



**PHYSICOCHEMICAL, PHYTOCHEMICAL & CHROMATOGRAPHY STUDY OF
DIFFERENT PARTS OF *BRAHMKAMAL* (*SAUSSUREA OBVALLATA* (DC.) EDGEW.) –
THE STATE FLOWER OF UTTARAKHAND**

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ABSTRACT

Brahmkamal {*Saussurea obvallata* (DC). Edgew.} is State Flower of Uttarakhand and also known as King of Himalayan flowers. The plant is distributed at an altitudinal range of 3000-4800 m. It is a very useful plant in prospective of traditional ayurvedic medicines, largely used by the natives residing in Himalayas. The plant has an undervalued status in the scientific community. *Brahmkamal* an ethnoreligious medicinal herb of Himalaya is selected for the present study to standardize the medicinal herb. A very limited work was done before, may be due to its difficult collection. Collection of plant is too difficult due to its presence in the alpine range of Himalaya at an altitude of 3000-4800 m only in period of mid monsoon this is also a reason for its ignored status in scientific community. Standardization of medicinal herb is quiet important. This present study is an attempt to find out potential measures such as Physicochemical, Phytochemical, Chromatography study of *Brahmkamal* to make protocol of standardization and quality evaluation. As the standardization protocol of *Brahmkamal* (*Saussurea obvallata*) is not mentioned in API, the obtained data would be helpful to standardize the drug for next study.

KEYWORDS: *Brahmkamal*, Physicochemical, Phytochemical, chromatography.

INTRODUCTION

Brahmkamal, the mysterious herb of Himalaya, botanically named as *Saussurea obvallata* (DC). Edgew. It belongs to Asteraceae family. In Hindu mythology *Brahmkamal* is believed to have divine powers.

Brahmkamal is highly valuable plant, beside its religious value, the plant is extensively harvested by local people for preparation of *traditional Ayurvedic medicines*. It has therapeutic potential to treat many diseases. Local people use it to treat many diseases in many forms.

Taxonomic Profile^[1,2]

Taxonomic profile of *Saussurea obvallata* is tabulated below [Table No. 1.1]

Kingdom	Plantae
Phylum	Tracheophyta
Class	Magnoliopsida
Order	Asterales
Family	Asteraceae
Tribe	Cynareae
Genus	<i>Saussurea</i>
Species	<i>Saussurea obvallata</i>
Binomial name	<i>Saussurea obvallata</i> (DC.) Edgew.
Synonyms	<i>Aplotaxis obvallata</i> DC. <i>Theodorea obvallata</i> (DC.) Kuntze

Botanical Description: Perennial herb; stems erect, well developed, simple; basal leaves obovate, elongated toothed, glabrous, petiolate; cauline leaves oblong; heads solitary or up to 6, large, hemispheric, glabrous, enclosed

by boat-shaped, papery, pale-yellow bracts; flowers purplish-white to pale brown; outer pappus bristles scabrid or absent.^[3]

Distribution^[4]: World: Himalayas.

India: Himalayas from Kashmir to Sikkim

Habitat: Along streams in sub-alpine fir and alpine scrub habitats between 3,500 to 4,000 meters elevation.^[5]

Propagation: It is propagated by seeds or rhizome cuttings.^[6]

Flowering & Fruiting: July to September.^[7,8]

Seed collection is from September – October.^[9]

Part Used

- Whole Plant, Root & Flower.^[10]
- Whole plant and Rhizome.^[11]
- Root – Rhizome.^[12]

MATERIAL AND METHODS

Collection: Genuine samples of *Brahmkamal* (*Saussurea obvallata*) and its different parts viz. Rhizome, Stem, Leaf, Flower were collected from Hemkund Sahib, Chamoli district, Uttarakhand (around 285 km from

Rishikesh), at an altitude of 15200 feet above sea level. (Fig 1.1).

Date of Collection: 09/09/2016

Authentication: A herbarium of *Brahmkamal* was prepared (fig.2.1) and authenticated at Botanical Survey of India, Dehradun.(fig. 2.2).

Physicochemical Study: Physicochemical parameters like foreign matter, moisture content, pH value, Ash value, Extractive values were recorded for different samples.

Phytochemical Study: Freshly prepared extracts were tested for the presence of various active phytochemicals like carbohydrates, alkaloids, amino acids, proteins, glycosides, phenolic compounds, saponins, flavonoids, tannins etc.

Chromatographic Study: Thin layer Chromatography (TLC) was performed and R_f values were calculated.

OBSERVATION AND RESULT

Physicochemical Study [Table No. 1.2]

Test	PHYSICOCHEMICAL STUDY OF ALL SAMPLES			
	Rhizome of <i>S.o</i>	Stem of <i>S.o</i>	Leaf of <i>S.o</i>	Flower of <i>S.o</i>
Foreign matter	0%	0%	0%	0%
Moisture Content	7.881%	5.85%	11.782%	12.3 %
pH	6.5	4.9	7.4	5.8
Total Ash	11.605%	7.87%	6.74%	6.72%
Acid Insoluble Ash	1.72%	3.45%	0%	2.54%
Water Soluble Ash	2.96 %	4.12%	3.99%	4.37%
Aqueous Extractive Value	2.658%	19.48%	3.82%	7.53%
Alcohol Extractive Value	6.73%	15.83%	2.16%	4.75%
Petroleum Ether Extractive	1.19%	2.19%	1.28%	2.74%

Phytochemical Study

1. Carbohydrate Test For Different Samples:- [Table No. 1.3]

Sample	Name of Test	Aqueous extract	Alcohol extract
Rhizome of <i>S.o</i>	Molisch test	+ve	+ve
	Benedict test	-ve	+ve
	Barfoed's test	-ve	-ve
	Fehling test	-ve	-ve
Stem of <i>S.o</i>	Molisch test	-ve	-ve
	Benedict test	+ve	+ve
	Barfoed's test	-ve	+ve
	Fehling test	-ve	+ve
Leaf of <i>S.o</i>	Molisch test	+ve	+ve
	Benedict test	+ve	+ve
	Barfoed's test	+ve	-ve
	Fehling test	-ve	-ve
Flower of <i>S.o</i>	Molisch test	+ve	+ve
	Benedict test	-ve	+ve
	Barfoed's test	-ve	-ve
	Fehling test	+ve	-ve

2. Alkaloid Analysis In Different Samples:- [Table No. 1.4]

Sample	Name of Test	Aqueous extract	Alcohol extract
Rhizome of <i>S.o</i>	Dragendorff test	-ve	-ve
	Wagner's test	-ve	-ve
	Hager's test	-ve	-ve
Stem of <i>S.o</i>	Dragendorff test	-ve	-ve
	Wagner's test	-ve	-ve
	Hager's test	-ve	-ve
Leaf of <i>S.o</i>	Dragendorff test	-ve	-ve
	Wagner's test	-ve	-ve
	Hager's test	-ve	-ve
Flower of <i>S.o</i>	Dragendorff test	+ve	-ve
	Wagner's test	-ve	-ve
	Hager's test	-ve	+ve

3. Amino Acid Analysis In Different Samples:- [Table No. 1.5]

Samples	Name of the Test	Aqueous extract	Alcohol extract
Rhizome of <i>S.o</i>	Ninhydrine test	-ve	+ve
Stem of <i>S.o</i>		+ve	+ve
Leaf of <i>S.o</i>		-ve	+ve
Flower of <i>S.o</i>		+ve	+ve

4. Analysis of Proteins In Different Samples:- [Table No. 1.6]

Samples	Name of the Test	Aqueous extract	Alcohol extract
Rhizome of <i>S.o</i>	Biuret test	-ve	+ve
	Xanthoprotic test	+ve	+ve
	Millon's test	-ve	-ve
Stem of <i>S.o</i>	Biuret test	+ve	-ve
	Xanthoprotic test	-ve	+ve
	Millon's test	+ve	-ve
Leaf of <i>S.o</i>	Biuret test	-ve	+ve
	Xanthoprotic test	+ve	-ve
	Millon's test	-ve	+ve
Flower of <i>S.o</i>	Biuret test	-ve	+ve
	Xanthoprotic test	+ve	+ve
	Millon's test	-ve	-ve

5. Analysis of Saponin In Different Samples:- [Table No. 1.7]

Samples	Name of the Test	Aqueous extract	Alcohol extract
Rhizome of <i>S.o</i>	Foam test	+ve	-ve
Stem of <i>S.o</i>		+ve	-ve
Leaf of <i>S.o</i>		-ve	+ve
Flower of <i>S.o</i>		+ve	-ve

6. Analysis of Glycosides In Different Samples:- [Table No. 1.8]

Samples	Name of the Test	Aqueous extract	Alcohol extract
Rhizome of <i>S.o</i>	Borntrager's test	-ve	-ve
Stem of <i>S.o</i>		-ve	-ve
Leaf of <i>S.o</i>		-ve	-ve
Flower of <i>S.o</i>		+ve	-ve

7. Analysis of Phenolic Compounds In Different Samples:- [Table No. 1.9]

Samples	Name of the Test	Aqueous extract	Alcohol extract
Rhizome of <i>S.o</i>	Phenolic test	-ve	-ve
Stem of <i>S.o</i>		+ve	+ve
Leaf of <i>S.o</i>		-ve	-ve
Flower of <i>S.o</i>		-ve	-ve

8. Analysis of Steroids In Different Samples:- [Table No. 1.10]

Samples	Name of the Test	Aqueous extract	Alcohol extract
Rhizome of <i>S.o</i>	Salkowski reaction	-ve	-ve
Stem of <i>S.o</i>		-ve	-ve
Leaf of <i>S.o</i>		-ve	-ve
Flower of <i>S.o</i>		-ve	-ve

9. Analysis of Tannin In Different Samples:- [Table No. 1.11]

Sample	Name of Test	Aqueous extract	Alcohol extract
Rhizome of <i>S.o</i>	FeCl ₃ test	-ve	-ve
	Lead acetate test	+ve	+ve
	Potassium dichromate test	-ve	-ve
Stem of <i>S.o</i>	FeCl ₃ test	+ve	+ve
	Lead acetate test	+ve	+ve
	Potassium dichromate test	+ve	+ve
Leaf of <i>S.o</i>	FeCl ₃ test	-ve	-ve
	Lead acetate test	+ve	+ve
	Potassium dichromate test	-ve	-ve
Flower of <i>S.o</i>	FeCl ₃ test	+ve	-ve
	Lead acetate test	+ve	+ve
	Potassium dichromate test	-ve	-ve

10. Analysis of Flavonoid In Different Samples:- [Table No. 1.12]

Samples	Name of the Test	Aqueous extract	Alcohol extract
Rhizome of <i>S.o</i>	Shinods test	+ve	+ve
Stem of <i>S.o</i>		-ve	+ve
Leaf of <i>S.o</i>		-ve	+ve
Flower of <i>S.o</i>		+ve	-ve

11. Analysis of Aflatoxin In Different Samples:- [Table No. 1.13]

Test for Aflatoxin	Rhizome of <i>S.o</i>	Stem of <i>S.o</i>	Leaf of <i>S.o</i>	Flower of <i>S.o</i>	Possible limit
Aflatoxin B1	Absent	Absent	Absent	Absent	0.5ppm
Aflatoxin B2	Absent	Absent	Absent	Absent	0.1ppm
Aflatoxin G1	Absent	Absent	Absent	Absent	0.5ppm
Aflatoxin G2	Absent	Absent	Absent	Absent	0.1ppm

12. Analysis of Microbial Limit In Different Samples:- [Table No. 1.14]

Test for Microbial limit	Rhizome of <i>S.o</i>	Stem of <i>S.o</i>	Leaf of <i>S.o</i>	Flower of <i>S.o</i>	Possible limit
Total Aerobic Microbial count	10 ⁴ /g	10 ³ /g	10 ² /g	10 ⁴ /g	10 ⁵ /g
Total yeast & Mould count	10 ³ /g	10 ¹ /g	10 ¹ /g	10 ² /g	10 ³ /g

E) Chromatographic Study

A. TLC (Thin Layer Chromatography) analysis for different samples: (fig.3.1)

Mobile solution

- Toluene: Ethyl acetate: Formic acid (6:3:1 ml) for rhizome & stem sample.
- Toluene: Ethyl acetate: Formic acid (6:3.5:0.5 ml) for flower sample.

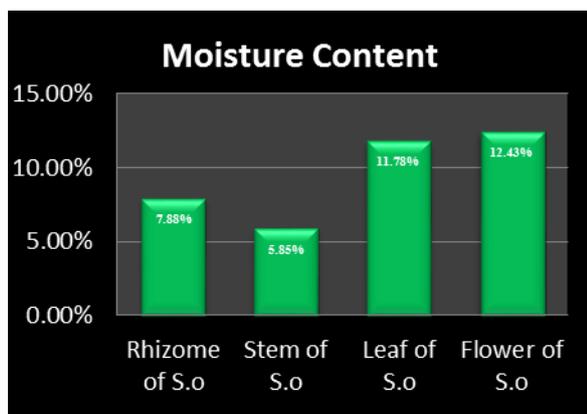
- Toluene: Ethyl acetate: Formic acid (7:2:1 ml) for leaf sample.

Visualization

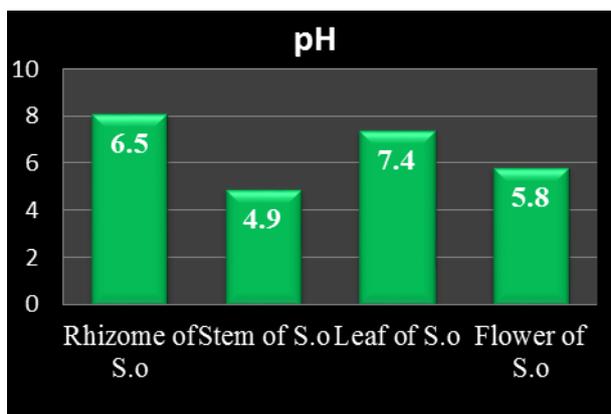
- Ultra Violet Flor for rhizome sample.
- Iodine Vapour for leaf sample.
- p-Anisaldehyde Sulphuric Acid for flower & stem sample.

TLC Profile of samples on Silica Gel G60F254 [Table No. 1.15]

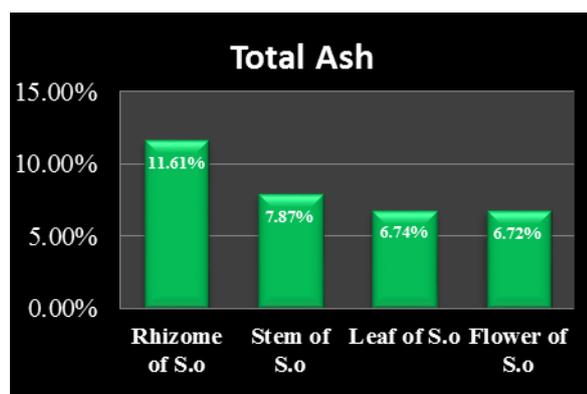
Samples	No. Of Spots	R _f Value
Rhizome of <i>S.o</i>	4	0.78, 0.83, 0.87, 0.92
Stem of <i>S.o</i>	4	0.24, 0.45, 0.67, 0.71
Leaf of <i>S.o</i>	5	0.26, 0.56, 0.67, 0.71, 0.89
Flower of <i>S.o</i>	6	0.12, 0.19, 0.52, 0.56, 0.72, 0.96



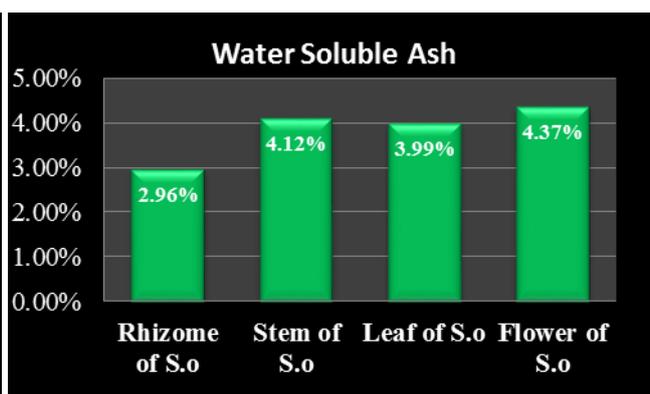
Graph 1.1



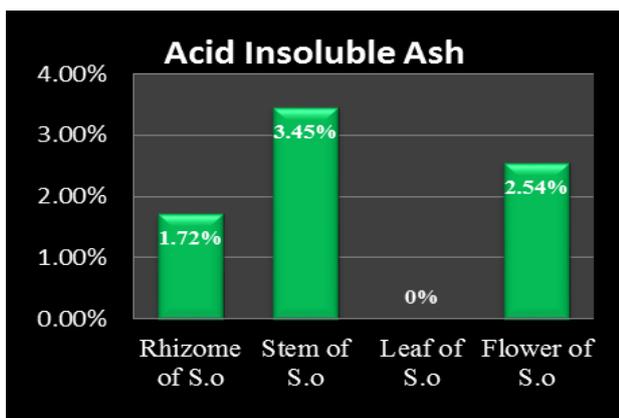
Graph 1.2



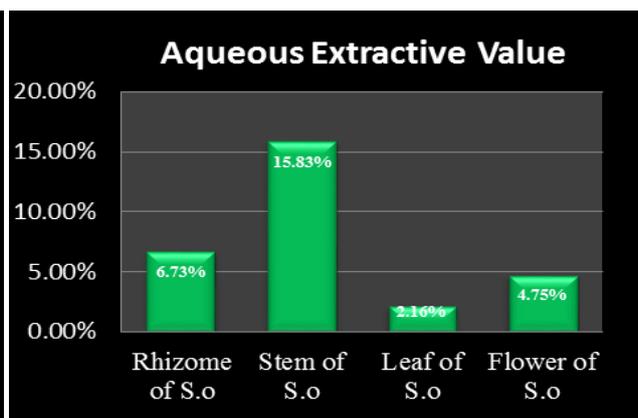
Graph 1.3



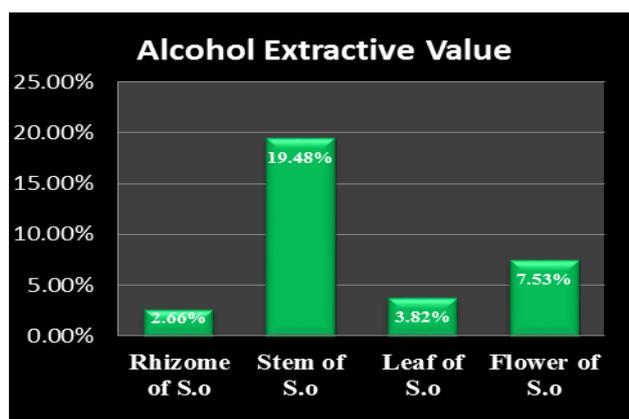
Graph 1.4



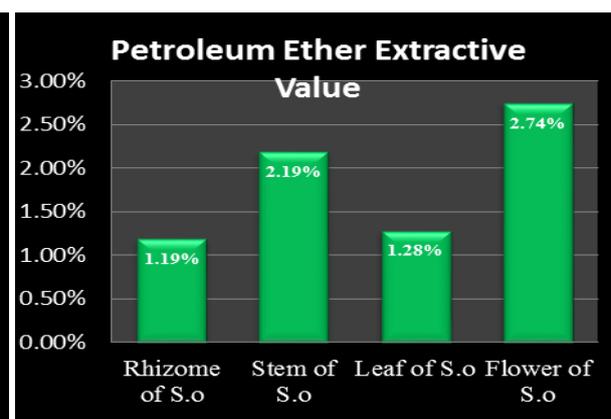
Graph 1.5



Graph 1.6



Graph 1.7



Graph 1.8

FIGURES



Fig. 1.1 – Collection of *Brahmkamal*

Fig. 2.1- Herbarium of *S. Obvallata*

Fig. 2.2- Authentication certificate

Samples	Rhizome	Stem	Leaf	Flower
TLC Plates				
R _f Value	0.78, 0.83, 0.87, 0.92	0.24, 0.45, 0.67, 0.71	0.26, 0.56, 0.67, 0.71, 0.89	0.12, 0.19, 0.52, 0.56, 0.72, 0.96

Fig. 3.1- Images of TLC plates.

DISCUSSION

The present study is a step towards the standardization of sacred medicinal herb. The standardization of *Brahmkamal* is not mentioned in API, this present study would help in future studies of *Brahmkamal*. This study include the collection of plant sample from alpine Himalayas followed by authentication of botanical identity *Saussurea obvallata* (DC.) Edgew. The Physicochemical study, Phytochemical study and Chromatography study of different parts of *Brahmkamal* provide measures for identification and quality evaluation.

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