

## PHARMACOGNOSTICAL AND PHYTOCHEMICAL INVESTIGATION OF *SALVADORA OLEOIDES* DECNE. STEM

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### ABSTRACT

Objective: Methajal is a member of Salvadoraceae family has been proven multipurpose tree. The objective of the research was to study standardization parameters such as Pharmacognostical studies which include macroscopic and microscopic evaluation. Physicochemical studies were carried out to confirm the identity of plant. Ash values and extractive values were also determined and recorded. Methods: Extraction was done by maceration process by using Chloroform, Methanol and water as solvents. Evaluation was done by using different parameters like ash value, extractive value, moisture content and foreign matter. Results: The extracts *Salvadora oleoides* stem shows the presence of phytoconstituents such as alkaloids, glycosides, terpenoids and Flavonoids and different values of standardization parameters. Conclusion: Above studied Pharmacognostical and Phytochemical parameters are used to check its quality, purity and for its identification.

**Keywords:** *Salvadora oleoides*, Microscopy, Phytochemistry, Pharmacognosy.

### INTRODUCTION

*Salvadora oleoides* Decne commonly called pilu/chhota pilu or meetha jal is a small shrub or ever green shrub with a bent trunk that is widely distributed in Arabia, India and Pakistan[1,2,3]. Leaves of *Salvadora oleoides* contain high concentration of phenolic compounds (25.7%) and stems contain high concentration of hydrocarbons (41.3%). Twenty three chemical constituents were common in the essential oil of both leaves and stems of *Salvadora oleoides*. Among all these compounds methoxy-4-vinylphenol (25.4%), (Z)-cis-3-Hexenyl benzoate (16.8%), phytol (13.9%), n-hexadecanoic acid (6.9%) and trans- $\beta$ -damascenone (2.1%) were the main constituents of the essential oil of leaves whereas stems contain high concentration of 2-methoxy-4-vinylphenol (21.6%), phytol (12.9%), n-hexadecanoic acid (3.6%), octacosane (7.9%), nonacosane (7.3%), 1-octadecene (5.8%), heptacosane (5.9%), hexacosane (4.5%), pentacosane (3.4%), squalene (3.9%) and trans- $\beta$ -damascenone (2.3%)[4]. This species contains other constituents such as salvadorine, salvadoura, vitamin C, alkaloids, salts mostly as chlorides. Methanolic extract of aerial parts contains 4-hydroxy benzoic acid, stearic acid, lupeol,  $\beta$  - amyryl, Ursolic acid, oleanolic acid,  $\beta$  - sitosterol-3- O-  $\beta$  - D- glucoside, 3 $\beta$ -erythrodiol, Trimethylamine, saponins, resins, tannins, proteins, carbohydrates, fatty acids, amino acids, dibenzylurea, tetracosone, n-octacosanol,  $\beta$ -sitosterol, flavonoids, mucilage and gums[5,6,7].

It showed Hypoglycemic and hypolipidemic activity in normal and Alloxan induced type-2 diabetes in rats[8]. Its roots and stems showed shows antimicrobial activity when compared with standard drug streptomycin[9].

### MATERIAL AND METHODS

#### Procurement and authentication of crude drugs

The fresh stems were collected in the month of July-August 2007 from Agricultural University Campus, Bikaner and authenticated by Dr. Shekhar Bhargava, Head of the Botany Department, Rajasthan University. After the collection, stems dried in shade at temperature 25-30 °C for 10 days and were crushed to obtain coarse powder which could pass through sieve number 40.

#### Evaluation parameters

#### Pharmacognostical examination[10,11]

#### Macroscopic examination

It is a shrub or small tree, attaining 6-9 m height under favorable

conditions; Leaves glaucous, linear-or ovate lanceolate, coriaceous and somewhat fleshy, dark greenish-yellow when young, grey when mature. Flowers sessile, greenish-white, calyx cup-shaped, in 4 rounded, obtuse lobes. Fruit is drupe, globose, about 2-4 cm in diameter, usually yellow, dark brown or red when dry. Seeds greenish-yellow, about 3 mm in diameter.

#### Microscopic examination

Transverse section of stem bark shows epidermis. Epidermis consists of single layer of cells which are closely packed. The walls are thickened and covered with a thin waterproof layer called the cuticle. Stomata with guard cells are found in the epidermis. Collenchyma with three to four layers of cells with cell walls thickened present at the corners. Beneath the collenchyma cells are a few layers of thin-walled cells, parenchyma, with intercellular spaces. The phloem is located towards the outside of the bundle and the xylem towards the center. The pith occupies the large central part of the stem.

#### Extraction procedure

The dried powder was extracted with methanol (80%) and Chloroform in a soxhlet apparatus separately. Aqueous extract was prepared by cold maceration process by using separate quantity of powder. The solvents were removed by distillation under reduced pressure and the resulting semisolid mass was vacuum dried using rotary flash evaporator.

#### Physical evaluation[12]

#### Determination of solvent extractive value[13]

#### Determination of water soluble extractive value

5gm of powdered drug was macerated with 100ml of water closed flask for 2hr and was occasionally shaken with 6hr time period and was allowed to stand for 18hr. After filtration the 25ml of the filtrate evaporated to dryness in a tarred flat bottomed shallow dish. Dried at 105°C and weighed. Percentage of water soluble extractive value was calculated with reference to the air dried drug. (Table-1)

#### Determination of alcohol soluble extractive value

Alcohol is an ideal solvent for extraction of various chemicals like tannins, alkaloids, resins etc. Ethyl alcohol (95% v/v) was used for determination of alcohol soluble extractive.

Five gm of powdered drug was macerated with 100ml of ethanol closed flask for 24hr and was occasionally shaken with 6hr time period and was

allowed to stand for 18hr. After filtration the 25ml of the filtrate evaporated to dryness in a tared flat bottomed shallow dish. Dried at 105°C and weighed. Percentage of ethanol soluble extractive value was calculated with reference to the air dried drug. (Table-1)

#### Determination of ether soluble extractive value

100 gm of coarsely powdered air dried drug was extracted in soxhlet apparatus with solvent ether for six hours. The extract is filtered into a tared evaporating dish and evaporates off the solvent on a water bath. The residue is dried at 105°C to constant weight. The

percentage of ether extractive was calculated with reference to air dried drug (Table-1)

#### Determination of Moisture Content [12,13]

The percentage of active constituents in crude drug is mentioned on air dried bases. Hence, the moisture content of the crude drugs should be determined and should also be controlled. The moisture content should be minimized in order to prevent decomposition of crude drugs either due to chemical changes or microbial contamination. (Table-1)

Table 1: Solvent Extractive Value

Drug	Water soluble extractive value(% w/w)	Alcohol soluble extractive value(%w/w)	Ether soluble extractive value (%w/w)	Moisture content (%w/w)
<i>Salvadora oleoides</i>	21.05	10.20	4.75	3.1

Table 2: Physico-chemical Characteristics

Drug	Foreign organic matter (% w/w)	Total ash value (%w/w)	Acid insoluble ash value (%w/w)	Water soluble ash value (%)
<i>Salvadora oleoides</i>	0.7	4.1	0.5	0.8

The powdered sample of stems of *Salvadora persica* weighed 5gm accurately and kept in IR moisture balance. The loss in wt. was recorded as percentage (%) moisture with respect to air-dried sample of crude drug.

#### Determination of foreign matter

About 10 gm of the sample was weighed and spread on a white tile uniformly without overlapping. Then the sample was inspected by means of 5x lens and the foreign organic matter was separated. After complete separation the matter was weighed and percentage w/w was determined.(Table-2)

#### Determination of Ash value [13,14]

The residue remaining after incineration is the ash content of the drug, which simply represents inorganic salts, naturally occurring in drugs or adhering to it or deliberately added to it as a form of adulteration. Many a time the crude drugs are admixed with various mineral substances like sand, soil, calcium oxalate, chalk powder or other drugs with different inorganic content. Ash value is a criterion to judge the purity of crude drugs. Generally either total ash value or acid-insoluble ash value or both is determined. Total ash usually consists of phosphates, silicates and silica. On the other hand, acid-insoluble ash, which is a part of total ash insoluble in dilute hydrochloric acid, contains adhering dirt and sand.

#### Determination of total ash

Total ash was determined by weighing 2 gm of the air dried crude drug in the tared platinum or silica dish and incinerated at a temperature not exceeding 450°C until free from carbon and then was cooled and weighed. (Table-2)

#### Determination of acid insoluble ash

The ash obtained from the previous process was boiled with 25ml of 2M HCl for 5 min. and the insoluble matter was collected on ash-less filter paper and was washed with hot water, ignited, cooled in a desiccator and weighed. Percentage of acid insoluble ash was calculated with reference to the air dried drug. (Table-2)

#### Determination of water soluble ash

The ash was boiled with 25ml of water for 5 min. and the insoluble matter was collected on ash-less filter paper and was washed with hot water, ignited for 15min. at a temperature not exceeding 450°C. The weight of the insoluble matter was subtracted from the weight of the ash and this represents the water soluble ash. Percentage of water soluble ash was calculated with reference to the air dried drug. (Table-2)

#### Qualitative Phytochemical analysis [10,11,15]

The extracts obtained were subjected to various qualitative tests to reveal the presence or absence of common Phytopharmaceuticals by using standard tests. (Table-3)

Table 3: Qualitative Phytochemical tests

Sr. no	Test for	Water extract	Alcohol extract	Chloroform extract
1	Alkaloids	-	+	+
2	Glycosides	-	+	-
3	Volatile oil	+	+	-
4	Gum & Resins	+	+	+
5	Tannins	+	+	-
6	Flavanoids	+	+	+
7	Proteins	+	+	-
8	Steroids	-	+	-
9	Carbohydrates	+	+	-

#### RESULT & DISCUSSION

It is a shrub or small tree, attaining 6-9 m height; Leaves are linear and lanceolate, dark greenish-yellow when young, grey when mature. Flowers are sessile and greenish-white, calyx cup-shaped, in 4 rounded, obtuse lobes. Fruit globose yellow when ripe, dark brown or red when dry. Seeds greenish-yellow, about 3 mm in diameter. Microscopy shows the presence of epidermis layer which is made up of number of cells. Cuticle layer is present for the protection. Stoma is present on the epidermis but type of stomata is not clear. Parenchyma is present with intercellular spaces. Pith is present in the central part of the stem.

Water, alcohol and ether extractive value were found to be 21.05% (w/w), 10.20% (w/w) and 4.75% (w/w) respectively. Moisture content and foreign organic matter were 3.1 % (w/w) and 0.7 % respectively. Total ash value, acid insoluble ash and water soluble ash were found to be 4.1%, 0.5%, and 0.8% respectively as shown in table 1 and 2.

Qualitative Phytochemical tests showed the presence of different phytochemicals in water, alcohol and chloroform extract of stems of *Salvadora oleoides* as shown in table 3.

#### CONCLUSION

*S.oleoides* is a medicinal herb belongs to the family Salvadoraceae. It is commonly known as methajal. Above studied Pharmacognostical

and Phytochemical parameters are used to check its quality, purity and for its identification. A number of secondary plant metabolites were isolated from stems of *S.oleoides*. But still there is scope for its chemical profile. Its chemotherapeutic value has not been fully substantiated and the mode of action of its bioactive compounds against diseases has not yet been established. Alkaloids, flavonoids, tannins, glycosides which are present in this plant may be responsible for its pharmacological activities. The road ahead is to establish specific bioactive molecules, which might be responsible for these actions. Therefore the cultivation, collection, and further pharmacological exploration of *S.oleoides* are essential.

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