire
Base	Cuneate
Venation	Pinnate reticulate
Surface	Pubescent (Young leaves)-Glabrous (Mature leaves)

Table 2. Macroscopic characteristics of *Adansonia digitata* stem bark

Macroscopic parameter	Description
Outer bark color	Grey
Slash/inner bark	Spongy texture Pink to brown color and contains latex
Fracture	Granular
Outer bark texture	Rough
Appearance of outer bark	Creased

alternately arranged and stipulate at each node (Fig. 2) Leaflets of matured leaves have a dark green color while those of younger leaves are of a lighter green shade. The lamina is elliptic to oblanceolate in shape with an entire margin, acuminate apex and cuneate base, as well as pinnate reticulate venation. Younger leaves are pubescent while older leaf surfaces are glabrous. The stem bark is grey with a rough texture and appearing to have folds and creases. The inner bark and slash are pink to brown and laticiferous (Fig. 3). The fracture is granular. Tables 1 and 2 summarize the macroscopic features of the leaves and stem bark respectively.

Microscopic description

The microscopic study of the cleared leaf surface revealed a regular polygonal reticulate vein pattern

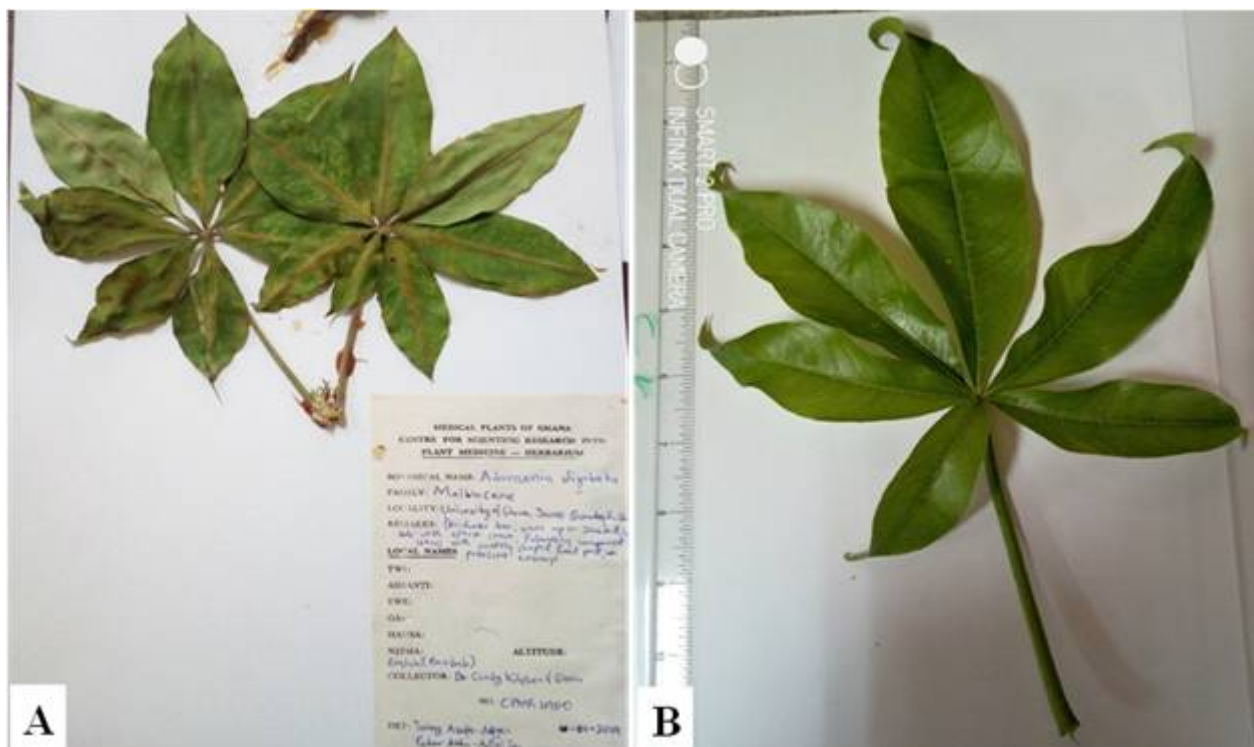


Fig. 2. Leaves of *A. digitata*; A. Mounted voucher specimen of *A. digitata* leaves B. Multifoliate palmate compound leaf.

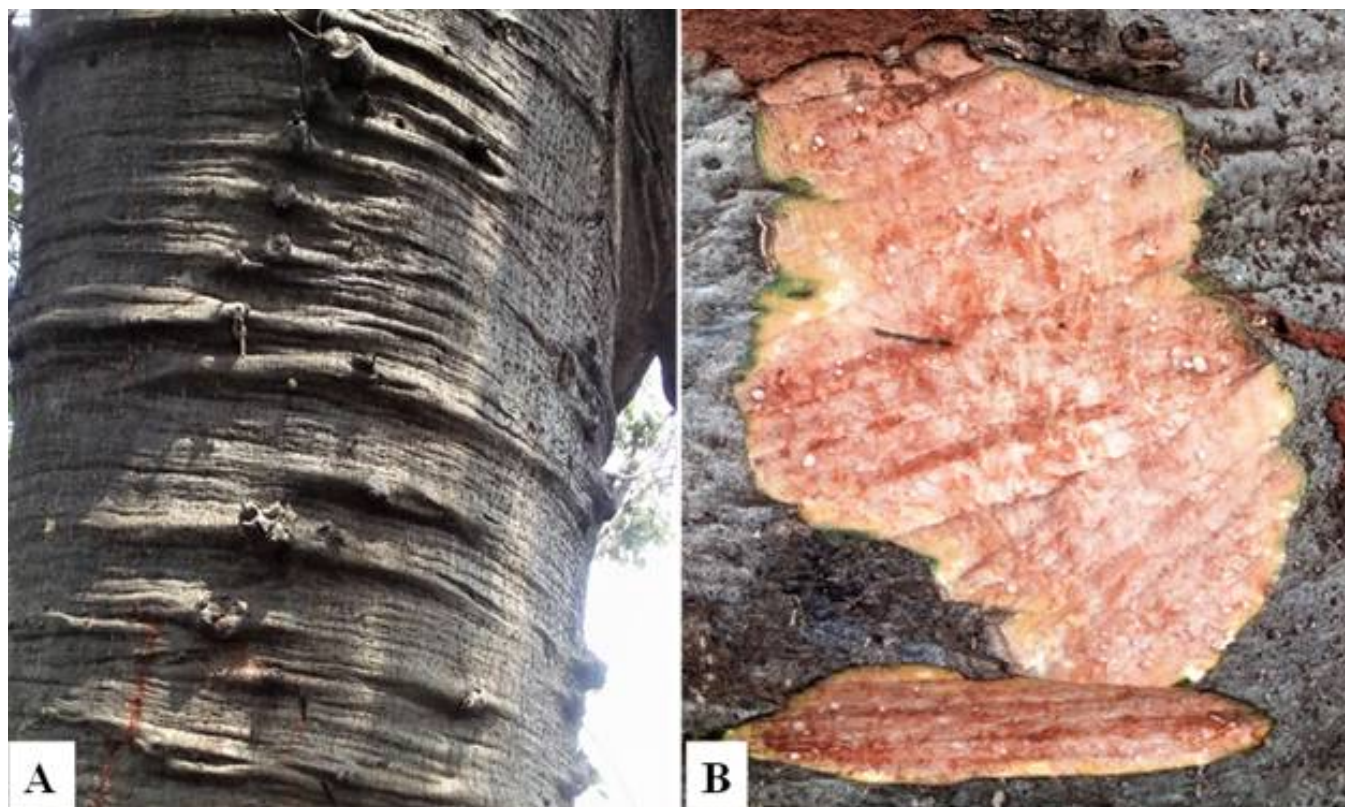


Fig. 3. Stem bark of *A. digitata* A. Grey colored outer stem bark showing folds and creases B. Spongy pink to brown slash/inner bark with laticiferous exudate.

with well-developed four to five-sided areoles (vein islets) and two or more branched freely ending ultimate veins (veinlet terminations) (Fig. 4A). Unicellular stellate trichomes were also observed along the tertiary veins of younger leaves (Fig. 4B). Irregularly shaped epidermal cells, anomocytic stomata and large round mucilaginous cells were also observed on the lower epidermal layer (Fig. 4C). No stomata were observed on the upper epidermis. An investigation of the cross section of the midrib showed the epidermal layer, cortex, distinct grey bundle sheath which surrounded the vascular tissues. The xylem and phloem were separated by the interfascicular cambium. The pith was observed to be hollow and mucilaginous, embedded with abundant rosette calcium oxalate crystals (Fig. 5). The powdered stem bark contained prismatic calcium oxalate crystals and brachysclereids (stone cells) (Fig. 6).

Quantitative microscopy

The quantitative microscopic data also known as leaf constants showed the vein islet number and veinlet termination number ranging from 7 to 9 per squared millimeter area and 9 to 10 per squared millimeter area respectively. Stomata number ranged from 4 to 9 per squared millimeter and stomata index was calculated to be 12% (Table 3).

Physico-chemical analysis

Table 4 presents the details of the physicochemical results. It also shows that the petroleum ether, ethyl acetate and ethanol extractive values were higher for the leaves than the stem bark.

Phytochemical screening

The preliminary phytochemical analyses showed the presence of saponins, tannins, flavonoids, alkaloids

Table 3. Quantitative microscopy of *Adansonia digitata* leaves

Quantitative parameter	Values
Vein islet number	7-9/mm ²
Veinlet termination number	9-10/mm ²
Stomatal number	4-9/mm ²
Stomatal index	12%

Table 4. Physicochemical properties of *Adansonia digitata* leaves and stem bark (%w/w)

Parameters	Leaf	Stem bark
Moisture content	3.23±0.01	7.35±0.06
Total ash	11.25±0.3	10.00±0.5
Water soluble ash	5.25±0.5	4.75±1.3
Acid insoluble ash	6.50±0.5	7.25±0.3
Petroleum ether extractive	2.8	0.4
Ethyl acetate extractive	16	3.0
Ethanol extractive	19.6	6.8

Table 5. Results of fluorescence analyses of *Adansonia digitata* leaves

Powdered sample + solvent	Visible light	Short UV	Long UV
		wavelength (254 nm)	wavelength (365 nm)
Distilled water	Light green	Dark green	Dark green
1N H ₂ SO ₄	Light green	Light green	Light green
1N HCl	Yellowish green	Yellowish green	Yellowish green
Glacial acetic acid	Light green	Light green	Light green
1N Acetic acid	Colorless	Colorless	Colorless
1N NaOH	Light green	Light green	Dark green
Ethanol	Light green	Pink	Light green
Ethyl acetate	Light green	Reddish pink	Light green
Chloroform	Light green	Pink	Light green

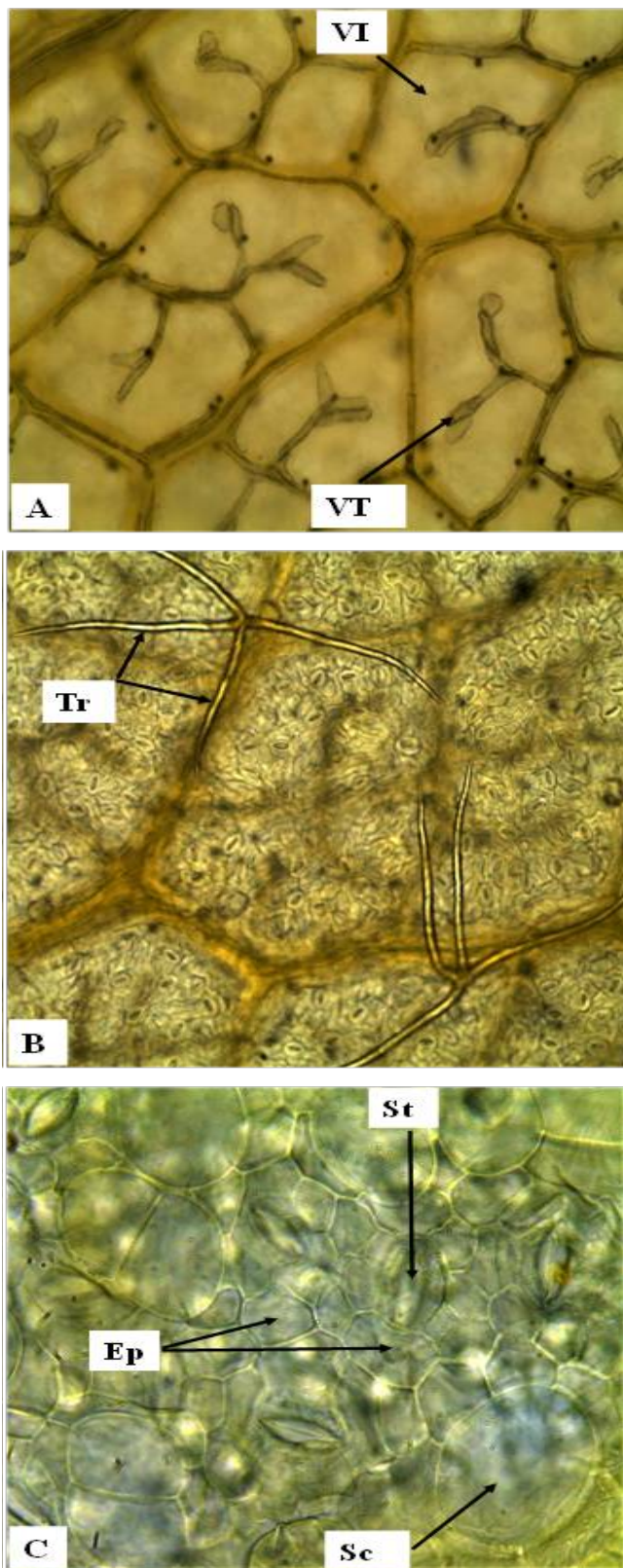


Fig. 4. Leaf surface characteristics of *A. digitata* A. Vein islets (VI) and veinlet termination (VT) B. Unicellular stellate trichomes (Tr) C. Anomocytic stomata (St); Irregularly shaped epidermal cells (Ep); Secretory cells (Sc).

and reducing sugars in both the leaves and stem bark of *A. digitata*.

Fluorescence analysis

The fluorescence characteristics of the powdered leaf and stem bark in different reagents under visible light and UV light are detailed in Tables 5 and 6.

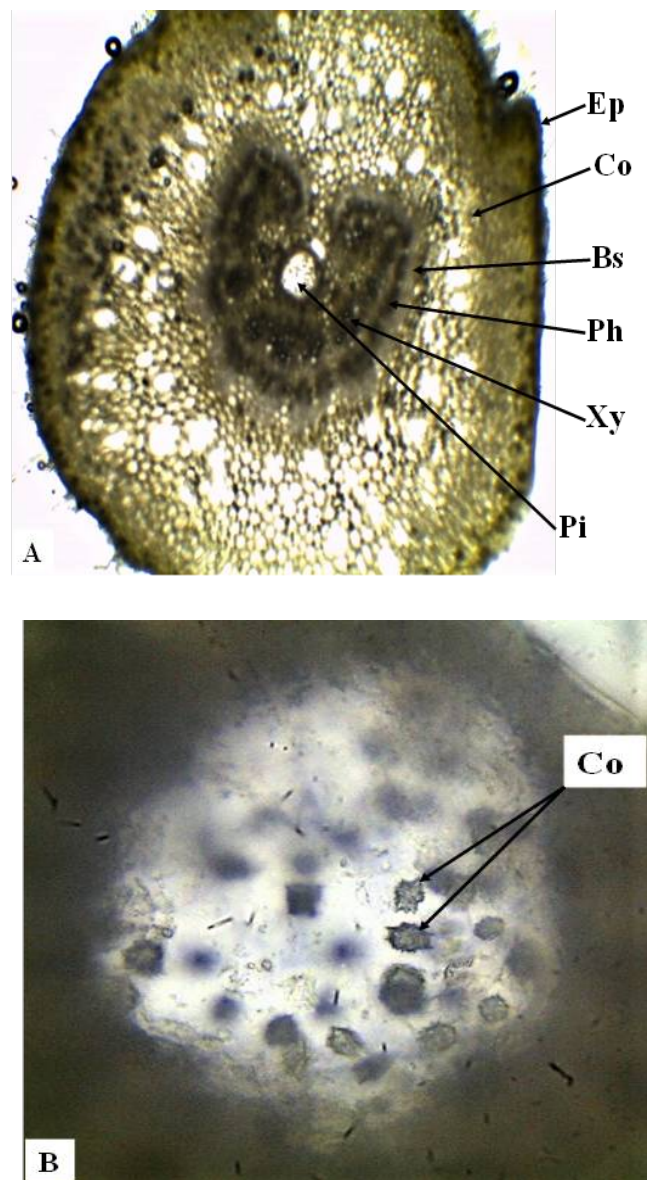


Fig. 5. Midrib cross section of *A. digitata* A. Cross section of midrib showing epidermis (Ep), cortex (Co), bundle sheath (Bs), phloem (Ph), xylem (Xy) and pith region (Pi) B. Magnified Mucilaginous pith containing rosette calcium oxalate crystals (Co).

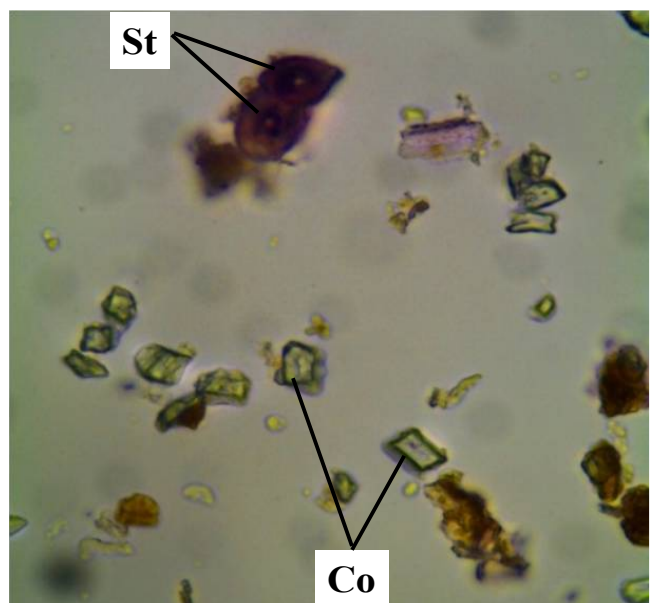


Fig. 6. Prismatic calcium oxalate crystals (Co) and brachysclereids (stone cells (St)) in powdered stem bark.

Elemental analysis

The elemental analyses showed high concentrations of mineral elements such as magnesium, silicon, potassium and calcium. Heavy metals such as lead and mercury were absent. Detailed results of the elemental analyses is presented in Table 7.

Discussion

The use of herbal medicines require that the identity and quality of the crude drug are established as ways of ensuring safety and efficacy (45). The present study aimed at investigating and establishing the diagnostic characteristics of *Adansonia digitata*, an indigenous medicinal plant by employing standard pharmacognostic protocols.

The macroscopic and microscopic characteristics of medicinal plants play important roles in their taxonomic classification and identification. Features such as palmate compound leaves, mucilaginous epidermis, adaxial stomata and stellate trichomes observed in this study are similar to published reports (7, 46).

Extractive values serve as important indicators of the nature of phytoconstituents in a crude drug (36). From the results (Table 4), a higher percentage of extract was obtained with ethanol and may therefore be the preferred solvent for extraction for both leaves and stem bark of *A. digitata*. It also gives the indication that the leaf and stem bark may contain polar constituents.

The moisture content of a crude drug is usually a determinant of the drug's stability and the chances of microbial contamination (47). The moisture contents of both leaves and stem bark (Table 4) fell within acceptable range for vegetable drugs, thus, the powdered sample can be stored for a long period with low probability of microbial attack (48).

Ash content show the amount of inorganic matter present and is an indication of the purity or possible adulteration of the drug (49). Total ash values for both leaves and stem bark powder were within close range of each other (Table 4). Water insoluble ash, which represents the amount of cellulosic substances were 5.25% and 4.75% for the leaves and stem bark respectively. Acid insoluble ash values were higher in the powdered materials than water soluble ash, indicating the presence of siliceous substances (45).

Preliminary phytochemical investigation of the leaves and stem bark of *A. digitata* showed the presence of alkaloids, flavonoids and tannins. Similar findings have been reported in earlier studies (50). The presence of these phytoconstituents may contribute to reported pharmacological effects and traditional uses of the plant for medicinal purposes (see section on traditional medicinal uses and pharmacological activities).

The fluorescence analyses of the leaves and stem bark powder of *A. digitata* showed their

Table 6. Results of fluorescence analyses of *Adansonia digitata* stem bark

Powdered sample + solvent	Visible light	Short UV wavelength (254 nm)	Long UV wavelength (365 nm)
Distilled water	Light brown	Dark brown	Dark brown
1N H ₂ SO ₄	Deep brown	Dark brown	Green
1N HCl	Amber	Colorless	Green
Glacial acetic acid	Yellowish green	Straw colored	Green
1N Acetic acid	Yellowish green	Straw colored	Green
1N NaOH	Light brown	Pale yellow	Pale yellow
Ethanol	Pale yellow	Colorless	White
Ethyl acetate	Pale yellow	Colorless	Colorless
Chloroform	Straw colored	Straw colored	Straw colored

Table 7. Average elemental composition of the leaf and stem bark of *A. digitata*

Element	Concentration (ppm)	
	Leaf	Stem bark
Magnesium (Mg)	8336.00 ±497.07	9141.00±424.70
Aluminum (Al)	1999.67±272.22	1583.00±77.44
Silicon (Si)	64645.33±54.68	63458.33±7.02
Phosphorus (P)	1346.33±9.07	1040.00±20.42
Sulphur (S)	1247.00±19.67	2499.00±15.52
Potassium (K)	13811.00±20.22	25645.00±18.52
Calcium (Ca)	32859.67±67.28	32502.33±88.55
Lead (Pb)	0.00±0.00	0.00±0.00
Manganese (Mn)	0.00±0.00	86.33±7.57
Iron (Fe)	1052.00±13.08	2998.00±23.64
Mercury (Hg)	0.00±0.00	0.00±0.00
Copper (Cu)	1.33±2.31	7.67±1.15
Zinc (Zn)	1.33±2.31	19.67±1.15
Rubidium (Rb)	15.00±0.00	27.00±0.00
Cadmium (Cd)	19.00±1.00	21.00±4.58

characteristic color behavior when treated with different organic and inorganic solvents and observed under different wavelengths of UV light (Tables 5 and 6). This may provide an idea about their chemical nature and can also be useful in the detection of adulterants in liquid preparations from the leaf and stem bark (36, 45).

Levels of elemental concentration in a plant material may be attributable to mineral composition of the soil, habitat, different climatic and environmental conditions as well as human activities (51). Previous studies have reported high concentrations of potassium, calcium and magnesium in the seeds and fruit pulp of *A. digitata* (32), similar to the results of the present study. It must however be noted that, essential mineral content in baobab vary significantly among different geographical locations (37). This factor must be considered during collection for medicinal and nutritional purposes. Macro and micronutrients play important roles in disease management and prevention (51). They also provide consumers with essential minerals for optimum body function (52). Heavy metals such as lead and mercury were not detected in the plant materials. Since such heavy metals can be hepato and nephrotoxic, it is imperative to make sure plant materials for medicinal or nutritional use are free of these.

Conclusion

The diagnostic characteristics of the leaves and stem bark of *A. digitata* reported in this study will serve as useful data for identification, quality control and research purposes. Also, findings from this research, may be of use in the preparation of a monograph.

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Authors' contributions

CK conceived the study, participated in its design and coordination and drafted the manuscript. NAM-G participated in the design of the study, supervised the phytochemical analysis. EOB participated in the physico-chemical and fluorescence analyses. JAS participated in the plant collection and participated in the design and coordination of the study. SFM carried out the elemental analyses. GT participated in the macroscopic and microscopic evaluation of the plant. MM participated in the phytochemical, physico-chemical and fluorescence analyses. TAA authenticated the plant sample. All authors read and approved the final manuscript.

Conflict of interests

The authors declare that they have no competing interests.

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