Pharmacognostical Standardization of 'Mushkdana' Seed (*Abelmoschus moschatus* Medic)

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Abstract

Mushkdana which is botanically identified as *Abelmoschus moschatus* Medic. (syn. *Hibiscus abelmoschus* Linn.) is an important medicinal plant of Indian traditional systems of medicine. The plant is cultivated for its musk-scented seeds, useful in perfumery and medicine. The seeds also known as Ambrette seeds are valued for the volatile oil present in the seed coat. They have been used as a tonic, stimulant, carminative, as flavouring agent for food and as a substitute for musk. Present paper deals with the pharmacognostical standardization of Muskhkdana seed, unstudied so far, in order to lay down the standards for its quality assurance. The study includes macro and microscopic features of the drug, besides maceration, powder analysis, measurement of cells/tissues, reaction with different chemical reagents and fluorescence analysis of the crude drug powder. Other parameters worked out are physiochemical constants such as ash value, alcohol and water soluble matter and thin layer chromatography.

Keywords: *Abelmoschus moschatus* Medic., Seeds, Pharmacognostical standardization

Introduction

In view of the fact that there still exists controversial status of many of the Unani and Ayurvedic drugs, there is a need to fix-up their pharmacopoeial standards in order to ensure authenticity of pharmaceutical materials used in therapeutic formulations. In this direction some 250 such Unani drugs have been earlier investigated for their pharmacognostical standardization (Anonymous, 1987; 1992; 1997; 2006a & 2006b). However, there appears no work done on 'Mushkdana' seeds thus far, whereas, leaves & roots of this drug have been investigated (Anonymous, 2006b). Hence, the present studies.

Abelmoschus moschatus Medic. (Family : Malvaceae) is an aromatic and medicinal plant native to India. It is commonly known as Mushkdana in Hindi; Ambrette plant and Musk mellow in English; Habbulmishk or Habbulmushk in Arabic; Mushkdana in Persian; Latakasturika in Sanskrit and Mushkadanah in Urdu. The plant is an erect, herbaceous annual, grown in the hotter parts of India. Leaves polymorphous, lower ovate, acute, upper palmately 3-7 lobed, flowers bright yellow, large, axillary, capsules 6.5 – 7.5 cm. long, ovate, acute, hispid (Anonymous, 1985; Chopra & Chopra, 1969; Purohit &Vyas, 2004; Verma *et al*., 1993; Agharkar, 1991; Lindley, 1985).

The plant is generally cultivated for their musk-scented seeds useful in perfumery and medicine(Singh *et al*., 1996).Seed analysis report 11.1% moisture, 31.5%

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crude fiber; 14.5% lipids, 13.4% starch, 2.3% protein, volatile oil (0.2-0.6%) and 5% resin (Srivastava, 1995). The crushed seeds on steam-distillation yield a volatile oil of commerce noted for their rich, sweet, floral-mushkyodour. Nee and Carts Pollard (1986) showed that the essential oil of these seeds is localized in the outer layer of the seed coat, whereas fatty oil is concentrated in the embryo and the endosperm. The oil from the seeds is rich in linoleic acid and contains a-cephalin, phosphatidylserine, its plasmalogen and phosphatidylcholine plasmalogen (Ghani, 2003). The characteristic musk-like odor of the seed oil is mainly due to the presence of a ketone, ambrettelide, a lactone of ambrettolic acid. The chemical constituents of the essential oil of the seeds are: trans-2-trans-6-farnesyl acetate and ambrettolide, cis-2-cis-6- farnesyl acetate, cis-2-trans-6-farnesyl acetate, ethyl hexadecanate, ethyl laurate and trans-2-trans-6-farnesol (Du *et al*., 2008).

The seeds are said to be stimulant, antiseptic, cooling, tonic, carminative and aphrodisiac. They are effective to treat intestinal complaints, skin diseases, stomatitis, dyspepsia, alley thirst and checking vomiting (James *et al*., 1993; Peter, 2007; Rival *et al.,* 2009). Powdered seeds are inhaled for dryness and hoarness of throat. The paste of pulverized seed used to treat leucoderma, heat and itch (Panda, 2004). A decoction or infusion or tincture of the seeds is useful in nervous disability, hysteria and other nervous disorders. In Unani system of medicine the seeds are valued for their diuretic, demulcent and stomachic properties. (Kirtikar & Basu, 1988; Nadkarni, 1986) Considering the medicinal importance, it was felt desirable to undertake pharmacognostic standardization of Mushkdana seed in detail so as to find out the reliable data for distinguishing the drug from its possible adulterants.

Material and Method

Seeds were collected from local market, New Delhi; identified and authenticated with the help of standard floras (Hooker, 1875; Cooke, 1903). Its macroscopical characters were studied. For microscopical studies, slides were prepared and stained in different reagents like saffranine, iodine solution, ferric chloride solution etc. and mounted in glycerine to study various cells/tissues/cell contents (Johansen, 1940; Trease and Evans, 1983). The representative diagrams were drawn using camera lucida. Measurement of the individual cell / tissues of the various parts of the seed were recorded. The powder and its behaviour on treatment with different chemical reagents were studied and the physico-chemical constants were determined. Fluorescence characters of the powdered drug were observed under U.V. according to the method described by Kokoski *et al. (*1958). Standard analytical methods were followed for chemical analysis (Anonymous, 1998).

Observations

Macroscopic characters

Seeds greyish brown, reniform, compressed, 0.4 mm. long with striations concentric about the hilum. Odour - delicate musk like; Taste – bitter and sharp. Average wt. of 100 seeds is 1.45gm.

Microscopic characters (Fig. 1 – 3)

Ovule campylotropus; bitegmic; testaregulose. In T.S. seed shows following characters:-

Seed coat

Outermost single layered epidermis, thick walled cells, lignified on the ridges followed by a palisade of malpighian cells which are radially elongated and lignified in inner part. Then a few layers of somewhat longitudinally elongated cells with thickened, brownish often pitted radial walls, somewhat lignified. Perisperm with thin parenchyma cells.

Endosperm

Thin walled, several layered, hexagonal to polygonal parenchyma cells.

Cotyledon

Outermost single layered epidermis followed by single layer palisade cells and few layers of spongy parenchyma cells and then inner epidermis.

All the cells of endosperm and cotyledon are filled with very minute, spherical starch grains; aleurone grains and oil globules.

Powder analysis

Greyish brown powder with delicate musk like odour and sharp, bitter taste. Microscopic examination of the powder in 40 mesh shows epidermal cells of testa, malpighian cells 112.5µ in length and 13.5µ in width, sclerenchymatous cells, fragment of cotyledon,oil globules, very minute, spherical starch grains 11.25µ - 22.50µ in diameter.

Maceration

Maceration of the seed with conc. Nitric acid shows epidermal cells, elongated cells lignified in inner part, thin walled parenchyma and palisade cells of cotyledon.

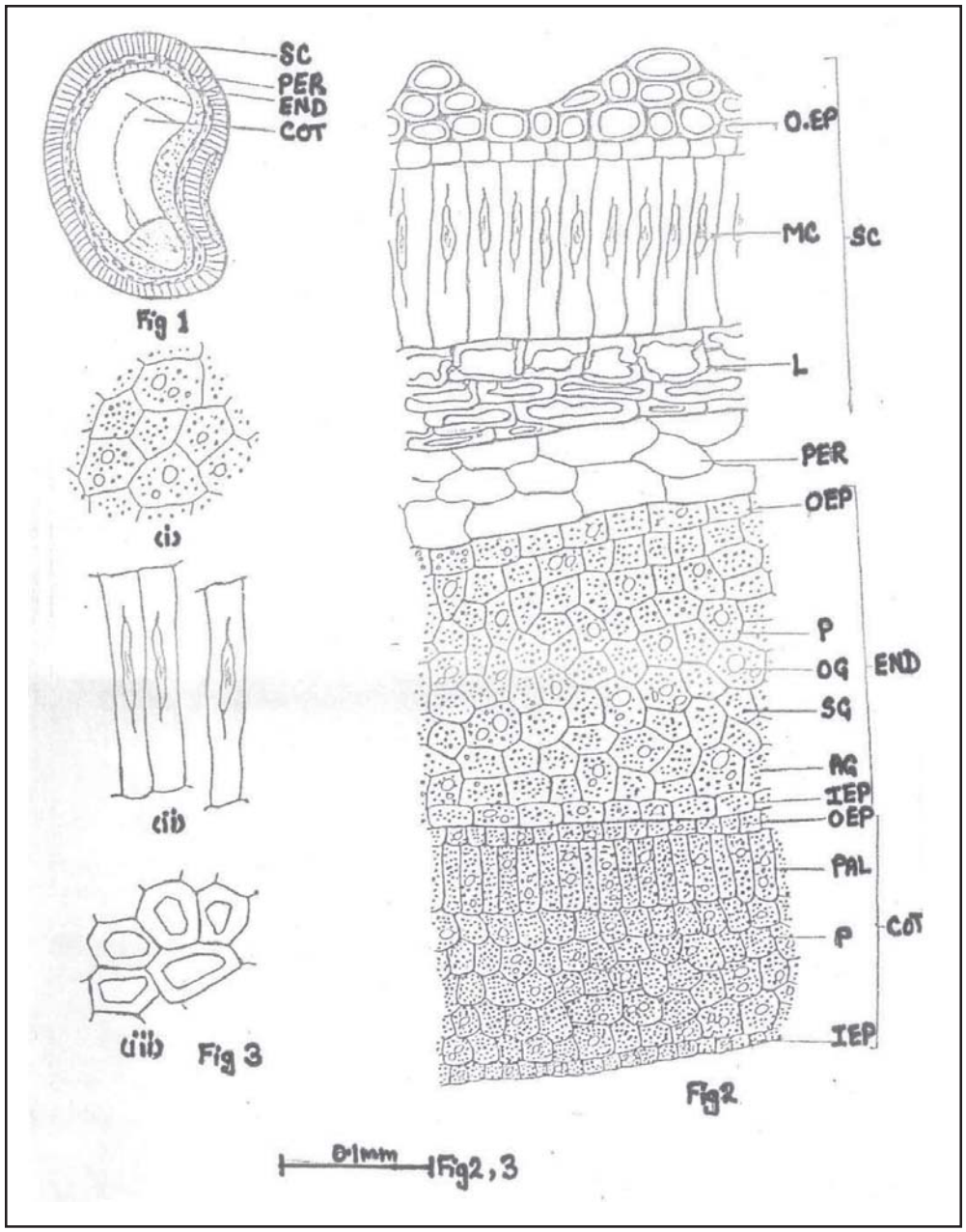

Fig. 1 – 3 : *Abelmoschus moschatus* Medic. (seed)

Fig. 1: L.S. of seed (Diagrammatic)

Fig. 2: T.S. of seed

Fig. 3: Surface view in powder and macerate

(i) Parenchyma cells of endosperm with oil globules and starch grains

(ii) Malpighian cells of seed coat

(iii) Lignified cells of seed coat

Abbreviations: AG : Aleurone grains; COT : Cotyledon; END : Endosperm; IEP : Inner Epidermis; L : Lignified cells; MC : Malpighian cells; OEP : Outer Epidermis; OG : Oil Globules; P : Parenchyma; PAL : Palisade cells; PER : Perisperm; SC : Seed coat; SG : Starch grains

Chemical reaction and Fluorescence analysis

Results of acid/chemical reagent reaction and fluorescence analysis are given in Table 1 & 2.

Table 1: Acid /chemical reagent reaction with powder

S.No.	Acid/Chemical Reagent	Observation
1.	Conc. Sulphuric Acid	Reddish black
2.	Conc. Hydrochloric Acid	Chocolate brown
3.	Conc. Nitric Acid	Orange
4.	Glacial Acetic Acid	Dark brown
5.	Picric Acid	No change
6.	Iodine Solution	Brown
7.	Ferric chloride Solution (aq.)	Bluish green
8.	Sodium hydroxide Solution (5%)	Dark brown & reaction with fumes
9.	Potassium hydroxide Solution (5%)	Dark brown & reaction with fumes
10.	Powder as such	Greyish brown

Table 2: Fluorescence analysis

S. No.	Reagent	Colour in Day-Light	Observation under U.V. Light		
			Modifying Colour	Quality of colour	Degree of radiance
1	Mounted in Nitro-Cellulose	Coffee brown	Brown	Dark	Bright
2	1N Sodium hydroxide in methanol	Dark brown	Green	Dark	Bright
3	Treated with 1N Sodium hydroxide in methanol & mounted in Nitro-Cellulose	Coffee brown	Brown	Dark	Bright
4	1N Hydrochloric Acid	Chocolate brown	Brown	Light	Bright
5	Treated with1N Hydrochloric Acid &Mounted in Nitro-Cellulose	Dark brown	Green	Dark	Bright
6	1N Sodium hydroxide in Water	Dark brown	Green	Dark	Bright
7	Treated with 1N Sodium hydroxide in water & mounted in Nitro-Cellulose	Brown	Coffee brown	Dark	Bright
8	Dilute Nitric Acid (1:1)	Orange	Orange	Dark	Bright
9	Dilute Sulphuric Acid (1:1)	Dark brown	Green	Dark	Bright
10	Powder as such	Greyish brown	Chocolate brown	Dark	Dull

Identity, purity and strength

Analytical values of different physico-chemical constants are shown in Table-3.

Table 3:

Parameter	Analytical Value
Foreigh Matter	Not more than 2 percent
Total Ash %, W/W	4.9
Acid Inslouble Ash %, W/W	1.4
Alcohol Soluble Matter %, W/W	13.2
Water Soluble Matter %, W/W	9.2

TLC profile

Thin layer chromatography of the alcoholic extract was carried out using Toluene: Ethylacetate (9:1) as mobile phase. On spraying with 5% Vanilin-Sulphuric acid and heating the plate for ten minutes at 110°C five spots were visualized at Rf .0.19; 0.31; 0.53; 0.71 and 0.93.

Conclusion

The musk scented seeds are valued for the volatile oil present in the seed coat. The present work provides reliable data on pharmacognostic standardization of this important Unani drug and has brought out many diagnostic key characters covering morphological, anatomical as well as physico-chemical aspects on the basis of which the drug can easily be identified. Salient morphological features include greyish brown reniform seeds compressed with striations concentric about the hilum. Lignified epidermal cells of the testa, palisade like malpighian cells, thin walled parenchyma cells of the cotyledon filled with oil globules and minute, spherical starch grains are some important anatomical characters that are helpful in identification and authentication of the drug either in fresh, dry or in powder form. Physio- chemical data i.e. ash value, water and alcohol soluble matter help in checking the quality and purity of the drug. Rf values and number of spots also provide reliable diagnostic characters.

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