



**A RESEARCH ARTICLE ON PHARMACOGNOSTICAL AND PHYTOCHEMICAL
STUDY OF *SELAGINELLA BRYOPTERIS* (L.) BAKER (*SANJEEVANI/AGNIJARI*)**

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ABSTRACT

Selaginella bryopteris (L.) Baker, locally known as ‘*Sanjeevani*’ means one that infuses Life. *Selaginella bryopteris* is the Indian endemic resurrection pteridophytic plant belongs to the family *selaginellaceae* and is known for its remarkable resurrection capabilities. In Sanskrit it is known as *Sanjeevani booti*, a lithophytic plant which grows on slopes of the hilly tropical areas of Himalayan region. This study includes the collection of plant sample from Kedarnath valley of Himalayan region followed by authentication of botanical identity of *Selaginella bryopteris* (L.) Baker. *Selaginella* has been used traditionally to treat many diseases such as irregular menstruation, uterine disorders for easy delivery, jaundice, heat stroke, phenols and other internal injuries etc. *Selaginella* contains a variety of secondary metabolites such as alkaloids, phenol and Flavonoids etc. due which it can act as antioxidants, anti-inflammatory, anti-cancer, anti-allergic, antimicrobial, antifungal, antibacterial, antiviral etc. It is also used as a health tonic to improve fitness and to expand life span. The Pharmacognostical, Physicochemical, Phytochemical and Chromatography study of *Sanjeevani* provide measures for identification and quality evaluation.

KEYWORDS; *Sanjeevani*, *Agnijari*, *Selaginella bryopteris*, Resurrection, Pharmacognosy, lithophytic. Physicochemical.

INTRODUCTION

The Himalayan region is particularly well endowed with medicinal plant species. This is largely because of the diverse agro-climatic conditions which exist in the area.^[1] *Selaginella bryopteris* (L) Baker (*Sanjeevani/Agnijari*) is a medicinal plant that has been widely used in indigenous system of medicine. Small amounts of the species are also used as ornamental plants, Family *Selaginellaceae* has only one genus, *Selaginella* consisting of 700-750 species and widespread in a cosmopolitan way^[2,3] It is represented by about 62 species in India.^[4] *Selaginella bryopteris* (L.) Baker. is an endemic pteridophytic species of India and Nepal distributed in warm hilly slopes of Himalayan region between 400-1000m altitude.

The herb infusion taken orally for curing burning sensation during urination, menstrual irregularities, for easy delivery and for curing jaundice.^[5] The herb is extensively exploited in very large scale and sold-brought in various markets of the country under different trade names viz. *Sanjeevani*, *Sanjiwani Booti*, *Lakshanam Booti*, *Mrit-sanjeevani*, *Pathar Chatta*, *Sanjeevan* etc.^[6] Therefore the exploitation of the herb should be check immediately for its conservation;

otherwise this magical endemic herb may be lost completely from nature in the near future.

Despite its traditional medicinal uses, the plant has an undervalued status in the scientific community. Till date, there is least focus on Pharmacological and Pharmaceutical studies and to approach a document to evaluate true clinical potential of the *Selaginella bryopteris* (*Sanjeevani/Agnijari*). Standardization & Quality evaluation of herbal drugs is quite important. This present study is an attempt to find out potential measures such as Pharmacognosy and Physicochemical, Phytochemical, Chromatographic study of *Selaginella bryopteris* (*Sanjeevani/Agnijari*) to make protocol of standardization and quality evaluation. As the standardization protocol of *Selaginella bryopteris* (*Sanjeevani/Agnijari*) is not mentioned in API and other *Ayurvedic Samhitas* the obtained data would be helpful to standardize the drug and for future studies.

The plant materials taken for the study was-
***Selaginella bryopteris* (L) Baker (*Sanjeevani/Agnijari*)
Family *Selaginellaceae***

MATERIALS AND METHODS**Collection of drug and authentication of sample**

- The genuine samples of *Selaginella bryopteris* L. was collected from the Kedarnath Dham trekking way near by Mandakini River, Rudraprayag District, Uttarakhand, at an altitude of 11755 feet above sea level, Herbarium were made and authenticated at BSI Dehradun.
- All the samples collected were studied macroscopically, organoleptically & microscopically to furnish identification tools of *Selaginella bryopteris* L. (*sanjeevani/agnijari*).
- Physicochemical study was done of *Selaginella bryopteris*(*sanjeevani/agnijari*).for quality & safety assurance.
- Phytochemical analysis of *Selaginella bryopteris*(*sanjeevani/agnijari*)was done to confirm the presence of different phytochemicals which contribute to the physiological properties.
- The TLC was done of *Selaginella bryopteris*(*sanjeevani/agnijari*), the R_f value of the sample is also a identification tool which helps in isolation of compounds.

RESULTS AND DISCUSSION**Pharmacognostical study**

A. Macroscopic study: The macroscopic characters of *Selaginella bryopteris* are discussed below in the tabular form

Features	<i>Selaginella bryopteris</i> L. (SANJEEVANI/AGNIJARI)
Habitat	Xerophytic heterophyllous herbs of Himalayas
Size of Plant	6-25 cm, differentiated in to nodes and internodes, erect or suberect
Leaf	Isomorphic on the main stem, green, distant, up 3 x 2 mm
Stem	Creeping, Stolonerous .
Rhizophores	leafless, prop like cylindrical, structure,originate from the stem at the point of branching
Root	Adventitious in nature and dichotomously branched
Sporophylls	uniform, ovate, acuminate, cuspidate, entire to minutely denticulate.

B. Organoleptic study

Different Organoleptic features of *Selaginella bryopteris* is tabulated below

C. Microscopic study

Parts	Colour	Odour	Touch	RASA	Shape	Size
Root of <i>S.bryoptris</i>	Dark Brown	Odorless	Coarse	Madhur	Tapering, covered with leaf remnants	1-4 cm
Stem of <i>S.b</i>	Brown	Odorless	Coarse	Madhur	erect or suberect	6-25 cm
Leaf of <i>S.b</i>	Green	Odorless	Coarse	Madhur	ovate-lanceotale, denticulate	3x2 mm

a) Transverse section

The anatomy of the mature stem is very distinct and is differentiated into an outer epidermis, middle cortex and centrally located stele. The outer cell walls of the epidermis are cutinised. It is devoid of stomata and hairs. In many species there are several layers of thick-walled cells (hypodermis) beneath the epidermis, which merge gradually with thin-walled chlorophyllous cells of the cortex.

A T.S. of a root shows a very simple arrangement with a centrally located protostele covered by parenchymatous cortex and bounded externally by cuticularised epidermis. In some species, the outer layers of cortex become sclerenchymatous and form hypodermis. The stele consists of a small, exarch and monarch xylem.

b) Powder microscopy of *selaginella bryopteris*

Starch, Tracheids, Tracheids reticulate vessels, Cork cells, Fibre, Calcium Oxalates crystal, Pitted vessels, Parenchymatous cells, Leaf Fragment, Fragment of xylem, Fragments of endocarp are present.

Physicochemical study

Test	PHYSICOCHEMICAL STUDY OF <i>Selaginella bryopteris</i> L. Baker (SANJEEVANI/AGNIJARI)
Foreign matter	0 %
Moisture Content	10.82 %
pH	5.9
Total Ash	5.80 %
Acid Insoluble Ash	4.08 %
Water Soluble Ash	14.45 %
Aqueous soluble Extract (%)	9.795%
Alcohol soluble Extract (%)	10.32%
Hydro alcohol soluble Extract (%)	14.83%
Ethyl acetate soluble Extract (%)	4.07%
Acetone soluble Extract (%)	4.43%
n-hexane soluble Extract (%)	1.97%
Chloroform soluble Extract (%)	3.09%

Phytochemical study**1. Test for carbohydrate**

- Benedict test showed the presence of carbohydrate in aqueous and hydro alcoholic extract of *Selaginella bryopteris*.
- Fehling test showed the presence of carbohydrate in Hydro alcoholic extract of *Selaginella bryopteris* (Sanjeevani / Agnijari)
- Molisch test and Barfoed's test is negative in all three extracts.

2. Test for alkaloids

- Mayer's test was positive in alcoholic and hydro alcoholic extract of *Selaginella bryopteris*.
- Dragendorff test showed the presence of alkaloid in aqueous, alcoholic, Hydro alcoholic and chloroform extract *Selaginella bryopteris*.
- Wagner's test showed the presence of alkaloid in alcoholic and hydro alcoholic extract of *Selaginella bryopteris*.
- Hager's test was also positive in alcoholic and hydro alcoholic extract of *Selaginella bryopteris*.

3. Amino acid analysis

- Ninhydrine test was negative in all extracts of *Selaginella bryopteris*.

4. Protein analysis

- Biuret test showed the presence of protein in alcoholic extract of *Selaginella bryopteris*.
- Xanthoprotic test showed the presence of protein in alcoholic extract of *Selaginella bryopteris*.
- Millon test also showed the presence of protein in alcoholic extract of *Selaginella bryopteris*.

5. Test for saponin

- Foam test showed the presence of saponin in aqueous, alcoholic hydro alcoholic, acetone, n-hexane and chloroform extract of *Selaginella bryopteris*.

6. Glycoside analysis

- Killer-killian test showed the presence of glycoside in n-hexane and chloroform extract of *Selaginella bryopteris*.
- Cumerine glycosides test the presence of glycoside in aqueous, alcoholic hydro alcoholic, acetone, and chloroform extract of *Selaginella bryopteris*.
- Borntrager's test showed the presence of glycosides aqueous, alcoholic hydro alcoholic *Selaginella bryopteris*.

7. Phenolic compound analysis

- Phenolic test is positive only in alcoholic & hydro alcoholic extract of *Selaginella bryopteris*.

8. Steroid analysis

- Salkowski reaction showed the presence of steroids in alcoholic and chloroform extracts of *Selaginella bryopteris*.

9. Tannin analysis

- FeCl₃ test showed the presence of tannin in alcoholic and hydro alcoholic extract of *Selaginella bryopteris*.
- Lead acetate test showed the presence of tannin in aqueous, alcoholic & hydro alcoholic extracts of *Selaginella bryopteris*.
- Potassium dichromate test showed the presence of tannin in aqueous, alcoholic & hydro alcoholic extracts of *Selaginella bryopteris*.

10. Flavonoid analysis

- Shinoid's test showed the presence of flavanoid in aqueous & hydro alcoholic extracts of *Selaginella bryopteris*.

Chromatographic study

TLC: establish phytochemical fingerprint profiling in drug for identity.

The alcoholic extracts of test samples was separated in mobile solution and visualized Iodine Vapour/conc.

Sulphuric Acid. which showed 6 spots having R_f values- 0.12, 0.15, 0.21, 0.27, 0.52, 0.76 ,

- The macroscopic study, microscopic study, R_f value of *Selaginella bryopteris* L. (Sanjeevani /Agnijari) was done to present identification parameters which is an essential tool for standardization.
- Physicochemical study of *Selaginella bryopteris* L. (Sanjeevani/Agnijari) was done to present parameters for quality, purity & safety assurance of the crude drug & the amount of active constituents in different types of extracts.
- **Phytochemical** (Qualitative Analysis) Phytochemical Qualitative analysis of *Selaginella bryopteris* L. (Sanjeevani /Agnijari) was done to present the parameters for presence of phytoconstituents in plant extracts which are known to exhibit medicinal as well as physiological activities.
- **Phytochemical** (Quantitative Analysis) Flavonoid, terpenoids, steroids, Alkaloids are secondary metabolites present in plants which are responsible for different actions on biological system.

Conclusion & future recomondation

- The Pharmacognostical studies help in the correct identification of the crude drugs which is an essential prerequisite. In the current investigation, it can be inferred that extractive values in hydro-alcohol and alcohol solvent as compared to water and other solvents was better.
- This implies that hydro-alcohol and alcohol are more effective solvents for extraction. Present study concludes that physicochemical and preliminary phytochemical investigation may be used for quality control, identification and to differentiate from other closely related species. Since scientific validation is essential for the acceptability of herbal drugs and it provides valuable information for further investigations.
- The above work was a step towards the intent of developing tools for standardization of *Selaginella bryopteris*. More research must be carried out to develop complete standard protocol of plant.
- It is suggested that further work should be carried out to isolate, purify & characterize the active constituents responsible for the activity of the plant.
- Clinical study should be taken up to establish antihepatotoxic and antimicrobial activities of *Selaginella bryopteris* drug in human model.
- *Selaginella bryopteris* is an endangered medicinal plant so further work must be done on the propagation & cultivation of endangered medicinal plant to promote conservation of endangered medicinal herbs.
- A comparative Pharmacognostical study of different species of *Selaginella* from different regions of India should be done.
- *Selaginella bryopteris* has been reported to have the highest degrees of drought resistance. It is observed that this fern can withstand years of drought very effectively, and still there is no adverse effect in its vigour. This is because of a drought resistant gene. There are also the possibilities of its use as a viable genetic tool to be engineered into drought tolerant plant for combating global warming and climate changes phenomenon.

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