Pharmaceutico-Analytical Study Of Tulasi (*Ocimum sanctum* Linn.) Arka (Hydro-distillate)

Tulasi is well known drug for respiratory disorders and it contain 0.4% volatile oil. Arka Kalpana (Hydro-distillation) is one of important method of extraction and it has more potency. Tulasi arka is used for shwasa, kasa and raktapitta. This study has revealed the organoleptic charcters, physicochemical analysis and GC-MS compositional analysis for use as tool for standardization of the Tulasi Arka.

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Short Communication

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ABSTRACT

Introduction: *Tulasi (Ocimum sanctum* Linn.) of Lamiaceae known as Holy Basillt is commonly available and cultivated across the India and it is having many therapeutic usages and *Arka*isan important dosage form because it has good patient compliance, long shelf life and increased potency. *TulasiArka* is one such *Arka* used for *Shwasa* (Asthma), *Kasa* (Cough) and *Hikka* (Hicough). ***Methods:*** *Tulasiarka* was prepared using leaves of *O.sanctum* as per the standard procedure. The prepared *Arka* was subjected to organoleptic, physiochemical and GCMS study. ***Results:*** Analysis of prepared *Arka* was carried out as per the protocols laid down by Ministry of AYUSH, Govt of India for *Arka*. 14 chemical compounds were detected in *TulasiArka* by GCMS Analysis. ***Conclusion:*** Organoleptic characters, physiochemical analysis, and GC-MS values can be used as supplement tool for standardization of the *TulasiArka*.

KEYWORDS Ayurvedic hydro-distillates, *TulasiArka*, GCMS.

PICTORIAL ABSTRACT

*T*ulasi (*Ocimum sanctum* Linn.) is perennial herb with an aromatic smell; known as holy basil. It is commonly available and cultivated across the India and it is having many therapeutic usages. Traditionally, the fresh fruit and leaf juice are commonly used in the treatment of cough, Upper respiratory tract infections, and worm infestations[1]. It is *Katu* (Pungent), *Tikta* (Bitter) *rasa, Laghu* (Light), *Ruksha* (Rough), *Guna, Usna* (Hot) *veery* and *KatuVipaka*. It has *Kaphavatashamaka*. It is useful in the disease such as *Hikka, Agnimandya, Kustha,Krimi, Aruchi, Chardi, Jwara, Mukhadourgandhya* and *Visavikara*, and also in *Parsvasula, Daha, Raktavikaraha*. It has extensive folklore use in *Shwasa, Kasa* and *Jwara* etc[2].

Ayurvedic pharmaceutics are dealt under the heading of *BhaishajyaKalpana*, which basically depends upon the *PanchavidhaKasayaKalpana*[3]. *ArkaKalpana* is described as one of the *Panchavidhakasayakalpna* in *Arkaprakasha*[4]. *Arka* is a liquid preparation obtained by distillation of certain liquids or of drugs soaked in water using the *Arkayantra* or any convenient distillation apparatus[5]. The *Arka* dosage form is one of the most sought after dosage forms, due to its reduced dose, increased patient compliance. *TulasiArka* is one such *Arka* used for *Shwasa* (Asthma), *Kasa* (Cough), *Raktapitta* (Bleeding disorders)[6]. In the present study an attempt has been made to standardize the method of *TulasiArka* and characterize the organoleptic, Physiochemical parameters. In addition, the components in the distillate have been tried to identify using GCMS.

1. Plant materials

The fresh *Tulasi* (*Ocimumsanctum*Linn.) leaves was collected from the Herbal Garden of Shri Dharmashtala Manjunatheshwara College of Ayurveda and Hospital, Hassan, Karnataka and authentication of the *Tulasi* leaf was done at department of Dravyaguna, Sri Dharmasthala Manjunatheswara College of Ayurveda and Hospital, Hassan based on macroscopic and organoleptic characters. The preparation of *TulasiArka* was

done at Department of *Rasashastra* and *Bhaishajyakalpana*, Sri Dharmasthala Manjunatheswara College of Ayurveda and Hospital, Hassan as per the reference of general method of preparation of *Arka.*

2. Preparation of Arka

In this study, It was prepared as per one of the ratio of *Arka* (distillate) is 1:2 ratio. For 50gms of raw *Tulasi* Leaves 2 times water was added and this mixture (drug and water) was continuously heated in the *ArkaYantra* (distillation apparatus) till 60% of distillate was collected. The vapour condensed and collected in a receiver. In the beginning the vapour consists of only steam and may not contain the essential principles of the drugs. So it was discarded. After cooling, the collected *Arka* (distillate) is preserved in air tight container.

3. Analytical Study

Arka was analyzed with organoleptic parameters like colour, taste, odour, state and physico-chemical characters like pH, specific gravity, refractive index, viscosity and volatile matter and Gcms analysis of *Arka.*

4. Physico-chemical parameters

The samples were analyzed for the organoleptic parameters like colour, taste, odour, state and physical parameters like pH, Specific gravity, and Refractive index by following the method prescribed in Ayurvedic Pharmacopeia of India (API) (Table 1).

Table 1. Standardization parameters of *TulasiArka*

Parameter	Results
Colour	Colourless
Odour	Aromatic
Taste	Pungent
State	Liquid
pH	5.6
Refractive index	1.33
Specific gravity	0.99
Viscosity	0.01
Volatile matter	0.04

5. GCMS Analysis

Gas Chromatography – Mass Spectrometry (GCMS) is the analysis method of choice for separation of Volatile substances or the volatile dervaties of certain non- volatile substances[7]. The identification of unknown compounds is achieved by comparison of their full mass spectrum with a mass spectral library or database. The Main limitation of GC-MS is, the compounds must be sufficiently volatile to allow transfer from the solid phase to the mobile carrier gas and thus elute from the analytical column to the detector. Large numbers of compound are still too polar or too large in size to be analyzed with this technique.

Instrument – GC-MS-5975C [AGILENT]. Column Oven Temperature – 70º C; Injector Temperature -250º C; Injection Mode – Split; Split Ratio – 10; Flow Control Mode – Linear Velocity; Column Flow -1.5ml /min; Carrier Gas – Helium 99.9995% purity; Injection volume-1microlitre; COLUMN –DB-5ms Agilent; Length -30.0m; Diameter – 0.25mm, Film Thickness – 0.25um.

Column oven temperature program -

Rate	Temperature (ºC)	Hold Time (min)
-	70.0	3.0
10	300	9.0 [35.0 mts total]

Ion source temp – 230º C; Interface temp – 240º C; Scan range- 40- 700m/z; Solvent cut time- 3mins; MS start time -3(min); MS end time -35 (min); Ionization- EI (-70ev); Scan speed – 2000;MS LIBRARY NIST -11

GCMS Spectrum profile of *TulasiArka* confirmed the presence of fourteen chemical compounds. All the parameters including name, retention time, and peak area were ascertained from this comparison. The major compound which accorded for 28.36% was unidentified along with 3 other minor constituents. Other major constituents identified were benzene 1,2,3-trimethyl (23.72%) and styrene (10.03%) (Table 2).

Table 2. GCMS Identified compounds of *TulasiArka.*

Peak	Retention Time(RT)	Area %	Name of the compound
1	3.402	10.03	Styrene
2	3.734	1.56	--
3	3.763	28.36	--
4	3.858	4.34	--
5	3.935	23.72	Benzene 1,2,3-trimethyl
6	4.077	3.83	Benzene,1-ethyl-4-methyl-
7	4.836	5.89	Isoborneol
8	5.969	4.06	Fumaric acid,2-2dichloroethyl tridecyl ester
9	6.413	6.00	Cycloheptasiloxane,tetradecamethyl-

10	6.793	1.10	3-phenyl-2H-chromene
11	7.676	4.14	1H-Imidazole-4-methanol
12	7.955	3.36	Benzoic acid,2,5bis(trimethylsiloxy)-,trimethylsilyl ester
13	8.275	1.13	--
14	10.131	2.46	Cyclohexane,1-1-(2-methyl-1,3propanediyl)bis-

The chemical composition of *TulasiArka* obtained in the present study is in agreement with the earlier findings reporting that Cyclohexane is found similar of volatile oil produced from the *O. sanctum*[8].

Tulasi is well known drug for respiratory disorders and it contain 0.4% volatile oil. *Arka kalpanā* is given specific importance and it has more potency and it can be used even upto one year. Gas Chromatography – Mass Spectrometry (GCMS) is the analysis method of choice for separation of Volatile substances. GCMS of *TulasiArka* identified 14 compounds. The analytical study gives a preliminary standard of *Tulasi Arka.*

CONFLICT OF INTEREST Nil

CONTRIBUTORS Dr Kavyashree contributed to the conceptualization of topic, intellectual study and design of the experiment. Dr Harini and Dr Prakash edited the manuscript, and made the write up suitable for publication. Dr KN Sunil Kumar planned and executed the study, interpreted the result and provided suitable suggestions.

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