Vol 14 | Issue 1| 2023 | 01-05.

e - ISSN 2249-7544
Print ISSN 2229-7464

INTERNATIONAL JOURNAL
OF
PHYTOPHARMACY RESEARCH
www.phytopharmacyresearch.com

POLYGONUM ARENASTRUM: PHARMACOGNOSTICS, PHYTOCHEMICALS, AND ANTHELMINTIC PHYSICIDS

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ABSTRACT
There are a number of people in India who use *Polygonum arenastrum* as an anthelmintic herb. The leaves of Polygonum arenastrum were studied pharmacognostically, phytochemically, and pharmacologically. Water soluble extractive value and alcohol soluble extractive value were determined, as well as physicochemical constants such as moisture content, ash values and extractive values. It was determined that the extract obtained from successive solvent extractions could contain a number of phytochemical compounds after a preliminary phytochemical analysis was carried out. Soxhlet apparatus was used to extract leaves of Polygonum arenastrum with solvents benzene, acetone, ethanol, and water. In order to determine the anthelmintic potential of the extract, Indian earthworms were used in the study. Polygonum arenastrum ethanol extract has been found to possess significant activity when compared to other ethanol extracts.

Keywords: *Polygonum arenastrum*, Ayurveda, Phytochemical constituents, Research.

INTRODUCTION

One of the most widely practiced systems of medicine in the world is the traditional system of medicine. Since ancient times, plants have been one of the most important sources of medicinal materials. Worldwide, 85 percent of people still use traditional/ethnic medicines according to a report by the World Health Organization. [1-5] There is classified information in ancient Indian literature about the use of plants for treating various human ailments. Many of the plants that grow in India are believed to possess medicinal properties, and there are approximately 50000 of them. Growing at an altitude of between 850 and 1900 meters, *Polygonum arenastrum* belongs to the family Polygonaceae.[6]

For anthelmintic and antidiarrheal treatments, Indian healers used plants' leaves juice. *Polygonum arenastrum* leaves have been studied for pharmacognostic and phytochemical properties as well as anthelmintic activity, taking into consideration these facts.[7]

METHODS AND MATERIALS

Collection of plants

A note book was used to record field data on the plant, such as its height, soil condition, and flower colour, which was brought from India in the month of October-November and washed in continuous water. A flowering plant from the selected collection was submitted for authentication to the Department of Botany, India.

Macroscopical Observation

It was observed macroscopically that the powdered drug was shaped, size, tasted, colored, odoured, and measured in relation to each of these attributes.

Microscopical analysis

T.S OF LEAVES

A clean glass slide was prepared by sectioning plants, putting glycerine water in the center, adding phloroglucinol and HCl (2:2), and then placing the sections in the glycerine water. The cover slip was placed on the slide with the thumb and finger of the left hand, with the edge of the cover slip resting on the left-hand edge of the drop at the left-hand edge of the slide. The right-hand edge of the cover slip should be inserted into the holes of a dissecting needle, allowing the latter to rest on the needle. As the coverslip is set on the drop of liquid, it is important that the space between the cover and the slide is filled exactly without any air bubbles getting trapped inside.[8]

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The coverslip can be placed on top of the liquid slowly. By using 15X and 50X lenses, observe the T.S. of the leaf by positioning the slide on the microscope stage.

OVERVIEW OF POWDER MICROSCOPY

To obtain powder samples, the whole plant was collected and thoroughly washed with water before being analyzed to ensure the presence of any unwanted matter was eliminated. As a final step, this was dried under shade for at least another hour. For identification of lignin, starch, and calcium oxalate crystals, it was powdered, then subjected to solutions of chloral hydrate, chloroglucinol, and conc. HCl (2:2).

Physicochemical constants

- **Calculation of Ash Value:** It is important to determine the ash content of a crude drug, particularly if it is powdered, in order to measure its quality and purity. There are several parameters such as total ash, acid insoluble ash, and water-soluble ash, which were determined following standard protocols.
- **Extractive Value Determination:** An extraction value was determined for the solution soluble in alcohol and the solution soluble in water. A crude drug's extractive values indicate how many active ingredients can be extracted from its plant material using solvents.
- **Moisture measurement:** A moist environment activates enzymes and provides a suitable environment for living microorganisms to multiply. When it comes to the quality and purity of a material, drying, as well as the process of drying, plays an important role.
- **Analyzing the fluorescence:** As far as the evaluation of drug crud is concerned, fluorescence analysis is considered to be one of the most important parameters. In an ordinary light and long wavelength with different reagents, powdered drug was examined. In order to prepare the powdered drugs, the reagents were used in combination with the powdered drugs in a petridish. A series of observations was made under different wavelengths of light, such as visible light, ultraviolet light, and infrared light (255 nm and 370 nm). As a result of observing and recording the various colour that were emitted, various results were obtained. [9]

Phytochemical Screening

Plant Extraction

To remove anything unwanted, we thoroughly rinsed the leaves with water. Further drying was carried out in the shade of a tree. It was then powdered after it had dried completely and stored in a container that was airtight once it had been dried. With the help of this air-dried powder, a Soxhlet apparatus was used to perform a Successive Solvent Extraction using the air-dried powder. [10] Benzene, Acetone, Ethanol and water were used as solvents of increasing polarity. Various chemical tests were conducted as per the standard methods to identify the constituents of the concentrated extracts redissolved in respective solvents.

Aspects of pharmacology

Six cm earthworms were placed in petridishes with 30 mg, 60 mg, and 99.9% drug concentrations of solution each. Normal saline was used as a control and Albendazole solution as a reference drug. A little finger tapping demonstrated motility in the live worms and non-motility in the dead ones. In order to return the motile worms to their petridishes, they were placed in individual flasks. [11]

Microscopical examination

- **Axial view of a leaf**

Using chloral hydrate, phloroglucinol, and dilute HCl, the transverse section (T.S.) of *Polygonum arenastrum* was initiated, stained with safranin, and iodine solution was applied to observe the following components of the leaves: Upper epidermis, Vascular bundle, Lower epidermis, Starch grains, Fragments of vessels, Fibres, Calcium oxalate crystals, Xylem, Phloem, Stomata.

- **Leaves examined by powder microscopy:**

Leaves powdered from *Polygonum arenastrum* have a distinctive odor and are light brown in color. Amounts of flattened starch grains, fragments of vessels, fibres, calcium oxalate crystals, xylem, phloem, and stomata were observed during mounting with chloral hydrate, phloroglucinol, and dil. HCl. [12]

Ash Value, the Extractive Value, and the Moisture Content

Typically, the ash value is a useful metric that can be used to determine the purity and quality of a crude drug, particularly one that is powdered. The ash formed by incineration of crude drugs consists of carbonates, phosphates, calcium, magnesium, sodium, potassium. Using solvents to extract active constituents from medicinal plant material determines an extractive value. The use of this technique is employed for substances for which it is not possible to conduct a chemical or biological analysis. [13] As much as is possible, moisture must be eliminated from crude drugs as much as is feasible in order to keep them as pure as possible. The drying process is one of the most important factors when it comes to determining both the quality and purity of the material.

Fluorescence analysis

A variety of wavelengths were used to observe these: visible rays, short wavelengths (255 nm) and long wavelengths (370 nm). It was observed that different colours were emitted.

EXTRACTION

Benzene was extracted first with 550 grams of powdered shade-dried leaves using a continuous hot percolation method, using Soxhlet apparatus, at 65 to 70°C. Starting with Benzene, Acetone, Ethanol, and water, solvents were selected in order of increasing polarity. It was decided to continue the extraction for a further three days. Vacuum distillation was employed to concentrate all extracts after extraction. In order to ensure that the final extracts are stored in an airtight container until they are

later used, the extracts were sealed in an airtight container and stored in the refrigerator.[14]

Table.1. An analysis of ash value, extractive value, moisture content

SPECIFICATIONS		
Amount of Ash	Amount of total ash	11% (weight-to-weight)
	Insoluble ash in acid	A 2% (weight-to-weight ratio)
	Ash that dissolves in water	3.0% (weight-to-weight)
Value of extraction	Obtaining extracts	Amount of extraction (% by weight)
	Extracts soluble in alcohol	7.77%
	Extracts that dissolve in water	9.22
Composition of moisture	13.33% (w/w)	

Table 2. Analyses of fluorescence

LEAVES OF *POLYGONUM ARENASTRUM* TREATED WITH POWDER	**RADIATION IN THE VISIBLE RANGE**	**INFRARED LIGHT**	
		A SHORT WAVELENGTH (255 NM)	**THE LONG WAVE RANGE (370 NM)**
A powder containing			
50% Sulfuric acid	The color is light brown	Almost black	A brown color
50% Nitric acid	A brown color	Almost black	A green color
Water at a cold temperature	The color is light brown	Almost black	A green color
Ferric chloride	A yellow color	Almost black	A green color
Solution containing 5% iodine	A dark brown color	A dark brown color	Almost black
A chloroform solution	A dark brown color	A dark brown color	Almost black

Table 3. Anthelmintic evaluation.

GROUPS	**PROCEDURES**	**CONCENTRATION (MG/ML)**	**DURATION OF PARALYSIS (MINUTES)**	**TIMING OF DEATH (MINUTES)**
1	Control (NS)	00	00	00
2	Albendazole	30	30 ±00	31 ±00
3	Ethanol extract of leaves	30	91 ±3	00
		55	61 ±2.3	121 ± 1.0
		99.9	36 ±0.50	47 ±0.50

Phytochemicals screening

Various chemical tests were performed on the concentrated extracts according to standard methods for identification of constituents after they were redissolved in the respective solvents. [15]

Analyzing of pharmacological effects

Based on the close anatomical and physiological similarities between a human intestinal roundworm parasite and the Indian earthworm *Pheretima posthuma*, we evaluated anthelmintic activity. *Pheretima posthuma* (earthworm) is more sensitive to ethanol extract from *Polygonum arenastrum* leaves. An analysis of anthelmintic activity was performed with albendazole as a reference standard.Indian earthworm (*Pheretima posthuma*) was significantly inhibited by *Polygonum arenastrum* leaves. It is ideal about selecting solvent to avoid paralysis or death of an earthworm in normal saline solution.[16-19]

There has been a focus on determining the pharmacognostic, phytochemical, and pharmacological properties of leaves of *Polygonum arenastrum* in order to extract the maximum benefit from the plant. Microscopically, *Polygonum arenastrum* leaves were found to be green with a characteristic odour. They were strongly acrid in taste and had a length and diameter of around 7 cm - 9 cm. Further examination of the fresh leaf into the microscopical area revealed that the fresh leaf contained fibres, calcium oxalate crystals, xylem, phloem, stomata, Strauch granules, upper epidermis, lower epidermis, and protective tissue. Physiochemical evaluation of powdered leaves included moisture content, ash value, extractive values like alcohol soluble extractives and water-soluble extractives.[20-23]

There are more amounts of water-soluble materials in the plants than in alcohol, indicating large amounts of water-soluble compounds.[24] It has been found that different solvents can be used to perform fluorescence analysis of powder drugs in different wavelengths.[25-28] Brown, yellow, black and green colours are observed under short and long wave lengths, respectively. Solvent extraction was used to extract the powder plant. Phytochemical analysis and anthelmintic activity against Indian earthworm (*Pheretima posthuma*) were performed using different concentrations of ethanolic extract at 30 mg/ml, 60 mg/ml, and 99.9 mg/ml. Study results indicate that almost all concentrations show anti-helmintic activity against Indian earthworms. Extracted extracts are more active with higher concentrations. According to phytochemical analysis, alkaloids,

carbohydrate, glycosides, proteins and amino acids, phytosterols, tannins are present.[29]

CONCLUSION

Polygonum arenastrum leaves are examined in this study to determine phytochemical content and medicinal value. Taking into consideration the above studies and obtained results, it seems likely that *Polygonum arenastrum* leaf extract shows potent anthelmintic properties. *Polygonum arenastrum* leaves have anthelmintic properties, but more research is required to determine exactly what phytochemicals and mechanisms are involved.

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