



ORIGINAL ARTICLE

# Phytochemical evaluation of the whole Sunishannaka plant (*Marsilea quadrifolia* Linn)

Jilu Joy<sup>1</sup>, Sara Monsy Oommen<sup>2</sup>, P Y Ansary<sup>3</sup> & Shincymol V V<sup>4</sup>

<sup>1</sup>Assistant Professor, Department of Dravyaguna Vijnana, Govt. Ayurveda College, Tripunithura, Kerala, India

<sup>2</sup>Professor, Department of Dravyaguna Vijnana, Govt. Ayurveda College, Tripunithura, Kerala, India

<sup>3</sup>Professor & HOD, Department of Dravyaguna Vijnana, Govt. Ayurveda College, Tripunithura, Kerala, India

<sup>4</sup>Associate Professor, Department of Dravyaguna Vijnana, Govt. Ayurveda College, Tripunithura, Kerala, India

\*Email: [drjilunoble@gmail.com](mailto:drjilunoble@gmail.com)

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## Abstract

A well-known perennial aquatic plant, *Sunishannaka* (*Marsilea quadrifolia* Linn) which is seen in moist and marshy lands, is being used extensively since *Samhita* period. Ample references about the plant is seen in *Nighantus* as well. A plant's medicinal value is assessed based on the biologically active components it contains and their concentration. The extraction, screening, and identification of the medicinally active compounds present in plants are referred to as phytochemical analysis. Due to their uses as medications, model molecules for drug production, or intermediates for synthetic pharmaceuticals, plant-derived chemicals are highly significant. Thus the study of phytochemicals reveals the potentials of a drug at its pharmacological aspects. Phytochemical screening of *Marsilea quadrifolia* Linn has revealed the presence of numerous chemical constituents like alkaloids, tannins, flavonoids, steroids, glycosides and saponins etc.

## Introduction

Pterydophytic fern Sunishannaka (*Marsilea quadrifolia* Linn), a member of the Marsileaceae family, is typically found in damp, marshy areas, flooded low lands, and along the banks of canals and rivers. It is commonly called as four leaved European water clover in English and as "Neeraraal or Nalilathamara" in Malayalam. In *Charaka Samhitha*, it is included under *saka varga* possessing *tridoshaghna* property<sup>[1]</sup>. It is included under *pathya saka varga* by Acharya Vagbhata<sup>[2]</sup>. *Acharyas* have highlighted the *tridoshahara* property and *laghuguna* of *Sunishannaka*. Many folkloric utility of the plant is being noticed in for insomnia, hypertension, stomach ulcer etc. Pharmacological claims of *Marsilea quadrifolia* Linn can be ascribed for its relevant bioactive phytoconstituents like tannin, flavonoid, saponin, alkaloids etc. Phytochemical screening promotes to isolate various active constituents present in plants for assessing their biological activity or medicinal utility. Different plant parts may contain various types and quantities of bioactive compounds. In essence, the distribution of bioactive compounds or active principles, which are more concentrated in particular areas of the plants, has a significant impact on the biological activity of plants<sup>[3]</sup>. This paper is intended to highlight the results of the preliminary phytochemical analysis of the plant.



## Materials and Methods

### 1. Collection of the plants:

The Sunishannaka plant, *Marsilea quadrifolia* Linn, was collected in its whole from the marshy areas and adjoining paddy fields in Kandanad, Ernakulam district. By using pharmacognostic examinations, which were carried out in the department of Dravyaguna vijnanam Government Ayurveda College, Tripunithura, the samples were verified as being genuine. The entire plants were completely cleansed in water to remove any physical impurities like soil, mud, etc., and then allowed to dry thoroughly in the shade. Once powdered, it was stored in airtight containers. Preliminary phytochemical testing on this powdered medication was carried out at the Drug Standardization Unit of the Department of Dravyagunavijnanam, Government Ayurveda College, Tripunithura.

### 2. Reagents and Apparatus used

Concentrated and dilute Hydrochloric acid, Xylene, Concentrated and dilute Sulphuric acid, Concentrated and dilute Nitric acid, Sodium hydroxide solution, Lead acetate solution,  $KMNO_4$  solution, Anhydrous Sodium carbonate, Petroleum ether, Cyclohexane, Acetone, Alcohol, Fehling's Solution A&B, Chloroform, Dragendroff's reagent, Mayer's reagent, Neutral ferric chloride, Magnesium ribbon, Methylene blue reagent, Sodium bicarbonate Copper Sulphate, Catechol, Folin catechu phenol reagent.

Round bottom flask, silica crucible, Dean and Stark's apparatus, Clevenger's apparatus, Soxhlet apparatus, water condensers, Buchner funnel, hot air oven, muffle furnace, bunsen burner, heating mantle, glass beakers, petri dishes, standard flask, measuring jars, conical flask, funnel, glass rods, watch glass, burettes, pipettes, shaker, centrifuge, Whatmann filter paper.

### 3. Procedure

#### 3.1 Physicochemical parameters

The powder of the whole plant of *Marsilea quadrifolia* Linn was assessed for various physico chemical parameters like foreign matter, total ash, acid insoluble ash, water insoluble ash, volatile oil, moisture content, fibre, tannin, total sugar, reducing sugar, phenol and pH.

#### 3.2 Qualitative analysis of Ash

The ash was subjected to qualitative analysis to confirm the presence of acid radicals- carbonate, phosphate, chloride, sulphate, and basic radical potassium.

#### 3.3 Determination of Extractive values

The cold alcohol soluble, hot alcohol soluble, cold water soluble and hot water soluble extractive values of powder

of the whole plant of *Marsilea quadrifolia* Linn was assessed in the study. Successive solvent extraction of the drug was also carried out using the solvents petroleum ether, cyclohexane, acetone and alcohol.

#### 3.4 Qualitative evaluation of phytochemical parameters

Qualitative phytochemical screening was done in the extracts of the whole plant *Marsilea quadrifolia* Linn to assess the presence or absence of phytochemical constituents like alkaloids, flavonoids, phenols, saponins, carbohydrates, proteins, steroids and tannins. The physical and preliminary phytochemical analysis was done according to the standard procedures mentioned in the Ayurvedic Pharmacopoeia of India<sup>[4]</sup>.

## Results

The following tables contains the findings from the initial phytochemical screening of the entire *Marsilea quadrifolia* Linn plant.

**Table: 1** Results of Sunishannaka's physicochemical parameters

Sl no	Parameters	<i>Sunishannaka</i>
1	Foreign matter	Nil
2	Total ash value	8%
3	Acid Insoluble Ash value	3.4%
4	Water Insoluble Ash value	4.7%
5	Moisture Content	6.82%
6	Volatile oil content	Nil
7	Fibre content	20.08%
8	Tannin Content	58.4%
9	Total sugar content	3.50%
10	Reducing sugar content	2.9%
11	Phenol content	37.6µg/g
12	pH	5.6 (blue litmus turned red)

**Table: 2** Extraction values for alcohol and water

Sl no	Form of Extractives	<i>Sunishannaka</i>
1	Cold Alcohol soluble value	8.6 %
2	Hot Alcohol soluble value	15.8 %
3	Cold water soluble value	3.9 %
4	Hot water soluble value	19.6 %

**Table: 3** Sunishannaka solvent extraction results

Sl.no	Solvent types	Successive solvent extraction
1	Petroleum ether	4.62%
2	Cyclohexane	6.61%
3	Acetone	5.25%
4	Alcohol	6.4%

**Table: 4** Sunishannaka phytochemical analysis results, qualitative

Sl.no	Tests	Sunishannaka
1)	Alkaloids	
a)	Dragendroff's test	-
b)	Meyer's test	+
2)	Flavonoids	+
3)	Saponins	+
4)	Carbohydrates	
a)	Fehling's test	++
b)	Benedict's test	++
5	Proteins	++
6)	Phenols	
a)	Ferric chloride test	+
b)	Lead acetate test	+
7)	Steroids	+
8)	Tannins	
a)	Ferric chloride test	+
b)	Lead acetate test	++

**Table: 5** Findings of qualitative evaluation of Sunishannaka extractives (Marsilea quadrifolia Linn)

Sl no:	Extract	Steroids	Alkaloids	Flavonoids	Phenols
1	Petroleum ether	+	++	+	-
2	Cyclohexane	-	++	-	-
3	Acetone	++	-	-	+
4	Alcohol	+	-	+	+

**Table: 6** Findings from qualitative analysis of Sunishannaka ash

Sl No	Tests	Sunishannaka
<b>Acid radicals</b>		
1	Carbonate	+
2	Phosphate	+
3	Chloride	+
4	Sulphate	++
<b>Basic radicals</b>		
5	Potassium	+

## Discussion

Preliminary phytochemical analysis was conducted on powder of whole plant of Marsilea quadrifolia Linn to analyse the physico-chemical property, extractive values and chemical constituents of the drug quantitatively and qualitatively. A number of physicochemical factors were estimated, including the amounts of foreign material, total ash, acid insoluble ash, water insoluble ash, moisture content, volatile oil content,

fibre content, tannin content, phenol content, total sugar, and reducing sugar. The current study demonstrated the lack of foreign material, which points to the purity of the medicine. Sunishannaka's overall ash content was discovered to be 8%. Total ash mostly refers to the residual of inorganic salts and impurities, such as sand, dirt, and other materials, left over after incinerated. This value was compared with the value obtained from an International journal and found to be lesser than that. This will ascertain the purity of the drug.

The presence of siliceous contaminants in a crude medication is indicated by acid-insoluble ash. Sunishannaka choornam's acid insoluble ash and water insoluble ash values were found to be 3.45% and 4.7%, respectively. Moisture content in the drug found to be 6.82% which minimizes the growth of micro organisms and promoting to increased shelf life of the drug.

The quantitative estimation of total sugar and reducing sugar, fiber content, tannin content and phenol content, have been estimated in this study according to standard procedures. *Sunishannaka choornam* showed 20.08% of fibre content, 58.4 % of tannin content, 115.95 microgram/gram of phenol content, 3.50% of total Sugar and 28.03% of reducing sugar content. Fiber content of a drug is helpful for removing toxins from the body, reduce excess weight around abdomen, improving health and beneficial to skin also. In Sunishannaka choornam, extractives that are soluble in both hot and cold alcohol as well as hot and cold water were estimated. Hot alcohol soluble extractive value was 15.8%, while hot water soluble extractive value was found to be 19.6%. The extractive values of cold alcohol and cold water were 3.9% and 8.6%, respectively. In the drug *Sunishannaka*, hot water soluble extractive value was more than alcohol soluble extractive values. So these values indicates that more amount of active principles are obtained in the water soluble extractives.

The extractive values of *Sunishannaka choornam* obtained in the soxhlet extraction were successively 4.62% for petroleum ether, 6.61% for cyclohexane, 5.25% for acetone and 6.4% for alcohol. This indicates that amount of extractives in polar solvents and in non polar solvents are almost similar. Qualitative analysis of the extractives of *Sunishannaka choornam* obtained in the successive solvents were analysed and it revealed that steroids, alkaloids and flavonoids were positive in petroleum ether extract. Alkaloids was highly positive in petroleum ether and cyclohexane extracts. Steroids, and phenols was positive in acetone extract and Steroids, flavanoids and phenols were positive in alcohol extract. The extractive values assist in determining which chemical components are present in the crude medicament

and which individual components are soluble in a certain solvent.

Alkaloids, Flavonoids, Saponins, Carbohydrates, Proteins, Phenols, Steroids, and Tannins were found in the drug's qualitative examination. Qualitative analysis of ash shows the presence of Carbonate, Phosphate, Chloride, Sulphate and Potassium. The pH was determined as 5.6 using digital pH meter for Sunishannaka choornam and the sample solution turned blue litmus turned red.

## Conclusion

Sunishannaka (*Marsilea quadrifolia* Linn) is an aquatic fern which is found to be rich in various phytochemicals which in turn proves its pharmacological as well as medicinal value. Therefore, early phytochemical analysis aids in identifying the components present in a crude medication and helps to de-

termine the active biochemical compounds and the likely pharmacological activities of a plant.

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