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Ayurveda

A COMPARATIVE PHARMACEUTICAL AND ANALYTICAL STUDY OF *GUDUCHI* GHRITA PREPARED FROM FRESH AND DRY *GUDUCHI* (*TINOSPORA CORDIFOLIA*)

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ABSTRACT Sneha kalpana is one of the by and large used and supported estimations sorts of Ayurvedic game plan of medication. It is a pharmaceutical technique which is sought after to convey an oleaginous medicament by the mix of kalka, Kwath, and distinctive dravyas, in express game plans by presenting them to a foreordained warming model and range. There are four sorts of sneha explained in Ayurveda classics.1)Ghrita, 2)Taila, 3)Vasa 4) Majja. Out of these 'Ghrita' is considered as 'the best' by standards of its tailing unique properties, it quickly solidifies the properties of various meds which collaborated with it and did not leave its own one of a kind trademark qualities when in contact with various drugs. Guduchi (Tinospora cordifolia) had been utilized in the indigenous arrangement of medication since the Vedic time frame. It was basic medication and was much of the time referenced in a large portion of the Samhitas, Nighantus and Ghranthas. It was considered as one among the mainstream sedate regarding accessibility, economy, and simplicity of organization. The botanical source of Guduchi throughout India is *Tinospora cordifolia (wild) Miers*, of the family Menispermaceae. As consent of any formulation it is said by ancient Acharyas that always Guduchi should be taken in fresh form but lack of availability and suitability dry Guduchi used more than fresh. Considering this, study had been planned to use fresh and dry Guduchi for its Ghrita preparation.

KEYWORDS : Sneha kalpana, Ghrita, Guduchi, Samhitas, Nighantus, Ghranthas

INTRODUCTION

Sneha kalpana is one of the widely used and preferred dosage forms of Ayurvedic system of medicine. It is a pharmaceutical procedure which is followed to produce an oleaginous medicament by the combination of kalka, Kwath, and other dravyas, in specific preparations by subjecting them to a specified heating pattern and duration[1]. There are four types of sneha explained in Ayurveda classics.1)Ghrita, 2)Taila, 3)Vasa 4) Majja[2]. Out of these 'Ghrita'is considered as'the best'by virtue of its following unique properties, it readily incorporates the properties of the other drugs which came in contact with it and did not leave its own natural qualities when in contact with other drugs[3].

Ghrita(medicated ghee) preparations containing the fat soluble components of the ingredients. Preparation involves boiling of ghrita with prescribed decoctions and a fine paste of the drug to where by the fat soluble principles are transferred to the Ghrita from the drug ingredients or decoction or expressed juice.

Guduchi (*Tinospora cordifolia*) had been used in the indigenous system of medicine since the Vedic period. It was very common drug and was quite frequently mentioned in most of the Samhitas, Nighantus and Ghranthas. It was considered as one among the popular drug in terms of availability, economy, and ease of administration. This plant was used in Ayurved as single drug in the form of Swarasa, Kalka, Kwath, Hima, Churna, Ghrita and also as one of the important ingredients in many other herbal and herbomineral formulation used for treating various ailments. The botanical source of Guduchi throughout India is *Tinospora cordifolia (wild) Miers*, of the family Menispermaceae.There was two types[4] of Guduchi available that was *Tinospora sinensis* and *Tinospora cordifolia* but for the present study *Tinospora cordifolia* had been taken into consideration which was easily procurable.

Till date, no work had been found regarding comparative study of fresh and dry Guduchi. As consent of any formulation it is said by ancient Acharyas that always Guduchi should be taken in fresh form[5] but lack of availability and suitability dry Guduchi used more than fresh. Considering this, study had been planned to use fresh and dry Guduchi for its Ghrita preparation.

OBJECTIVES

- To develop standard manufacturing procedure of Guduchi Ghrita prepared from Fresh and Dry Guduchi.
- To develop possible analytical profile of trial drugs.
- To compare both sample pharmaceutically and analytically.

METHODOLOGY

Collection of raw material

1. **Raw Drugs:** All the raw materials (haritaki, bibhitaki, amalaki, haridra, musta) used for this study were procured from Pharmacy, Parul institute of Ayurveda, Limda, Vadodara, Gujarat.
2. **Fresh Guduchi:** Fresh guduchi was collected from Waghodia.
3. **Dry Guduchi:** first this drug was procured from Waghodia in fresh form then it was kept for dry.
4. **Milk:** Freshly Go dugdha was collected from Damodara, Vadodara.
5. **Bijaura nimbu:** Bijaura nimbu was procured from khandera market of Vadodara. Authentification was done by senior experts.

Pharmaceutical study

Murcchita ghrita

Kalka dravya were taken in coarse powder form. Sufficient quantity of water was added to the powder of Kalka dravya and converted into balls.

Go Ghrita was taken in a vessel then put it in mild fire then added Kalka and water in it after that Phenashanti stage was observed. At this stage Ghrita becomes moisture free but some part of moisture remains in the Kalka. Then Mridupaka stage was observed while Madhyam paka was achieved then no moisture was found in the Kalka. It took total 15 hr duration to complete Ghrita murcchana. Then filter it with clean cotton cloth.

Table no.1 Murcchita Ghrita

Go Ghrita	5kg200gm
Kalka	1kg625gm (325gm each drug)
Jala	20lit800ml
obtain	4kg900gm
loss	300gm

Fresh guduchi ghrita

Taken 2 kg Murcchita Ghrita in a vessel and put it in Mridu agni then added kalka and Kwath simultaneously then boiling properly after that added go milk in it. That Phenashanti stage was observed. At this stage Ghrita becomes moisture free but some part of moisture remains in the kalka. Then Mridupaka stage was observed while Madhyam paka was achieved then no moisture was found in the Kalka. It took total 10 hr duration to complete fresh Ghrita preparation. Then filter it with clean cotton cloth.

Table no.2 Fresh Guduchi Ghrita

Go Ghrita	2kg
Kalka	500gm
Kwath	8lit
Milk	2lit
obtain	1kg 745gm
loss	255 gm

Dry guduchi ghrita

I took same procedure but taken dry Guduchi for preparation.

Table no.3 Dry Guduchi Ghrita

Go Ghrita	2kg
Kalka	500gm
Kwath	8lit
Milk	2lit
obtain	1kg 742gm
loss	258gm

OBSERVATION

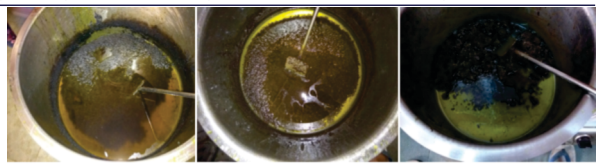
Table no.4 Observation

S.No.		Dry Guduchi Ghrita	Dry Guduchi Ghrita
1	Starting Temp.	27.2°c	27.2°c
2	Temp. Kalka Added	50°c	50°c
3	Temp.Guduchi Kwath Added	60°c	60°c
4	Temp.:- Godugdha added	75.1°c	75.1°c
5	Temp. For Phenashanti	90.7°c	90.7°c
6	Temp. For Mridupaka	75°c	75°c
7	Temp. For Madhyam Paka	80°c	80°c
8	Maximum temperature obtained during Ghrita Paka (in 0 C)	112°c	112°c
9	Total Time for the Process-Hrs	10 hr	10 hr
10	In Days	3 days	3 days

INGREDIENTS

SNEHA SIDDHI LAKSHANA

Vartivatasnehakalka | Sabdahinoagninikshipta | Gandhavarnarasotpatti



Murcchita Ghrita | Fresh Guduchi Ghrita | Dry Guduchi Ghrita

ANALYTICAL STUDY

- Organoleptic characters
- Physico-chemical Characters
- HPTLC
- Stability test

ORGANOLEPTIC CHARACTERS

Table no.5 Organoleptic Characters

Parameters	Fresh guduchi ghrita	Dry guduchi ghrita
Colour	Greenish yellow	Golden yellow
odour	Bitter	Bitter
taste	Bitter	Bitter

PHYSICO-CHEMICAL CHARACTERS

Table no.6 Physico-Chemical Characters

S.No.	Parameters	Fresh guduchi ghrita	Dry guduchi Ghrita
1.	Specific gravity	0.912	0.918
2.	Refractive index	1.468	1.546
3.	Acid value	1.74	2.6
4.	free fatty acid	0.87	1.30
5.	Iodine value	34.86	33.6
6.	Saponification value	220.82	219.78
7.	Peroxide value	5.2	3.46
8.	Rancidity	Negative	Negative
9.	Loss on drying	0.99	1.02
10.	PH value	3-4	3-4

HPTLC

Preparation of test solutions (T):

Accurately weighed 5.0 g of sample individually in Iodine flask and add 30 mL methanol to it. Vortex it for 5 min, centrifuge 10mL of it for 5 minutes, filters it with Whatman filter paper no. 1 and then concentrate it on water bath up to 2mL. Filter again if required and use for HPTLC profiling.

Track 1: Guduchi Ghrita - Fresh (10 µL)
Track 2: Guduchi Ghrita - Dry (10 µL)

Preparation of Spray reagent [Anisaldehyde – sulphuric acid reagent]: 0.5 mL Anisaldehyde EP is mixed with 10 mL Glacial acetic acid AR, followed by 85 mL Methanol AR and 5 mL Sulphuric acid 98% GR.

Chromatographic Conditions:

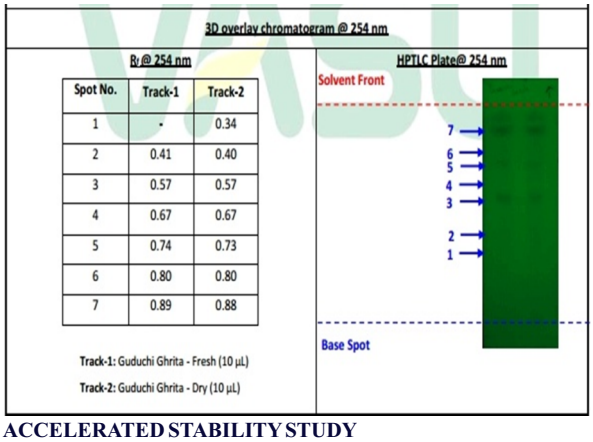
Table no.7 Chromatographic Conditions

Application Mode	CAMAG Linomat 5 – Applicator
Filtering System	Whatman filter paper No. 1
Stationary Phase	MERCK - TLC / HPTLC Silica gel 60 F254 on Aluminum sheets
Application (Y axis) Start Position	10 mm
Development (Y axis) End Position	90 mm from plate base
Space Between Band	10 mm
Sample Application Volume	10 µL
Development Mode	CAMAG TLC Twin Trough Chamber
Chamber Saturation Time	30 minutes
Mobile Phase (MP)	Toluene : Ethyl acetate : Formic acid (10:3:1)
Visualization	@254nm, @ 366nm and @ 540 nm (after derivatization)
Spray reagent	Anisaldehyde Sulphuric acid reagent

Derivatization mode	CAMAG – Dip tank for about 1 minute
Drying Mode, Temp. & Time	TLC Plate Heater Preheated at 100± 50C for 3 minutes

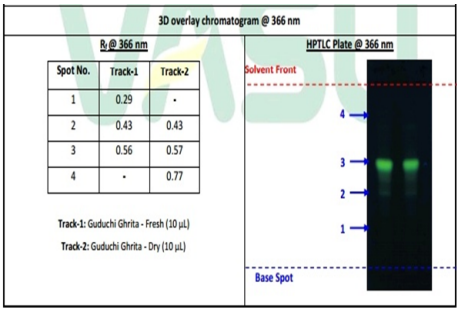
Observation:
After Derivatization, plate was examined visually for appearance of different bands at different Rf.

Table no.8 Refraction value at 254 Nm



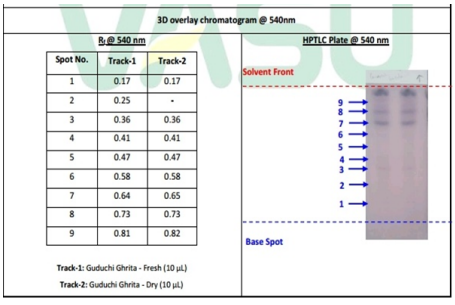
Spot No.	Track-1	Track-2
1	-	0.34
2	0.41	0.40
3	0.57	0.57
4	0.67	0.67
5	0.74	0.73
6	0.80	0.80
7	0.89	0.88

Table no.9 Refraction value at 366 nm



Spot No.	Track-1	Track-2
1	0.29	-
2	0.43	0.43
3	0.56	0.57
4	-	0.77

Table no.10 Refraction value at 540 nm



Spot No.	Track-1	Track-2
1	0.17	0.17
2	0.25	-
3	0.36	0.36
4	0.41	0.41
5	0.47	0.47
6	0.58	0.58
7	0.64	0.65
8	0.73	0.73
9	0.81	0.82

ACCELERATED STABILITY STUDY

Fresh Guduchi Ghrita

(Condition: Temp:40^{0}C ± 2, RH:75% ± 5)

Table no.11 Fresh Guduchi Ghrita

S.N.	Parameters	0 Month	1 Month	3 Month	6 Month
Organoleptic Parameters					
1	Colour	Golden yellow	Golden yellow	Golden yellow	Golden yellow
2	Odour	characteristic	Characteristic	characteristic	characteristic
3	Taste	characteristic	Characteristic	characteristic	Characteristic
Physico-chemical Parameters					
1	Specific Gravity	0.913	0.912	0.911	0.911
2	Refractive Index	1.469	1.466	1.469	1.469
3	Acid Value	0.88	1.15	1.59	2.432
4	Saponification Value	198.25	222.66	230.93	233.18
Microbial Limit Test					
1	Total Plate Count (NMT10^5-cfu/g)	<10cfu/gm	Not Applicable	<10cfu/gm	
2	Total Yeast & Mould Count (NMT10^{3}cfu/g)	Absent		Absent	
3	E.coli	Absent		Absent	
4	Salmonella	Absent		Absent	
5	S.aureus	Absent		Absent	
6	P. aeruginosa	Absent		Absent	
Heavy Metal Analysis					
1	Lead (NMT 10 ppm)	Not Detected	Not Applicable		
2	Cadmium (NMT 0.3 ppm)	Not Detected			
3	Mercury (NMT 1 ppm)	Not Detected			
4	Arsenic (NMT 3 ppm)	Not Detected			

Dry Guduchi Ghrita

(Condition: Temp:40^{0}C ± 2, RH:75% ± 5)

Table no.12 Dry Guduchi Ghrita

S.N.	Parameters	0 Month	1 Month	3 Month	6 Month
Organoleptic Parameters					
1	Colour	Golden yellow	Golden yellow	Golden yellow	Golden yellow
2	Odour	characteristic	Characteristic	characteristic	Characteristic
3	Taste	characteristic	Characteristic	characteristic	Characteristic
Physico-chemical Parameters					
1	Specific Gravity	0.917	0.910	0.909	0.896
2	Refractive Index	1.469	1.468	1.469	1.469
3	Acid Value	1.31	1.84	1.92	2.93
4	Saponification Value	226.31	228.43	246.32	248.15
Microbial Limit Test					
1	Total Plate Count (NMT10^{5}cfu/g)	<10cfu/gm	Not Applicable	<10cfu/gm	
2	Total Yeast & Mould Count (NMT10^{3}cfu/g)	Absent		Absent	
3	E.coli	Absent		Absent	
4	Salmonella	Absent		Absent	
5	S.aureus	Absent		Absent	

6	P. aeruginosa	Absent		Absent
Heavy Metal Analysis				
1	Lead (NMT 10 ppm)	Not Detected	Not Applicable	
	Cadmium (NMT 0.3 ppm)	Not Detected		
3	Mercury (NMT 1 ppm)	Not Detected		
4	Arsenic (NMT 3 ppm)	Not Detected		

DISCUSSION

Guduchi is ancient medicine which we used as a medicine. References regarding Guduchi are available from Atharvaveda[6]. Since Samhita kala Guduchi has been used as a medicine in various places. In Sharangdhar Samhita[7] there is reference that some drug should be taken in fresh form for medicine preparation ,Guduchi is one amongst them. But in present scenario Guduchi in dry form is used for medicine preparation. Fresh guduchi has complications such as difficulties were seen at the time of grinding the stem of the plant .

By screening the reference regards Guduchi Ghrita there are 11 reference found like Bhavprakash, Vrindamadhava, Chakradatta, Vangasena vatarakta rogadhikara, Bhavprakash khanda 2, Gadanigraha vatarakta rogadhikara, Charaka Samhita, Vangasena pandu rogadhikara, Gadanigraha pandu rogadhikara, yoga ratnakara. Many reference regarding the preparation are available which are almost similar, only one reference is found different by Acharya Bhavprakash[8] where the reference regarding the kwath dravya is not available hence the go dugdha which is to be used is taken to the ratio of 1;4 to that of go ghrita.

Reference regarding the preparation of Guduchi Ghrita was taken from Chakradatta[9]. According to given reference Guduchi Ghrita is prepared with Guduchi Kalka, Guduchi Kwath and milk. In this, reference regarding the drugs ratio is not explained. So for the preparation of Ghrita reference was taken from Chakradatta Jwar Chikitsa here we get the general methods of Ghrita preparation.

In the present study for the preparation of guduchi ghrita the reference for kalka preparation was taken from Sharangdhar Samhita And for the kwath preparation reference from Chakradatta Jwar Chikitsa was taken, but it was found that these references were suitable for the preparation of ghrita with fresh plant, so for the preparation of kwath from dry plant the ratio of water taken was 8 times because the fresh plant is comparative Mridu hence 4 times water is enough for the preparation where as in dry form it is not found sufficient hence 8 times water was taken as per the reference available in Sharangdhar Samhita.

PHARMACEUTICAL STUDY

Standard operative procedure (S.O.P.) is a written document which contains relevant information about the activity or procedures to be performed.

Here on attempts was made to develop SOP for Guduchi Ghrita with the help of comparative study of different samples of Guduchi Ghrita which prepared by fresh and dry Guduchi. Therefore some steps/stages fix for murcchana process, fresh Guduchi Ghrita, dry Guduchi Ghrita were mentioned in pharmaceutical study.

Therefore specification of vessel used, source of heat, drava, sneha, kalka and critical observation were noted at each step in regular intervals.

Patanjali Ghrita procured from the market for the study. sample were subjected to analytical parameter like Organoleptic character Ash Value**,** Specific gravity**,** Refractive index**,** Acid value, free fatty acid, Iodine value, Saponification value, Peroxide value**,** Rancidity**,** Loss on drying, PH value.

Murcchana dravya procured from the market. Fresh Guduchi and cow milk collected from Waghodia field. Procured fresh guduchi is dried completely and used for second sample.

ANALYTICAL STUDY

Raw material analysis

On observing the result of analytical parameter of sample of Patanjali Ghrita very minimum variations were found.

On observing the result of analytical parameter of all herbal drugs under API parameter.

In process analysis

On observing the result of analytical parameter of sample of Murcchita Ghrita very minimum variations were found.

Finish product analysis

On observing the result of analytical parameter of fresh Guduchi Ghrita very minimum variations were found.

On observing the result of analytical parameter of dry Guduchi Ghrita very minimum variations were found.

During comparative study between fresh and dry Guduchi Ghrita observed very minimum variations.

It might be possible because the base used for both the preparation is same that is ghrita. Hence prepared medicine will not show much difference analytically either in fresh or dry drug form. Whereas difference in organoleptic characters like colour and taste were found more prominent in freshly prepared medicine.

Qualitative HPTLC finger printing done for both fresh and dry Guduchi Ghrita. When it was observed under 254nm total 6 spot were found in fresh Guduchi Ghrita and 7 spot were found in dry Guduchi Ghrita after that when it was observed under 366nm total 3 spot were found in fresh Guduchi Ghrita and 3 spot were found in dry Guduchi Ghrita. After that When it was observed under 540nm total 9 spot were found in fresh Guduchi Ghrita and 8 spot were found in dry Guduchi Ghrita. So it is found out that Guduchi Ghrita prepared from both fresh and dry Guduchi is having almost similar active principle constituents.

As per the classical reference shelf life of Ghrita is 16 month[10]. As per new amendment year 2016 it is 2 year but the accelerated stability study was carried out to check the changes during 6 months at 40°C temperature and 75% humidity. Ghrita was kept for 6 months in this high temperature and pressure so can say that the shelf life of fresh and dry Guduchi Ghrita is up to 3 yr.

CONCLUSION

As SOP was one of the objectives, So Guduchi Ghrita prepared from both fresh and dry was carried out. For the same purpose three batches were prepared and analysed.

Guduchi Ghrita prepared from fresh Guduchi is greenish yellow in colour and prepared from dry Guduchi is golden yellow in colour.

Comparative analytical parameters of both the sample showed very negligible difference.

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