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PHYTOCHEMICAL AND PHARMACOGNOSTIC STUDIES OF *BUCHANANIA ANGUSTIFOLIA* ROXB. (ANACARDIACEAE) LEAVES

*D. Venkatanarayana and ¹S. Mohana Lakshmi

*Assistant Professor, Department of Pharmacognosy, Rao's College of Pharmacy, Nellore, Andhra Pradesh – 524320, India

¹Professor, Department of Pharmacognosy, Sree Vidyanikethan College of Pharmacy,
A. Rangampet, Tirupati, Andhra Pradesh – 517102, India.

ABSTRACT

Pharmacognostic investigation was carried out on the *Buchanania angustifolia* Roxb. leaves. The assignment such as macroscopy, anatomical studies and preliminary phytochemical screening were performed since the species was not noted for its pharmacognosy in part. Macroscopic studies is a technique of qualitative evaluation based on the study of morphological and sensory profiles of *Buchanania angustifolia* Roxb. leaf. Microscopic studies is a technique of qualitative evaluation and used to confirm the structural details of drugs from the *Buchanania angustifolia* Roxb. leaf and also study of the constituents by application of chemical methods to small quantities of drug of phoroglucinol and concentrated hydrochloric acid give red stain with lignin. Mucilage is strained pink with ruthenium red. The perusal of literature also revealed that no pharmacognostic work had been carried out on the plant of *Buchanania angustifolia* Roxb. For this reason we have investigated the pharmacognostic profiles of *Buchanania angustifolia* Roxb. leaves.

Keywords: *Buchanania angustifolia* Roxb., Phytochemical Screening, Pharmacognostic Studies.

INTRODUCTION

Plants have played a significant role in maintaining human health and improving the quality of human life for thousands of years and have served humans well as valuable components of medicines, seasonings, beverages, cosmetics and dyes. Herbal medicine is based on the premise that plants contain natural substances that can promote health and alleviate illness. In recent times, focus on plant research has increased all over the world and a large body of evidence has collected to show immense potential of medicinal plants used in various traditional systems. Today, we are witnessing a great deal of public interest in the use of herbal remedies [1].

The future development of pharmacognosy as well as herbal drug industry would be largely dependent upon the reliable methodologies for identification of marker compounds of the extracts and also upon the standardization and quality control of these extracts. Mother earth has given vast resources of medicinal flora and fauna both terrestrial and marine and it largely depends upon the forthcoming generations of

pharmacognosist and phytochemist to explore the wonder drug molecule from this unexploited wealth [2].

A thorough Pharmacognostic investigation was carried out on the *Buchanania angustifolia* Roxb. leaves. The assignment such as macroscopy, anatomical studies and preliminary phytochemical screening were performed since the species was not noted for its pharmacognosy in part. Macroscopic studies is a technique of qualitative evaluation based on the study of morphological and sensory profiles of *Buchanania angustifolia* Roxb. leaf. Microscopic studies is a technique of qualitative evaluation and used to confirm the structural details of drugs from the *Buchanania angustifolia* Roxb. leaf and also study of the constituents by application of chemical methods to small quantities of drug of phoroglucinol and concentrated hydrochloric acid give red stain with lignin. Mucilage is strained pink with ruthenium red. The perusal of literature also revealed that no pharmacognostic work had been carried out on the plant of *Buchanania angustifolia* Roxb. For this reason we have investigated the pharmacognostic profiles of *Buchanania angustifolia* Roxb. leaves.

Corresponding Author: **D. Venkatanarayana** Email:- venkatanarayana1978@gmail.com

Standardization problems arise from the complex composition of drugs which are used in the form of whole plant, parts of the plants, plant extracts. Standardization of presumed active compounds of drug in general does not reflect reality, only in few cases drug activity depends upon single component. Generally it is the results of concerted activity of several active components as well as the inert substance. Though these inert accompanying components do not directly affect pathological mechanism. It is responsible to use such components which might influence bioavailability and excretion of active component. Further by inert plant component might be increased and the rate of side effects be minimized. If these are different active components present in a plant drug, they might have synergistic or potentiation effect. Following parameters were observed while standardizing the plant *Buchanania angustifolia* Roxb. (Anacardiaceae) taken for present study.

MATERIALS AND METHODS

Collection and Authentication of the plant material

The plant *Buchanania angustifolia* Roxb. (Anacardiaceae) is widely found throughout India. For our project work the plant was collected from the forest area of the Tirupathi, Chittor district, Andhra Pradesh, India, during the month of August 2009 with help of botanist, care was taken to select healthy plant and for normal organs [3]. It was authenticated by Dr. P. Jayaraman, Director, Plant anatomy research center (PARC) west tambaram, Chennai. The plant was also compared with the herbarium specimen present at above mentioned centre (The flora of Tamilnadu zarmatic, Mathew). The voucher specimen (PARC/2009/346) was deposited at the college for further reference.

Phytochemical Investigation

Phytochemical investigation involves the following features,

- To extract the active constituents from plant material.
- To identify the phytochemical constituents of extracts.

Literature studies reveals that no phytochemical analysis has performed in *Buchanania angustifolia* Roxb. and we focussed on this studies on above mentioned features.

Materials : Petroleum ether, Soxhlet apparatus, Powered plant material.

Reagents : Mayer's reagents, Dragendroff's reagents, Wagner's reagents, Hager's reagents.

Chemicals : Sulphuric acid, Acetic anhydride, Hydrochloric acid, Sodium hydroxide, Sodium chloride, Sodium nitroprusside.

METHODOLOGY

Extraction of plant material

The fresh leaves of *Buchanania angustifolia* Roxb. (Anacardiaceae) were they were washed cleaned, dried and powdered in a grinder mixer to obtain a coarse powder and then passed through a 40 mesh sieve. About 120 g of powder drug was extracted with petroleum ether

(60-80°), methanol and water using soxhlet apparatus. The extraction was carried out until the extractive becomes colourless. The solvent was completely removed from the marc.

The solvent was removed from the extract by distillation under reduced pressure. The dried extract was kept in a dessicator and used for identifying their chemical groups present. The percentage yields of the extractive were mentioned in Table 1.

Phytochemical Tests

The following qualitative chemical tests, for identifying various phytoconstituents present, were carried out on various extracts of leaf of *Buchanania angustifolia* [4,5] (Table 2).

Tests for Alkaloids

- **Mayer's test:** (Potassium mercuric iodide solution). To extract/sample solution, add few drops of Mayer's reagent, creamy white precipitate is produced.
- **Dragendroff's test:** (Potassium bismuth iodide solution). To extract/sample solution, add few drops of Dragendroff's reagent, reddish brown precipitate is produced.
- **Wagner's test:** (Solution of Iodine in Potassium Iodide). To extract/sample solution, add few drops of Wagner's reagent, reddish brown precipitate is produced.
- **Hager's Test:** (Saturated solution of Picric acid) To extract/sample solution, add few drops of Hager's reagent, yellow precipitate is produced.

Tests for Carbohydrates

- **Molisch's test:** Treat the extract solution with few drops of alcoholic α -naphthol. Add 0.2 ml of concentrated H_2SO_4 slowly through the sides of the test tube, purple to violet color ring appears at the junction.
- **Benedict's test:** Treat the extract solution with few drops of Benedict's reagent (alkaline solution containing cupric citrate complex) and upon boiling on water bath, reddish brown precipitate forms if reducing sugars are present.
- **Barfoed's test:** General test for monosaccharides. Heat the test tube containing 1ml reagent and 1 ml of extract solution in a beaker of boiling water; if red cuprous oxide is formed within two minutes, a monosaccharide is present. Disaccharides on prolonged heating (about 10min) may also cause reduction, owing to partial hydrolysis to monosaccharides.
- **Fehling's test:** Equal volume of Fehling's A (Copper sulphate in distilled water) and Fehling's B (Potassium tartrate and Sodium hydroxide in distilled water) reagents are mixed along with few drops of extract solution, boiled, a brick red precipitate of cuprous oxide forms, if reducing sugars are present.

Tests for Proteins & Aminoacids

- **Millon's Test:** Extract solution + 2 ml of Millon's reagent (Mercuric nitrate in nitric acid containing traces of nitrous acid) white precipitate appears, which turns red upon gentle heating.

- **Ninhydrin Test:** Amino acids and proteins when boiled with 0.2% solution of Ninhydrin (Indane 1, 2, 3 trione hydrate) produces violet color.

Tests for Sterols and Triterpenoids

- **Liebermann-Burchard test:** Extract treated with few drops of acetic anhydride, boil and cool, concentrated sulphuric acid is added from the side of the test tube, A brown ring at the junction of two layers and the upper layer turns green indicates the presence of sterols and formation of deep red color indicates the presence of triterpenoids.
- **Salkowski's test:** Treat extract in chloroform with few drops of concentrated Sulfuric acid, shake well and allow to stand for some time, red color appears in the lower layer indicates the presence of sterols and formation of yellow colored lower layer indicating the presence of triterpenoids.

Tests for Phenolic Compounds

- **Ferric chloride test:** Extract solution gives blue-green color with few drops of $FeCl_3$.
- **Zinc-Hydrochloride reduction test:** To the extract solution, add a mixture of Zinc dust and conc. Hydrochloric acid. It gives yellowish, yellow- orange occasionally orange color after few minutes.

Tests for Flavonoids

- **Shinoda Test** (Magnesium Hydrochloride reduction test To the extract solution add few fragments of magnesium ribbon and concentrated Hydrochloric acid drop wise, pink scarlet, crimson red or occasionally green to blue color appears after few minutes.
- **Zinc-Hydrochloride reduction test:** To the extract solution, add a mixture of Zinc dust and conc. Hydrochloric acid. It gives red color after few minutes.
- **Alkaline reagent test:** To the extract solution, add few drops of Sodium hydroxide solution; formation of an intense yellow color that turns to color less and addition of few drops of dilute acetic acid indicates the presence of flavonoids.

Tests for Tannins

- **Gelatin test:** Extract solution with 1% gelatin solution containing 10% sodium chloride gives white precipitate.
- **Ferric chloride test:** Extract solution gives blue-green color precipitate with $FeCl_3$.
- **Vanillin Hydrochloride test:** Extract solution when treated with few drops of Vanillin Hydrochloride reagent gives purple red color.
- **Alkaline reagent test:** Extract solution with sodium hydroxide solution gives yellow to red precipitate within short time.

Test for glycosides

- **Legal test:** To the hydrolysate, 1ml of pyridine and few drops of sodium nitroprusside solution were added and it was made alkaline with sodium hydroxide. Appearance of pink to red colour, indicate the presence of glycoside.

- **Borntragers test:** hydrolysate was treated with chloroform and the chloroform layer was separated. To this, dilute ammonia solution was added. Pink colour in the ammonia solution indicates the presence of glycosides.

Test for fixed oil

- A small quantity of various extract was separately pressed between two filter papers. Appearance of stain in the paper indicates the presence of fixed oil.

Test for gums and mucilage

- A small quantity of extract was slowly added into a test tube containing alcohol with constant stirring. Formation of precipitate indicates the presence of gums and mucilage.

Test for saponin

- **Froth formation test:** The extracted was diluted with distilled water and agitated in a granulated cylinder for 15 minutes. The formation of 1cm layer of stable froth (foam) indicates the presence of saponins.

MACROSCOPICAL STUDIES

The fresh leaves of *Buchanania angustifolia* Roxb. (Anacardiaceae) was studied individually for its morphological characters (Evans 2008) such as color, odour, taste, shape and sizes etc. in the field and photographed under original environment and evaluated botanically (Table 3 & 4).

MICROSCOPICAL STUDIES

- **Preparation of specimens:** The fresh sample were cut into small pieces and fixed in FAA solution (Formalin 5ml + Glacial acetic acid 5ml + 70% Ethanol 90ml). After fixing, the specimens were dehydrated with graded series of tertiary butyl alcohol (TBA) as per the standard procedure. After complete dehydration, the specimens were embedded in paraffin wax.
- **Sectioning:** The paraffin embedded specimens were sectioned with the help of Rotary microtome (thickness 10-12 μ m). Dewaxing and staining of the sections were done by customary procedure. Sections were stained mostly with toluidine blue.

Staining: For anatomical studies the following staining schedules were followed 1. **Tannic Acid** – Ferric Chloride counterstained with 0.5% alcoholic safrain. This schedule was found to be quite satisfactory for all young plant tissues in which the primary walls were stained. 2. **Alcoholic safrain** (0.5%) counterstained with 0.25% fast green. This schedule gives good result for studying the histology of different tissues of the plant organs especially the cell inclusions. 3. **Toluidine Blue** – **O** stain was prepared by dissolving 0.25gm of the stain in the mixture of benzoic acid 0.25gm, sodium benzoate 0.29gm and distilled water 200ml with p^H of 4.2 – 4.4. Since Toluidine blue is a polychromatic stain, the staining results were remarkably good and the dye render pink colour to the cellulose walls, blue to the lignified cells, dark green to suberin, violet to the mucilage, blue to the protein bodies,

etc. After dewaxing, the slides were stained for 5 – 10 minutes and then dehydrated.

Photomicrograph: All permanent slides, after staining were dehydrated by using graded series of Ethanol + Xylol and mounted in DPX. Photomicrographs were done on NIKON – Labphot – 2 microscope using Konica colour film (100 ASA). For normal observations bright field was used. For the study of crystals and starch grains, the sections were photographed under polarized light. Magnifications of the figures are indicated by scale bars. Descriptive terms of various observations are as found in standard Anatomy books.

Powder microscope The leaf powder is boiled with chloral hydrate for 5-10 minutes, and then stained with phloroglucinol, Toluidine and observed for the microscopic features under high power (40 x) (Table 5) [6-8].

Physicochemical parameters

The determination of various physicochemical parameters such as total ash, acid insoluble ash, water soluble ash, water soluble extractive value, alcohol soluble extractive value, swelling index, loss on drying were calculated as per Indian Pharmacopoeia.

Determination of total ash: 3gm of accurately weighed quantity of the shade-dried leaves powdered drug was taken in a tarred silica crucible and incinerated at the temperature not exceeding 450⁰C until free from carbon, cooled and weighed.

Determination of Acid-insoluble ash: Total ash obtained was boiled for five minutes with 25 ml of dilute Hydrochloric acid. The insoluble matter was collected on an ash less filter paper, washed with hot water and ignited, cooled and weight. The percentage of acid insoluble ash was calculated with reference to shade-dried leaves powder.

Determination of Water-soluble ash: Total ash obtained was boiled for five minutes with 25ml of distilled water, cooled and collected the insoluble matter on an ash-less filter paper, washed with hot water and ignited for 15 minutes at temperature not exceeding 450⁰C. Subtracted the weight of the insoluble ash. The percentage of water-soluble ash was calculated with reference to Shade -dried leaves powder [9-11].

Extractive value

Water soluble extractive value: Shade dried *Buchanania angustifolia* Roxb (Anacardiaceae) leaves were pulverized and 100g of the crude drug powder was macerated with 0.7% chloroform water for 7 days with occasional shaking to get the aqueous extract. The aqueous extract was concentrated in a rotary flash evaporator and dried in desiccator over sodium sulfite.

Alcoholic soluble extractive value: Shade dried *Buchanania angustifolia* Roxb (Anacardiaceae) leaves were pulverized and 100gm of the crude drug powder was extracted with 95% ethanol in a soxhlet extractor. The liquid extract was concentrated in a rotary flash evaporator. The residue was dried in a desiccator over sodium sulfite.

Loss on drying

1 gm of the powdered sample was accurately weighed in a tarred petri dish, previously dried under specified conditions as per Indian Pharmacopoea (1996) [12]. The powder was distributed as evenly as practicable, by gently side shaking. The dish was dried in an oven at 100-105⁰ C for 1 hour. It was cooled in desiccator and again weighed. The loss on drying was calculated with reference to the air dried powder drug.

Swelling index

Swelling Index is the determination factor for gums, mucilage, pectin or hemicellulose. It's determination is based on addition of 25ml of water with 1gm of powder drug. Shaken for 1h for every 10 min and allow to stand for 24h. The volume (in ml) has to be measured.

Fluorescence

The powder prepared from the leaves were treated with various solvents like 1N Hydrochloric acid, Acetic acid, Iodine, Ferric chloride, Ammonia, and Water to evaluate the fluorescence analysis in visible/day light, long UV (365nm) and short UV (254nm) light. Various extract of leaves were also subjected to fluorescence analysis. This parameters were carried out according to the standard procedure as per Indian Pharmacopoea (1996) [12].

RESULTS AND DISCUSSION

The result of extractive value of powdered drug in different solvent such as petroleum ether, methanol and distilled water were shown in table-1. Methanol extract yield is higher than other extract, which indicates the presence of highly polar chemical constituents. All extracts subjected to qualitative chemical test and results were shown in Table-2. The result shows that maximum constituents found in methanol extract of *Buchanania angustifolia* Roxb. including carbohydrate, proteins flavonoids, phenols, steroids and saponins. Such preliminary phytochemical screening was helpful in prediction of nature of drugs and also useful for the detection of different constituents present in different polarity solvent. So it could be helpful to extract out particular constituents by solvent.

Macroscopical studies

The macroscopic characters were useful in quick identification of plant material and also serves as an important standardization parameter [13]. Organoleptic evaluations of leaves of *Buchanania angustifolia* Roxb were reported in table-1. The photograph of twig of *Buchanania angustifolia* Roxb are presented in figure 1.

Microscopical Studies

Anatomy of the leaf

The leaf has thick Plano-convex midrib with abaxial projection. It is 1.15 mm thick and 1 mm wide. The epidermal layer along the midrib consists of radially oblong thick walled cells with dark contents. The abaxial epidermal cells are slightly papillate. The ground tissue includes small, circular, thick walled cells towards the periphery and large, thin walled parenchyma cells in the centre. The vascular system is multi stranded. There is an "abaxial arc" of six or seven large, collateral vascular bundles; of these bundles the median bundle is the largest. There is another horizontal plate of five or more bundles the adaxial part. The vascular bundles have closely arranged, parallel lines of three to six xylem elements in each row (Fig 3). Phloem occurs in thick arcs along the outer end of the vascular bundle. Outer to the vascular bundles there are wide, circular, mucilage canals (Fig: 3). Tannin content is found in most of the ground parenchyma cells making the midrib tissues dark in appearance.

Cell inclusion in the midrib as seen under polarized light microscope

Calcium oxalate crystals are fairly abundant in the parenchyma cells of the midrib. The crystals are mostly druses; occasionally prismatic type (Fig: 4 & 5). The crystals are located in the ordinary parenchyma cells. The crystals are 15-20 μm in diameter.

Anatomy of the lamina

The lamina consists of a wide single layer or at certain places two layers of vertically elongated thick walled epidermal cells (Fig: 6 & 7). Some of the epidermal cells are filled with dense mucilage content. The abaxial epidermis is narrow, the cells being rectangular or square cells. The mesophyll tissue consists of broad zone of two layers of narrow cylindrical, palisade cells, which are loosely arranged. The spongy mesophyll comprises seven to five layers of lobed reticulate parenchyma cells.

The lamina is 200 μm thick. The marginal part of the lamina is broadly conical and slightly bent downwards. The mesophyll tissue of the marginal portion consists of heavily thick walled, lignified, mass of sclerenchyma cells (Fig: 8). The marginal portion is also 200 μm thick.

Epidermal Morphology

The surface features of epidermal cell and stomatal morphology were studied in Paradermal sections. The adaxial epidermis consists of small, angular, thin walled compact cells. Mucilage abundant in most of the epidermal cells. The adaxial epidermis is "apostomatic" (Fig: 9).

The abaxial epidermis has angular, thin walled, compact cells similar to adaxial epidermis. Stomata are abundant on the abaxial epidermis (Fig: 10).

The anticlinal walls are straight and slightly thick. Most of the stomata seem to be "actinocytic". A stoma has five or less radiating subsidiary cells all around the guard cells (Fig: 11).

Venation Pattern

Venation pattern was also studied from paradermal sections. The lateral veins and veinlets are thick and straight. The Vein-islets are small and squarish. The vein-terminations are not distinct. When the termination is distinct, they are short, thick and unbranched (Fig: 12, 13, 14) running along the veins there are long, narrow fibre-sclereids. The sclereids have no pits or secondary wall thickenings. They have thin wall and wide lumen.

Powder Microscopy of the leaf

Coarse powder of the leaf was examined under the microscope. Thick pieces of lamina, epidermal peeling and separated cells of the vein were seen under the microscope. The venation pattern is very distinct in surface view of cleared fragment of lamina (Fig: 15, 16, 17 & 18). The lateral veins are thick due to the presence of fibre sclereid which run all along the veins and vein-lets. The sclereids are wide, thin walled and slightly lobed. The sclereids also protrude into the vein-islets. The vein-terminations are short and thick.

Epidermal peeling

Both adaxial and abaxial epidermal peelings are seen in the powder. The adaxial epidermal cells are small polygonal and thin walled. They have mucilage filled wider cells (Fig: 19, 20). The abaxial epidermis consists of small, slightly thick walled cells. They have stomata which are 'actinocytic' or 'anamocytic' type. The anticlinal walls are straight.

Powder microscopy of the leaf

The powder also shows long, narrow and thick walled fibres (Fig 20). The lumen of the fibre is narrow. The fibre is 15 μm thick and several mm long. The sclereids are also seen in the powder. They vary in size and shape. Some of them narrow and elongated and some are highly lobed (Fig. 20, 21). The sclereids are thin walled with wide lumen.

Powder Microscopy

The various diagnostic characteristic of powder was coarse, dark green with characteristic odour and acrid taste. Microscopic examination of powder shows various characters such as anomocytic stomata, epidermal cells of petals, annular to spiral vascular bundle, compound vessels, epidermal cell are present [14].

Physicochemical parameters

The determination of physicochemical parameter is important in determination of adulterants and improper handling of drugs. Table- 5 shows the result of various physico chemical parameters of powdered drug carried out using standard methods. Moisture content of drugs could be at minimal level to discourage the growth of bacteria, yeast or fungi during storage. Ash values used to determine quality and purity of crude drug. It indicates the presence of various impurities like carbonate, oxalate and

silicate. The acid insoluble ash consist mainly silica and indicate contamination with earthy material. The water soluble ash is used to estimate the amount of inorganic elements present in drugs. The extractive values are useful to evaluate the chemical constituents present in the crude drug and also help in estimation of specific constituents soluble in a particular solvent [15,16].

Fluorescence Analysis

The fluorescences studies of the leaf of powder of *Buchanania angustifolia* Roxb. (Anacardiaceae) Showed the colour with different chemical reagents. These colours changes in different light were observed and mentioned in table-6. Fluorences analysis of various extract were evaluated and results shown in table-7.

Table 1. Percentage yield of various extracts of *Buchanania angustifolia* (anacardiaceae) Roxb

S. No.	Extract	Colour	Consistency	% w/w
1.	Petroleum Ether	Green	Semisolid	20.85
2.	Methanol	Dark brown	Semisolid	22.38
3.	Distilled water	Light brown	Sticky	12.29

Table 2. Chemical investigation of *Buchanania angustifolia* Roxb. (anacardiaceae) leaves extracts

S. No.	Chemical Test for	Petroleum Ether	Methanol	Aqueous
1.	Alkaloids	-	-	-
2.	Carbohydrates	-	+	+
3.	Proteins and amino acids	-	+	+
4.	Terpenoids	+	+	+
5.	Phenols	+	+	+
6.	Flavonoids	-	+	+
7.	Tannins	-	-	-
8.	Glycosides	-	+	+
9.	Gums and mucilage	-	+	+
10.	Saponins	-	+	+
11.	Steroids	+	+	-

“+” indicate positive response, “-“ indicate negative response

Table 3. Morphological features of fresh leaf of *Buchanania angustifolia* Roxb

Sl. No.	Features	Observation
1.	Condition	Fresh
2.	Colour	Green
3.	Odour	Characteristic
4.	Taste	Characteristic
5.	Size	13-15 cm length, 3-5 cm width
6.	Shape	Linear Oblong
7.	Texture	Firm

Table 4. Botanical evaluation of leaf of *Buchanania angustifolia* Roxb

Leaf portion	Observation
Condition	Fresh
Type	Simple
Apex	Obtuse
Margin	Entire
Lamina, Leaf Blade	
Shape	Lanceolate oblong
Size	10-15 cm length,
Venation	Curved
Base	Slightly decurrent into stalk
Midrib	Paraller
Petiole	
Size	¾- 1, ¼ in Long
Shape	Round
Surface	Smooth

Table 5. Microscopical evaluation of *Buchanania angustifolia* Roxb. Leaf

Sl. No	Features	Observations
1	Trichomes	Absent
2	Epidermis	Radially oblong thick walled cells
3	Vascular bundles	Xylem and Phloem
4	Collenchyma cells	Present
5	Parenchyma	Ground

Table 6. Powder characteristic of *Buchanania angustifolia* Roxb. (Anacardiaceae) leaves

Sl. No.	Features	Observation
1	Nature	Coarse powder
2	Color	Dark Green
3	Odour	Characteristic
4	Taste	Acrid
Microscopic characteristics		
5	Trichomes	Absent
6	Lamina	Present
7	Stomata	Anomocytic

Table 7. Physicochemical Characterization of *Buchanania angustifolia* Roxb. Leaf powder

S. No	Parameters	% W/W
1.	Ash values	
	a) Total ash	12.8
	b) Acid insoluble ash	4.2
	c) Water soluble ash	10.5
2.	Extractive values	
	a) Alcohol soluble Extractive	15.4
	b) water soluble Extractive	13.9
3	Loss on drying	11.82
4	Swelling index	20.9

Table 8. Fluorescence Analysis of *Buchanania angustifolia* Roxb. (Anacardiaceae) leaf Powder under visible and UV light.

S. No.	Treatment	Visible light	UV short 254 nm	UV long 365 nm
1.	Powder as such	Green	Brown	Green
2.	Powder + NaoH in methanol	Green	Dark brown	Brown fluorescence
3.	Powder + 1N hydrochloric acid	Green	Brown	Greenish brown
4.	Powder + Nitric acid	Green	Dark green	Brown
5.	Powder +Sulphuric acid	Green	Green	Brown
6.	Powder +Acetic acid	Green	Dark green	Brown
7.	Powder +Picric acid	Yellowish brown	Brown	Yellow

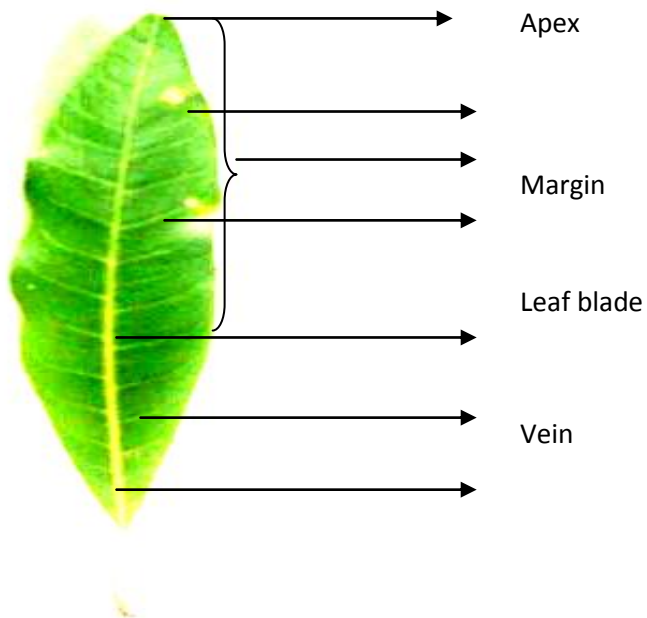
Table 9. Fluorescence analysis of various extracts of Leaves of *Buchanania angustifolia* Roxb

Extract	Day light	Long UV 365nm	Short UV 250 nm
Petroleum ether	Green	Red	Greenish brown
Methanol	Green	Orange	Green
Distilled water	light green	Light Green	Brown

Fig 1. Twig of *Buchanania angustifolia*



Fig 2. Botanical evaluation of leaf of *Buchanania angustifolia* Roxb



Figures of microscopical studies of *Buchanania angustifolia* Roxb. (Anacardiaceae) leaf
Fig 3.1. Transverse Section of leaf through midrib with lamina

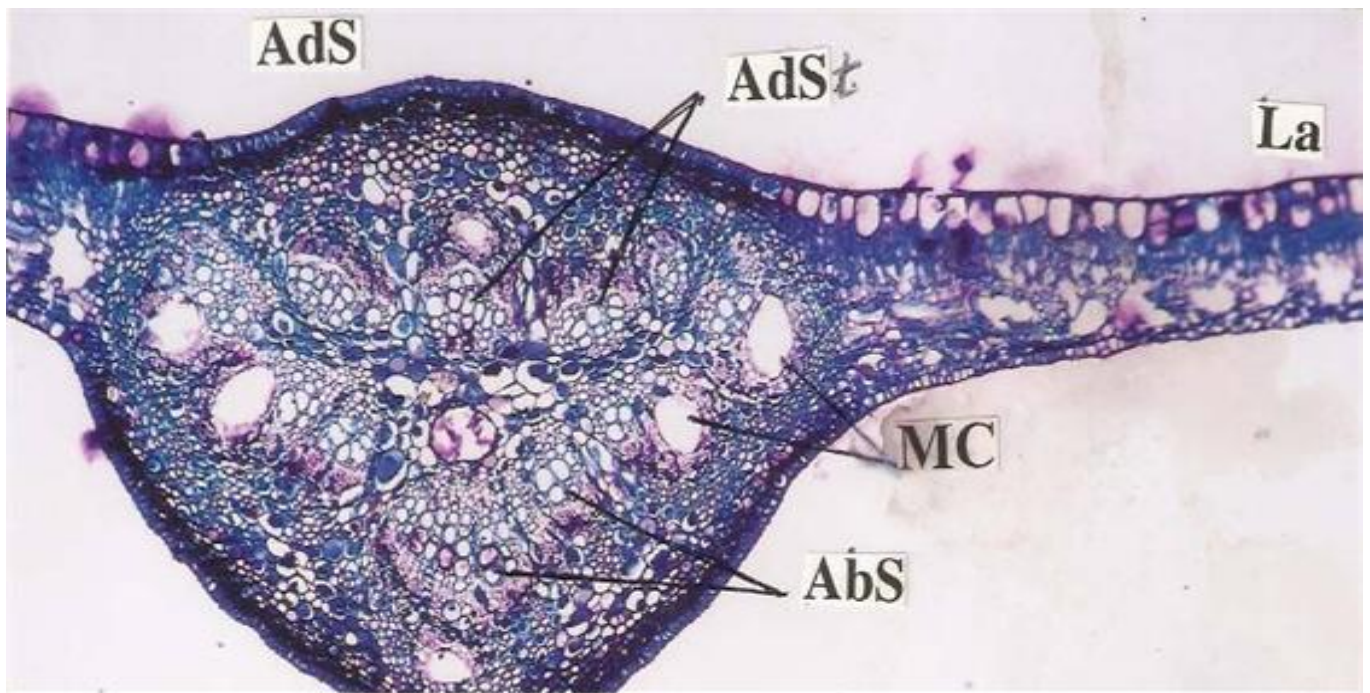
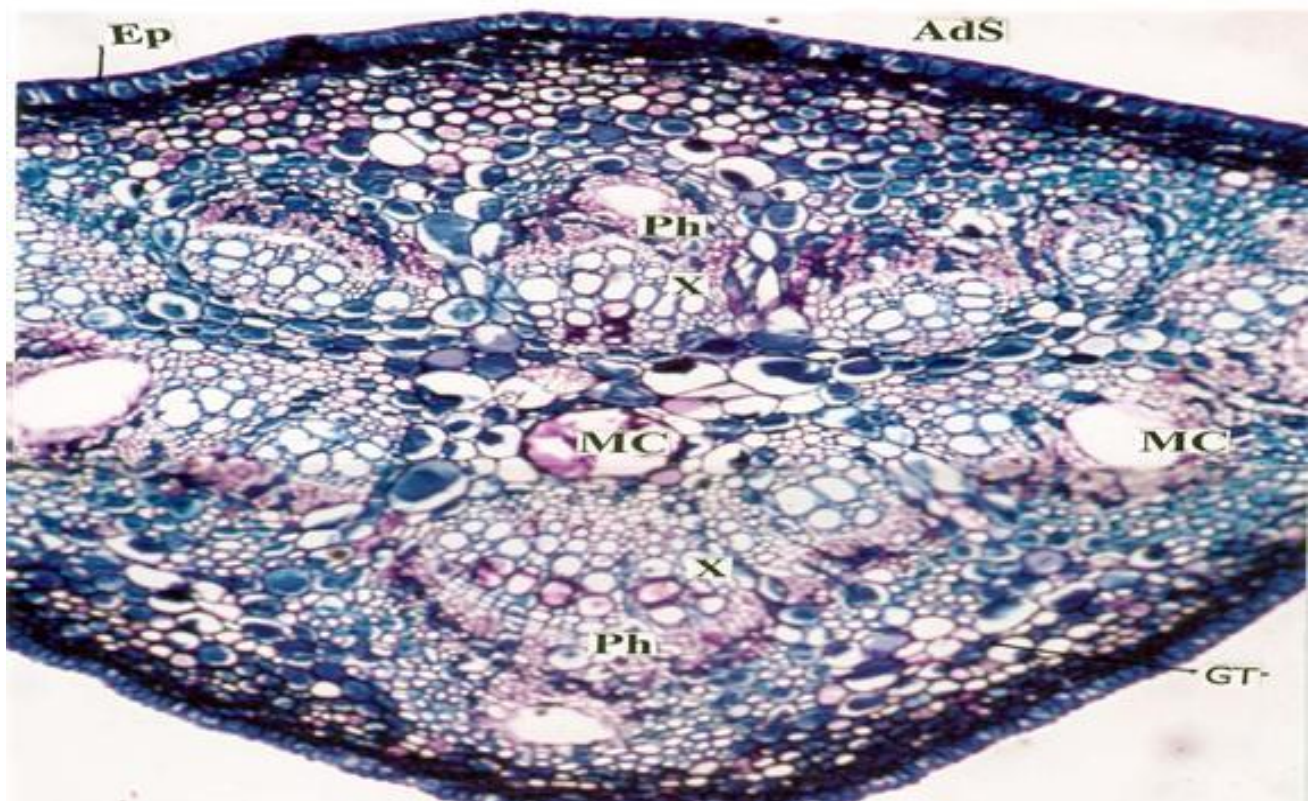


Fig 3.2. Transverse Section of midrib – enlarged



(Abs – Abaxial side ; Ads – Adaxial side; Adst – Adaxial strand; Ep – Epidermis;
GT – Ground tissue; La – Lamina; MC – Mucilage – Cavity; Ph – Phloem; X – Xylem)

Fig 4. Transverse Section of leaf showing crystals in the ground tissue

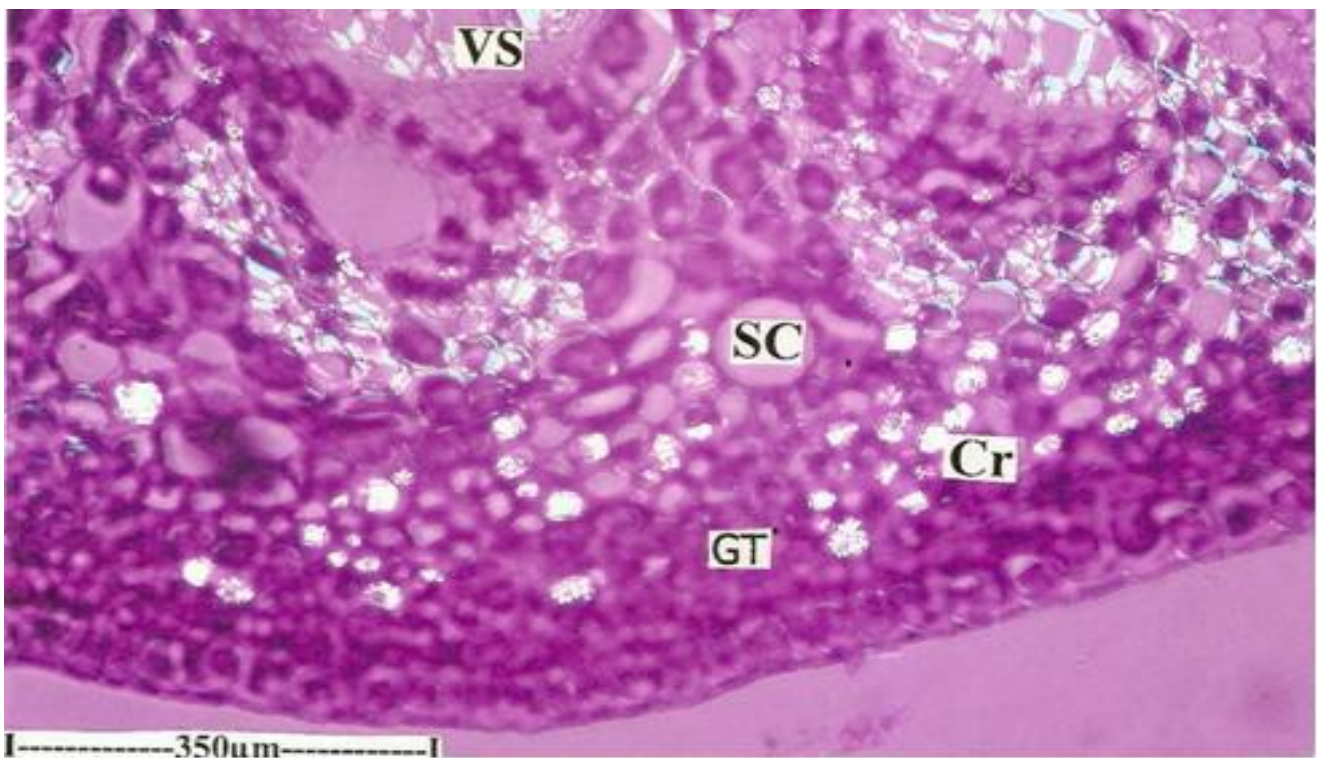
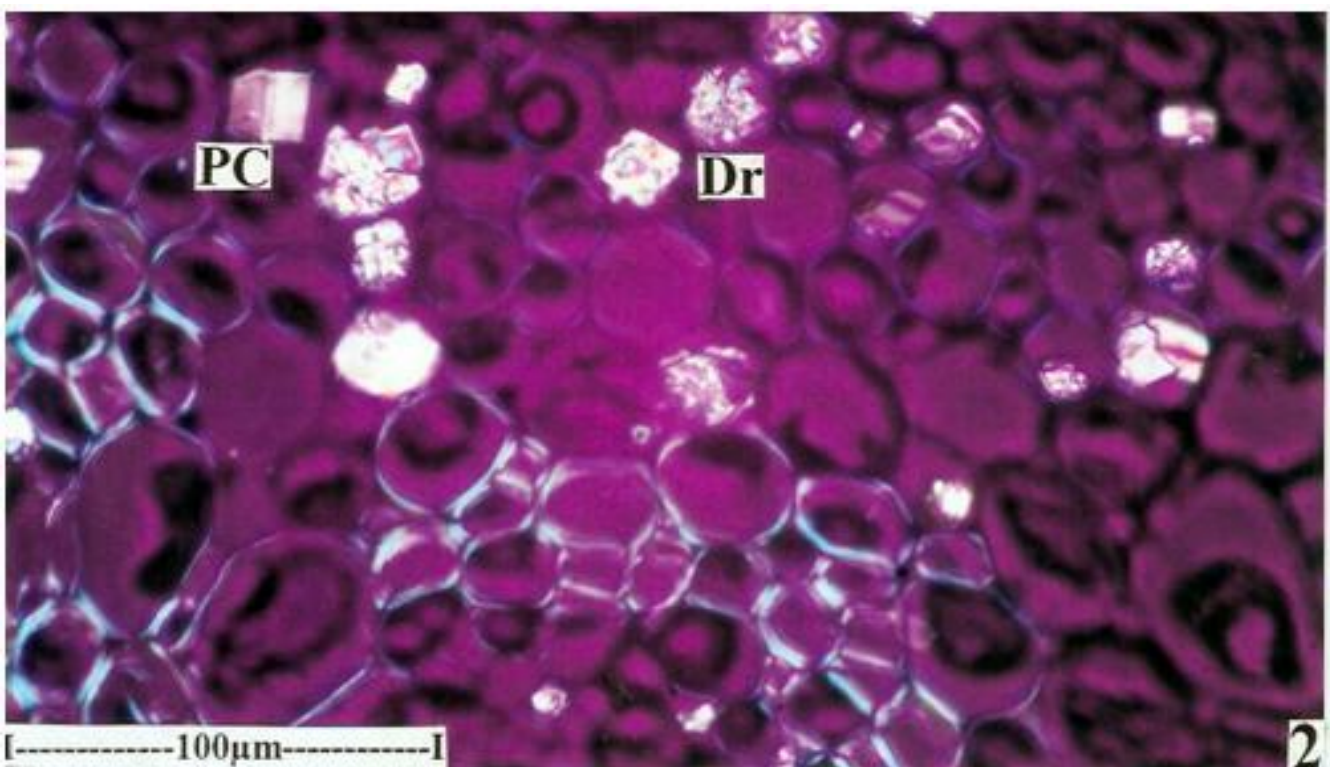


Fig 5. Transverse Section of leaf showing crystals in the ground tissue Enlarged



(Cr – crystals; Dr – Druses; GT – Ground tissue; PC – Prismatic Crystals; SC – Secretary Cavity; VS – Vascular strand).

Fig 6. Transverse Section of lamina through lateral vein

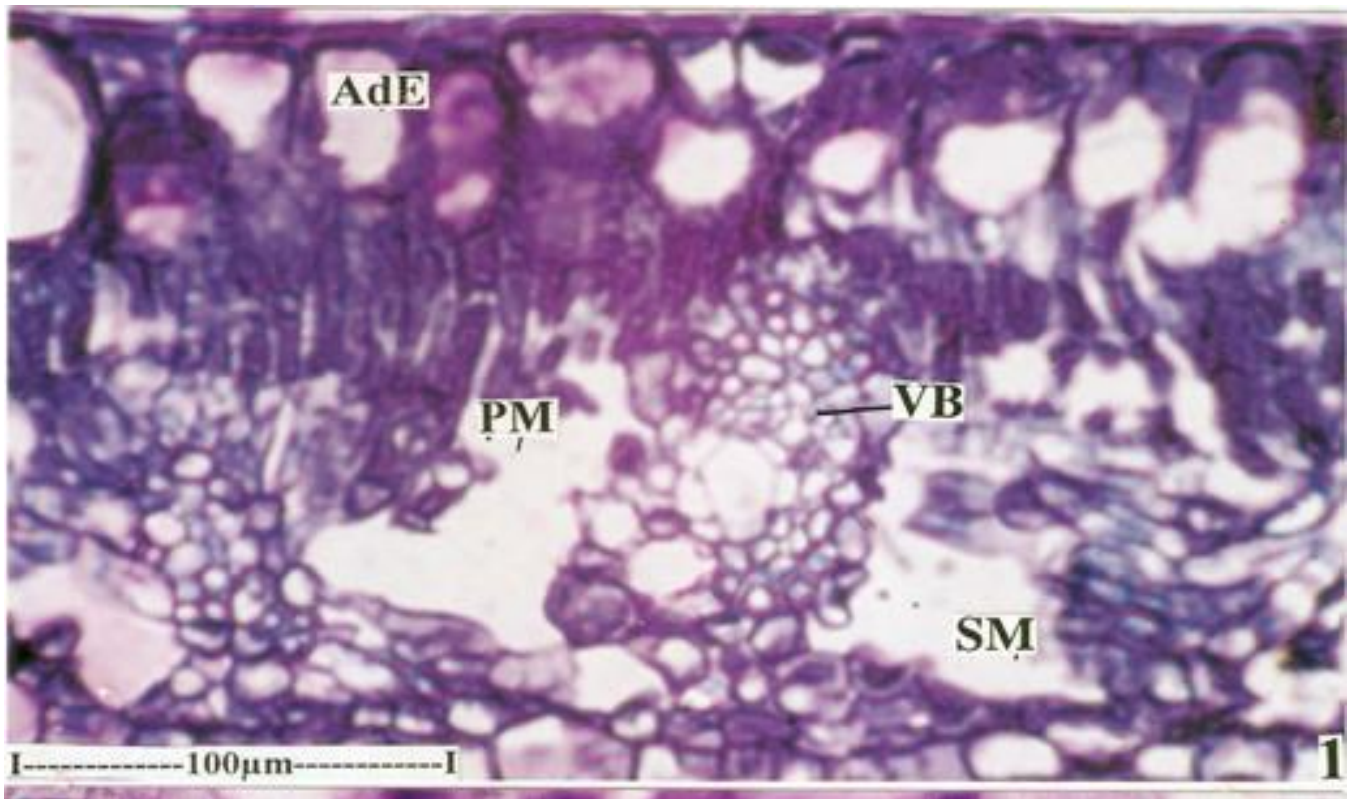


Fig 7. Transverse Section of lamina showing mucilage cavities

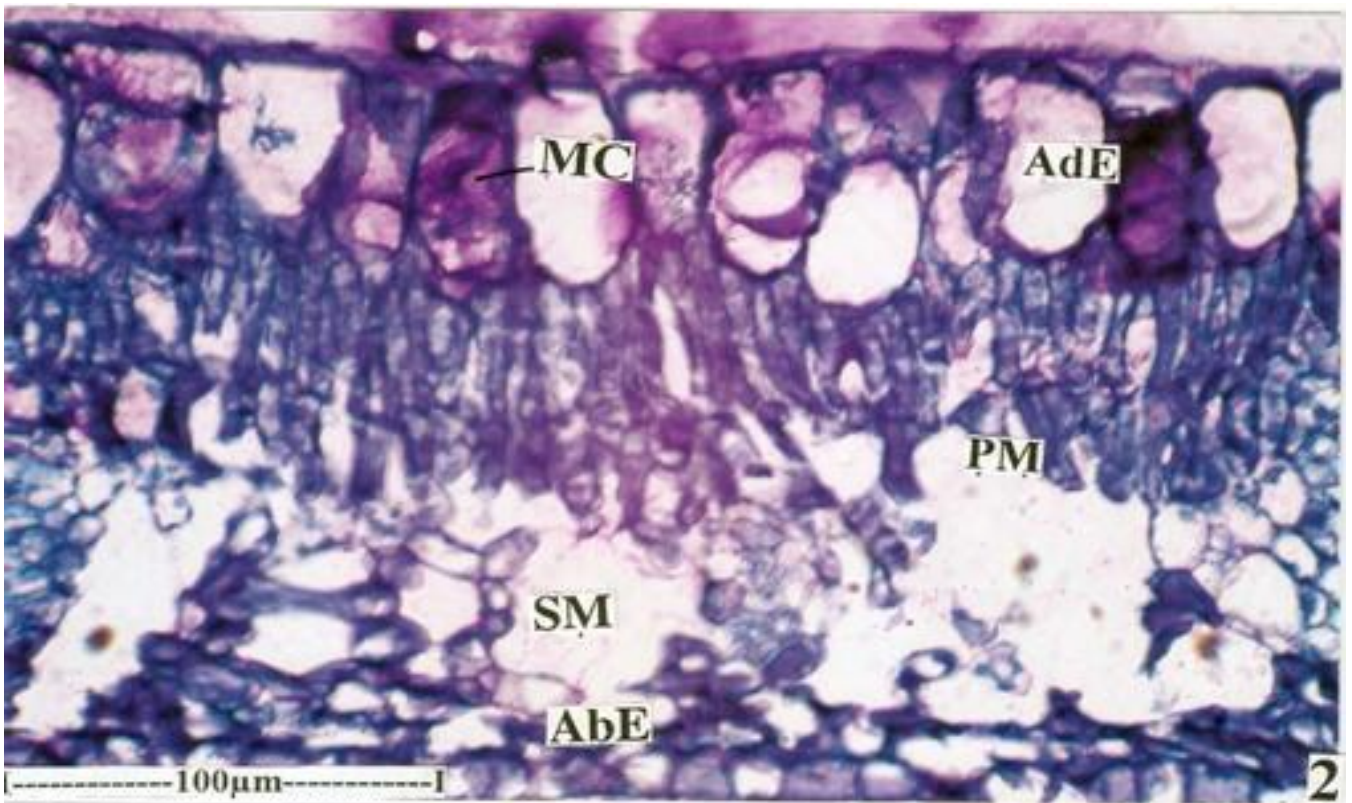
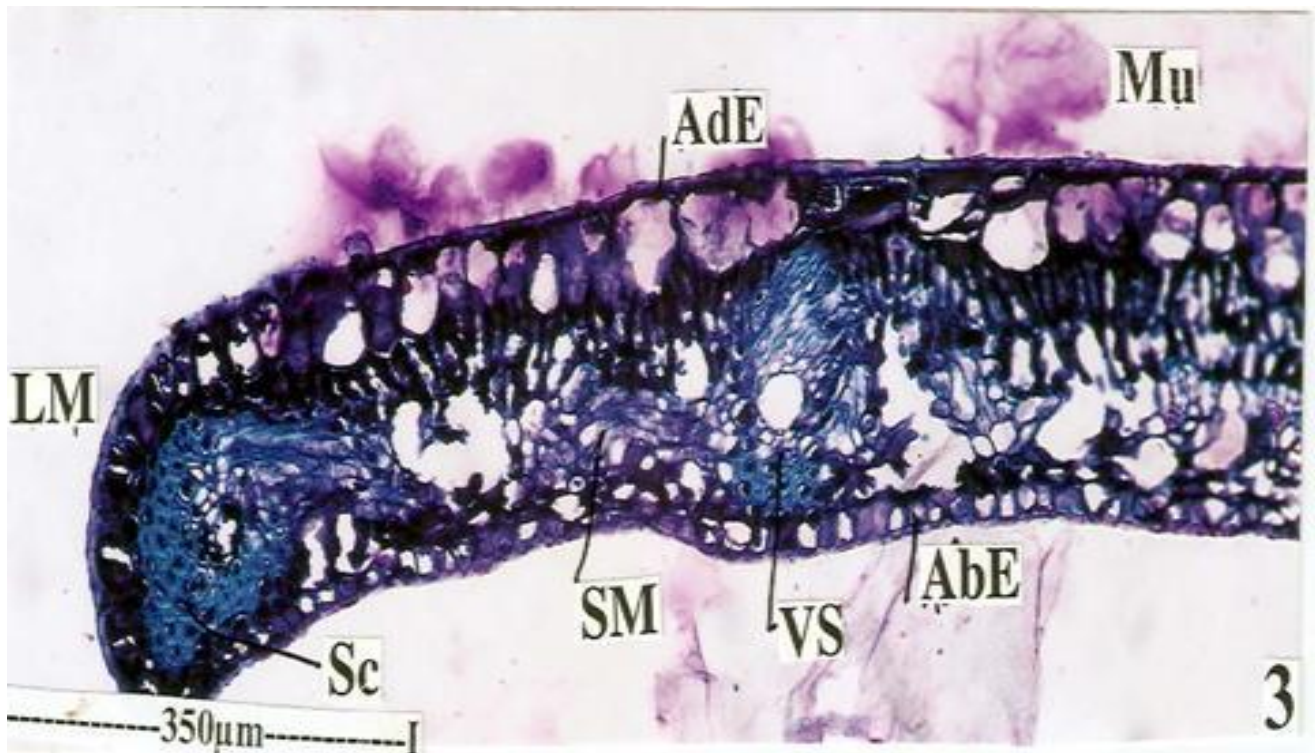


Fig 8. Transverse Section of leaf margin



AbE – Abaxial Epidermis; AdE – Adaxial Epidermis; LM – Leaf margin; MC – Mucilage cavity; Mu – Mucilage; PM – Palisade mesophyll; SC – Sclerenchyma; SM – Spongy mesophyll; VB Vascular bundle; VS – vascular stand.

Fig 9. Paradermal Section showing Adxial epidermis

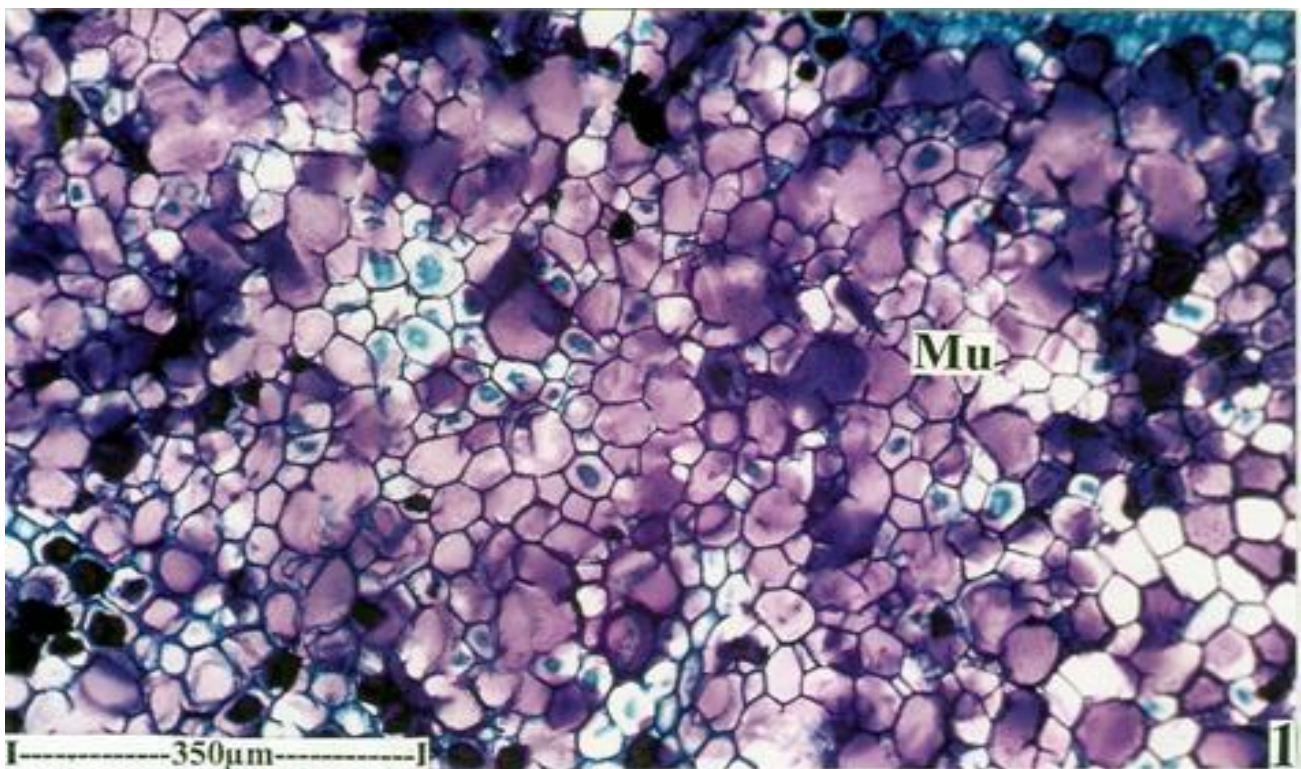


Fig 10. Paradermal Section showing Abaxial Epidermis with stomata

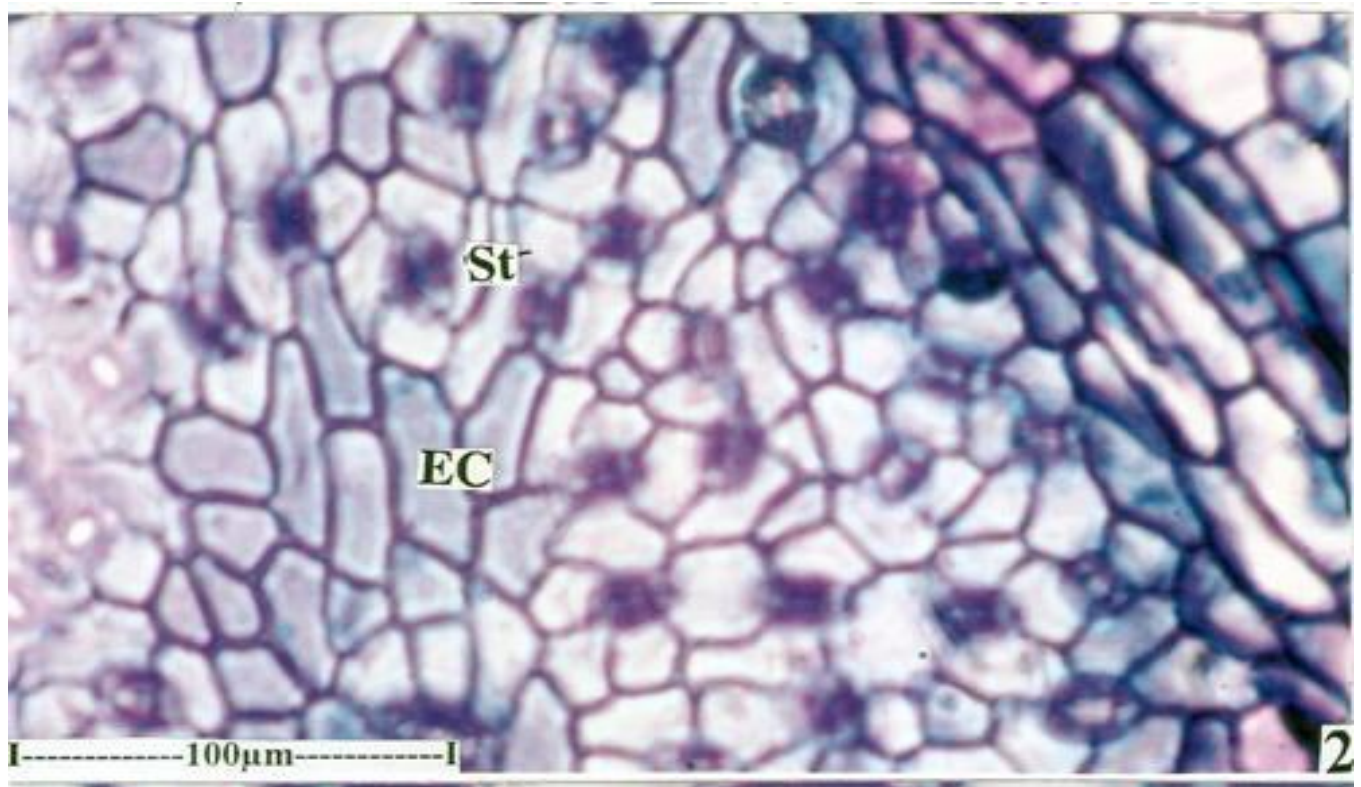
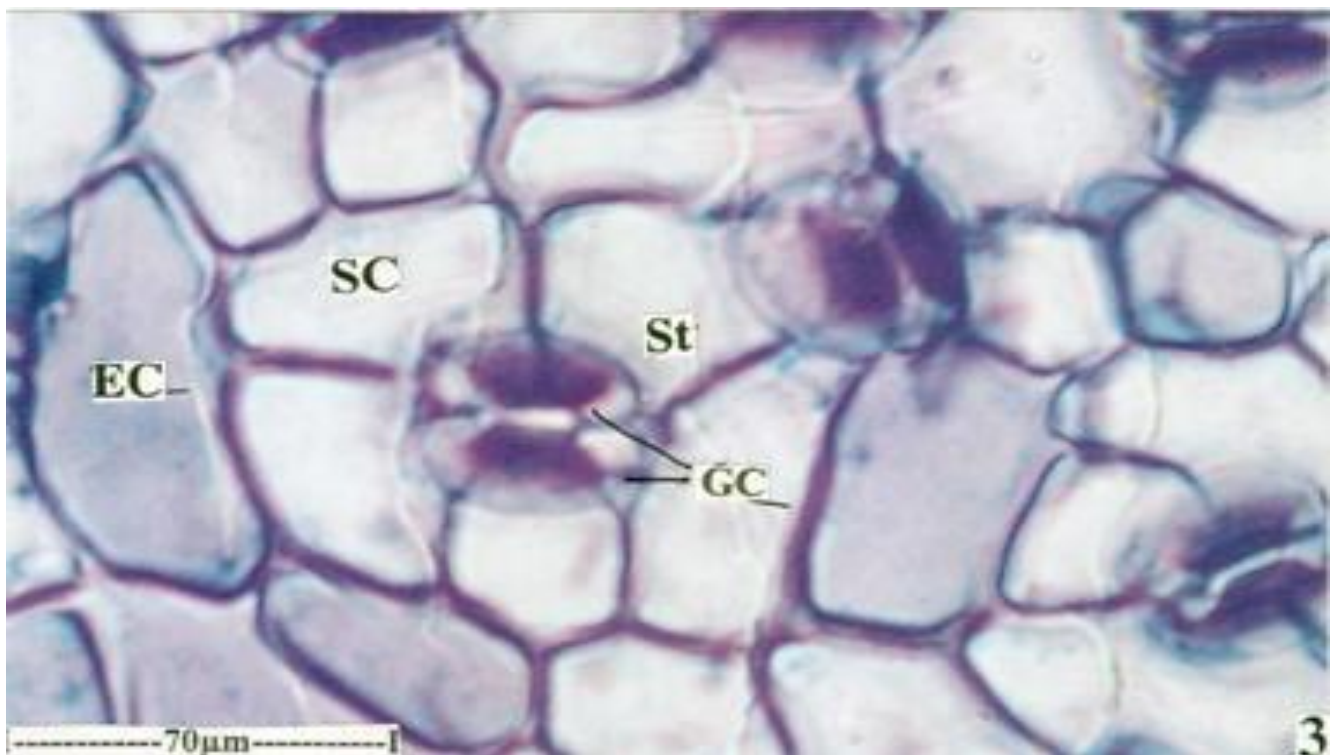


Fig 11. Paradermal Section showing Abaxial Epidermis with stomata enlarged



(EC – Epidermal cell; GC – Guard cell; Mu – Mucilage; SC – Subsidiary cell; St – stomata).

Fig 12. Paradermal section showing vein-islets and vein-termination

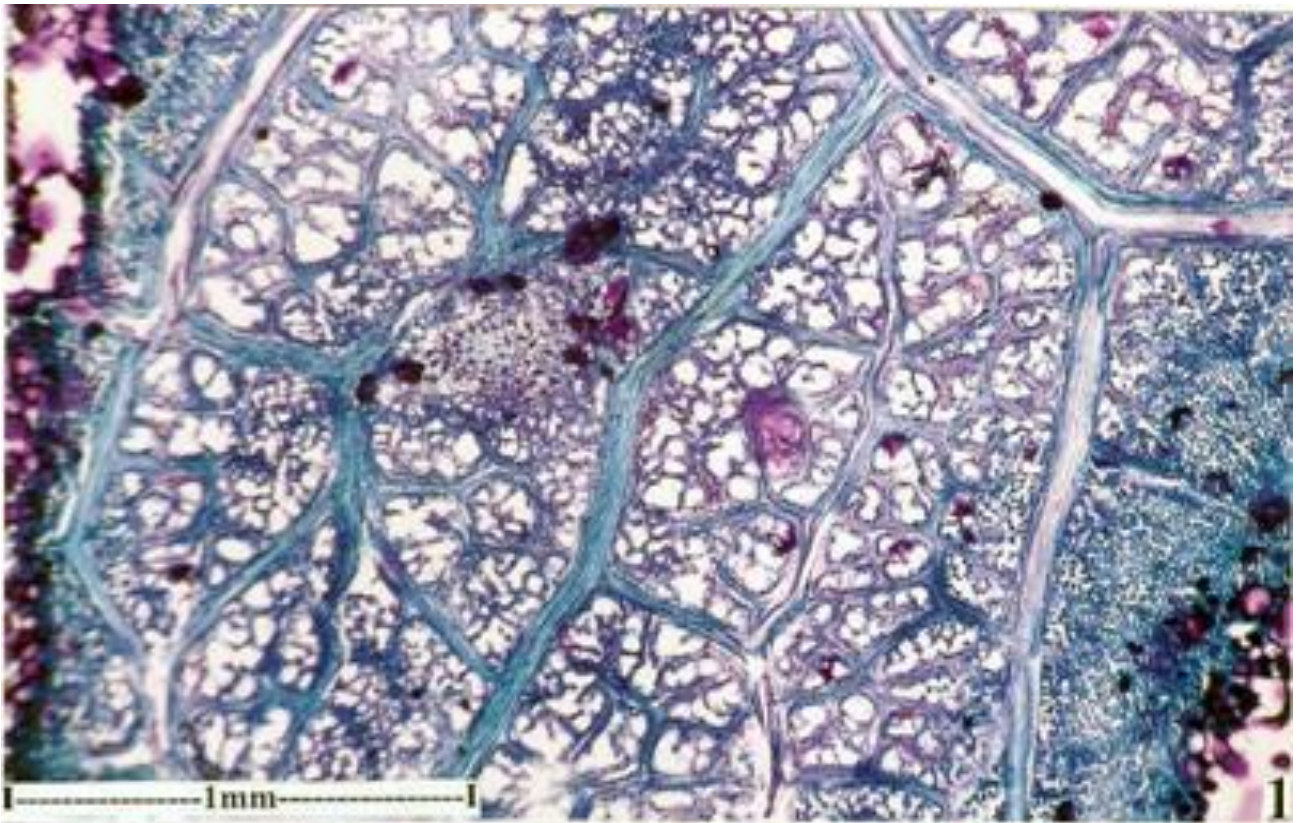


Fig 13. 1.Paradermal section showing vein-islets and vein-termination enlarged

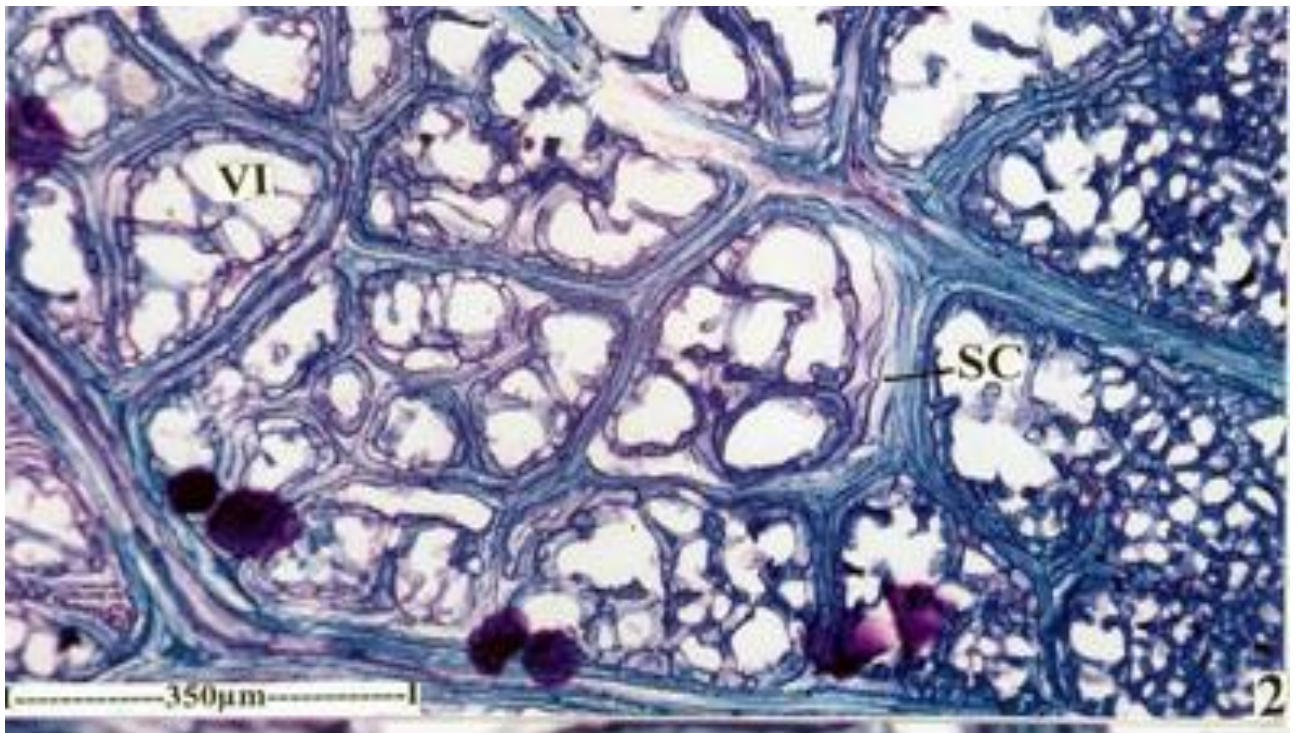


Fig 13.2. One vein-islets and vein-termination enlarged



(SC -sclereids; VI – Vein – islets, VT – Vein – termination)

Fig 14. Cleared leaf showing vein-islets and vein-termination

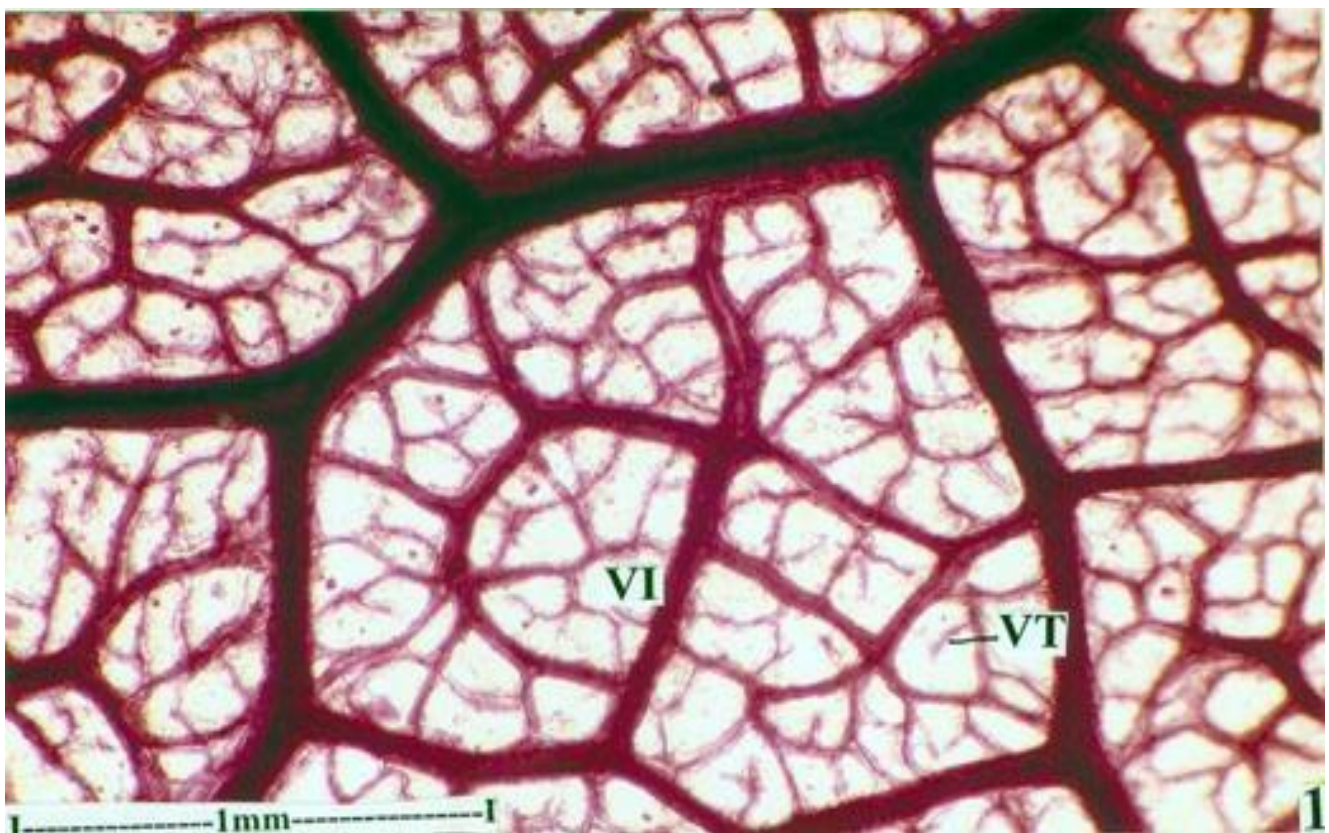
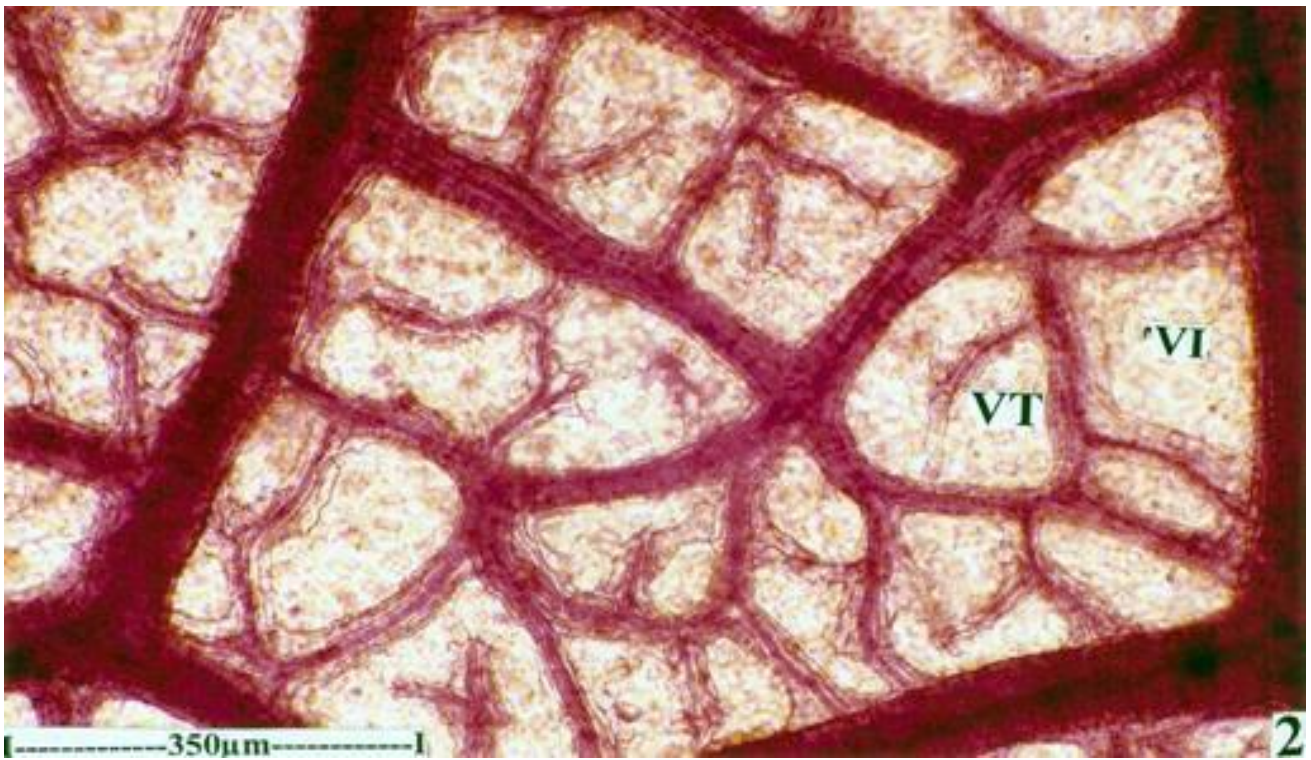


Fig 15. Cleared leaf showing vein-islets and vein-termination – a portion magnified



(VI – Vein-Islets, VT – Vein – termination)

Fig 16. Vein – islets with vein – termination

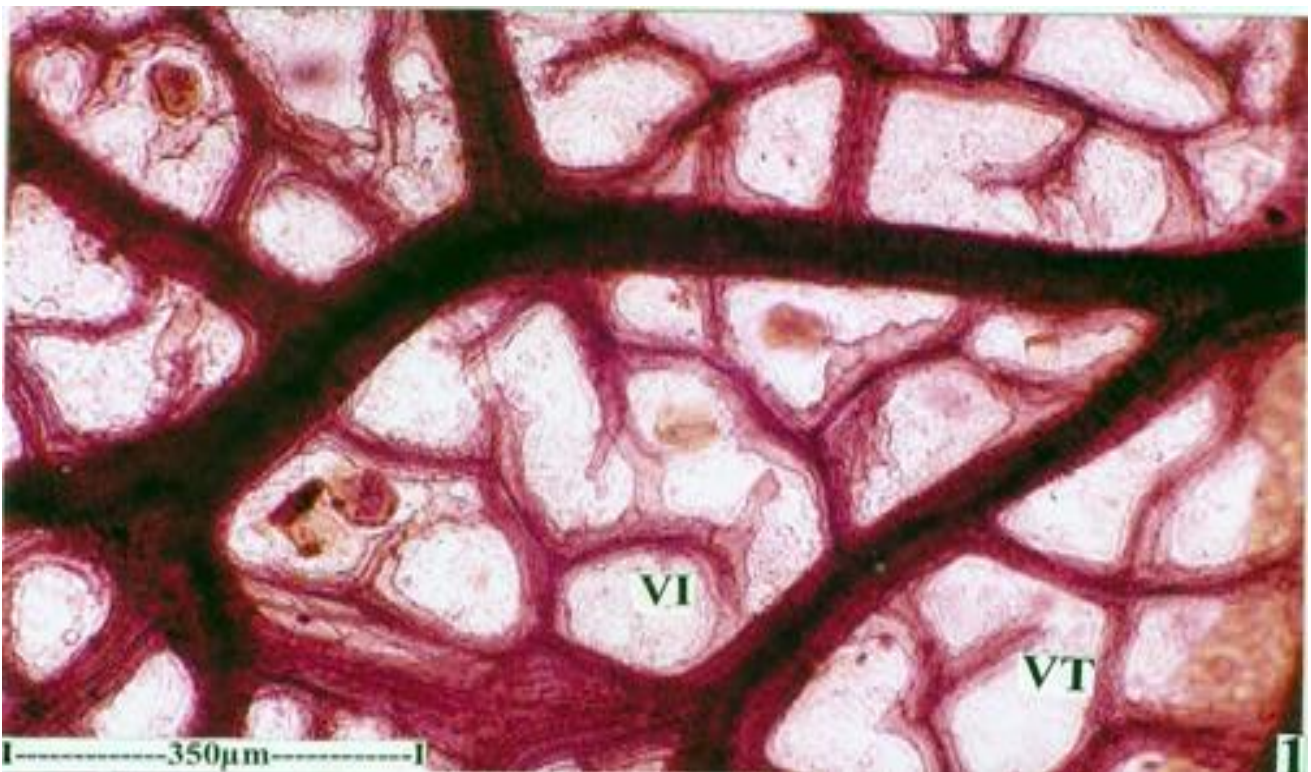
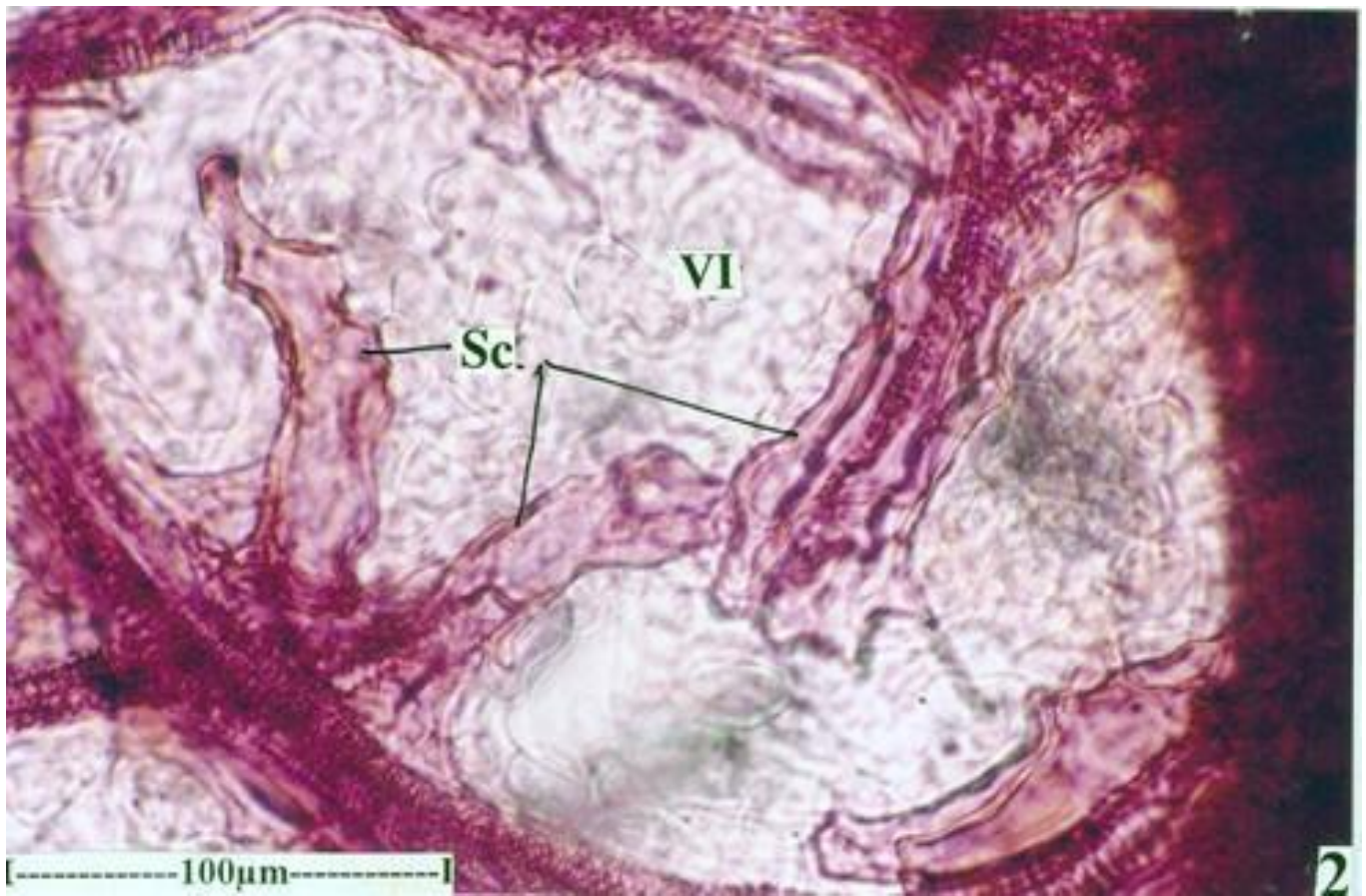


Fig 17. Sclerotic vein termination enlarged

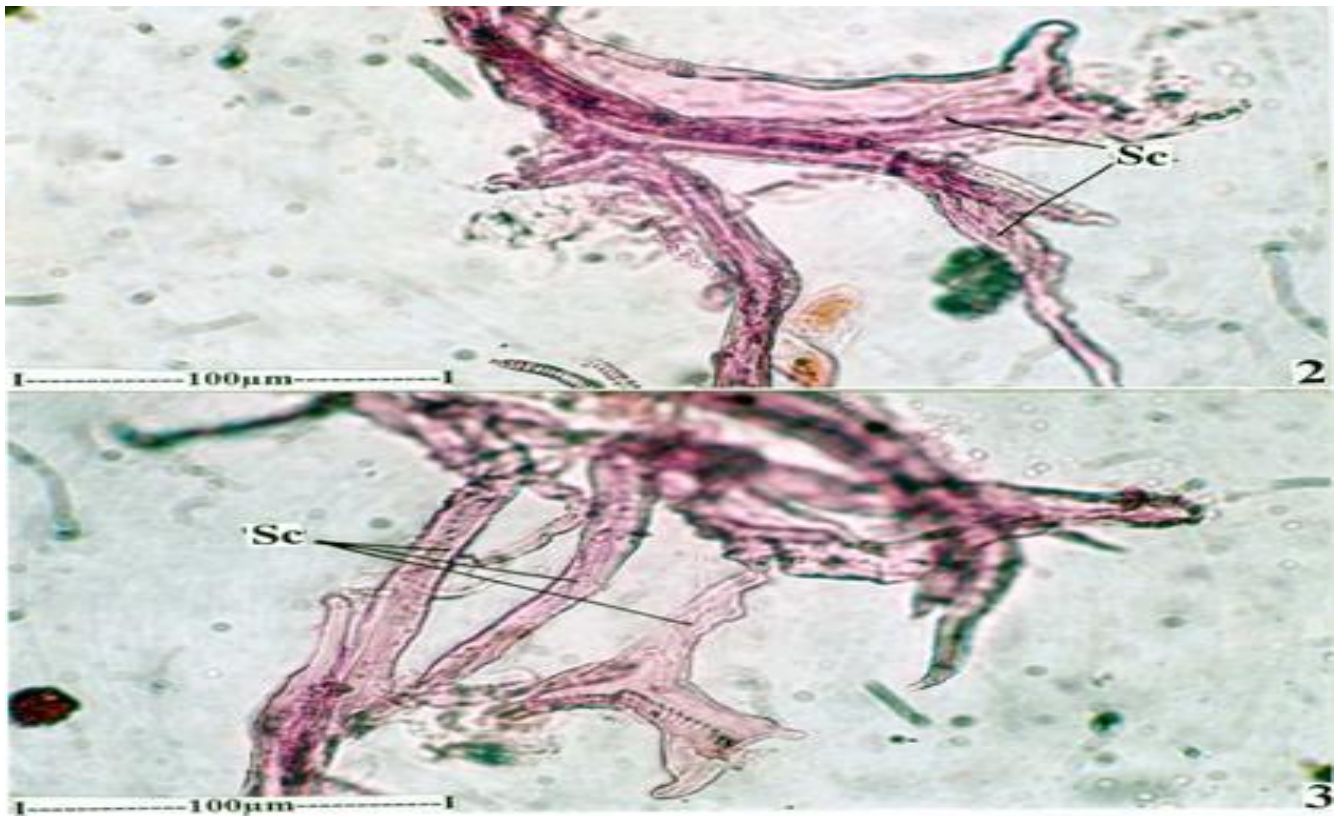


(Fi – Fibre; Sc – Sclereids; VI-Vein-islets; VT- vein- Termination)

Fig 18. Leaf powder showing a fibre



Fig 19. Sclereids in the leaf powder



(Fi – Fibre, SC – Sclereids)

Fig 20. Fragment of leaf showing adaxial epidermis

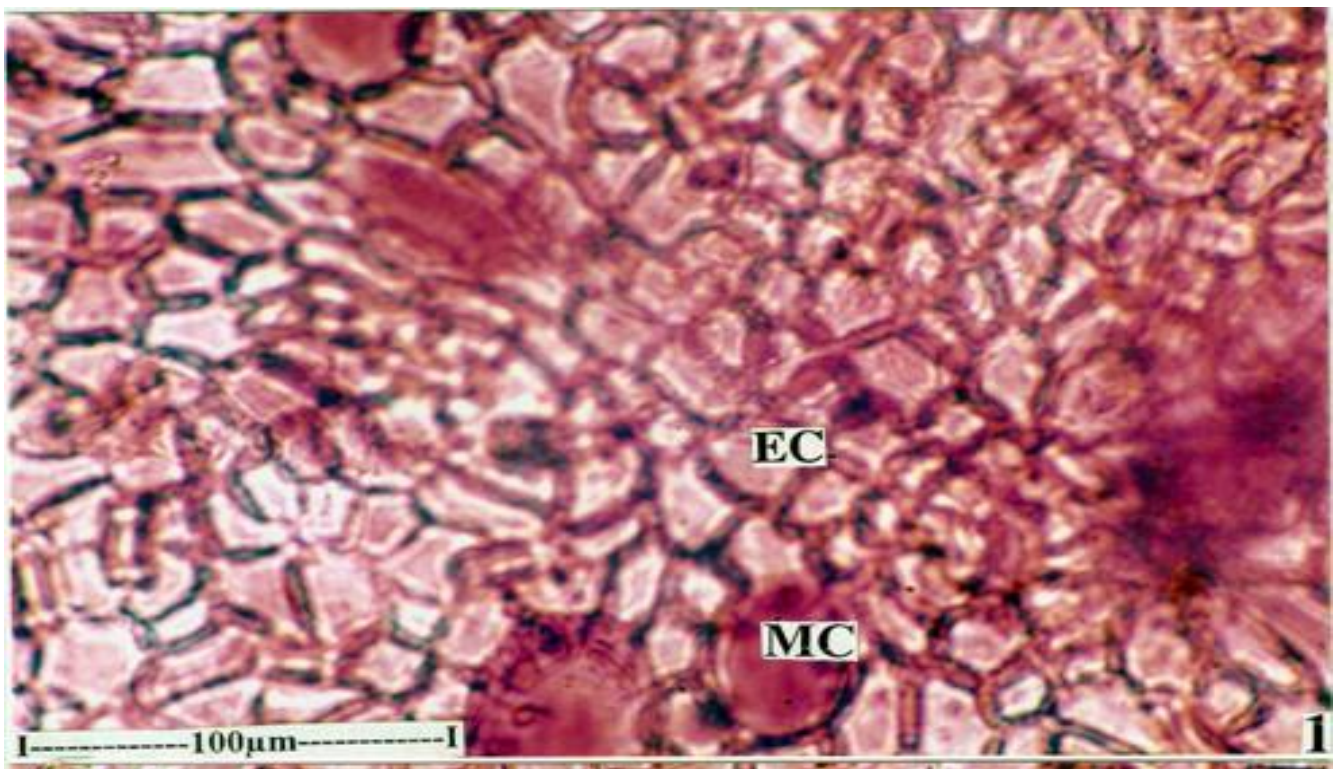
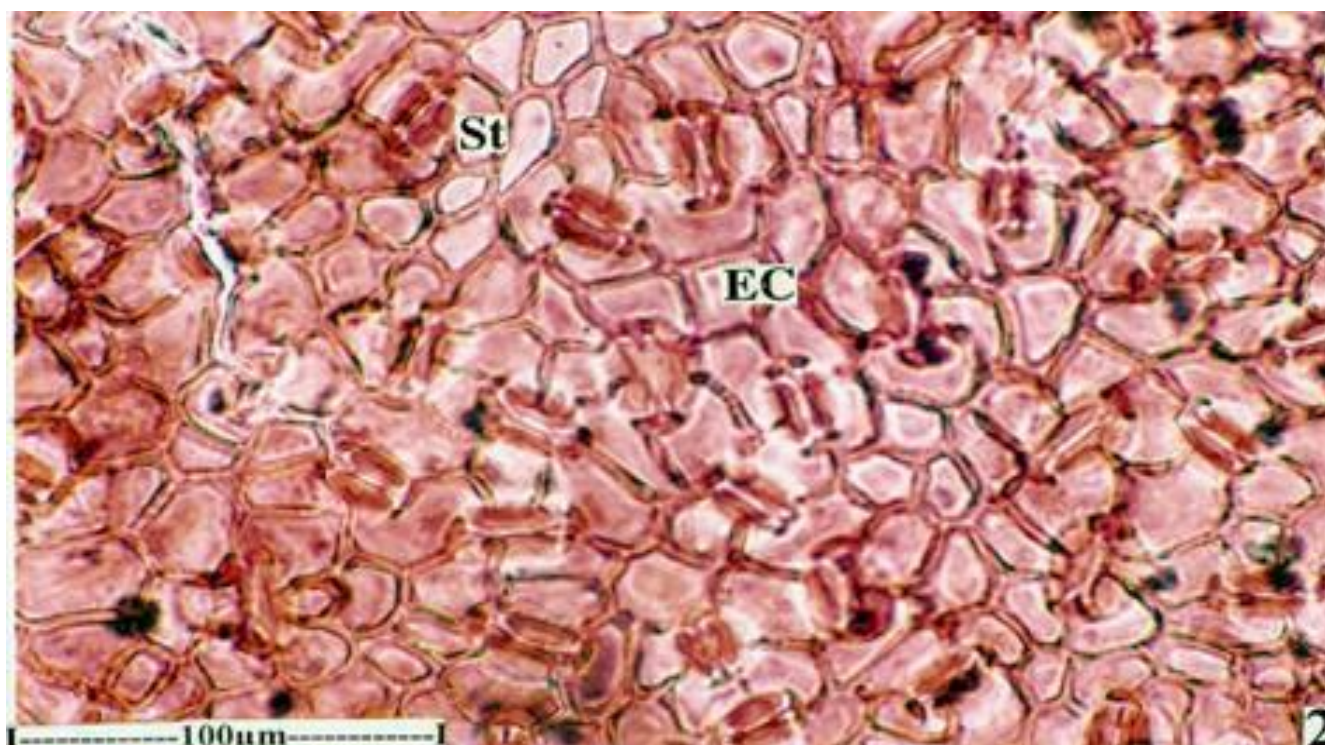


Fig 21. Abaxial epidermis with stomata



(EC – Epidermal Cell, MC – Mucilage cavity; St – Stomata)

CONCLUSION

Phytochemical evaluation of *Buchanania angustifolia* Roxb. (Anacardiaceae) Leaves provided useful information like the presence of various phytoconstituents such as carbohydrates, proteins, terpenoids, phenols, glycosides, gums and mucilage, saponins and steroids. The colour consistency and the percentage yield of the extracts were also determined and reported.

Pharmacognostic evaluation of *Buchanania angustifolia* Roxb. leaves provided useful information regarding its correct identity and helped it from the closely related other species of *Buchanania angustifolia* Roxb. The other parameters observed are also useful for the future identification of the plant, and serves as a standard monograph for identification and evaluation of plant.

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