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## COMPARATIVE EVALUATION OF PHYTO-CHEMICAL STUDY OF SHIGRUPATRA SWARASA & ARKA

**KHOSE AA<sup>1</sup>, BIRADAR AB<sup>2\*</sup> AND HIREMATH RS<sup>3</sup>**

**1:** Final year PG Scholar, Department of Shalakyatantra, KAHER's Shri B M K Ayurveda Mahavidyalaya, KLE Academy of Higher Education and Research, Deemed-to-be-university, Belagavi – 590 003, Karnataka, India

**2:** Professor and Guide, Department of Shalakyatantra, KAHER's Shri B M K Ayurveda Mahavidyalaya, KLE Academy of Higher Education and Research, Deemed-to-be-university, Belagavi – 590 003, Karnataka, India

**3:** Professor and HOD, Department of Rasashastra and Bhaishajya Kalpana, KAHER's Shri B M K Ayurveda Mahavidyalaya, KLE Academy of Higher Education and Research, Deemed-to-be-university, Belagavi – 590 003, Karnataka, India

**\*Corresponding Author: Dr. A. B. Biradar: E Mail: [arunkumar.biradar@gmail.com](mailto:arunkumar.biradar@gmail.com)**

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### ABSTRACT

**Background:** Herbs have been used by all civilizations as a source of medicines since ages. In *Ayurveda*, *Bhaishajya Kalpana* deals with most potent form of formulations with their therapeutic utility. *Arka* & *Swarasa* are two different forms of extraction of active principles. *Shigru* (*Moringa Oleifera*) is widely used in various commercial ophthalmic preparations as a major ingredient. It's various forms like *Swarasa*, *Arka*, *Lepa*, *Rasakriya* are being advocated in treatment of many *Netraroga*. Hence, this drug was chosen for this investigative study.

**Aim:** To compare phyto-chemical study of *Shigrupatra Swarasa* and *Arka*

**Objective:** To study *Shigrupatra Swarasa* on analytical and pharmacological parameters

To study *Shigrupatra Arka* on analytical and pharmacological parameters

**Methods:** Fresh *Shigru Patra* were collected and subjected for authentication. *Shigrupatra Swarasa* & *Arka* were prepared as per classical guidelines. Then subjected for Organoleptic,

physicochemical, microbiological, and phytochemical analysis as per Standard Operative Procedure of CRF.

**Results & Discussion:** A comparative analytical study of both formulations were conducted. Significant variations were found in the phytochemical components, specific gravity, and p<sup>H</sup>. The microbiological and sterility requirements were met by both formulations.

**Conclusion:** This study contributes to understand the phyto-chemical differences in two different forms of a single drug. (i.e., *Shigrupatra Swarasa & Arka*).

**Keywords:** *Shigrupatra, Arka Kalpana, Swarasa Kalpana, pharmaceutical analysis, phytochemical screening, Microbial study, Organoleptic study*

## INTRODUCTION

Moringa is the only genus, found in the Moringaceae family of flowering plants [1]. *Bhaishajya Kalpana* (i.e., pharmaceutical science of *Ayurveda*) emphasize on development and use of herbal formulations customized for particular medical situations [2]. *Arka Kalpana* (distillate preparations) [3] and *Swarasa Kalpana* (fresh juice preparations) are widely used in various clinical condition. Both the preparations were prepared using reference of *Sharangadhara Samhita* [4, 5].

*Shigrupatra* (*Moringa Oleifera*) is known for its rich nutritional profile and therapeutic qualities, has been often cited in variety of illnesses including *Netra Rogas* (eye problems), also referred as *Chakshushya* [6]. *Sushruta Samhita* [7] and *Bhaishajya Ratnawali* [8] emphasizes more on use of *Shigru* in various formulations concerned with eyes diseases. It has *Tikshna Guna* (penetrative nature), *Katu Rasa* (pungent taste), and *Ushna Virya* (hot potency) make it good at lowering the

*Kapha* and *Vata doshas*, which are frequently linked to eye diseases [9].

*Sushruta Uttartantra* claims that, various eye diseases caused by an imbalance in *Dosha, Dhatu, and Mala* [10]. These diseases include such as *Abhishyanda, Arma, Timira, Akshipaka*, etc which includes inflammatory disorders, degenerative disorders, conjunctivitis, refractive errors, vitreo-retinal, neuro-ophthalmic diseases, etc.

*Shigrupatra Arka* is prepared out of aqueous distillation process which makes make it sterile, useful for ailments like allergies, infections, redness, and irritation. It has cooling, antibacterial, and calming qualities [11].

*Shigrupatra Swarasa*, is made from fresh drumstick leaves, high in phytochemicals including flavonoids, phenolics, and vitamins that have anti-inflammatory and antioxidant properties, crucial for controlling inflammation and oxidative stress [12].

This study was planned to evaluate & compare phyto-chemical properties of both formulations based on modern analytical tools.

## MATERIALS & METHOD

### Standard Operating Procedure (SOP) for *Shirupatra Arka* Preparation

#### Ingredients:

- *Shigrupatra* (fresh leaves): 80 g (**Figure 1**)
- Distilled water: 800 ml
- Equipment: Aqueous distillation apparatus, sterile eye drops bottles

#### Procurement and Authentication:

A licensed AYUSH drug testing lab verified the ingredients, which were obtained from the institution's herbal garden. Microbial load tests, TLC, and first phytochemical analysis were among the quality evaluations.

#### Sterilization Protocol:

Drug were prepared in teaching pharmacy of *Rasashastra Bhaishajya Kalpana* of our institution. Each empty eye drop bottle were packed in individual triple layer air tight polythene zipper pack and subjected for Gamma radiation with 25 kGy strength of radiation in a certified sterilization unit.

All the glass equipment's of distillation apparatus are cleaned thoroughly and then used for drug preparation. 3 Beakers used for collection of prepared drugs, aluminium foil paper and dispenser / pipette for filling the eye drop bottles were

wrapped in a cotton cloth and subjected for autoclave in standard steam chamber for 45 minutes. Sterilisation of the above equipment's verified with sterilization indicator strips placed over the wrapped cloth and sterilization bean. Once the autoclave bean is brought to normal temperature aluminium foil is wrapped over the mouth / opening of prepared drug collection utensil without leaving gap. This procedure is preferably performed in sterilised ophthalmic operation theatre / aseptic zone under all aseptic precautions.

#### Distillation procedure – (**Figure 2**)

Once the things are ready then final drug is prepared under a septic condition. Both the ingredients are placed in boiling chamber in 1:10 proportion (i.e,80gms of raw drug and 800ml of distilled water). Once the boiling chamber start giving the steam, it is passed through a condensation chamber. Condensed steam is collected in sterilization collection beaker.

Fraction Collection (**Figure 3**) – Finished product is categorised in 3 segments. Initial 20% (i.e,60ml) of condensed finished product, middle 60% (i.e.,480ml) of condensed finished product and last remaining 20% (i.e.,160ml) of the finished product. Initial 160ml of finished product is collected in first beaker. Middle 480ml of finished product is collected in second beaker. Last 160ml of finished product is collected in third beaker.

As a thumb rule initial and last beaker finished product are discarded and middle 60% of finished used in therapeutic usage.

### Finished product packaging, labelling and sterilization

Once the desired quantity of drug is obtained, it is taken to sterile ophthalmic operation theatre or sterile zone (Figure 4). Under all aseptic precautions finished product is filled in eye drop bottles. Proper labelling is done as per the standard protocol and guidelines (Figure 5).

Then medicine filled each eye drop bottles were packed in individual triple layered polythene air tight zipper packs. Packed finished product is again sent for

sterilization with Gamma radiation with 25 kGy strength of radiation in a certified sterilization unit.

Once the finished product is received from sterilization, then subjected for quality control and then submitted to MRC for conducting clinical trial.

### Quality control checking of finished product –

Finished product were subjected for repeated MLT, Total fungal count, culture and other analytical testing to ascertain sterility and asepsis. Finished product were subjected for above testing prior to drug submission in MRC for clinical trial.

## RESULTS

### Analytical study:

Table 1: Test for specified Micro-organisms (Qualitative)

<i>Shigrupatra swarasa</i>			<i>Shigrupatra Arka</i>		
	LIMITS (As per IP)	Results		LIMITS (As per IP)	Results
<i>E coli</i>	Absent/100ml	Absent	<i>E coli</i>	Absent/100ml	Absent
<i>S aureus</i>	Absent/100ml	Absent	<i>S aureus</i>	Absent/100ml	Absent
<i>P aeruginosa</i>	Absent/100ml	Absent	<i>P aeruginosa</i>	Absent/100ml	Absent
<i>S abony</i>	Absent/100ml	Absent	<i>S abony</i>	Absent/100ml	Absent

Table 2: Microbial limit test (Quantitative)

<i>Shigrupatra swarasa</i>			<i>Shigrupatra Arka</i>		
	LIMITS (As per IP)	Results		LIMITS (As per IP)	Results
Total Bacterial Count	30-300 cfu/ml	No growth	Total Bacterial Count	30-300 cfu/ml	No growth
Total Fungal count	10-100 cfu/ml	10 cfu/ml	Total Fungal count	10-100 cfu/ml	No growth

Table 3: Organoleptic Characters

<i>Shigrupatra swarasa</i>		<i>Shigrupatra Arka</i>	
Tests	Results	Tests	Results
Form	Liquid	Form	Leaves
Colour	Greenish	Colour	Greenish-grey to pale green
Odour	Characteristics	Odour	Not distinct

Table 4: Physicochemical Standards

<i>Shigrupatra swarasa</i>		<i>Shigrupatra Arka</i>	
Tests	Results	Tests	Results
Specific Gravity	1.002	Specific gravity	0.999%
p <sup>H</sup>	5.27	pH Value	4.65
Total solids	2.676%	Volatile content	Absent

**Table 5: Preliminary Phytochemical Screening**

<i>Shigrupatra swarasa</i>		<i>Shigrupatra Arka</i>	
Tests	Results	Tests	Results
Test for Carbohydrates	Positive	Test for Carbohydrates	Positive
Test for Reducing sugar	Positive	Test for Reducing sugar	Negative
Test for Monosaccharides	Positive	Test for Monosaccharides	Negative
Test for Pentose sugar	Negative	Test for Pentose sugar	Negative
Test for Hexose sugar	Negative	Test for Hexose sugar	Negative
Test for Non reducing sugar	Negative	Test for Non reducing sugar	Negative
Test for Proteins	Positive	Test for Proteins	Negative
Test for Amino acids	Positive	Test for Amino acids	Negative
Test for Steroids	Negative	Test for Steroids	Negative
Test for Flavonoids	Positive	Test for Flavonoids	Negative
Test for Alkaloids	Negative	Test for Alkaloids	Negative
Test for Tannins	Positive	Test for Tannins	Negative

**Table 6: Test for Glycosides**

<i>Shigrupatra swarasa</i>		<i>Shigrupatra Arka</i>	
A. Cardiac Glycoside	Positive	A. Cardiac Glycoside	Negative
B. Anthraquinone glycosides	Negative	B. Anthraquinone glycosides	Negative
C. Saponin glycosides	Positive	C. Saponin glycosides	Positive

**Table 7: TLC (Alcohol Extract) Mobile phase- Toluene: Ethyl acetate**

Ratio: 7:3	Rf Values		
	Short wave	Long wave	Day Light
	0.18, 0.35, 0.68, 0.74, 0.82, 0.89, 0.92, 0.96	0.16, 0.35, 0.47, 0.68, 0.71, 0.83, 0.88, 0.91, 0.98	0.30, 0.35, 0.67, 0.71, 0.88, 0.94, 0.97



**Figure 1: Collected Raw drug Shigru patra**



**Figure 2: Aqueous distillation of Shigru patra**



**Figure 3: Collected Shigru patra Arka**



**Figure 4: Sterile filling of Shigru patra Arka**



Figure 5: Shigru patra Arka eye drop bottles

## DISCUSSION

All pharmaceutical analytical investigation were done and results recorded for *Shigrupatra Swarasa & Arka*.

*Shigrupatra Swarasa* (sample 1) has an acidic environment with an average  $p^H$  of 5.27 with specific gravity of 1.002. It has considered sterile because of below approved microbial limits. As a result, the prepared *Swarasa* was suitable for *Aschyotana*.

*Shigrupatra Arka* (sample 1) has an acidic environment with an average of  $p^H$  of 4.65 with specific gravity of 0.999. It has considered sterile because of below approved microbial limits. As a result, the prepared *Swarasa* was suitable for *Aschyotana*.

The alcoholic extract of sample 1 contained proteins, amino acids, flavonoids, tannins, cardiac glycosides, reducing sugars, carbohydrates, monosaccharides, and saponin glycosides, according to a phytochemical examination. Using a toluene-ethyl acetate (7:3) solvent mixture, the TLC examination of sample 1 (alcoholic

extract) on a silica gel plate revealed 8 spots under short wave UV light, 9 spots under long wave UV light, and 7 spots under daylight.

The alcoholic extract of sample 2 was not subjected to TLC analysis because of the limitations of aqueous solutions in Thin Layer Chromatography. Water's polarity and other problems with bioactive chemicals' solubility could lead to inadequate separation or inefficient spotting on the TLC plate.

Phytochemical examination of aqueous extract of sample 2 revealed the presence of saponin glycosides and carbohydrates. Saponin glycosides and carbohydrates were present in both the alcoholic and aqueous extracts. Potential anti-cancerous activity is suggested by the presence of carbohydrates in the water extracts of both samples [13]. Flavonoids, which have antiviral, antiallergic, antiplatelet, anti-inflammatory, antitumor, and antioxidant qualities, were more prevalent in sample 1 than in sample 2 [14].

Both samples include saponins, which have antibacterial properties. Saponins have anti-inflammatory, anti-cancer, antioxidant, and weight-loss effects. They are also utilized in intracellular histochemistry labelling to allow antibodies to access intracellular proteins [14]. Testing on physical characteristics like colour and odour as well as physicochemical elements like sterility, microbiological limits, and qualitative testing for particular microorganisms were part of the analytical investigation of *Shigrupatra Arka*. These metrics are essential for proving the drug's efficacy and safety.

#### CONCLUSION

The *Swarasa Kalpana* has been described primarily in classical texts, including *Sharangadhara Samhita*, while *Arka Kalpana* are mainly mentioned in later period of ayurvedic literature like *Arka Prakasha*. This study successfully analysed and standardized *Shigrupatra Arka* and *Swarasa* which suggesting their potential use in *Netrarogas* formulations; differences in  $p^H$ , specific gravity, and phytoconstituents suggest different therapeutic applications for each and both preparations were found suitable for clinical evaluation.

However, *Swarasa* has limitation of fresh herbs, clean equipment for preparation, portability and sterility issues. Whereas *Arka* is sterile, storable form,

portability and ready to use with good shelf-life.

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