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PHARMACOGNOSTICAL STUDIES ON THE STEM BARK OF "ANNONA GLABRA"

R. Sinchana *, T. Tamizh Mani, T. Pavithra and L. Shiju

Department of Pharmacognosy, Bharathi College of Pharmacy, Bharathinagara, Mandya - 571422, Karnataka, India.

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Correspondence to Author: R. Sinchana

Department of Pharmacognosy, Bharathi College of Pharmacy, Bharathinagara, Mandya - 571422, Karnataka, India.

E-mail: sinchanagowda802@gmail.com

ABSTRACT: In the present study, an attempt was made to investigate Pharmacognostical studies on the stem bark of Annona glabra (Annonaceae). The plant was identified and authenticated by Dr. V. Rama Rao, Research officer (Botany), Central Ayurveda Research Institute, Bengaluru. The macroscopical studies have been carried out on the stem bark. The Characters of transverse section of stem bark shows periderm, consisting of 3-4 layers of cork, 2-3 layers of phellogen and 6-7 layers of Phelloderm. The cortex region shows the presence of parenchymatous cells. Group of fibres in patches in the outer and inner cortex. The secondary cortex consists of moderately thick-walled, parenchymatous cells. Medullary rays were present, these are bi, tri or multiseriates in the region of the inner secondary phloem. Starch grains are present. The longitudinal section shows multiseriate ray initials and single, long, septate fusiform initials, ends are arranged at different levels fusiform initials or non-stratified cambium. Starch grains are also present in the parenchyma cells. Powder shows reddish brown in colour, fragments of parenchymatous cells embedded with brownish content; fragments of cork cells; Group of Prismatic Crystals; Pitted tracheid; Scalari form vessel; Xylem fibres, trichomes with tannin content. Physicochemical parameters such as moisture content, total ash value, sulphated ash value, Acid insoluble ash value, water-soluble ash value and extractive value were determined. These can serve in qualifying and differentiating the plant. This research provides valuable insights that will benefit future researchers in their endeavors.

INTRODUCTION: Nature has been one of the main sources for the traditional medicine for thousands of years, and large number of modern drugs have been isolated from natural sources ¹. Plant-derived natural products have extremely high potential to developed as medicines ².



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One such plant is *Annona glabra*, belonging to the genus Annona and the family Annonaceae. It is a very large family of plants, comprising about 120 genera and more than 2000 species ³.

Annona glabra L., is commonly known as pond Apple, alligator apple, monkey apple, swamp apple, cow apple, cork wood, bob wood ⁴. It is a natural introduction to the mangroves of southern Kerala and has widely spread along the backwaters. A. glabra is a small-woody tree that grows up to 3– 12 m hight ¹. A. glabra is made up of small plants (shrubs or trees), is fruitful when ripened, the fruit changes its colour from green to either yellow or orange. Stems are grey with prominent lenticels, the leaves are arranged alternately and are oblongepithelial, with acute or shortly acuminate tips, Flowers are short-lived and rarely noticed, 2-3 cm in diameter, pale-yellow to cream with three leathery outer petals and three smaller inner petals ⁵. Many active compounds have been found in A. glabra, mainly flavonoids, glycosides, saponins, acidic compounds, tannins, steroids, anthraquinones ¹. The A. glabra fruit is edible and can be made into jam. In the Maldives, it is a popular ingredient in fresh fruit drinks. The crushed seed was cooked with coconut oil and applied to get rid of head lice in older days. A. glabra is used in traditional medicines against several ailments, such as fever, constipation, ulcers, and tumours, including cancer. It is a kind of survival food ⁶. The leaves and young stems, sometimes combined with the leaves and stems of Passiflora foetida, are boiled to make a tea, which is drunk to destroy worms and nematodes. The bark and leaves, combined with the bark and leaves of Annona squamosa, are used as sedatives and cardiotonic infusions. The wood is used to make bottle caps, oars, and as a substitute for cork in fishing nets. Seeds and leaves are insecticidal, leaves placed in hen nests kill lice on the fowl. Auseful fibre is obtained from the bark. It is sometimes used locally ⁷. Annona plants have several scientifically proven pharmacological effects, such as anticancer, antidiabetic, antidiarrhea, antiulcer, antimalarial,

antioxidant, antileishmanial, anti-inflammatory, antibacterial, antifungal, antidepressant, antinociceptive, anticonvulsant. anti-acetyl cholinesterase, and dengue vector control activity 8. As there is no scientific data on Annona glabra bark, here in the present study plant was taken for macroscopical, microscopical studies quantitative evaluations were also carried out. In microscopical studies T.S, L.S and powder drug analysis of bark were carried out. In quantitative evaluations, moisture content, water and alcohol soluble extractive value, total ash, water soluble ash, acid insoluble ash, sulphated ash values were carried out for crude dried powder drug.

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Taxonomy 9:

Kingdom = Plantae

Phylum = Tracheophyta

Subphylum = Angiospermae

Class = Magnoliopsida – Dicotyledons

Order = Magnoliales

Family = Annonaceae

Genus = Annona

Species = Annona glabra

Botanical name = *Annona glabra Linn*.

Vernacular Names 4:

Pond apple, Alligator apple, Bob wood, Corkwood, Cow apple, Mangrove Annona, Monkey apple English

Malayalam Kattathi, Kattu-Aatha, and Kadalatha

Kannada Hanumapala

Germany Alligatorapfel, Alligator-Binnbaum, Annone

Netherlands Zuurzak

Brazil Araticum bravo, Araticum do brejo, Araticum-caca, Araticupana

China Yuan Hua Fan Li Zhi, Niu xin guo

France Anone des marais, Bois flot, Cachiman cochon, and Corossol des narais

Spanish Anona Lisa, Anon Liso, Anon de Puerco, and Anonillo Cabuye

Japanese Pondo appuru

Jaca de pobre, Araticum do brejo, and Araticurana Portuguese

MATERIALS AND METHODS:

Collection of Plant Material: The plant material was collected from H.G doddi, Mandya district, Karnataka, India in the month of march 2024. The plant was identified and authenticated by Dr. V. Rama Rao, Research officer (Botany), Central Ayurveda Research Institute, Bengaluru. An herbarium voucher specimen was preserved in the department of Pharmacognosy, Bharathi College of Pharmacy, Bharathinagar for further reference.

Drying and size Reduction of Bark ¹⁰: The bark of Annona glabra were subjected to shade drying and further crushed to powder, and then the powder

is passed through the sieve no. 80, and stored in air tight container for further use.

Experimental Procedure:

Macroscopical Studies ¹¹: Stem bark of *Annona glabra* was studied macroscopically for examining its colour, odour, taste, size, shape, fracture and texture. Macroscopic examination of crude drug was carried out by naked eye by placing the individual raw materials on a white paper surface.

Microscopical Studies ¹²: Microscopical study has been carried out by taking free-hand sections of fresh stem bark. Thin sections were cleared with chloral hydrate solution followed by water and stained with safranin and observed under the microscope. As well as the dried bark powder was evaluated by treated with chloral hydrate solution followed by water and stained with safranin and

observed under the microscope. Microphotographs have taken by using CatCam microscope camera fixed with the microscope (Model: OLYMPUS CX31).

Physicochemical Constants ¹³: Physicochemical constants such as the percentage of moisture content, total ash, acid insoluble ash, water soluble ash, sulphated ash, water and alcohol soluble extractives, loss of weight on drying, were calculated based upon standard procedures prescribed in Indian Pharmacopoeia.

Preliminary Phytochemical Studies ¹⁴: Preliminary phytochemical test for stem bark of *Annona glabra* were performed and chemical constutients were determined by using standard procedures described by Kokate C.K., Purohit A.P., and Gokhale S.B.

RESULTSAND DISCUSSION: Macroscopical Studies:



FIG. 1: A. ANNONA GLABRA PLANT, B. BARK POWDER, C. BARK

TABLE 1: MACROSCOPICAL CHARACTER OF STEM BARK OF ANNONA GLABRA INCLUDES

Colour	Grayish-brown			
Odour	Characteristic			
Taste	Woody			
Size	3-60 ft. tall, 2 to 20 ft. wide (varies)			
Shape	Flat			
Fracture	Fibrous			
Texture	Smooth			

Microscopical Character:

Transverse Section of Stem Bark: The transverse section preparation of the stem bark was studied under the microscope and the following inclusions were recorded. It shows periderm, consisting of 3-4 layers of cork, 2-3 layers of phellogen and 6-7

layers of Phelloderm. The cortex region shows the presence of compact, thin-walled, medium-sized parenchymatous cells. Group of fibres in patches in the outer and inner cortex, inner cortex consists of thin-walled parenchymatous cells embedded with a group of fibres and reddish content. The secondary of moderately thick-walled, cortex consists parenchymatous cells. Medullary rays were present, thin-walled, elongated, rectangular and gradually increasing in dimension towards the outer ends. These are bi, tri or multiseriates in the region of the inner secondary phloem. Starch grains are present Fig. 2-3.

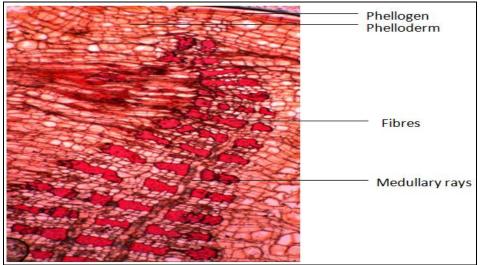


FIG. 2: TRANSVERSE SECTION OF STEM BARK

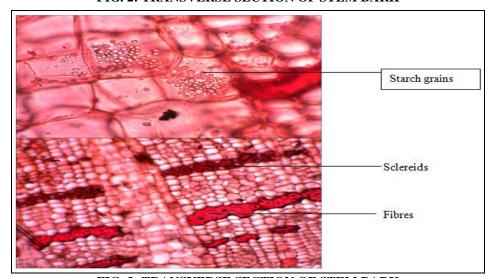


FIG. 3: TRANSVERSE SECTION OF STEM BARK

Longitudinal Section of Stem Bark- It shows multiseriate ray initials and single, long, septate fusiform initials, ends are arranged at different levels, non-storied fusiform initials or non-stratified cambium. Starch grains are also present in the parenchyma cells **Fig. 4.**

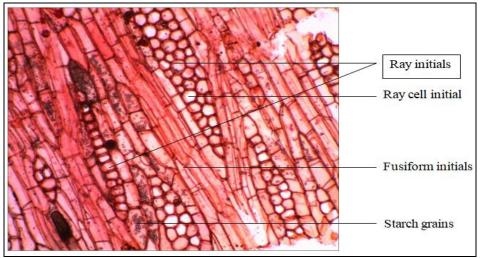


FIG. 4: LONGITUDINAL SECTION OF STEM BARK

Powder Microscopy of Stem Bark: Powder shows reddish brown in colour **Fig. 5B**, fragments of parenchymatous cells embedded with brownish

content; fragments of cork cells; Group of Prismatic Crystals; Pitted tracheid; Scalariform vessel; Xylem fibres.

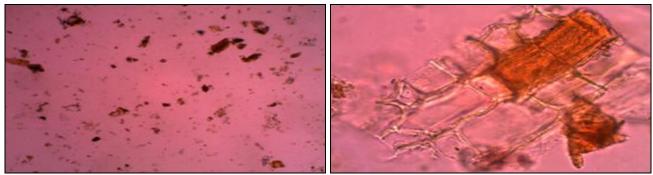


FIG. 5: DIFFERENT FRAGMENTS OF TISSUE IN THE BARK

FIG. 6: CELLS WITH BROWN IN THE BARK

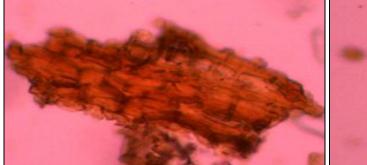


FIG. 7: CORK CELLS



FIG. 8: GROUP OF PRISMATIC CRYSTALS

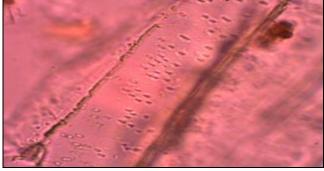


FIG. 9: PITTED TRACHEID

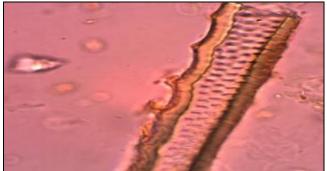


FIG. 10: SCALARIFORM VESSEL

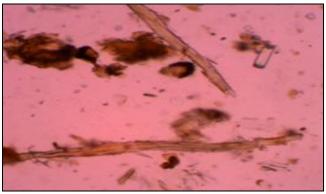


FIG. 11: XYLEM FIBRE

Physicochemical Parameters: The results were reported in Table 2 & 3. Moisture content determination is important for maintaining the Pharmacopoeial standards and it indicates the stability of the drug. Equally important in the evaluation of crude drugs is the total ash values. The total ash is particularly important in the

evaluation of purity of crude drugs, i.e. the presence or absence of foreign inorganic matter such as Metallic salts and/or silica. The alcohol yielded higher extractive value (18.4% W/W) when compared with water (12% W/W). The alcohol and water-soluble extractives were indicators of the total solvent soluble component.

TABLE 2: SHOWING RESULTS FOR QUANTITATIVE EVALUATION OF THE STEM BARK OF ANNONA GLABRA

Evaluation parameters (% W/W)	Stem bark (% W/W)
Moisture content	62.3
Total ash value	6.3
Acid insoluble ash value	0.5
Water soluble ash value	3.1
Sulphated ash value	12.3

TABLE 3: EXTRACTIVE VALUES OF STEM BARK OF ANNONA GLABRA

Evaluation parameters (%	Stem bark	
W/V)	(% W/V)	
Alcohol soluble extractive value	18.4	
Water soluble extractive value	12	

Preliminary Phytochemical Studies: The preliminary phytochemical investigation of the

petroleum ether, chloroform, ethyl acetate, methanol extract of stem bark of *Annona glabra* showed the presence of alkaloids, glycosides, saponin, flavonoid, carbohydrate, fat and oil, proteins and amino acid, steroid, triterpenoid presented in **Table 4.**

TABLE 4: QUALITATIVE ANALYSIS OF PHYTOCHEMICALS IN STEM BARK OF ANNONA GLABRA

Phytoconstutients	PE	CL	EA	ME
Alkaloids	+++	+	=	-
Glycosides	-	-	-	+++
Saponin	-	-	-	+++
Flavanoid	-	-	-	+++
Carbohydrate	-	-	++	-
Fat and oil	+	-	-	-
Proteins & amino acid	-	++	-	-
Steroid	++	+	-	-
Triterpenoid	++	+	-	-

Note: + =Present: - =Absent

CONCLUSION: Standardization of a crude drug is the current topic of interest so as to set a distinctive identity and quality of a crude drug. Most of the standard parameters were revealed

based on the microscopy and physicochemical constants of a plant. Before the plant enters in to Pharmacopoeia its standards should be established. So, to ensure the quality of the crude drug the standardization is utmost important so will be the medication including it. In the present work attempts to establish the Pharmacognostical standards for the same like macroscopical and microscopical features, physicochemical constants were done and hope this would help enrich the ayurvedic medicine and to potentiate the effective use of folklore medicinal practices. These standards might initiate the scientists who are keen and sincere to investigate the traditional claims of plant drugs.

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CONFLICTS OF INTEREST: No conflicts of interest.

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