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Pharmacognostical studies of *Moringa oleifera* Lam. seeds

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Abstract
Moringa oleifera Lam. fruit are used as vegetables. It is commonly known as Drumstick/ Shevgha (Family Moringaceae). The seeds yield oil commercially known as 'Ben oil' which resembles olive oil. The seeds have antibacterial properties, used in stomach pain, ulcers, joint pain and sunburns. In the present investigation, pharmacognostical studies of dry seeds were carried out. It involves macroscopy, microscopy, histochemical analysis, powder study, physicochemical evaluation and phytochemical analysis. The dry seeds are brownish in colour with three papery wings. The transverse section of seeds shows, papery wings with outer and inner epidermis. The mesophyll regions are with reticulate parenchyma and oil globules. The testa consists of outer and inner epidermis, reticulate parenchyma, oil globules and sclereids. The cotyledon has thin walled parenchyma smaller in size and filled with oil globules. Powder microscopy showed the presence of oil globules, reticulate parenchyma, thin walled parenchyma and annular vessels. The physicochemical study revealed total ash 3.45 % w/w, water soluble ash 2.25% w/w, acid insoluble ash 0.35% w/w, water soluble extractive value 6.5% w/w and alcohol soluble extractive value 13.8% w/w. Histochemistry and Phytochemical screening of aqueous and alcoholic extracts showed presence of alkaloids, terpenoids, saponins, oil, proteins, etc. The diagnostic characters of seeds would provide necessary information regarding its identity. The phytochemical evaluation provided information of the phytoconstituents present in this crude drug. These will help in identification, authenticity and to lay down the pharmacopoeial standards of the said plant part.

Keywords: Pharmacognosy, *Moringa oleifera*, Moringaceae, drumstick, seeds, pharmacopeia

Introduction
Moringa oleifera Lam. a magic tree as all its parts are used in vegetables as well as in curing various ailments. It belongs to family Moringaceae. *Moringa oleifera* is a small, graceful, deciduous tree with sparse foliage, tripinnate compound leaf; flowers in loose axillary panicles, fruit long with papery winged seeds. [1, 2] It is well known as Drumstick/ Shobhanjana/ Shevga, etc. The use of leaves and fruit of *Moringa* is known throughout the world. The use of seeds is less known. Folk medicine uses raw or crushed *M. oleifera* seeds as a decoction for treating stomach pain, ulcers, poor vision, joint pains and for aiding digestion. [3] The seeds of *M. oleifera* have been found to be good antioxidants, able to reduce oxidative damage associated with aging and cancer. It also cures sunburn [4] The seed yield oil known commercially as 'Ben oil'. It is a good substitute for olive oil [5] Hence the present study includes macroscopic, microscopic, histochemical evaluation, determination of physicochemical constants and preliminary phytochemical screening of *Moringa oleifera* dry seeds. Preparation of herbal whitening sun cream from *Moringa* seeds is studied [6] Anti-pyretic potentials of *Moringa* seeds were investigated [7] The pharmacognosy of *Moringa oleifera* seed is not studied yet hence the present study is carried out. The investigation intended to bring the salient; morphological characters of the seeds so as to lay down the standards which are of utmost importance to authentify a crude drug.

Material and Methods
Collection of plant material
The matured dried seeds of *Moringa oleifera* Lam. were procured from A1 Oil India, Borivali (Mumbai), Maharashtra, India. The botanical identity was confirmed from Department of Botany, Mithibai College, Mumbai. The dried seeds were soaked in water and studied for its anatomical structures. The remaining dried seeds were ground into powder which was sieved through mesh no. 710 with 0.710 mm size of aperture. The voucher specimens of the authentic drug were deposited at Research Laboratory, Botany Department, Mithibai College.

Pharmacognostic Studies
Pharmacognosy of the entire seeds was carried out using standard methodology.

Macroscopy
The entire seeds were studied for its morphological characters using appropriate techniques [8 – 9]

Microscopy
Ransverse hand cut sections were taken and made permanent with suitable stains [10] Quantification and photomicrographs were taken of the permanent preparations. The cell contents were measured using stage and ocular micrometer [11-12]

Histochemistry
The histochemical studies for the cell content were done by staining the hand cut sections with different reagents. [13]

Powder study
The dried powder of seeds was treated with chloral hydrate solution followed by staining in 1% safranin for 5-10 min and mounted in 50% glycerine [14]

Proximate analysis
The physicochemical parameters like ash values (total ash, water soluble ash and acid insoluble ash) and extractive values (water and alcohol extractive values) were established using powdered drug [15]

Preliminary phytochemical screening
A known quantity of dried powder was extracted with alcohol and water. These extracts were tested for different constituents [16-18]

Results
Macroscopy
Each seed has a brownish, semi-permeable hull with three papery wing set around it at 120^0 intervals. The wings are thin and hyaline. The seed is globular 1 cm in diameter. The wings are produced at the base of the seed to apex 2 – 2.5 cm long and 0.4 – 0.7 cm wide. The seeds are non-endospermic. The embryo is straight with zig zag micropyle. (Figure 1)

Fig 1: Entire seeds of *Moringa oleifera*

Organoleptic study: Colour – brownish, Odour – characteristic, Taste – characteristic.

Microscopy
T.S of seed passing through papery wing:
Outer epidermis is single layered, tangentially arranged measuring 30 – 45 µm breadth and 75 - 225 µm length and covered with cuticle. The inner epidermis is similar to outer epidermis. The mesophyll is made of thin walled polygonal parenchymatous cells measuring 15 -75 µm in width. Each parenchyma cell has reticulate network and also filled with oil globules. (Figure 2)

T.S of seed passing through testa:
Outer epidermis is singled layered tangentially arranged measuring 45 - 120 µm in length and 30 – 45 µm in breadth, covered with cuticle. Below the epidermis is polygonal parenchymatous cells with reticulate network measuring 15 – 30 µm in width. It is filled with oil globules at intervals. The parenchymatous cells are in continuation with 4 – 5 layers of sclereids measuring 45 - 120 µm in diameter. The Inner epidermis goes concurrent with outer epidermis. (Figure 3 and 4)

T.S of seed passing through cotyledon:
The epidermis has outer and inner epidermis. The epidermis is tangentially arranged measuring 45 µm in length and 15 - 30 µm in breadth. It is covered with cuticle. The epidermis is followed by thin walled polygonal to rounded parenchymatous cells measuring 30 – 105 µm in diameter. The cells are filled with oil globules. (Figure 5)

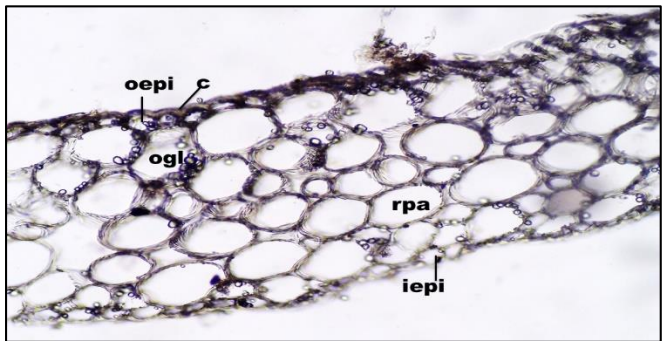

Fig 2: T.S of *Moringa oleifera* seeds passing through papery wings (c- cuticle, oepi- outer epidermis, iepi- inner epidermis, ogl- oil globules, rpa- reticulate parenchyma cell)

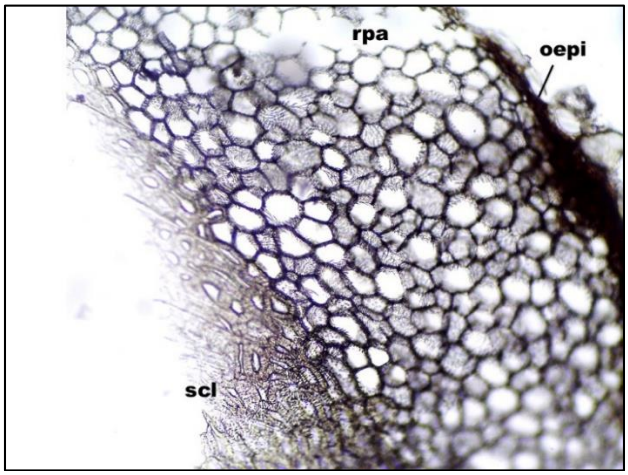

Fig 3: T.S of *Moringa oleifera* seeds passing through testa (oepi- outer epidermis, rpa- reticulate parenchyma cell, scl- sclereids)

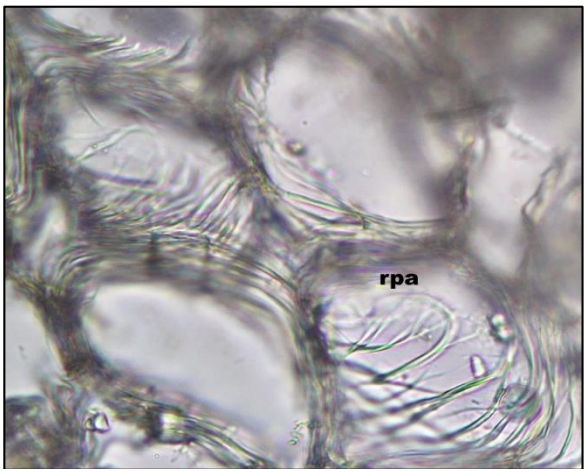

Fig 4: Parenchyma cells of testa showing reticulate network in T.S of *Moringa oleifera* seeds

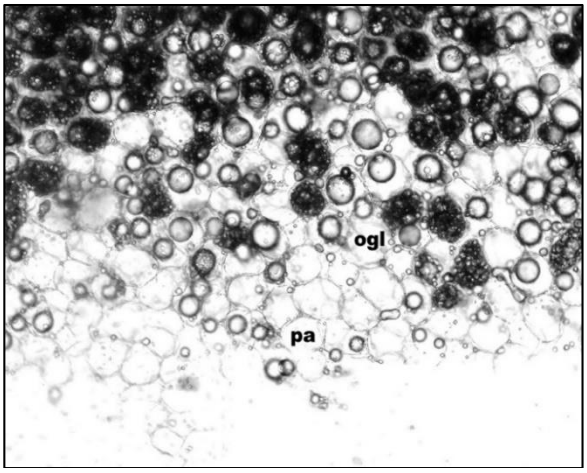

Fig 5: T.S of *Moringa oleifera* seeds passing through cotyledon

Powder study

The *Moringa oleifera* seed powder

The seed powder on treatment with chloral hydrate solution followed by staining in 1% safranin for 5-10 min and mounted in 50% glycerine exhibited fragments of parenchymatous cells, abundant oil globules (12 - 15 µm in diameter 10X), parenchyma cells with reticulate network (60 - 66 µm in diameter 45X), sclereids (300 µm in length and 135 µm in width 45X), annular vessels (16.5 µm in length and 6.6 µm in breadth 45X), fibre (225 µm in length and 15 µm in breadth 10X). (Figure 6)

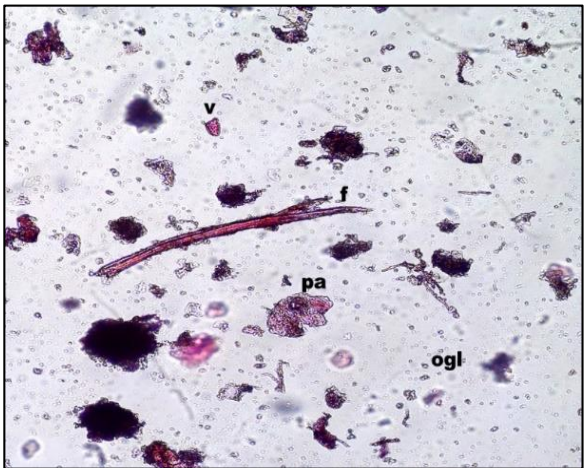

Fig 6: Powder study of *Moringa oleifera* seeds (f- fibre; pa- parenchyma cells)

Histochemical Analysis

The histochemical analysis using various reagents showed the presence of primary and secondary metabolites like proteins, saponins, oil globules, alkaloids and terpenoids respectively.

Physicochemical evaluation

The physicochemical constants such as ash values showed total ash 3.45% w/w, water soluble ash 2.25% w/w and acid insoluble ash 0.35% w/w. Thus, the acid insoluble ash value states the presence of least amount of silica in seed powder. The extractive value of water is 6.5% w/w and ethanol 13.8% w/w the above extractive values determine that more chemical constituents are soluble in the water.

Preliminary phytochemical studies

The preliminary phytochemical studies revealed the presence of proteins, saponins, oil globules, alkaloids and terpenoids in water and ethanol extracts Table 1.

Table 1: Preliminary phytochemical screening of *Moringa oleifera* seeds

Test for phytoconstituents	Water extract	Ethanol extract
Test for Starch	-	-
Test for Terpenoids	+	+
Test for Proteins	+	+
Test for Amino acid	+	+
Test for Mucilage	-	-
Test for Alkaloids	+	+
Test for Anthraquinone glycoside	+	+
Test for Cardiac glycoside	+	+
Test for Saponin	+	-
Test for Tannins	-	-
Test for Steroids	_	_
Test for Flavonoids	-	-

Discussion

The current research work focuses on, pharmacopoeial standards of seeds of *Moringa oleifera* Lam. Macroscopic observations are useful for gross identification of the drug. Anatomical features like unicellular, parenchymatous cells with reticulate network, parenchyma cells thin walled smaller than the parenchymatous cells of testa and wings, and oil globules are of significance in recognition of seeds parts. Authentication of powdered drug can be reliably done on the basis of diagnostic characters of parenchyma cells with reticulate network, oil globules, sclereids, fibres, annular vessels, etc. Physicochemical parameters of ash and extractive values are of help in detection of adulteration if any. The qualitative phytochemical and histochemical screening revealed the presence of diverse types of phytochemicals namely, alkaloids, terpenoids, oil, saponins, etc. They give clue about therapeutic potential of the drug. In brief, all these findings are highly essential for the drug manufacturers in thorough assessment of quality drug.

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