



**PHARMACOGNOSTICAL, PHYTOCHEMICAL AND PHARMACOLOGICAL  
SCREENING OF *LAGENARIA SICERARIA* LEAVES**

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**ABSTRACT**

In the present study leaves of *Lagenaria siceraria* was subjected to pharmacognostical studies such as macroscopic, microscopic and micromeritics parameters were also observed. Physicochemical studies such as ash values, extractive values of plant part were carried out to confirm the identity of plant. Ash values such as total ash, acid insoluble ash and water-soluble ash were determined and recorded. Extractive values such as alcohol soluble extractives and water-soluble extractive values were also determined. The leaves of *Lagenaria siceraria* shows the presence of phytoconstituents such as carbohydrates, flavonoids, terpenoids, flavanoids and tannins. This article also covered important pharmacological profile of the plant *Lagenaria siceraria*.

**Keywords:** *Lagenaria siceraria*, Microscopy, Pharmacognosy, Phytochemistry

**INTRODUCTION**

*Lagenaria siceraria* (Molina) standley (LS) (Family: Cucurbitaceae) is an annual herbaceous climbing plant with a long history of traditional medicinal uses in many countries, especially in tropical and subtropical regions. Since ancient times the climber has been known for its curative properties, and has been utilized for

treatment of various ailments, including jaundice, diabetes, ulcer, piles, colitis, insanity, hypertension, congestive cardiac failure (CCF), and skin diseases. Its fruit pulp is used both as an emetic and purgative, and for its cooling, diuretic, antibilious, and pectoral properties. Boiled in oil this pulp is used to treat rheumatism

and insomnia. A wide range of chemical compounds including sterols, terpenoids, flavonoids, and saponins have been isolated from the species. Its extracts have been found to possess various pharmacological activities [1-2].

## MATERIAL AND METHODS

### 1. Procurement and authentication of Crude drugs:

The leaves of *Lagenaria siceraria* were procured from local market of Indore (M.P.) and authenticated from Department of Botany, Devi Ahilya Vishvavidhyalaya,

Indore (Voucher No. MODERN/MIPS/PHARM/2022/76. The leaves were then allowed to dry in air and crushed in small pieces for extraction and extractive values.

### 2. Evaluation Parameters:

(A) Pharmacognostic examination: [3, 4, 5]

(i) Macroscopic examination: [3,4,5]

The morphological studies such as size, shape, color, odour and taste of *Lagenaria siceraria* leaf were carried out (Table 1).



Figure 1: Leaves of *Lagenaria siceraria*

Table 1: Morphological Evaluation

|          |  |
|----------|--|
| Size     | 7.9 - 11.5 -15.5cm in length.<br>3.8 - 4.5 - 5.7cm in breadth. |
| Shape    | Elliptical   |
| Margin   | Entire   |
| Venation | Parallel   |
| Apex     | Acute to acuminate   |
| Base     | Sub sessile to cuneate   |
| Surface  | Leathery to coriaceous   |
| Texture  | Firm, flexible, slightly succulent                             |
| Colour   | Dark green adaxially, light green abaxially                    |
| Taste    | Bitter   |
| Odour    | Characteristic   |

**(ii) Microscopic examination: [3, 4, 5]**

Fresh leaves of *Lagenaria siceraria* were taken and transverse sections of leaves were cutted and were observed under microscope. Transverse section of *Lagenaria siceraria* leaf showed following features. Upper epidermis consists of elongated parenchymatous cells, covered by cuticle. It shows few stomata, which are of anisocytic type. Palisade cells are present at upper and lower epidermis. It shows hexagonal to polygonal, large, thin-walled colourless cells, may be water storing. Mesophyll – Mesophyll is made up

of 3-4 layered chloroplast containing, compactly arranged, oval to circular cells. It is interrupted by vascular bundles of various sizes. Vascular bundles - Vascular bundles are surrounded by 2-3 layered sclerenchyma. They are conjoint, collateral and closed. Xylem is placed towards upper epidermis and phloem towards lower epidermis. Lower epidermis – contains elongated wavy walled parenchymatous cells covered by cuticle. Number of Covering and collapsed trichomes are present, while very few glandular trichomes are also present (**Figure 1**).



**Figure 1:** Transverse section of *Lagenaria siceraria* leaf

**(B) Micromeretic parameters: [6]**

**(i) Angle of repose:** Angle of repose is the maximum angle possible between the surface of a pile of the powder and the horizontal plane. Procedure: A glass funnel is held in place with a clamp on a ring support over a glass plate. Approximately 50 gm of powder is transferred in to the funnel by keeping the orifice of the funnel

blocked by the thumb. As the thumb is removed, the labjack is adjusted so as to lower the plate and maintain about a 6.4 mm gap between the bottom of the funnel stem and the top of the powder pile. When the powder is emptied from the funnel, the angle of the heap to the horizontal plane is measured with the protractor and calculated by following formula (**Table 3**).

$$\tan \theta = h/r$$

$$\theta = \tan^{-1} h/r$$

Where,

h = height of pile,

$\theta$  = angle of repose

r = radius of the base of the pile

#### Bulk Density:

Bulk density is the mass of powder divided by its bulk volume. Powder of *Lagenaria siceraria* leaf was passed through a standard sieve no.20 was introduced in to the bulk density apparatus and the timer knob was set for 100 tapping. The volume occupied by the powder was noted. Further, another 50 taps were continued and the process of tapping was continued until concurrent volume was achieved as bulk volume. Then bulk density was calculated by using this equation (Table 3).

$$\text{Bulk density } (\rho) = \frac{\text{Mass of powder}}{\text{Bulk volume}}$$

#### Tapped Density:

Tapped density was achieved by mechanically tapping a measuring cylinder containing a powder sample. After observing the initial volume, the cylinder was mechanically tapped, and volume readings were taken until no further volume change was observed (Table 3).

$$\text{Tapped density } (\rho) = \frac{\text{Mass of powder}}{\text{Tapped volume}}$$

#### Extraction:

50gm of coarsely powdered drugs was kept for maceration in 200ml alcohol for 7 days. It was remacerated and obtained

extracts were further used for chemical evaluation.

### 3. Physical Evaluation: [7, 8]

#### A. 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 3).

#### B. Determination of solvent extractive value:

##### i) Determination of water soluble extractive value: [8]

Five gm of powdered drug was macerated with 100 ml of water in a closed flask for 2hr and was occasionally shaken for 6hr time period and was allowed to stand for 18hr. After filtration 25 ml 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 3).

##### ii) Determination of alcohol soluble extractive value: [8]

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 3).

#### C. Determination of Moisture Content: [8, 9]

The percentage of active constituents in crude drug is mentioned on air dried basis. Hence, the moisture content of the crude drug 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.

**Procedure:** The powdered sample of leaves of *Lagenaria siceraria* was 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 (Table 3).

#### D. Determination of Ash value: [8, 9]

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 creation 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.

##### i) 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 3).

##### ii) 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 dessicator and weighed. Percentage of acid insoluble ash was calculated with reference to the air dried drug (Table 3).

##### iii) 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 3).

#### 4. Qualitative Phytochemical analysis [5, 8, 9]

The extracts obtained were subjected to various qualitative tests to reveal the presence or absence of common phytopharmaceuticals (Table 4).

##### A. Alkaloids

Small portion of alcoholic extract was stirred separately with a few drops of dilute hydrochloric acid and then filtered. The filtrate was then tested carefully with various alkaloid reagents such as:

##### (a) Mayer's test

Alkaloids give precipitate with Mayer's reagents. One ml of Mayer's reagent (Potassium mercuric iodide solution) was added to 1 ml extract, whitish yellow or cream colored precipitate indicated the presence of alkaloids.

##### (b) Dragendroff's test

Alkaloids give orange brown precipitate with Dragendroff's reagents. One ml of Dragendroff's reagent (Potassium bismuth iodide solution) was added to 1 ml extract, an orange-red precipitate indicated the presence of alkaloids.

##### (c) Hager's test

Alkaloids give yellow colored precipitate with Hager's reagents. In to the 1 ml

extract, 3 ml of Hager's reagent (saturated aqueous solution of picric acid) was added, a yellow colored precipitate indicated the presence of alkaloids.

##### (d) Wagner's test

Alkaloids give reddish brown precipitate with Wagner's reagents. In to 1 ml extract, 2 ml of Wagner's reagent (iodine in potassium iodide) was added and the formation of reddish-brown precipitate indicated the presence of alkaloids.

##### B. Carbohydrates and glycosides:

Small quantity of extract was dissolved in distilled water and filtered. The filtrate was subjected to the following test for Carbohydrates.

##### (a) Molisch's Test

One ml of  $\alpha$ -naphthol solution and concentrated sulphuric acid was added in 2 ml of the extract, through the side of the test tube. The formation of purple or reddish violet color at the junction of the two liquids reveals the presence of carbohydrates.

##### (b) Fehling's solution test

Equal volume of Fehling's A (copper sulphate in distilled water) and Fehling's B (potassium tartrate and sodium hydroxide in distilled water) reagent was mixed along with few drops of extract solution and boiled, a brick red precipitate of cuprous oxide forms.

##### (c) Benedict's test

Extract solution was treated with few drops of Benedict reagent (alkaline Solution containing cupric citrate complex) and upon boiling on water bath, reddish brown precipitate forms, if reducing sugar is present.

#### C. Test for proteins and free amino acids:

Small quantity of alcoholic extract was dissolved in to the few ml of water and subjected to Millon's, Biuret and Ninhydrin tests.

#### Test for gums and mucilage:

About 10 ml of extract was slowly added to the 25ml of absolute alcohol with constant stirring, filtered, dried in air and examine for its swelling properties.

#### D. Terpenoids:

Alcoholic solution of Sudan III was added to the thin section of the drug, red color shows the presence of terpenoids.

#### E. Volatile oil:

50 gm of coarse powder was taken in a Clevenger's apparatus and subjected to hydro distillation. The distillate was observed for the presence of volatile oil.

#### F. Tannins:

Small quantities of alcoholic extract was taken separately in water and tested for the presence of phenolic compounds and tannins with dilute ferric chloride solution (5%), 1% solution of gelatin containing 10% sodium chloride, 10% lead acetate and aqueous bromine solutions.

### 5. PHARMACOLOGICAL ACTIVITIES

**a. Antidepressant Activity [10]** Prajapati *et al.* evaluated the antidepressant activity of methanolic extract of *Lagenaria siceraria* fruits in rats by using forced swim (behavior despair) model. The extract was used in doses of (50, 100 and 200 mg/kg, orally). The extract possesses antidepressant activity in dose dependent manner. The activity may be due to presence of flavanoids, triterpenoids, sterols and saponins.

**b. Antistress Activity** Lakshmi *et al.* evaluated the antistress activity of ethanolic extract of *Lagenaria siceraria* in mice by using anoxia tolerance and chronic cold restraint stress models. The extract was evaluated in doses of (100, 200 and 400 mg/kg, per oral). The extract prolonged the onset of clonic convulsion, and amelioration of cold stress induced changes, thereby suggested an antistress and adaptogenic property. The activity was due to presence of flavonoids, glycosides, saponins, triterpenes, cucurbitans and steroids [11].

**c. Hypolipidemic and Antihyperlipidemic Activity** Ghule *et al.* evaluated the hypolipidemic and antihyperlipidemic effect of different extract viz, Petroleum ether, Chloroform, Alcoholic and Aqueous extract of *Lagenaria siceraria* fruit in Triton- induced hyperlipidemic rats. Among the extract chloroform and alcoholic extract at two

doses (200 and 400 mg/kg, per oral) showed significant effects in lowering total cholesterol (TC), triglyceride (TG) and low density lipoprotein (LDL) along with an increase in HDL level. The activity was due to presence of sterols (campesterol and fucosterol) [12].

**d. Diuretic Activity** Ghule *et al.* evaluated *Lagenaria siceraria* fruit for its diuretic activity in albino rats. The rats treated with vacuum dried fruit juice extract and methanol extract (100-200 mg/kg; per oral) showed higher urine volume when compared to the respective control. The activity indicated that vacuum dried fruit juice extract and methanolic extract act as effective hypernatremic, hyperchloremic and hyperkalemic diuretics (increased Na<sup>+</sup>, K<sup>+</sup> and Clexcretion values) [13] .

**e. Analgesic and Anti-inflammatory Activity** Ghule *et al.* investigated analgesic and antiinflammatory effects of *Lagenaria siceraria* (Molina) Stand. Fruit juice extract (LSFJE) in rats and mice. LSFJE was studied for its analgesic effect on acetic acid-induced writhing and formalin pain tests in mice. The anti-inflammatory effects were investigated employing the acute inflammatory models, i.e. ethyl phenyl propionate induced ear edema, carrageenan, and arachidonic acid-induced hind paw edema and also the albumin-induced paw edema in rats. The activity was due to the presence of flavonoids [14].

**f. Antiulcer Activity** Srivastava *et al.* evaluated antiulcer activity of methanolic extract of the fruit of *Lagenaria siceraria* against Aspirin, Pylorus ligation and Ethanol induced ulcer models. The extract were used in dose of (100 and 200 mg/kg, per oral) and proved that the methanolic extract of the fruit in a dose (100-200 mg/kg, per oral) possesses potent antiulcer activity in rats [15] .

**g. Antidiabetic Activity** Saha *et al.* evaluated the methanolic extract of *Lagenaria siceraria* aerial parts for Antidiabetic activity using Streptozotocin induced diabetic rats. The extract of were used in dose of (200 and 400mg/kg, per oral). Streptozotocin induced diabetic rats treated with methanolic extract showed the significant reduction in blood sugar levels and proved that the aerial part of the *Lagenaria siceraria* possess potent Antihyperglycemic activity, which is probably due to its rich flavanoid and polyphenolic contents [16].

**h. Anticancer Activity** Saha *et al.* evaluated the methanolic extract of *Lagenaria siceraria* (Mol.) Standly aerial parts for anticancer activity against Ehrlich Ascites carcinoma (EAC) model in mice. The extract was used in dose of (200 and 400 mg/kg, per oral). In EAC tumor bearing mice, a regular rapid increase in ascites tumor volume was observed. Ascitic fluid is the direct nutritional source for

tumor cells and a rapid increase in ascetic fluid with tumor growth would be the means to meet the nutritional requirement of tumor cells. The extract significantly inhibited the tumor volume, packed cell volume and viable tumor cell count, increasing the non viable cell count. The extract possesses significant anticancer activity which may be due to presence of flavonoid [17].

**i. Cardioprotective Activity** Upaganlawar *et al.* evaluated the cardioprotective activity of *Lagenaria siceraria* fruit powder extract against Isoproterenol induced Myocardial Infarction induced in Wistar male rats and proved that the *Lagenaria siceraria* possessed cardioprotective effect against Isoproterenol induced Myocardial infarction in rats. These effects might be due to the high conc. of polyphenolic components in the fruit of *Lagenaria siceraria* [18] .

**j. Hepatoprotective Activity** Deshpande *et al.* evaluated hepatoprotective activity of the ethanolic extract of the *Lagenaria siceraria* fruit epicarp against the carbon tetra chloride induced hepatotoxicity. The extract of the fruit epicarp was used in doses of (100 and 200 mg/ kg, per oral). Silymarin was used as standard in dose of (100 mg/kg, per oral). The extract in a dose of (200 mg/kg/, per oral) showed significant prevention of elevated levels of serum glutamate oxaloacetate transferase

(SGOT), serum glutamate pyruvate transaminase (SGPT), alkaline phosphatase (ALP), acid phosphatase (ACP) and bilirubin. The activity was due to presence of sterols (campesterol and fucosterol) [19].

**k. Antioxidant Activity** Deshpande *et al.* investigated the antioxidant activity of ethanolic extract of the *Lagenaria siceraria* fruit epicarp against the CCl<sub>4</sub>- induced hepatotoxicity. The extract of fruit epicarp was used in doses of (100 and 200 mg/ kg, per oral). Silymarin was used a standard in dose of (100 mg/kg, per oral). The extract in a dose of (200 mg/kg) shows significant prevention of elevated levels of serum glutamate oxaloacetate transferase (SGOT), serum glutamate pyruvate transaminase (SGPT), alkaline phosphatase (ALP), acid phosphatase (ACP) and bilirubin. The activity was due to presence of sterols (campesterol and fucosterol) [19].

**l. Immunomodulatory Activity** Sathaye *et al.* evaluated the immunomodulatory effect of fresh fruit juice of *Lagenaria vulgaris* against Pyrogallol induced immunesuppression in rats. They observed that fresh fruit juice cause stimulation of both humoral and cellular immune response in immunecompromised condition. Various non specific immunity parameters were also measured such as Neutrophils and total Leukocyte count. There was increase in Neutrophils and total Leukocyte count,

which could be responsible for stimulation of non specific immunity [20].

**m. Anthelmintic and Antimicrobial Activity** Badmanaban *et al.* evaluated the anthelmintic and antimicrobial activity of the different leaf extract of *Lagenaria siceraria*. Ethanolic leaf extract showed the

presence of carbohydrates, flavonoid, tannins and sterols. It is quite apparent from the studies that the sterols present in the ethanolic extract possess significant anthelmintic activity and antimicrobial activity [21].

**Table 2: Pharmacological activities of different parts of *Lagenaria siceraria* [22]**

| Sr. No. | Activity                             | Plant part                          |
|---------|--------------------------------------|-------------------------------------|
| 1.      | Hepatoprotective                     | Fruit                               |
| 2.      | Immunomodulatory                     | Fruit, Fruit Juice                  |
| 3.      | Diuretic                             | Fruit                               |
| 4.      | Antidepressant                       | Fruit                               |
| 5.      | Hypolipidemic and Antihyperlipidemic | Fruit                               |
| 6.      | Cardioprotective                     | Fruit                               |
| 7.      | Antiulcer activity                   | Fruit                               |
| 8.      | Antihyperglycemic                    | Aerial parts, Fruit epicarp         |
| 9.      | Anticancer                           | Aerial parts, Stem, Fruit           |
| 10.     | Antibacterial activity               | Leaf                                |
| 11.     | Anthelmintic activity                | Leaf                                |
| 12.     | Analgesic                            | Fruit                               |
| 13.     | Anti-inflammatory                    | Root                                |
| 14.     | Antistress activity                  | Fruit                               |
| 15.     | Antioxidant activity                 | Leaves, Fruit Fresh and dried Fruit |

## RESULT & DISCUSSION

Macroscopic study reveals that the *Lagenaria siceraria* leaf were dark green adaxially, light green abaxially, elliptical in shape, having characteristic odour and having bitter taste (Table 1). Microscopic study of *Lagenaria siceraria* leaf showed upper epidermis consists of elongated parenchymatous cells, covered by cuticle. It shows few stomata, which are of anisocytic type. Palisade cells are present at upper and lower epidermis. It shows hexagonal to polygonal, large, thin-walled colourless cells, may be water storing.

Micromeretic parameters such as angle of repose, bulk density and tapped density

were found to be 37.09, 0.25, and 0.27 respectively. Water soluble extractive value and alcohol soluble extractive value were found to be 4.04 % (w/w) and 5.36 % (w/w) respectively. Moisture content and foreign organic matter were 4.60 % (w/w) and 2 % respectively. Total ash value, acid insoluble ash and water-soluble ash were found to be 2.5 %, 1 % and 2.1 % respectively. Qualitative phytochemical tests showed the presence of carbohydrates, flavonoids, terpenoids, flavonoids and tannins in ethanolic extract of *Lagenaria siceraria* leaf (Table 4).

Table 3: Results of different parameters of *Lagenaria siceraria* leaf.

| S. No. | Particulars                              | Result |
|--------|--|--------|
| 1.     | Angle of repose                          | 37.09  |
| 2.     | Bulk Density                             | 0.25   |
| 3.     | Tapped Density                           | 0.27   |
| 4.     | Foreign Organic Matter                   | 2 %    |
| 5.     | Water soluble Extractive value (% w/w)   | 4.04   |
| 6.     | Alcohol soluble Extractive value (% w/w) | 5.36   |
| 7.     | Moisture content (% w/w)                 | 4.60   |
| 8.     | Total Ash Value (%)                      | 2.5    |
| 9.     | Acid Insoluble Ash Value (%)             | 1      |
| 10.    | Water Soluble Ash Value (%)              | 2.1    |

Table 4: Qualitative phytochemical tests

| S. No. | Test for      | Result |
|--------|---------------|--------|
| 1.     | Alkaloids     | -      |
| 2.     | Carbohydrates | +      |
| 3.     | Glycosides    | -      |
| 4.     | Volatile oil  | -      |
| 5.     | Proteins      | -      |
| 6.     | Terpenoids    | +      |
| 7.     | Gums & resins | -      |
| 8.     | Tannins       | +      |
| 9.     | Flavanoids    | +      |
| 10.    | Saponins      | +      |

## CONCLUSION

From the above studies it can be concluded that the various parameters such as pharmacognostical, phytochemical and micromeritics parameters of the leaves of *Lagenaria siceraria* may be utilized for its identification and differentiation from other species. Phytochemical investigation reveals the presence of triterpenoids and tannins in etanolic extract of *Lagenaria siceraria* leaf. Due to presence of these compounds in the leaves of *Lagenaria siceraria*, it showed variety of pharmacological activities like Hepatoprotective, Immunomodulatory, Diuretic, Antidepressant, Hypolipidemic and Antihyperlipidemic, Cardioprotective, Antiulcer activity, Antihyperglycemic, Anticancer, Antibacterial activity,

Anthelmintic activity, Analgesic, Anti-inflammatory, Antistress activity and Antioxidant activity. Thus, the *Lagenaria siceraria* leaf may be a good choice for the futuristic research on such activities.

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