



## ***Punica Granatum*:- Study on Its Pharmacognostical and Phytochemical Properties**

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### **ABSTRACT:**

*Punica granatum* is a deciduous shrub or a small tree of *Punicaceae* family, found in almost all parts of India. This plant is used to cure a wide range of diseases across different civilization and cultures. It has great nutritional values and health benefits. Pomegranate has been used in natural and holistic medicine to treat sore throat, coughs, urinary infections, digestive disorders, skin disorders, arthritis and to expel tapeworms. In folklore practice, bark of *Punica granatum* is used to treat *Shwetapradara*. Importance of Pharmacognosy has been widely felt in recent times. Pharmacognostical and Phytochemical studies ensure plant identity, lays down standardisation parameters which will help and prevent adulteration. This study is important and lays down parameters for standardisation and authentication of *Punica granatum* with the help of which adulteration and substitution can be proved.

**Key words:-** *Punica granatum*, Pharmacognostical studies, Phytochemical studies,

### **INTRODUCTION:-**

In Ayurveda, plants have been the basis for medical treatments through much of human history, and such traditional medicine is still widely practiced today. While some people may dismiss herbal remedies as quackery, the use of plant is well rooted in medical practice. Practitioners from ancient periods methodically collected information about herbs and developed well-defined pharmacopoeias to treat a variety of ailments<sup>1</sup>.

Herbal drugs have found extensively used in several countries not only because they are easily accessible and are cheaper but due to an important reason that they are safer than synthetic drugs which may not always be accurate. Medicinal plants played a vital role in the detection of novel and useful drugs used in contemporary medicine. Currently we have a number of drugs useful and lifesaving and also drugs which can provide direct healing benefit.

The drug *Punica granatum* is being used for thousands of years to cure a wide range of diseases across different civilization and cultures. It has great nutritional value and health benefits. Pomegranate has been used to treat sore throat, cough, urinary infections, digestive disorders, skin disorders, arthritis and to expel tape-worms. It might prevent heart diseases, heart attacks and stroke<sup>1,2,3</sup>.

Therapeutic efficacy of medicinal plants depends upon the quality and quantity of chemical constituents. The misuse of herbal medicines starts with wrong identification. All these problems can be solved by Pharmacognostical and phytochemical studies.

## MATERIALS AND METHODS

- Collection of Sample: The Botanically identified samples of *Punica granatum* (Linn.) fruit were collected from Calicut District.
- Place of work: Pharmacognostical study was carried out at Kottakkal Arya Vaidya Sala, Centre for Research in Ayurveda and Allied Sciences, Kottakkal, Malappuram [Kerala].
- Phytochemical study was carried out in P G Department of *Dravyaguna Vijnana*, Alva's Ayurveda Medical College, Moodbidri.

### A. Pharmacognostical Study<sup>4,5</sup>

#### **A.1. Macroscopy of *Punica granatum* (Linn.) Fruit rind**

Procedure: The external features of the test samples – *Punica granatum* (Linn.) Fruit rind were documented using Canon IXUS digital camera.

#### **A.2. Microscopy of *Punica granatum* (Linn.) Fruit rind**

Procedure: Sample was preserved in fixative solution. The fixative used was FAA (Formalin-5ml + Acetic acid-5ml + 70% Ethyl alcohol-90ml). The materials were left in FAA for more than 48 hours. The preserved specimens were cut into thin transverse section using a sharp blade and the sections were stained with safranin. Transverse sections were photographed using Zeiss AXIO trinocular microscope attached with Zeiss Axiom Cam camera under bright field light. Magnifications of the figures are indicated by the scale-bars.

#### **A.3. Powder microscopy of *Punica granatum* (Linn.) Fruit rind**

Procedure: The *Punica granatum* (Linn.) were collected, dried in sun light, pounded and sieved in mesh to get fine powder and kept separately. A pinch of the sample was mounted on a microscopic slide with a drop of glycerine water. Characters were observed using Zeiss Axiom trinocular microscope attached with Zeiss Axiom camera under bright field light. Magnifications of the figures are indicated by the pre-calibrated scale bars using Zeiss Axiom vision software.

### B. Physico Chemical Study<sup>6</sup>

The dried *Punica granatum* (Linn.) Fruit rind is powdered finely and subjected to various analyses, such as determination of moisture content, ash value, acid insoluble ash, water soluble ash etc were recorded. The extractive value in various solvents and ash value are important in identification and standardization of single drugs.

### C. Preliminary Phytochemical Study<sup>7</sup>

Freshly prepared extracts were tested for the presence of various active phytochemical compounds like carbohydrates, alkaloids, amino acids, proteins, glycosides, phenolic compounds, saponins, flavonoids, tannins, steroids etc.

### D. Chromatographic Studies

#### **D.1. Thin Layer Chromatography**

Thin layer chromatography is a technique in which a solute undergoes distribution between two phases, a stationary phase acting through adsorption and a mobile phase in the form of a liquid. The adsorbent is a uniform layer of dry finely powdered material applied to a glass plate. Separation may also be achieved on the basis of partition or a combination of partition and adsorption depending on the particular type of support, its preparation and its use with different solvent. Identification can be effected by observation of

spots of identical  $R_f$  value. A visual comparison of the size and intensity of the spots usually serves for semi-quantitative estimation.

## D.2 HPTLC Fingerprint Profile

HPTLC is a planar chromatography where the separation of sample components can be achieved on high performance layers with detection and data acquisition using an advanced workstation.

## OBSERVATION AND RESULT

### Macroscopic study:

- The pomegranate fruit is of sub-globular shaped and brownish yellow to dull crimson red coloured. The fruit was deemed to be a large berry around 2.5 to 5 inches in diameter crowned with a large, deeply five-toothed, tubular, coriaceous calyx, within which are the remains of the stamens and style. Fruit itself gives rise to three parts: the seeds, having weight 4% without juice. the juice, about 34% of the fruit weight, and the peels contributing 56% fruit weight (weights reported are average value). The peel of fruit was composed of tough and glossy leathery skin that is variously referred to as the rind, husk, or pericarp and the interior membranous walls and white spongy pith referred as carpels or arils of the fruit which forms locules to hold seeds. The peel of the fruit, separated from the seeds, occurs in thin curved fragments, granular and brownish yellow or reddish on the outer surface, uniformly yellowish within, and there exhibiting distinct, shallow, angular depressions left by the seeds; to some fragments the remains of the coriaceous calyx are attached. Thickness of the peel was around 2.5 to 3.0 mm with granular, thick, hard, and coriaceous outer surface, while inner surface was with large cavities divided by thin, membranous dissepiments into a number of cells, each of which is completely filled with numerous seeds. Peel breaks with a short granular fracture, has little odour, but a powerfully astringent taste. The intensity of acrid- astringent taste was more promising in arils compared to pericarp.

### Microscopic study:

- The Transverse section of the peels showed presence of a cuticle layer on the outer surface (pericarps) of pomegranate fruits followed by epidermal cells organized by one layer immediately underneath the cuticle. The epidermis forms the outer most cell layer on the structure of pomegranate fruit peel. Epidermis has showed evenly distributed lenticels which function as stomata were observed on the peel of the pomegranate fruit. The observed lenticels were mostly in the shape of a lens and were characterized as extending from loosely lined complementary cell surface. In addition to this transverse section has shown presence of pigment layer followed to epidermis. Below pigment layer transverse section had shown presence of several cell thick spongy parenchyma layers. These Parenchyma cells are in the isodiametric shape. Vascular bundles and sclerenchyma cells were located between parenchyma cells. It was found that protective sclerenchyma cells were densely distributed on the peel of fruit. In addition to the above T.S of Fruit rind showed the presence of Stone cells, Prismatic and Cluster Crystals and Scalariform Tracheid's.

**Table no: 1** Physio Chemical Studies

Percentage of Moisture content	12.06%
Total ash	3.11%
Acid insoluble ash	0.54%

**Table no:2** Preliminary Phytochemicals

PHYTOCHEMICALS PRESENT	PHYTOCHEMICALS ABSENT
Terpenoides	Alkaloids
Flavonoids	Steroids
Phenolic compound	Phlobatannins
Tannins	Saponins

Water soluble ash	22%	Liginin	Insulin
		Fat & Oil	Anthroquinones
PH value	3.92	Proteins	Glycosides
		Carbohydrates	Amino acids
Extractive values in different solvents	Ether(4.45%),Chloro form(6.06%,alcohol(14.4%), Water(19.70%)		

## DISCUSSION

TS of the rind shows 3-4 layered collenchymatous epicarp covered with thick cuticle. The innermost few cells occasionally contain reddish to orange brown coloured pigments. Underneath layers lies many layered wide Parenchymatous mesocarp traversed by groups of stone cells and vascular strands. Stone cells are of varying size, shape and thickness. Prismatic crystals of calcium oxalate. Simple and compound starch grains and dark brown pigment Travers throughout Parenchymatous cells of mesocarp.

Powder microscopic study shows isolated or groups of stone cells, Cluster and Prismatic crystals of calcium oxalate and Starch grains

### Physico chemical study:

Physical analysis revealed that, Total ash 3.11%, Water soluble ash 22%, Acid insoluble ash 0.54% and Moisture content 12.06%, the Total ash of a formulation represents the inorganic residue left after the incineration.

The Preliminary phytochemical study of *Punica granatum Linn* shows the presence of Proteins, Carbohydrates, Flavonoids, Phenolic compound, Terpenoides, Tannins, Liginins, Fat and Oil,

Ash analysis of *Punica granatum Linn* showed the presence of Potassium, Sodium, Iron, Manganese and Zinc.

HPTLC and TLC documentation of drug had been carried out. In HPTLC documentation at 254 nm 6 peaks, at 366 nm 5 peaks were obtained respectively.

## CONCLUSION:

- *Punica granatum (Linn.)* is a drug which is easily available in almost all part of India.
- Presence of Sandy particle, Cluster and Prismatic Crystals are the Characteristic features of the Fruit Rind.
- The amount of extractable matter was found to be maximum proportion with Water as solvent (19.70%).
- Phytochemical analysis on *Punica granatum* fruit rind showed the presence of Proteins, Carbohydrates, Flavonoids, Starch, Tannin, Fat & Oil and Terpenoides
- Ash analysis showed the presence of Potassium, Sodium, Iron, manganese, Zinc.

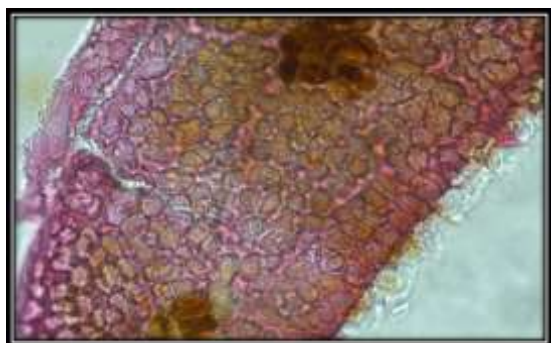


Fig no: 01 Epidermis in SurfaceView

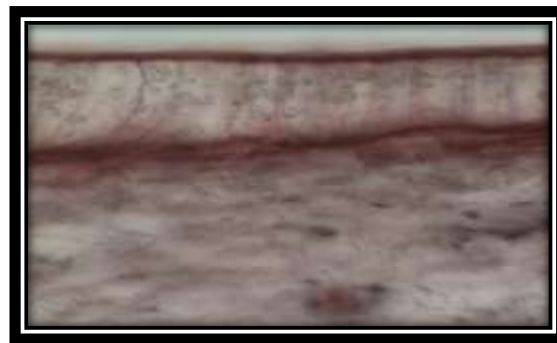


Fig no: 02 Epidermis with Sandy Particle



Fig no: 03 Pericarp with CC-Cluster and PC-Prismatic crystals



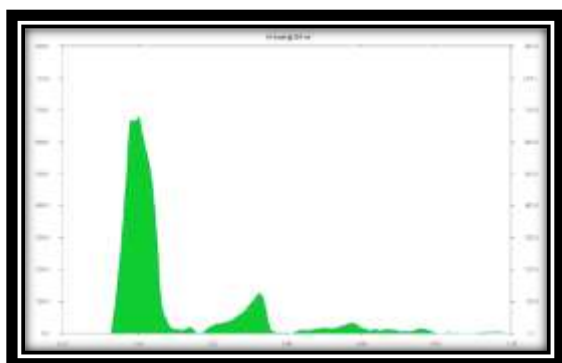
Fig no: 04 Stone Cells



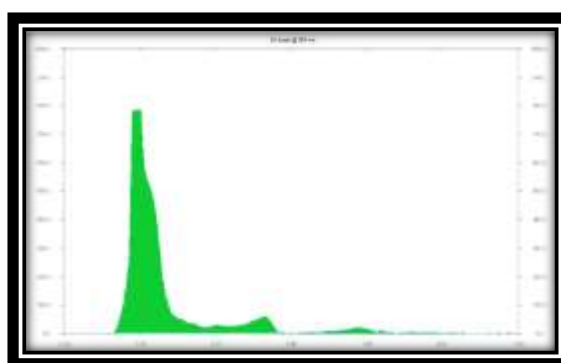
Fig no: 05 stone cells with narrow lumen



Fig no:06ScalariformTracheid's



Graph no: 01 HPTLC densitometric scan at 254nm



Graph no: 02 HPTLC densitometric scan at 366 nm

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