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ORIGINAL RESEARCH ARTICLE-EXPERIMENTAL OPEN ACCESS

PHARMACEUTICO-ANALYTICAL STUDY ON *BRIHATMADHUSNUHIRASAYANA*

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Abstract

The classical textual compendium Sahasrayoga explains a unique formulation –*Brihatmadhusnuhirasayana*, under *Lehyakalpana*(lictus preparations).Madhusnihi- *Smialx china* being the prime drug of this recipe also possesses *Guggulu* and *Gandhaka* which are also appreciated well for their potent *kushthaghna (*skin disease pacifying medicaments*), vranaropaka*(wound healing) and *rasayana*(rejuvinative)action.The formulation is widely used even today while during the treatment of *arsha-bhagandara-vrana (*haemorrhoids-fistula-ulcers) etc.The formulation is a well known recipe in the cure of the deadly diseases like non healing ulcers and syphilis, chronic degenerative arthritis etc.The formulation was prepared as per the classical method of *lehyakalpana* by taking the ingredients as referred in classical mentioning. The pharmaceutico-analytical findings of the test drug appear valuable in determining the quality of the drug and hence may be used as an important tool to establish its effective molecules in selective diseases too.

Key words: Brihatmadshusnuhirasayana, Pharmaceutical study, Analytical study, Physico- chemical analysis

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INTRODUCTION

Lehyakalpanas[1] are well known dosage forms of Ayurvedic system of medicine. They are known well for their potent rejuvenative benefit, being in semisolid form of medicine. The recipes belonging to this kind of dosage from are appreciated well throughout and hence most of the pharmaceutical companies do prepare varieties of *lehya kalpanas*.

*Brihatmadhusnuhirasayana*is one such formulation sited in Sahasrayoga under Lehyakalpana[2].

The formulation contains 26 minor ingredients like *triphala, trikatu, trijataka, jeeraka, dhanyaka, trivrit*etc. and major three ingredients- *Shuddhagandhaka, Madhusnuhi* and *Shuddha guggulu*.

Ghee is the oleaginous substance included in this formulation and sugar candy is the sweetening substance. Honey is mentioned as the augmenting agent referred as *prakshepaka dravya*.

Materials and Methods

The formulation *Brihatmadhusnuhirasayana* was prepared in three batches by keeping each ingredient as constants in all the batches. The method of preparation of *Lehya*, as per Acharya Sharngadhara(3) is taken as the standard classical reference for the study. The formulation was prepared in the laboratory associated with Post graduate department of Rasashastra and bhaishajya kalpana, Alva's Ayurveda Medical college, Vidyagiri ,Moodabidire, Karnataka during 23/3/2017 and 21/8/2017.

The ingredients used and their quantity are shown in **Table No.1.**

Table No1. Ingredients and their quantity

Sl no	**Drug**	**Practical I,II,II each**
I 1.	**Sweetening substance-**Sitopala(Sugar candy)	2.880 Kg
	Ashuddhaguggulu	1.500Kg
II 1.	**Medicinal substances-** ShuddhaGuggulu	960 gram
2.	Madhusnuhi/Chopachini	960 gram
	AshuddhaGandhaka	1.500 Kg
4.	ShuddhaGandhaka(Purified sulphur)	960 gram
5.	Ghrita(Cow's ghee)	2.880Kg+1.200 Kg = 4.080 Kg
6.	Madhu(Honey)	3.840 Kg

	Prakshepaka drugs:	
7-9	**Trikatu-** Shunthi Pippali Maricha	90 g
10-12	**Triphala-** Hareetaki Vibheetaki Amalaki	90 g
13-15	**Trijataka-** Twak Ela Patra	90 g
16	Jatiphala(nutmeg)	30 g
17	Patra	30 g
18	Chitraka (Shuddha)	30 g
19	Lavanga	30 g
20	Dhanyaka	30 g
21	Godhuma(Wheat)	30 g
22	Yava(Barley)	30 g
23	Ajamoda	30 g
24	Vidanga	30 g
25	Chavya	30 g
26	Kushtha	30 g
27	Trivrit	30 g
28	Pippalimoola	30 g
29	Ashwagandha	30 g
30	Bharngi	30 g
31	Jyotishmati	30 g
32	Nagakesara	30 g
33	Cow's milk	6.000 L+9.000L = 15 Litre

General standard operative procedures of lehya kalpana are adopted in all the study samples.

The prepared samples were subjected for physico- chemical analysis and instrumental analysis in accredited Pharmaceutico-

analytical laboratories-Biotechnology department of Alva's College, Moodabidire, Karnataka, Nagarjuna herbal concentrates, Thodupuza, Kerala and Vaidyaratnam Oushhadhashala , Private limited, Thrissur, Kerala.

Observation

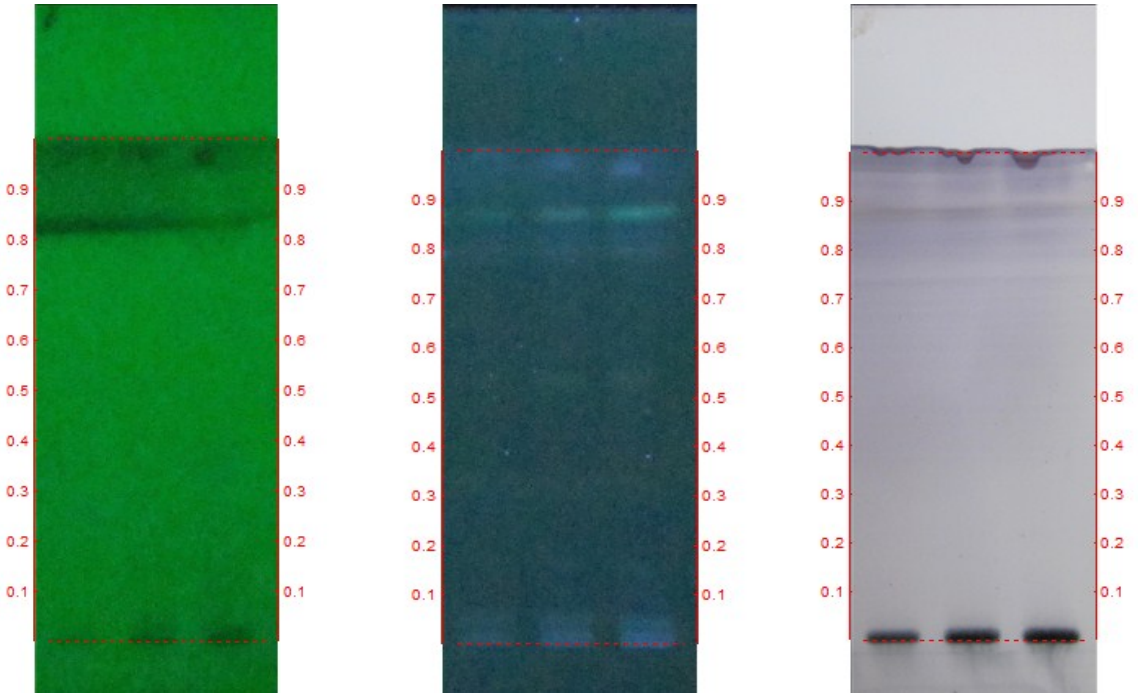
The average(n=3) Loss on drying at105°C is reported to be 10.124% w/w, Total solids-84.877% w/w,Total ash-1.008%w/w,Acid insoluble ash 0.199%w/w,Water soluble ash 1.133%w/w,Alcohol soluble extractive value 25.899%w/w,Water soluble extractive value 74.225%w/w, Reducing sugar 17.849%w/w, Total sugar 54.592%w/w and pH 5.25.

Photo-documentation of TLC reports and corresponding Rf values are summarized in the **Figures No 1. 2 and 3 respectively.**

The Table 2 denotes R_f values of the sample *Brihatmadhusnuhirasayana.*

Table No.3 represents the Results of the functional groups as per Chemical Analysis of three different batches of *Brihatmadhusnuhirasayana*

Table No.4 presents the Results of the Phytochemical Analysis of three different batches of *Brihatmadhusnuhirasayana*



At 254 nm **At 366nm** **After Post Derivatisation**

Figure 1. TLC photo documentation of *n*-Butanol extract of *Brihatmadhusnuhirasayana*

Track 1- *Brihatmadhusnuhirasayana*– 3 µl

Track 2– *Brihatmadhusnuhirasayana* – 6 µl

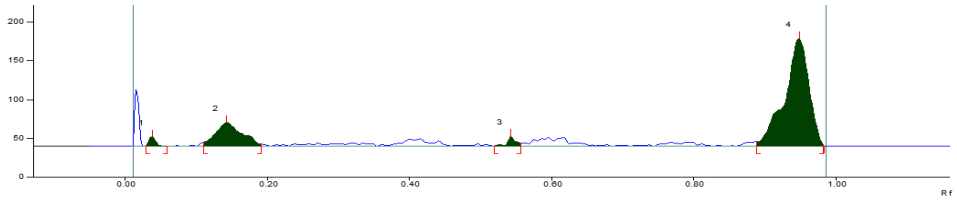
Track 3– *Brihatmadhusnuhirasayana* – 9 µl

Solvent system: Toluene: Ethyl Acetate:Acetic Acid: Water (3:2:0.8:0.1)

Table 2: R_f values of the sample *Brihatmadhusnuhirasayana*

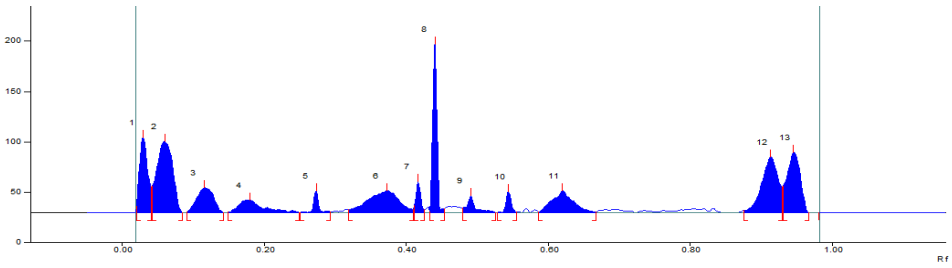
At 254 nm	At 366 nm	After Post Derivatisation
-	0.05(F L Violet)	-
-	0.17(F L Green)	-
-	0.34(F L Green)	-
-	0.55(F L Green)	-
-	0.71(F L Green)	-
-	0.80(F L Green)	-
-	-	0.84(L Violet)
-	0.88(F Green)	0.88(L Violet)

***L-Light, F-Fluorescence**



Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.03 Rf	1.1 AU	0.04 Rf	12.1 AU	6.28 %	0.06 Rf	0.0 AU	82.5 AU	1.88 %
2	0.11 Rf	5.2 AU	0.14 Rf	30.2 AU	15.69 %	0.19 Rf	2.4 AU	812.7 AU	18.56 %
3	0.52 Rf	0.2 AU	0.54 Rf	12.5 AU	6.49 %	0.56 Rf	3.5 AU	104.7 AU	2.39 %
4	0.89 Rf	5.6 AU	0.95 Rf	137.8 AU	71.55 %	0.98 Rf	1.4 AU	3378.7 AU	77.16 %

Figure 2. Densitometric Scan of the sample *Brihatmadhusnuhirasayana*



Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.02 Rf	6.4 AU	0.03 Rf	74.7 AU	12.45 %	0.04 Rf	25.8 AU	631.3 AU	9.59 %
2	0.04 Rf	26.7 AU	0.06 Rf	70.6 AU	11.78 %	0.09 Rf	0.2 AU	1125.3 AU	17.09 %
3	0.09 Rf	0.4 AU	0.12 Rf	24.8 AU	4.13 %	0.14 Rf	0.0 AU	422.4 AU	6.42 %
4	0.15 Rf	0.5 AU	0.18 Rf	12.6 AU	2.10 %	0.25 Rf	0.2 AU	313.2 AU	4.76 %
5	0.25 Rf	0.3 AU	0.27 Rf	21.4 AU	3.58 %	0.29 Rf	0.7 AU	106.8 AU	1.62 %
6	0.32 Rf	3.0 AU	0.37 Rf	22.0 AU	3.67 %	0.41 Rf	3.2 AU	685.1 AU	10.40 %
7	0.41 Rf	7.4 AU	0.42 Rf	30.8 AU	5.14 %	0.43 Rf	2.8 AU	141.9 AU	2.15 %
8	0.43 Rf	0.8 AU	0.44 Rf	167.3 AU	27.91 %	0.45 Rf	4.1 AU	718.1 AU	10.90 %
9	0.48 Rf	4.9 AU	0.49 Rf	17.2 AU	2.88 %	0.53 Rf	0.3 AU	142.7 AU	2.17 %
10	0.53 Rf	0.3 AU	0.55 Rf	20.9 AU	3.49 %	0.56 Rf	0.8 AU	110.3 AU	1.68 %
11	0.59 Rf	0.9 AU	0.62 Rf	22.1 AU	3.69 %	0.67 Rf	0.8 AU	464.3 AU	7.05 %
12	0.88 Rf	2.2 AU	0.91 Rf	55.6 AU	9.27 %	0.93 Rf	25.7 AU	919.5 AU	13.96 %
13	0.93 Rf	26.2 AU	0.95 Rf	59.6 AU	9.93 %	0.97 Rf	0.1 AU	804.2 AU	12.21 %

Fig 3.a At 254nm

Table No.3: The Results of the Functional groups as per chemical Analysis of three different batches of *Brihatmadhusnuhirasayana*

Sl No	Name of theTests	Results		
		Sample A	**Sample B**	**Sample C**
1	***Proteins***			
	Biuret test	Present	Present	Present
	Millon's test	Present	Present	Present
2	**Carbohydrate test**			
	Benedict's test (Reducing sugar)	Present	Present	Present
	Benedict's test (Non-reducing sugar)	**Present**	Present	Present
3	**Tannins**	Present	Present	Present
4	**Saponin**			
	Foam test	Absent	Absent	Absent
5	**Flavanoids**			
	Flavanoid test	Present	Present	Present
6	**Phenols**			
	Phenol test	Absent	Absent	Absent
7	**Steroids**			
	Salkowski's test	Absent	Absent	Absent
8	**Alkaloids**			
	Mayer's test	Absent	Absent	Absent
9	Tritrepenoides	Absent	Absent	Absent
10	Starch	Present	Present	Present

Table No.4: The Results of the Phytochemical Analysis of three different batches of *Brihatmadhusnuhirasayana*

	Sample 1	Sample 2	Sample 3
Refractive index	1.45844	1.43844	1.44844
Specific gravity	0.91533	0.90823	0.89643
Viscosity	40 minutes/50 ml	38 minutes/50 ml	39 minutes/50 ml
Saponification value	147.26mg/ml	148.36mg/ml	146.56mg/ml
Iodine value	5.153g/ml	5.213g/ml	5.123g/ml
Acid value	0.871g/ml	0.761g/ml	0.791g/ml
Ester value	136.465g/ml	138.455g/ml	137.64g/ml

Unsaponifiable matter	0.6426g	0.6936g	0.6756g
Total fatty matter	39.357m Eq/L	38.646m Eq/L	39.3974 Eq/L
Free fatty acid	19.257 m Eq/L	19.658 m Eq/L	19.452 m Eq/L

DISCUSSION

The classical *lehya* formulation *Brihatmadhusnuhirasayana* is having wider scope for its utility in ayurvedic therapeutics due to its appropriate consistency and good prospective of therapeutic claim. In this background when the formulation was prepared and analyzed physic chemically, the product was found with its stable mode of presentation and appearance by all the physical criteria like color, consistency, texture, appearance etc.

When all the three successful samples were prepared by using the standard criterion, good quality outcomes were possible to arrive at. The physico-chemical values were appear within the normal range of values as per the guidelines of Ayurvedic Formulary of India[4].

The analytical test reports for various functional groups of all the three study samples have shown uniform pattern of the constituents for their presence and absence. This helps to validate the study samples for their operative procedures as well as finished product qualitative assement (Table No.3). Iodine value and Acid values of all the three study samples were within the narrow range of the prepared samples further denote the standard parameters of the operative protocol of the preparation *Brihatmadhusnuhi rasayana* (Table No.4.).Meanwhile the test samples were found enough scope of stability and hence longer shelf life too.

CONCLUSION

The physico-chemical values like Loss on drying at105°C - 10.124% w/w, Total solids-84.877% w/w,Total ash-1.008%w/w,Acid insoluble ash 0.199%w/w,Water soluble ash 1.133%w/w,Alcohol soluble extractive value 25.899%w/w,Water soluble extractive value 74.225%w/w, Reducing sugar 17.849%w/w, Total sugar 54.592%w/w and pH 5.25 can be used as standardization markers for the test drug-BrihartMadhusnuhirasayana.

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