

Pharmacognostical Study of a Unani Herbal Drug 'Parsiaoshan' (*Adiantum venustum* D.Don)

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Abstract

The unani herbal drug available as 'Parsiaoshan' has been identified as the aerial parts of the plant *Adiantum venustum* D.Don. Apart from being an important drug in the Unani as well as other herbal systems of medicine, the plant has assumed great significance due to recent findings of its anti cancerous, antimicrobial and antiinflammatory activities. In order to provide criteria and to set standards for ascertaining genuineness and quality of the drug, pharmacognostical study has been carried out on the dried sample of the drug. The work describes in detail morphological and anatomical features, study of the powdered drug and its analysis under UV light, qualitative determination of chemical constituents, some physico - chemical parameters and results of thin layer chromatography. The salient features of the drug have been discussed.

Key words: Herbal drug, Parsiaoshan, Pharmacopoeial standardisation, Quality assurance.

Introduction

The drug Parsiaoshan in Unani system is reputed for its antipyretic, demulcent, expectorant, diuretic, emmenagogue, desiccant and resolvent actions. It is used in all types of fevers; in catarrh, coryza and asthma; also useful in anuria, dysuria and amenorrhoea. Its decoction is used to remove dandruff. (Ali, 1979; Khan, 1913; Kirtikar, 1935). Though the drug in Unani system is attributed to the plant *Adiantum capillus-veneris* L. (Anonymous, 1981), it has also been mentioned as *Adiantum venustum* D.Don by some authors (Kirtikar, 1935; Watt, 1889) and in practice, use of various other species of *Adiantum*, particularly *A. venustum* is common. The plant is known as 'Hansraj' and most of the drug available in commerce belongs to this plant (Anonymous, 1985; Watt, 1889). Medicinal properties attributed to this plant have remarkable similarity with those described for the drug 'Parsiaoshan'. The plant has been reported as anodyne in bronchitis; diuretic and emmenagogue; fronds reported to be astringent, aromatic, emetic in large doses, tonic, febrifuge, expectorant and deobstruent; used in the treatment of biliousness, inflammatory diseases of the chest, ophthalmia, hydrophobia, colds and headache; also as hair tonic. (Anonymous, 1985; Gupta, 2004).

The samples of "Parsiaoshan" obtained for the present study have also been identified as the plant *Adiantum venustum* D.Don.

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The plant is a graceful fern found very commonly in North-East Himalayas, Kashmir and in Simla at altitudes of 1,350 to 3150m in shaded forest beds; also common in Punjab; having dark glossy stipes; fronds 3-4 pinnate, pinnules obovate-cuneate, striated, 2-3 lobed, finely dentate- serrate; fertile lobes with two, rarely three notches, each notch bearing a rather large sorus at the bottom. (Anonymous, 1985; Kirtikar and Basu, 1935).

Recent studies on the plant *A. venustum* have revealed that it has remarkable anti-inflammatory, anti-microbial and anti-cancerous activities which greatly add to its future therapeutic potential. (Hussain, 2008; Meenakshi, 2008; Devmrari, 2010)

A review of the up to date published works, reported on the subject indicated that no pharmacognostical work on this drug source plant was available. (Anonymous, 1986-2011; 2002; 2003-2005; Farooq, 2005; Gupta and Tanden, 2004; Mitra, 1985).

Material and Methods

Samples of the drug consisting of dried aerial parts were obtained from several separate sources in the market. All were found to be alike and identified, based on the descriptions available, (Anonymous, 1985; Kirtikar, 1935; Watt, 1888) as well as by comparing with live plant available at the Botany department, AMU, Aligarh. Free hand sections were used for microscopic study. The diagrams were sketched using a camera lucida and measurements of cells were done by using a standardized eyepiece micrometer. Method described by Johansen (1940) was followed for maceration of the material. Fluorescence analysis was done according to method described by Kokoski et al. (1958). Physico-chemical parameters were done according to standard methods while extracts of the drug obtained from successive extraction were used for TLC analysis which was performed using pre-coated aluminium plate of silica gel 60 F-254.

Observations

Macroscopic Characteristics

The drug consists of dried fronds having smooth, shining, dark brown, glabrous petioles; 3-4 pinnate (Fig.1). The segments (pinnae) are bright green, shortly petiolulate, glabrous, 6-9 mm long and 3-5 mm broad in the middle. The blade has prominent non reticulate branching venation; not cleft or lobed (Fig.2). The upper margin is rounded, dentate while it is cuneate below. Fertile segments

bear linear, rather large sori, formed by the revolute upper margins of the pinnae (Fig.3). It gives a slight aromatic odour and has no distinguishable taste.

Microscopic Characteristics

Petiole:

A cross section of the petiole shows almost a circular outline with a small notch on one side (Fig.6). The outer most tissue is a single layered epidermis, covered with a smooth thin cuticle and composed of dark coloured thick walled parenchyma; followed by 3-4 seriate layer of sclerenchyma, composed of dark coloured thick walled fibres. This is followed by a fairly large compact zone of cortex, consisting of parenchymatous cells having abundant starch grains and showing little or no inter-cellular spaces (Fig.7). A single stele, enclosed within a layer of endodermis and pericycle occupies the centre (Fig.8). The xylem consisting of tracheids is situated in the centre in the form of a large single band. The metaxylem is in the middle with two opposite protoxylem points, surrounded by phloem, consisting of sieve cells and parenchyma.

The petiole tissue macerated with 20% nitric acid shows an abundance of fibres of varying lengths (Fig.11 A & B) and parenchymatous cells. The xylem consists of quite long tracheids having annular simple pits (Fig.11 C) and spiral thickenings (Fig.11 D).

Measurement of isolated cells (microns)

Tracheids (Length) : 482.00; 940.00; 1012.00

Fibres (Length) : 552.00; 779.00; 1123.00

Table 1: Histo-chemical colour tests

S. No.	Reactions	Observations	Inference
i	Section placed in 10% Aq. FeCl ₃ + little Na ₂ CO ₃	No Blue green colour	Tannins absent
ii	Section directly placed in a drop of H ₂ SO ₄	No colour	Lipids and Saponins absent
iii	Section placed in a weak iodine solution	Colour appears on small rounded bodies	Starch present
iv	Section placed in 5% Tartaric acid in 95% ethyl alcohol for 2 days, washed and then a solution of iodine added	No colour	Proteins absent

S. No.	Reactions	Observations	Inference
v	Section placed in a drop of 1:2500 resorcinol blue for 15 minutes	No blue colour	Callose absent
vi	Section placed in a drop of Phloroglucinol + a drop of HCl	Violet-red colour appears	Lignin present

Pinna

The upper and lower surfaces are glabrous with scattered anomocytic stomata, present on lower surface only (Fig.5). The stomatal number ranges from 8 to 12 with a mean value of 10. The epidermal cells are much more in length and have wavy outlines (Fig. 4 & 5). A transverse section of pinna shows simple, thin upper and lower epidermal layers with only 2-3 seriate loosely arranged chlorenchyma present in between. Vascular areas show 4-7 xylery elements surrounded by phloem (Fig. 9 & 10).

Macerated leaf tissue shows an abundance of epidermal parenchyma having irregular outlines and chlorenchyma (Fig.12 B & C). Tracheids are much shorter and have simple pits (Fig.12A).

Measurement of isolated cells (microns)

Tracheids (Length) : 94.00; 100.00; 106.00
 Epidermal Cells : 12.0 x 79.0; 18.0 x 69.0; 19.0 x 88.0
 Chlorenchyma : 31.0 x 63.0; 67.0 x 71.0; 43.0 75.0

Powdered drug

The powder is dark brownish green in colour, homogenous, a bit fluffy in texture. It gives a slight aromatic odour and has no distinguishable taste. The powder after being cleared in chloral hydrate, was observed under microscope which showed mostly fragments of pinnae and petiole; sporangial wall tissue with characteristic transverse thickenings and triangular spore tetrads.

The other studies comprising colour reaction of powder with different reagents, fluorescence analysis, physico-chemical parameters and thin layer chromatography have been provided in respective tables.

Table 2: Reaction of powder on treatment with different reagents

S.No.	Treatment	Observations
i	Powder triturated with water	An emulsion formed.
li	Powder shaken with water in a test tube	No frothing occurs.
lii	Powder treated with 66% H ₂ SO ₄	Turns dark blackish brown.
lv	Powder treated with 5% NaOH	Turns dark chocolate brown.
v	Powder treated with 5% FeCl ₃	Turns dark green.
vi	Powder pressed between two filter papers for 24 hours	No oil stain appears.

Table 3: Fluorescence analysis of the powdered drug (After Kokoski *et al.*, 1958)

S. No.	Treatment	Observation under	
		Ordinary Light	U.V. light
i	Powder as such	Dark dull Green	Colourless
ii	Powder treated with 1N NaOH in methanol	Dark Brown	Colourless
iii	Powder treated with 1N NaOH in water	Dark Brown	Colourless
iv	Powder treated with 1N HCl	Brown	Colourless
v	Powder treated with 50% HNO ₃	Dark bright Brown	Colourless
vi	Powder treated with 50% H ₂ SO ₄	Dark Brown	Colourless
vii	Powder mounted in Nitrocellulose in Amyl acetate	Dark blackish Brown	Colourless
viii	Powder treated with 1N NaOH in methanol, dried and then mounted in Nitrocellulose in Amyl acetate	Dark chocolate brown	Colourless
ix	Powder treated with 1N NaOH in water, dried and then mounted in Nitrocellulose in Amyl acetate	Dark Brown	Colourless
x	Powder treated with 1N HCl, dried and then mounted in Nitrocellulose in Amyl acetate	Dark Brown	colourless

Note: Reactions (ii) to (vi) observed immediately after treatment, within one minute while reactions (vii) to (x) are observed after being allowed to dry.

Table 4: Physico chemical studies

1	Chemical Constituents (Qualitative)	Organic: Inorganic: Heavy Metals:	Carbohydrates, Glycosides, Phenolics, Steroids / Terpenes and Resins. Aluminium, Calcium, Iron, Magnesium, Potassium and sodium Mercury, Lead, Cadmium and Arsenic not detected.
2	Ash Values (%)	Total Ash Acid insoluble (10% HCl) Water soluble	7.60 - 8.20 4.20 - 4.60 0.50 - 0.70
3	Loss on drying at 105oC(%)		7.00 - 8.00
4	Solid Contents (%)		74.00 - 76.00
5	Successive extractives values (%)	Petroleum Ether (60-80o): Chloroform: Acetone: Alcohol: Water:	4.40 - 4.60 3.00 - 3.40 4.50 - 4.80 9.00 - 9.50 14.00 - 14.60

Table 5: Thin Layer Chromatography

Extract	Solvent System	Treatment	No. of Spots	Rf Values
Petroleum	Petroleum	Exposed to Iodine	8	
Ether	Ether(40-60o):	Vapours	5	0.96
(60-80o)	Solvent Ether	Spraying with 2%		0.90
Absolute	(4:1)	H2SO4 in Ethanol &		0.71
Alcohol	Toluene: Ethyl	heating the plate for		0.62
	formate: Formic	about ten minutes at		0.55
	Acid (5:4:1)	105oc in an oven		0.49
				0.42
				0.28
				0.63
				0.58
				0.56
				0.50
				0.07

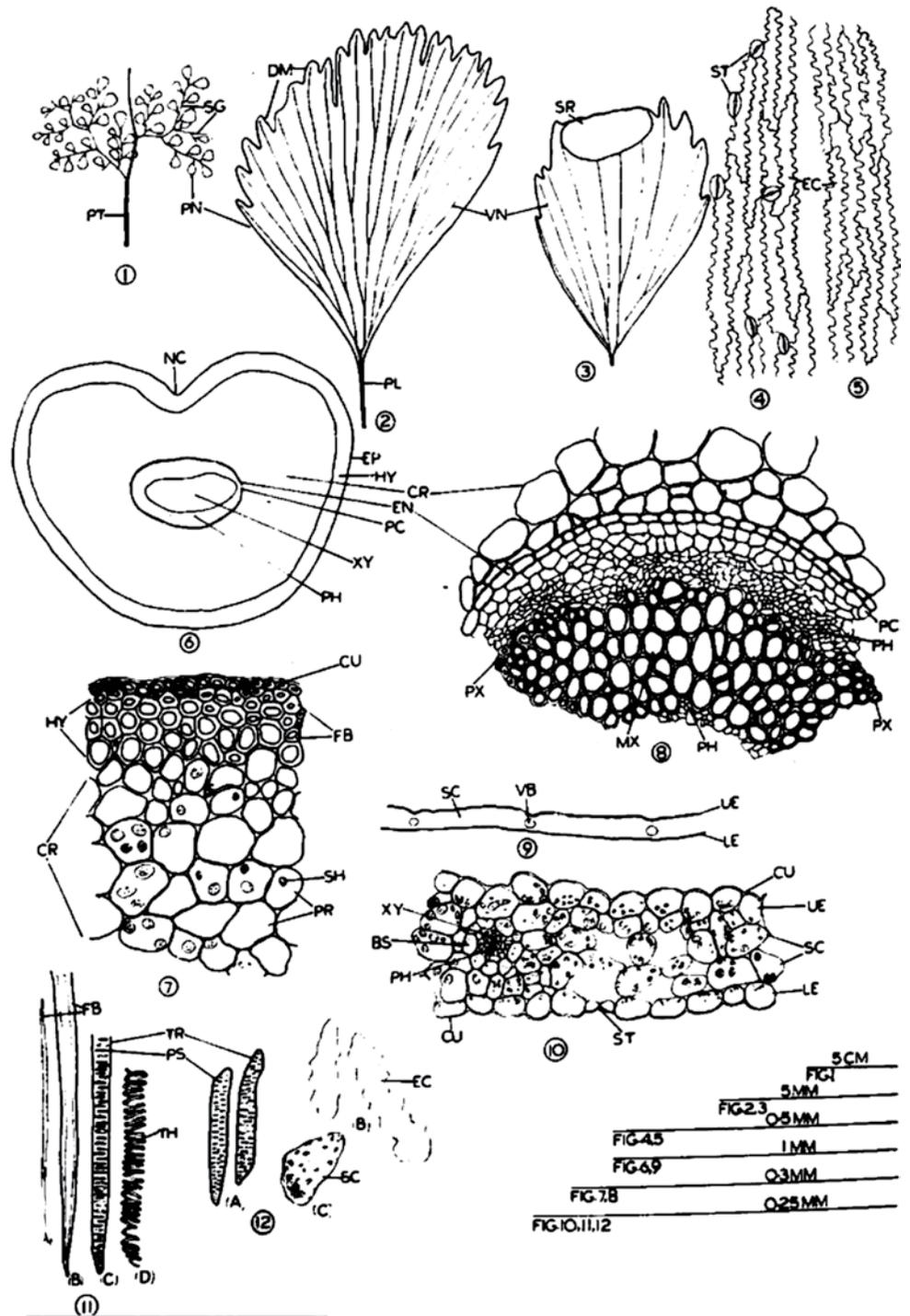
Conclusion

The study has provided a detailed description of the drug based on morphological and anatomical features along with some important physico-

Chemical characteristics. As a result, certain specific key characters have been worked out on the basis of which, the drug samples can be checked for genuineness and quality. Distinguishing morphological features include dark, shining, smooth stipe; 3-4 pinnate frond; rounded, deltoid-cuneate, dentate segments having prominent free, forking veins; fertile segments having sub marginal sori and inrolled upper margins. Internally the stipe shows a sclerenchymatous hypodermis and stele with a band of xylem tissue having opposite protoxylem. The macerated tissue shows long tracheids while triangular spore tetrads are present in the powder. Apart from these prominent structural features, other details worked out under macro-microscopic studies, fluorescence analysis, physico-chemical studies and thin layer chromatography constitute definite specifications for the drug.



Fig. 1: Habitat of *Adiantum venustum* D.Don.



5CM
 FIG.1
 5MM
 FIG-2,3
 0.5MM
 FIG-4,5
 1MM
 FIG-6,9
 0.3MM
 FIG-7,8
 0.25MM
 FIG-10,11,12

Fig. 2: Adiantum Venustum Don: Pharmacognostic details

Explanation of Figures

1. The Frond
2. A pinna

3. Pinna with sorus
4. Pinna: Upper surface view
5. Pinna: Lower surface view
6. Petiole: Transverse section (Diagrammatic)
7. Petiole: Transverse section showing Hypodermis & Cortex
8. Petiole: Transverse section showing Stele
9. Pinna: Transverse section (Diagrammatic)
10. Pinna: Transverse section showing cellular details
11. Macerates (Rachis): Fibre under low magnification (A); Fibre under high magnification (B); Portion of Tracheid (C); Spiral thickenings (D)
12. Macerates (Pinna): Tracheids (A); Epidermal Cells (B); Chlorenchyma (C).

Abbreviations

BS = Bundle sheath; CR = Cortex; CU = Cuticle; DM = Dentate margin;
 EC = Epidermal cells; EN = Endodermis; EP = Epidermis; FB = Fibre;
 HY = Hypodermis; LE = Lower Epidermis; MX = Metaxylem; NC = Notch;
 PC = Pericycle; PH = Phloem; PL = Petiolule; PN= Pinna;
 PR = Parenchyma; PS = Pits; PT = Petiole; PX = Protoxylem;
 SC = Spongy Chlorenchyma; SG = Segments; SH = Starch grains;
 SR = Sorus; ST = Stomata; TH = Thickening; TR = Tracheids;
 UE = Upper Epidermis; VB = Vascular bundle; VN = Veins; XY = Xylem.

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