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Original Research Article



Pharmacognostical Evaluation of Annona squamosa Linn.

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ABSTRACT

Annona sqamosa (Annonaceae) is known as sitaphal in Hindi and custard apple in English is an important medicinal plant. It has been used traditionally in diarrhea, dysentery, cold, as abortifacient, insecticidal drug etc., so would be more suitable to evaluate the drug. This paper deals with the detailed pharmacognostic evaluation of the crude drug. Morphoanatomy of the entire plant have been studied with the aim to aid pharmacognostic and taxonomic species identification using WHO recommended physico-chemical determinations and microscopical characters that will provide referential information for checking adulteration. The physico-chemical and histological parameters presented in this paper may be proposed as parameters to establish authenticity of *Annona squamosa* and can possibly help to differentiate the drug from its other species. Many important diagnostic characters such as bicollateral vascular bundle, paracytic stomata, pitted and spiral vessel, presence of stone cells, starch grains, lignified xylem and phloem will certainly help in identification of drug.

Keywords: Annona squamosa, Annonaceae, Physico-chemical characters, Microscopical characters

Introduction

Annona squamosa Linn. (Annonaceae) is a small tropical tree distributed throughout India. It is native to South America. Traditionally, a bark decoction is used to stop diarrhoea, while the root is used in the treatment of dysentry. A decoction of the leaves is used as a cold remedy and to The fruits of Annona are clarify urine. haematinic, cooling, sedative, stimulant. expectorant, maturant, and tonic. They are useful in anaemia, burning sensation. The seeds are abortifacient and insecticidal and are useful in destroying lice in the hair.

Scientific investigations have shown that the crude extract possesses anxiolytic [1], miticidal [2], antifeedant [3] and antidiabetic [4,5] activities. In spite of the numerous medicinal uses attributed to this plant, pharmacognostic information about this plant has not been published. Hence the present investigation is an attempt in this direction and includes

determination of physico-chemical constants and histological evaluation.

Material and Methods

Plant Material

The plant *Annona squamosa* Linn. were collected from the field of Punjabrao Krishi Vidyapith, Nagpur, India, in the 1st week of August. It was authenticated from Department of Botany, Nagpur University, Nagpur. Its herbarium is deposited in the above department. (Voucher specimen no.9089). The fresh leaves were used for microscopic study. Collected leaves were shade dried and coarsely powdered. This coarse powder was used for the physico-chemical analysis.

Chemicals and Instruments

Glass sides, cover slips, watch glass, muffle furnace, silica crucible and other common glasswares were the basic apparatus for the study. The microscopy of authenticated plant *Annona squamosa* was done with the help of binocular compound microscope facilitated with Motic image plus 2.0 (Software).

Experimental Design

a) Microscopic Analysis

The stem, petiole, and leaf were studied for microscopic characters. Sections were taken manually, stained (with phloroglucinol and starch) and mounted following prescribed techniques and representative diagrams were taken with the help of binocular microscope [6-8].

b) Physico-chemical Analysis

Physico-chemical characters i.e. percentage of ash values and extractive values were determined according to the WHO guidelines on the quality control methods for medicinal plant materials [9-13].

Result and Discussion

a) Exomorphology

Annona squamosa is a small, semi-(or late) deciduous, much branched shrub or small tree 3 metre to 8 metre tall with a broad, open crown or irregularly spreading branches and a short trunk, not buttressed at base. The sugar apple is a fruit, which is pale green. The pulp is typical of the Annonaceae family, creamy-white, fragrant and sweet. The fruit contains numerous black seeds which are ellipsoid to obovoid, 1-1.4 cm in size while flowers are yellowish green.

Annona squamosa is willing to grow at altitudes of 0 metre to 2,000 metre and does well in hot dry climates; at much lower altitudes than many of the other fruit bearers in its family.

b) Macroscopy

Thin leaves occur singly, 5 centimetre (2.0 in) to 17 centimetre (6.7 in) long and 2 centimetre

(0.79 in) to 6 centimetre (2.4 in) wide; rounded at the base and pointed at the tip. Pale green on both surfaces and mostly hairless with slight hairs on the underside when young. The sides sometimes are slightly unequal and the leaf edges are without teeth, inconspicuously hairy when young. Leaf stalks are 0.4 centimetre (0.16 in) to 2.2 centimetre (0.87 in) long, green, sparsely pubescent.

c) Microscopy

Leaf

Annona squamosa leaf is dorsiventral (Figure1b). Single layer of palisade cells are present below upper epidermis. Stomata are of paracytic type (Figure 4d), found in lower epidermis. Mesophyll consists of 3-4 layers of spongy parenchyma with many intercellular spaces (Figure1b, 1e). The region shows collenchyma below midrib epidermis on both surfaces. Parenchymatous cells occupy the space between collenchyma and vascular bundle. The vascular bundle consist of lignified xylem and phloem that are arranged in collateral-open type (layer of cambium is separating xylem and phloem). Sclereides are present below collenchymatous cells of upper epidermis (Figure 1d).



Figure 1a) Transverse section of leaf through midrib stained with phloroglucinol showing



Figure 1b) T.S of leaf through lamina showing dorsiventral leaf



Figure 1c) Collateral open type lignified vascular bundles



Figure 1d) lignified sclereides



Figure 1e) Empty vessels

Petiole

Transverse section of petiole shows an outer epidermal layer. A region of collenchymatous cells, composed of many layers, is present below epidermis. Vascular bundles are arranged in a broad zone of parenchyma, in such a manner that a ring is formed. Sclereides or stone cells are present in the collenchymatous cells (Figure 2b).



Figure 2a) Transverse section of petiole under 5x



Figure 2b) Stone cells under 45x

Stem

Transverse section of stem shows collenchymatous cells below epidermis, followed by pericyclic fibers, xylem, phloem, and parenchymatous cells. Xylem is surrounded by mounted with phloroglucinol and HCl, the following elements were observed.

• Epidermal cells with characteristic



Figure 3a) Transverse section of stem under 5x



Figure 3b) T.S of stem under 10x stained with iodine solution giving blue color to starch grains

starch grains and pith contains lignified stone cells (Figure 3b). Starch grains are oval or ellipsoid, turning blue when treated with iodine.

Powder Characteristics (Figure 4)

The powder is green in color with characteristic odor and bitter taste. When the powder was

- stomata (Figure 4d).
- Fragments of pitted and spiral vessels are found scattered (Figure 4a, 4b).
- Fibers are long, narrow, elongated with tapered ends (Figure 4c).



Figure 4a) Pitted vessels



Figure 4b) Spiral vessels



Figure 4c) Fiber



Figure 4d) Paracytic stomata

d) Physico-chemical Analysis

The Physico-chemical characters of powdered leaves of *Annona squamosa* such as ash values and extractive values are given in Table 1.The result shows higher water soluble extractive value indicating the presence of high sugar content. The percentage yield of successive extractive values of powder were petroleum ether 3.85, chloroform 2.33, ethyl acetate 2.39, Acetone1.2, methanol 7.07 % w/w.

Table 1: Ash Values and Extractive Values ofLeaves of Annona squamosa

Ash value $(\% w/w)$	Total ash	10.5
(,,,,,,,)	Acid insoluble ash	1.3
Extractive value (% w/w)	Water soluble	18.4
	Alcohol soluble	2.4

Conclusion

As there is no pharmacognostical anatomical work on records for this traditionally valued herb, the present study is taken up in the view to lay down the physico-chemical and microscopic standards. Microscopical and physico-chemical standards discussed here can be considered as the identifying parameters to substantiate and authenticate the drug and could be useful in the preparation of herbal monograph for its evaluation.

Authors' contributions

1) MA, PI, AP, AK, JY have made substantial contributions in conception and design, acquisition of data.

2) MA, AK, JY participated in analysis and interpretation of data.

3) MA, YA have been involved in drafting the manuscript or revising it critically for important intellectual content.

4) MA and YA have given final approval of the version to be published.

All authors read and approved the final manuscript. All contributors who do not meet the

criteria for authorship should be listed in an acknowledgements section.

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