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The Pharmaceutico-Analytical Study of *Shatavari Ghrita*

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ABSTRACT

Background

In Pharmaceutical preparation of Ayurveda the *Snehakalpana* was mentioned since *samhita* period and has its own importance. S*amskaraanuvarthitwa* produces the *Ghritakalpana* superior in *Sneha Kalpana. Shatavari Ghrita*is one of the formulations mentioned for the treatment of *Amlapitta*. The ingredients of *Shatavari Ghrita* are *Murchita Ghrita, Shatavari Mula Swarasa, Shatavari Mula Kalka, Go-Dugdha* and *Jala*. *Murchita Ghrita* includes commonly available six drugs that are *Haritaki, Vibhitaki Amalaki, Haridra, Musta* and *Nimbu*

Aim

To carry out the pharmaceutical preparation of *Shatavari Ghrita* and to analyze the prepared *Shatavari Ghrita*

Materials and Methods

Shatavari Ghrita is a unique formulation which comes under *snehakalpana* prepared by the general method of *sneha* preparation. It is indicated in *Amlapitta*

Results

Pharmaceutical study showed that there is no pharmaceutical error in preparation of *Shatavari Ghrita.* Analytical parameters could generate preliminary standards of *Shatavari Ghrita* as per the protocol of testing. GCMS technique helps to make an account of active components present in a given sample

KEYWORDS

Snehakalpana, Shatavari Ghrita, Pharmaceutical Study, Analytical Study

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INTRODUCTION

*Kalpana*is defined as the processes through which the *Dravya*is converted into a suitable prescription form. The form thus obtained is called as *Bhaishajya*. The *Dravya* which overcomes the fear of the disease is called as *Bhaishajya*. *Bhaishajya Kalpana Vijana*is the study of the art of preparing and standardizing of the medicine, which are intended for therapeutic administration. The basic *Kalpana*[1] were *Swarasa*, *Kalka*, *Kwatha*, *Hima*and *Phanta*. In due course of time many new concepts and formulations were formulated.

*Ghrita*is one such *Kalpana*, which is a considerate pharmaceutical procedure in Ayurvedic pharmacies to obtain semi solid oleaginous dosage form used in different diseases for systemic or topical application. By subjecting *Ghrita* to a particular heat pattern with *Kalka* (paste) and *Drava*(any liquid medium, whether it could be juice, decoction, cold or hot infusion, milk etc.) in prescribed formula[2]

Shatavari Ghrita is mentioned in *Bhaishajya Ratnavali*[3] in *Amlapitta Chikitsa Prakarana* and also mentioned in *Chakradatta*, which can be used in the treatment of *Amlapitta* owing easy availability of ingredient since ancient time. The taste, smell, shelf life and acceptability of the *Shatavari Ghrita* has made to the patient. In the present study, the pharmaceutical study of *Shatavari Ghrita* along with analytical study was assessed

MATERIALS AND METHODS

Pharmaceutical study

The pharmaceutical study deals with the whole process of preparation of medicine, beginning from collection of drugs to obtaining the final product. It is divided into the following sections

A. Collection of the drug

B. Authentification of the raw drugs

C. Murchana of Go-Ghrita

D. Preparation of *Shatavari Ghrita*

A. Collection of the drug –

The raw drugs required for the preparation of medicine were procured from Teaching Pharmacy S.D.M.C.A.H. Hassan on 06 – 05 – 2016. And *Nimbu*, *ShatavariMula*were collected from the area of Thanneruhalla, Hassan, Karnataka.

B. Authentification of raw drugs –

The Authentication of all the raw drugs was done at the Department of *Dravyaguna*, Sri Dharmasthala Manjunatheswara College of Ayurveda and Hospital, Hassan.

C. Preparation of Murchanaof Go –Ghrita[4]

Ingredients-

1. *Haritaki*- 25 gms 2. *Bibhitaki*- 25 gms 3. *Amalaki*- 25gms 4. *Musta*- 25gms

5. *Haridra*- 25gms 6. *NimbuSwarasa*- QS (80 ml) 7. *Ghrita*- 600 ml

8. *Jala*- 2400 ml

Preparation of Kalka –

Haritaki, Bibhitaki, Amalaki, Musta and *Haridra* were taken 25 g each and made coarsely powdered in *Khalva Yantra*and converted in fine powder with the help of mixer grinder. The particles of fine powder were of the sieve size number (80 -100), 80 ml of *Nimbu Swarasa* was taken for making the Kalka into a bolus form. Total quantity of *Kalka* taken was 187.5gms

Process of Ghrita Paka–

● A stainless steel vessel was kept over LPG stove and mild flame was maintained during the process. Plain uncooked *Ghrita* (600ml) was taken in the vessel and for *Drava Dravya* 2400 ml *Jala was* mixed to the *Ghrita*. *Kalka was* added to the mixture. The proportion (*Kalka: Sneha: Drava Dravya*) was 1/3.2: 1: 4. The whole mixture was stirred till it becomes homogenous. The mixture was bolied until all *Sneha Siddhi Lakshana*s appeared. The total time was taken for *Paka* of *Ghrita* 5 hrs and 10 mins. The prepared *Ghrita*was filtered through a clean cloth, obtained quantity was 570ml. *Ghrita* was packed in an air tight glass bottle after cooling. The obtained *Ghrita* was used to prepare *Shatavari Ghrita.*

OBSERVATION

Changes observed in the *GhritaMurchana*as seen in Table 1

Table 1 Changes observed in the *GhritaMurchana*

Time	Temperature (^{0}C)	Changes observed	Color
11.55 AM	40^0 C	Initially *Ghrita*smell	Yellow
12.00 PM	90^{0}C	Added Water	Yellow
12.05 PM	60^{0}C	Added *Kalka*	Brownish yellow
12.20 PM	80^{0}C	Smell of a *Ghrita*	-do-
12.35 PM	90^{0}C	Smell of a *Ghrita*	-do-
12.50 PM	90^{0}C	*Kalka* is mixed completely in *Drava*	-do-
1.05 PM	94^{0}C	Odor of *Ghrita*reduced	-do-
1.20 PM	94^{0}C	-do-	Brown
1.35 PM	94^{0}C	-do-	Dark Brown
1.50 PM	94^{0}C	Slight froth was observed	-do-
2.20 PM	94^{0}C	-do-	-do-
2.35 PM	94^{0}C	-do-	-do-
2.50 PM	94^{0}C	Disappears froth	-do-
3.05 PM	92^{0}C	-do-	Black
3.20 PM	93^{0}C	Separation of *Kalka* started	-do-
3.35 PM	93^{0}C	-do-	-do-
3.50 PM	93^{0}C	-do-	-do-
4.05 PM	90^{0}C	*Kalka* separated, disappears froth	-do-
4.20 PM	90^{0}C	Checked for *Varti*formation	Blackish Green
4.35 PM	90^{0}C	-do-	Colour of kalka- Black
4.50 PM	90^{0}C	Formation of *Varti*of *Kalka*	-do-
5.10 PM	90^{0}C	*Agni Pareeksha*positive &*Gandha Varna* of the *Ghrita*was appreciated	-do-

RESULT

The results of the study are given in Table 2

Table 2 Tabulation of pharmaceutical work for *GhritaMurchana*

Paka Day	*Ghrita*Taken	*Kalka*	Water Taken	Duration	Outcome	Difference	% of loss
1	600 ml	187 gms	2400 ml	5hr.45min	570 ml	30 ml	5%

D. Preparation of Shatavari Ghrita[3]

Ingredients –

(1)S*hatavariMulaSwarasa*- 500 ml

(2)*ShatavariMula Kalka* - 62.5 gms

(3)*Murchhita Go-Ghrita*- 500 ml

(4)*Go-Dugdha*- 2000 ml

(5)Jala. - 2000 ml

Method-

Day 1

The preparation was started at 03:25 pm. A stainless steel vessel was kept over LPG stove and mild flame was maintained during the process. *Murchita Ghrita*(500ml) was poured in the vessel. After liquefy of *Ghrita*, 500 ml *Shatavari Swarasa* was added. *Go-Dugdha* (2000ml) is mixed with the above mixture. 2000 ml water was poured in the mixture and 62.5g of *Kalka*is added with the mixture. The proportion was 1/8:1:4. The whole mixture was stirred till it gets homogeneity i.e. all the ingredients are well distributed throughout. The temperature maintained in this preparation was *Mandagni*. The whole process was done byContinuous stirring. Heating was stopped after 2 hour and 20 mins. Vessel was covered with a cloth.

Day 2

2nd day Paka was started at 08:42 am. The whole mixture was stirred until it turned homogenous i.e. all the ingredients are well distributed throughout. *Mandagni* was maintained during the whole process. The temperature changes were noted at every 15 min throughout theprocess. Heating was stopped at 4.37 pm after it attained *Sneha Siddhi Lakshana*.The total time taken for *Paka* of *Ghrita w*as 10 hrs and 15 mins. The *Shatavari Ghrita* was filtered through a clean cloth, Obtained quantity was 350ml. This *Ghrita*was used for Analytical and Experimental study.

OBSERVATION

Changes observed in the preparation *of Shatavari Ghrita* as seen in Table3

Table 3 Changes observed in the *Shatavari Ghrita* (Day-1)

Time	Temperature	Observation	Colour
3.30 pm	50^0C	Odour of *MurchhitaGhrita*	Golden Yellow
3.35 pm	60^0C	Added *ShatavariSwarasa*	-do-
3.38 pm	70^0C	Added *Go-Dugdha*	-do-
3.40 pm	70^0C	Added *Jala*	-do-

3.42 pm	60^{0}C	Added *Kalka*	-do-
3.50 pm	70^{0}C	Odour of *Ghrita*reduced	Yellow
4.05 pm	94^{0}C	Kalka is mixed properly in *Drava*	Whitish Yellow
4.20 pm	94^{0}C	Slight white creamy part observed	-do-
4.35 pm	94^{0}C	-do-	-do-
4.50 pm	94^{0}C	Boiling started	-do-
5.05 pm	94^{0}C	-do-	-do-
5.20 pm	94^{0}C	-do-	-do-

Table 4 Changes observed in the *Shatavari Ghrita* (Day-2)

Time	Temperature	Observation	Color
8.45 am	50^{0}C	white creamy part observed	Whitish yellow
9.00 am	60^{0}C	-do-	-do-
9.15 am	80^{0}C	-do-	-do-
9.30 am	94^{0}C	-do-	-do-
9.45 am	94^{0}C	Boiling started	-do-
10.00 am	94^{0}C	-do-	-do-
10.15 am	94^{0}C	-do-	-do-
10.30 am	94^{0}C	-do-	-do-
10.45 am	94^{0}C	White creamy part observed	-do-
11.00 am	94^{0}C	-do-	-do-
11.15 am	94^{0}C	-do-	-do-
11.30 am	94^{0}C	-do-	-do-
11.45 am	94^{0}C	Increasing consistency of the mixture	Brownish yellow
12.00 pm	94^{0}C	-do-	-do-
12.15 pm	92^{0}C	-do-	Brown
12.30 pm	94^{0}C	-do-	-do-
12.45 pm	94^{0}C	-do-	-do-
1.00 pm	93^{0}C	-do-	-do-
1.15 pm	95^{0}C	Slight Bubbles started appearing	-do-
1.30 pm	94^{0}C	-do-	-do-
1.45 pm	94^{0}C	Disappeared bubble	-do-
2.00 pm	92^{0}C	Turned in thicker consistency	-do-
2.15 pm	94^{0}C	-do-	-do-
2.30 pm	94^{0}C	Absorbing *Ghrita* by *kalka*	-do-
2.45 pm	94^{0}C	-do-	-do-
3.00 pm	94^{0}C	Absorbed *Ghrita*	Deep brown
3.15 pm	92^{0}C	Started Separation of *Ghrita*from *Kalka*	-do-
3.30 pm	92^{0}C	-do-	-do-
3.45 pm	92^{0}C	Kalka separated	-do-
4.00 pm	92^{0}C	-do-	-do-
4.15 pm	92^{0}C	Formation of *Varti*of *Kalka*	-do-
4.30 pm	92^{0}C	*Agni Pareeksha*positive &*GandhaVarna* of the *Ghrita*was appreciated	-do-

RESULTS

The results of the study are given in Table5

Table 5 Tabulation of pharmaceutical work for *Shatavari Ghrita*

Paka Day	*Shatavari Swarasa*	*Shatavari Mula Kalka*	*Ghrita* Taken	Milk & Water taken	Duration	Outc-ome	Diffe-rence	% of loss
2	500ml	62.5 gms	500 ml	2000 ml Each	10 hr. 10 min	350	150 ml	30%

ANALYTICAL STUDY

RESULT

Organoleptic findings

Organoleptic characteristics for various sensory characters like color, taste, odor was carefully noted down.(As seen in Table 6)

Table 6 Result of Organoleptic character

Parameters	*ShatavariGhritha*
Color	Yellow
Odor	Characteristic
Taste	Pungent

Pharmaceutical Evaluation

Physico-chemical parameters of *Shatavari Ghrita* like Refractive index, Specific gravity Rancidity, Acid value, Saponification value, Iodine value and Peroxide valuewere assessed. Details are being given in Table 7

Table 7 Results of physicochemical parameters of Shatavari Ghrita

Parameter	*Results n = 3 %w/w*
	Shatavari Ghrita
Refractive index	1.45817
Specific gravity	0.9426
Rancidity	Fat is not oxidized
Acid value	0.55
Saponification value	212.1085
Iodine value	25.58
Peroxide value	0

GCMS

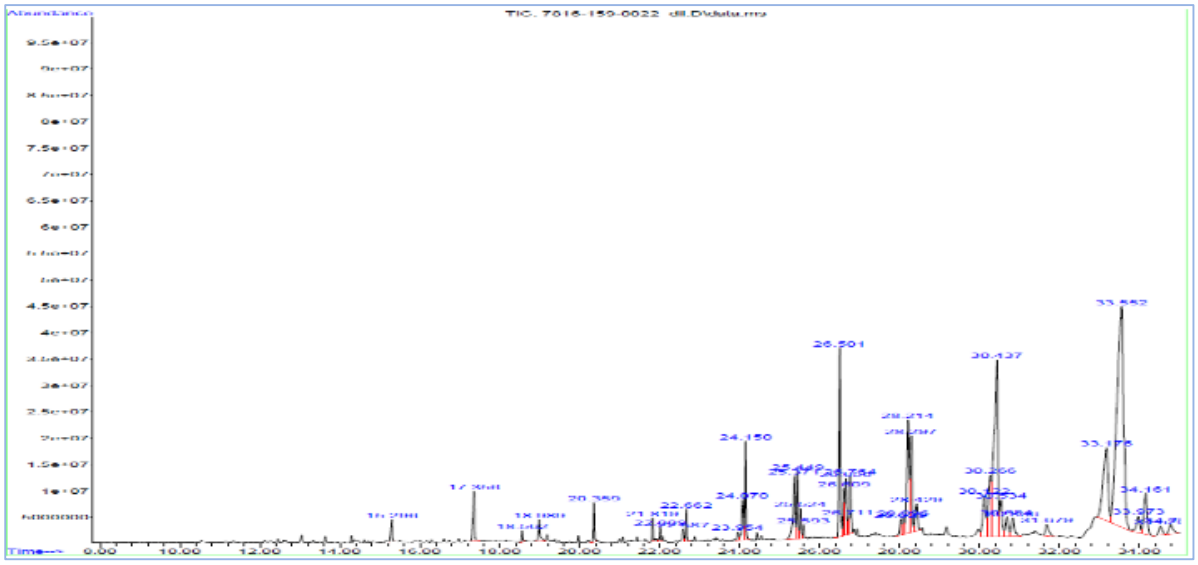
The details of Chromatogram of General Profiling of *Shatavari Ghrita* are discussed in Graph 1 and Table 8

Table 8- Showing the GCMS General Profiling contents present *Shatavari Ghrita*

PK	RT	Area	Name of the Compound	%
1	15.297	0.59	Tetradecanoic acid	99
2	17.354	1.59	n-Hexadecanoic acid	99
3	18.565	0.22	- -	42
4	18.989	0.70	Oleic acid	99
5	20.356	0.75	- -	41
6	21.819	0.46	9-Octadecenal	90
7	22.012	0.31	- -	64
8	22.584	0.44	- -	30
9	22.666	0.62	- -	14
10	23.951	0.20	- -	52
11	24.070	1.51	- -	14
12	24.152	2.17	- -	14
13	25.370	2.75	- -	27
14	25.452	1.68	- -	22
15	25.526	0.67	- -	22
16	25.593	0.27	- -	30
17	26.499	5.14	17-(1,5-Dimethylhexyl)-10,13-Dimethyl-2, 3, 4, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17-tetradecahydro-1H-cyclopenta [a] phenanthren-3-o1	99
18	26.610	1.20	- -	42
19	26.655	1.47	- -	38
20	26.714	0.32	- -	43
21	26.766	1.95	Napthalene -1-carboxamide, N- (4-methyl-1-piperazinyl)	90
22	28.029	0.60	- -	50
23	28.096	0.68	- -	41
24	28.215	5.04	- -	25
25	28.297	3.53	- -	38
26	28.430	1.28	- -	43
27	30.124	2.54	- -	38
28	30.265	3.59	- -	41

29	30.436	12.67	- -	38
30	30.533	1.90	- -	38
31	30.681	1.05	- -	25
32	30.837	1.09	- -	43
33	31.677	0.78	- -	47
34	33.177	7.10	- -	46
35	33.549	28.03	- -	15
36	33.972	1.08	- -	42
37	34.158	2.72	- -	38
38	34.522	0.62	- -	25
39	34.775	0.72	- -	25

'- -' Unidentified



Graph 1 Chromatogram of General Profiling of *Shatavari Ghrita*

DISCUSSION

Discussion on *Ghrita murchana*[4]

Even though the*Murchana* procedure is not mentioned in *Samhita Granthas*.Later *granthas* like *BhaishajyaRatnavali*, etc has specificinformation on the *Murchana* procedure. *Amadosha* may be considered asunwanted components in the raw *Ghrita*, like intermediate chemical constituents, dissolved gases, adulterants, plant toxins and moisture present in raw *Ghrita* or developed due to long time storage. *Dourgandha* may be caused due to the long term storage of the *Ghrita*, before the preparation it is ensured that only pure and potent *Ghrita* is taken for *Siddha Ghrita* preparation. Through the process of *Murchana* the capacity of the *Ghrita* to absorb the active components of the drug is increased. *Murchana* helps in maintaining the necessary ratio of unsaturated and saturated fats suitable for human physiology.

Discussion on Preparation of *Shatavari Ghrita*[3]

The reference of *Shatavari Ghrita* was taken from *ChakradattaAmlapittaChikitsa.* This reference was selected because the

method of preparation was easy compared to other references and ingredients were also less. Here the study was carried out with *Kalka Dravya*, *SnehaDravya* and *Drava Dravya*. The preparation was done with 2 days duration.Wide mouthed vessel was used for the preparation of the *Ghrita*. This was done for easy stirring and for proper evaporation. After *Ghrita* (500 ml), *ShatavariMulaSwarasa* (500 ml) was added, then followed by *Go-Dugdha* (2000 ml) and *Jala*(2000 ml) instead of *KalkaDravya* (62.5 gms). It was done to avoid the burning of *Kalka* and proper mixing with the *Sneha* and *Drava Dravya*. *Jala* and *Go-Dugdha* was added to extract the water and lipid soluble active principle . The consistency of *Ghrita* was reduced soon after the addition of *Drava Dravya* as the *Drava Dravy* was less viscous than *Ghrita*. Continuous stirring was done during the preparation so as to avoid charring of the *Kalka Dravya*. Temperature was maintained in *Mandagni* only to avoid the degradation of the phyto constituents of the drug. The reduction in the quantity and the increased consistency was due to the evaporation of the water content from the mixture. The duration of *Paka* was 2 days because the quantity of *Go-Dugdha* was more than *Swarasa.PhenaShanthi* was observed due to the generation of lower fatty acids Soon after attaining the *Paka*, vessel was taken out from the stove to avoid the risk of proceeding of *Paka*to further stage. Filtration was done when it was still in hot so as to attain maximum yield of *Ghrita* through squeezing. Volume of the *Ghrita* obtained was 350 ml. the loss was 150 ml, due to the 2 days *Paka* of *Ghrita* during preparation and due to the absorption of *Ghrita* by *Kalka*. The color of *Shatavari Ghrita* was golden yellow in color.

Discussion on Analytical Study

The analytical study was done to assess the standard parameters mentioned for the formulation as per guidelines of CCRAS.

A. *Organoleptic characters*

 Color – The *Ghrita color* was yellow. The color may be due to *Ghrita*

 Odor– Preparation was having characteristic strong smell

 Taste – The preparation were having pungent taste.

B. *Physico- chemical parameters*

1. *Refractive Index*[6]:

The Refractive index measurement can be used for qualitative and quantitative analysis as well as structural study. It is an inherent property of a substance. Hence it is used to determine the identity and purity of a chemical. It is also useful in controlling the analysis of commercial products and in identifying unknown substance. Also help to measure its consistency. So, it is an

important parameter for differentiating the *Snehas*.

The increase in refractive index value indicates the factors which are responsible for the refraction of light through *Ghrita* sample. Refractive index of sample is 1.45817

2.*Specific gravity*[7]*:*

The presence of dissolved substances in *Sneha*is expected to change its specific gravity. So it is considered to be an important parameter for analyzing medicated *Sneha*. This helps us to access the molecular information in a non-invasive way.

The data showed that the specific gravity of *Shatavari Ghrita* was 0.9426, which indicated active constituents present in it.

3. *Determination of Acid value*[9]*:*

The free fatty acids are responsible for the rancidity of the compound. Higher the free fatty acid more the rancidity of *Ghrita*. Decreased percentage of fatty acid or stable number of fatty acids decreases the rancidity of the compound. The edibility of a fat is inversely proportional to the acid number.

Acid value of sample is 0.55, Acid value signifies the presence of free acids and used to indicate the rancid state. Rancidity causes free acid liberation.

4.*Determination of Saponification value*[10]*:*

Medicated *Ghrita* with high saponification value has a better absorption. The molecular size can be determined by this method. It is inversely proportional to the molecular weight of fat. High saponification value indicates the presence of fatty acids of low molecular weight (molecules are in simple form). Low saponification value indicates that the molecules are in complex form. This value is high in fats containing a short chain fatty acids.

The amount of alkali needed to saponify a given amount of fat will depend upon the number of – COOH group present. Thus fats containing short chain fatty acids will take up more -COOH groups per gram than long chain fatty acids and this will take up more alkali and hence will have higher saponification number.

Saponification value of *Shatavari Ghrita* was 212.1085 With this it can be understood that *Shatavari Ghrita* has more stability.

5. *Iodine value*[11]*:*

The Iodine number is a measure of degree of unsaturation of fat. The more the Iodine number, more the unsaturated fatty acid bonds are present. When more Iodine is attached, higher the Iodine value and the more reactive, less stable and more susceptible to oxidation. The susceptibility to rancidity increase with Iodine value. A

high Iodine number indicates a high degree of unsaturation of the fatty acids in fat.

The Iodine value of *Shatavari Ghrita was 25.58*, indicating less chances of rancidity and otherwise also indicating more stability of *Shatavari Ghrita* .

7.*Rancidity*[8]*:*

Shatavari Ghrita was not oxidized, Rancidity determines the level of oxidation and Helps to determine the shelf life of *Ghrita*

6. *Peroxide value*[12]*:*

It is a measurement of peroxides present in the *Ghrita.* Peroxide value signifies the percentage of oxidation of the *Ghrita* and *Taila*. It helps us to find the stability of the sample. If the peroxide value is more, it shows more oxidation and chances of attaining rancidity is also more.

Peroxide value of *Shatavari Ghrita* was found to be 0, which indicatesthat*Shatavari Ghrita* hasmore stability.

GCMS

GCMS technique helps to quantitative analysis of active components present in a given sample. *Shatavari Ghrita* is having total 39 components, among them 6 are identified. These 6 components are having the properties like Antioxidant, Lubricant, Immunostimulant and anti-inflamatory which helps to reduce the gastric ulcer. (As seen in Table 9)

Table 9 Showing summary of Components in *Shatavari Ghrita*

Sl No	Peak Name	Nature Compound	Biological Activities
1	Tetradecanoic acid	Saturated fatty acid	Antioxidant, Lubricant
2	N-Hexadecanoic acid	Saturated fatty acid	Anti-inflamatory , Antioxidant
3	Oleic acid	Unsaturated fatty acid	Protect cell membrane from free radicals, Antioxidant
4	9-Octadecenal	Saturated fatty acid	Immunostimulant Anti-tumour
5	17-(1,5 Dimethylhexyl)-10,13-dimethyl-2, 3, 4, 7, 8, 9, 10, 11, 12, 13, 14, 15,16, 17- tetradecahydro-1H-cyclopenta [a] phenanthren-3-ol	Cholesterol	Essential to maintain both membranes structural integrity and fluidity
6	Naphthalene – 1- Carboxyamide	Sebacic Acid	Lubricants

CONCLUTION

Genuine raw materials of study formulations are easily and abundantly available. There is no pharmaceutical constraint in preparation of *Shatavari Ghrita* . Analytical studies including GC-MS have helped to generate preliminary standard for *Shatavari Ghrita* . The analysis of the values suggests that the formulation is within the limit of standard parameters. This suggests that the preparation was done in an authenticated manner. The analytical

values obtained by this study can be considered as preliminary standards for *Shatavari Ghrita*. All the six drugs of *Murchita Ghrita* and one drug of *Shatavari Ghrita* individually possess anti-ulcer activity.

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