

PRELIMINARY ANALYTICAL STUDY OF PANCHTIKTA GHRITA

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ABSTRACT :

Panchtikta Ghrita was processed as per the process of Snehapaka procedure described Ayurvedic formulary of India. It contained Panchtikta that is Nimba (Azadirachta indica), Patola (Luffa acutangula), Vyaghri, Guduchi, Vasa (Adhatoda vasica). They are used in the form of decoction in the preparation of Panchtikta Ghrita. Paste of Triphala – Haritaki (Terminalia chebula), Vibhitaki (Terminalia bellirica) Amla (Emblica officinalis) is also prepared and used in Panchtikta ghrita. Mixture were mixed in Purana Ghrita and heated for three hours at 100°C every day for two days. On the second day Ghrita was filtered to obtain the finished product. In this manner, three samples of Panchtikta Ghrita were prepared. To understand the changes that occurred during the preparation, Panchtikta Ghrita and Purana Ghrita were analyzed by using modern parameters such as Acid value, Saponification value, iodine value, peroxide value and so on.

Key words: Panchtikta Ghrita, Acid value, Saponification value, Iodine value.

INTRODUCTION: Sneha kalpana is widely described secondary dosage form in Ayurvedic pharmaceutics under which medicated oil and ghrita are prepared. Ghrita is obtained from the class mammalian of the animal Kingdom. Ayurveda recommends the Goghrita as best. Ghrita alleviates pitta and vata, is beneficial for rasa, semen and ojas, cooling, softening and improves voice and complexion ^{1,2}. Ghrita preparations manufactured in Ayurvedic pharmaceutics are used broadly for medicinal purposes. It is one of the eye-catching techniques in Ayurvedic drug industry to achieve both fat soluble and water soluble extractives into the ghrita media acts on liposomal level ³. Panchtikta Ghrita ⁴ is commonly used ghrita kalpana used as Ayurvedic

medicine for the treatment of skin disorders. As the herbs used in the manufacturing of Panchtikta ghrita are considered best among rakta shodhak (blood purifier) drugs.

MATERIALS AND METHODS:

Method of Preparation of Panchtikata ghrita:⁵ It was prepared as per reference of Ayurvedic Formulary of India part I.

Ingredients:

Nimba – Azadirachta indica – 240 g
Patola – Luffa acutangula – 240 g
Vyaghri – Solanum xanthocarpum – 2480 g
Guduchi – Tinospora cordifolia – 240 g
Vasa – Adhatoda vasica – 240 g
water for decoction – 6.144 liters boiled and reduced to 1.5 liters
Triphala – Haritaki (Terminalia chebula), Vibhitaki (Terminalia bellirica) Amla

(*Emblica officinalis*) – 64 g each for kalka (paste)

Ghrita – ghee – 384 ml..

Cow's Ghrita (Old) was taken and subjected to Murcchana samskar as per reference of Bhaishjya Ratnavalli.

Process of Ghrita Murcchana⁶

Ingredients: Pathya , Dhatri, Vibhitaki, Musta (Jaladha) , Rajani , Matulunga swarasa

From Pathya to Matulunga swarasa each I Pala = 48gram

Cow's Ghrita 1 Prastha(64tola)= 788grams

Jala (Water)4Prastha(256tola)= 3.072liters

- Heat the Ghrita till it's become free from froth. Add 4 part of water along with powder of all the drugs.. Boil it on moderate heat till the Ghrita became free from water. Filter it and use for the preparation of medicated Ghrita.
- By this process unpleasant odour of the Ghrita is removed. It obtains good colour and fragrance.

First of all the ingredients of Panchtikta ghrita were taken. Wash and dry all the herbal raw materials thoroughly. Pulverize ingredients numbered 1 to 5 (drugs for decoction) to coarse powder, add specified quantity of water, heat and reduce the volume to one-fourth. Filter with muslin cloth to obtain Panchtikta kwatha (decoction). Take the other ingredients (kalka dravya) numbered 7 to 9 in the formulation composition, Powder and pass through sieve number 85. Transfer the powdered ingredients to wet grinder and grind with sufficient quantity of water to prepare a homogeneous paste (Kalka). Take Murchhita ghrita in a stainless steel vessel and heat mildly. Add increments of Kalka. Stir thoroughly while adding kwatha.

Heat for 3 hour with constant stirring maintaining the temperature around 100⁰c

.Stop heating and allow to stand overnight. Start heating next day and observe the boiling mixture sneha siddhi lakshana⁷ e.g. for subsidence of froth (phena shanti) and constantly check the kalka for formation of varti. Expose the varti to flame and confirm the absence of crackling sound indicating absence of moisture. Stop heating when the kalka forms a varti and the froth subsides. Filter while slight hot through a muslin cloth and allow to cool. Pack it in tightly closed glass containers to protect from light and moisture. The above combination is heated till Panchtikta ghrit is prepared.

Three samples of panchtikta ghrita were prepared for the purpose of standardization and coded as PG 1, PG 2 AND PG 3. Sample of Murcchit ghrita was also preserved and coded as MG.

Analytical techniques: To ensure reproducibility of the *Panchtikta Ghrita*, the analytical methods were applied to three samples, which were prepared with the same ingredients, following the same manufacturing method. These were analyzed to obtain parameters, such as, organoleptic study, acid value, saponification value, iodine value, and refractive index, and peroxide value according to the Quality Control Manual of Ayurveda, Siddha, and Unani Medicine (the standard protocol mentioned in books).⁸

The test was done as per the standard pharmaceutical laboratory process given in Appendix 3 (Physical test determination) of the Ayurvedic Pharmacopeia of India.⁹

Measurement of Specific gravity¹⁰: Specific gravity of a substance is the weight of the substance in grams at a specific temperature compared with the weight of the same volume of water in grams at a same temperature.

1. A clean and dried 25ml capacity of specific gravity bottle kept in hot air oven was taken in desiccator and weighed empty. Then it was filled with water and weighed at room temp.

2. Again the bottle was clean and dried. Kept it in hot air oven and then took in dessicator. Filled the Panchtikta ghrita sample up to the mark and weighed at the same temp.

Specific gravity of the sample = Wt. of Sample/ Wt. of Water = Weight of (oil) sample in grams/ weight of same volume of water at same temp in grams.

Determination of Refractive index ¹¹:

The refractive index (or index of refraction) of a medium is a measure for how much the speed of light (or other waves such as sound waves) is reduced inside the medium. It is the ratio of the velocity of light in a vacuum to its velocity in the substance.

Abbe's Refractometer was used to determine the Refractive Index. First the mirror of the Abbe's Refractometer was adjusted to 45°c. Then the sample of Panchtikta ghrita was inserted in the prism box by using a pipette. After each sample refractometer was cleaned with petroleum ether followed by the distilled water. Different color bands were observed in the right eye piece. These color bands were removed with the help of compensator knob in such a way that only the black and white portion should be seen in the right eye piece. The black and white portion are accustomed to the cross wire with the help of lever. Finally the result was noted on the scale through left eye piece. Both samples were analyzed by this way.

Measurement of Saponification value

¹²: Saponification value (or "saponification number", also referred to as "sap" in short) represents the number of milligrams of

potassium hydroxide or sodium hydroxide required to saponify 1g of fat under the conditions specified

Initially 500ml capacity of round bottom flask is fitted with a reflux condenser. Then 4gms of Panchtikta ghrita sample with 50ml of 0.5N KOH was taken into the round bottom flask. 2-3 pieces of pumice stones were put into the same flask and the mixture was boiled on water bath at 40°c for 30 min. Than after it was taken out from water bath and 1 ml of phenolphthalein solution (indicator) was added to it. Titration was done immediately with 0.5N HCl. The burette reading was noted (a). Process was repeated out without taking the ghrita sample, i.e. a blank test under same conditions and burette reading was noted (b). Both the samples were analyzed by this method.

Saponification value was determined as per following formula.

Saponification value = $\{(b-a) \times 28.05\} / W$.
*W=Weight of the substance in gms.

Determination of Acid value ¹³ : Acid value (or "neutralization number" or "acid number" or "acidity") is the mass of potassium hydroxide (KOH) in milligrams that is required to neutralize one gram of chemical substance.

First of all a solvent is prepared by adding 50ml alcohol and 50 ml ether in a container. Then 20 grams of Panchtiktaghrita sample was mixed in 100ml of solvent which was prepared earlier. Now 2 ml of Phenolphthalein indicator was added to it and titration was done with 0.1 N Sodium hydroxide (NaOH) until the solution remained faintly pink for 30 sec. even after shaking. Finally the reading of the barrette was noted.

Acid value was calculated as per following formula

Acid value = (Nx5.61)/W

*N= Number of ml of 0.1NaOH required and *W =Weight of sample in gms.

Determination of Peroxide Value ¹⁴: The peroxide value is the number of milliequivalents of active oxygen that expresses the amount of peroxide contained in 1000 g of the substance.

1. First of 5 grams of panchtikta ghrita is weighed accurately
2. Transferred it into a 250-ml glass-stoppered conical flask
3. Add 30 ml of a mixture of 3 N glacial acetic acid and 2 N chloroform, swirl until dissolved and add 0.5ml volumes of saturated potassium iodide soluton.
4. Allow to stand for exactly 1 minute, with occasional shaking, add 30 ml of water and titrate gradually, with continuous and vigorous shaking, with 0.01M sodium thiosulphate until the yellow colour almost disappears.
5. Add 0.5 ml of starch solution and continue the titration, shaking vigorously until the blue colour just disappears (a ml).
6. Repeat the operation omitting the substance being examined (b ml).
7. The volume of 0.01M sodium thiosulphate in the blank determination must not exceed 0.1 ml.
8. Calculate the peroxide value from the expression Peroxide Value = $10(a - b)/W$ Where W = weight, in g, of the substance.

Determination of Iodine Value ¹⁵ : The Iodine value of a substance is the weight of iodine absorbed by 100 parts by weight of the substance

1. First take an Iodine flasks have a nominal capacity of 250 ml and kept it in hot oven for 15 minutes
2. Place the substance accurately weighed to 5 grams, in dry iodine flask,
3. Add 10 ml of carbon tetrachloride, and dissolve.
4. Add 20 ml of iodine monochloride solution insert the stopper, previously moistened with solution of potassium iodine and allow to stand in a dark place at a temperature of about 17^0 c for thirty minutes.
5. Add 15 ml of solution of potassium iodine and 100 ml water; shake, and titrate with 0.1 N sodium thiosulphate, using solution of starch as indicator.
6. Note the number of ml required (a).
7. Repeat the operation in exactly the same manner, but without the substance being tested, and note the number of ml of 0.1 N sodium thiosulphate required (b).
8. Calculate the iodine value from the formula- Iodine Value = $(b - a) \times 0.01269 \times 100/W$ Where 'W' is the weight in g of the substance taken.

RESULTS:

Organoleptic Properties

There is difference in color of Murchhit ghrita and prepared Panchtikta ghrita. The organoleptic characters of both the *Ghritas* are mentioned in Table 1. *Murchhit Ghrita* was of yellow in color and had a peculiar smell. All three samples, that is, Samples PG 1, PG 2 and PG 3 of the *Panchtikta Ghrita* had Greenish yellow Color and a peculiar smell.

Analytical Parameters:

Result of these parameters is tabulated in table 2.

Table 1 Showing Organoleptic Properties

Sample	Color	Taste	Odour
MG	Yellow	Bitter	Characteristic
PG 1	Greenish yellow	Bitter	Characteristic
PG 2	Greenish yellow	Bitter	Characteristic
PG 3	Greenish yellow	Bitter	Characteristic

Table 2 Showing Analytical Result

S.No.	Panchtikta Ghrita Sample	Specific Gravity at melted stage	Refractive Index at 40 ⁰ c	Saponification value	Acid value	Peroxide Value	Iodine Value
1.	MG	0.8828	1.4436	220.4	0.64	2.64	29.64
2.	PG 1	0.9124	1.5126	222.6	1.58	2.76	30.24
3.	PG 2	0.9196	1.5094	223.2	1.98	2.98	30.78
4.	PG 3	0.9212	1.5210	228.8	2.46	3.24	31.92

DISCUSSIONS:

Specific Gravity: Specific gravity of ghrita is indication of the solid to liquid ratio into ghrita. Specific gravity of all the sample of Panchtikta ghrita was found more in compare to Murchhit ghrita. It may be due to solid extractives of the kalka (paste) drugs transferred to murchhit ghrita in the preparation of Panchtikta drugs.

So increased Specific gravity of Panchtikta ghrita than Murchhit ghrita reveals that solid content is raised into the prepared ghrita formulation in compare of liquid content which increases the life span of formulations and thus its therapeutic value.

Refractive Index: Refractive index is the ratio of the velocity of light in a vacuum to its velocity in the substance. It is a fundamental physical property of a substance often used to identify a particular substance, confirm its purity, or measure its concentration. Increased Refractive Index in all the samples of Panchtikta ghrita than Murchhit ghrita reveals that that some active substances

of ingredients used in the preparation of Panchtikta ghrita were incorporated into Murchhit ghrita.

Saponification value : Saponification value is a measure of the average molecular weight (or chain length) of all the fatty acids present in sample. Saponification value is the directly proportional to the fatty matter content. Long chain fatty acid found in fat that have low saponification value. Short-chain fatty acids (SCFAs) have a high saponification value. Short chain fatty acids are readily absorbed than Long-chain fatty acids. Increased saponification value of all the samples of Panchtikta ghrita shows that *Panchtita ghrita* has greater short chain fatty acid than Murchhit *ghrita*. Thus, *Panchtikta ghrita* is easily absorbed and digested in the body.

Acid value: The acid number is a measure of the amount of carboxylic acid groups in a chemical compound, such as a fatty acid. As oil-fats started to rancidify, triglycerides are converted into fatty acids and glycerol, causing an increase in acid value. Less acid value denotes the less

chance of decomposition of the composition of Ghrita thus increasing both life span and therapeutic value. Increased Acid value of all the samples of prepared formulation than Murchhit ghrita indicates that during the process of *Snehapaka*, hydrolysis of *Ghrita* takes place, which may be promoted by the reaction of the triglycerides in the *Ghrita* with the active ingredients present in *Panchtikta ghrita*, resulting in glycerol and free fatty acids. High amounts of free fatty acid (Acid Value) shows that stability, and shelf life of *Panchtikta ghrita* is less than that of Murchhit ghrita.

Peroxide value: The peroxide value is defined as the amount of peroxide oxygen per 1 kilogram of fat or oil. The Peroxide value of fat is used as a measurement of the extent to which rancidity reactions have occurred during storage. The double bonds found in fats play a role in Auto oxidation. The best test for auto oxidation (oxidative rancidity) is determination of the peroxide value. Peroxides are intermediates in the auto oxidation reaction. Auto oxidation is a free radical reaction involving oxygen that leads to deterioration of fats and oils which form off-flavours and off-odours. Results of Peroxide value confirm the importance of the finding of Acid value.

Iodine value: The iodine value (or "iodine adsorption value" or "iodine number" or "iodine index") in chemistry is the mass of iodine in grams that is consumed by 100 grams of a chemical substance. One of the important applications of the iodine number is the determination of the amount Unsaturated contains in fatty acids. The higher the iodine number, the more unsaturated fatty acid bonds are present in a fat. Unsaturated fat supplementation increases the total dietary energy intake to

the recommended levels, and it has no adverse impact on the blood lipids. It also improves the nutritional status of diet. Iodine value of all the samples of Panchtikta ghrita is more than Murchhit ghrita denoting its high therapeutic value.

CONCLUSION: *Murchhit Ghrita* and other raw materials were taken in the measured quantities for all three samples (Samples PG 1, PG 2 and PG3) of *Panchtikta Ghrita* and prepared by the *Snehapaka* process. From the present study it was found that the acid value, saponification value, iodine value, and refractive index of all three samples (1, 2 and 3) of *Panchtikta Ghrita* were higher than those of the *Murchhit ghrita*, but sample 3 of *Panchtikta Ghrita* had the highest acid value among all the samples of *Panchtikta Ghrita* and this change was not significant. It was concluded that the analytical values, namely, the acid value, saponification value, and iodine value of *Panchtikta Ghrita* were higher than those of *Murchhit ghrita*. These variations in their values indicate that various active components of the ingredients used in the preparation of *Panchtikta Ghrita* were incorporated in it. From the present study it was also revealed that, there was no significant variation in the analytical values among all three samples of *Panchtikta Ghrita*. Hence, the average values of the analytical parameters may be used for quality assessment and standardization of *Panchtikta Ghrita*.

REFERENCES:

- 1.Pt. Kashinath Shastri, Acharya Charaka's Charaka Samhita, Part I, Chaukhamba Sanskrit Samsthana, 8th Edition.
- 2.Pt. Kashinath Shastri, Acharya Charaka's Charaka Samhita, Part

I,Chaukhamba Sanskrit Samsthana, 8th Edition .

3.Neetu Singh etall, A comparative review study of *Sneha Kalpana (Paka)* vis-a-vis liposome, Ayu.2011 Jan-Mar; 32(1):103–108.

4.Dr.Shailaja Srivastava, Acharya Sarngadhar's Sgarngdhar Samhita, Chaukhamba Orientallie, 3rd Edition, 2003

5.Ayurvedic Formulary of India, Part I, New Delhi: Govt. of India Ministry of Health and Family Welfare, Dept. of AYUSH, 2005

6.Pt. Ambika Dutt Shastri, , Kaviraj Govind das Sen's Bhaishjya Ratnavalli, Chaukhamba Prakashan, Reprint 2014

7.Dr.Shailaja Srivastava, Acharya Sarngadhar's Sgarngdhar Samhita, Chaukhamba Orientallie, 3rd Edition, 2003

8. Lohar . Ghaziabad: Protocal for Testing of Ayurveda, Siddha and Unani medicine page 124-126,Department of Ayush, Ministry of health and family welfare, pharmacopoeial laboratory for Indian medicines;

9.First ed. Vol. 2. New Delhi: Govt. of India Ministry of Health and Family Welfare, Dept. of AYUSH; 2008. The Pharmacopeia of India.Part II (formulations). Appendices 1 to 5.

10.Hough, J.S., Briggs, D.E., Stevens, R and Young, T.W. Malting and Brewing Science, Vol. II Hopped Wort and Beer, Chapman and Hall, London, 1991, p. 881

11.Kinsler, Lawrence E. Fundamentals of Acoustics. John Wiley., 2000, p. 136.

12. Remington's The Science and Practice of Pharmacy, 21st Edition, 2005, Page no. 425

13.Kardash, E. and Tur'yan, Y. I.etall, Acid Value Determination in Vegetable Oils by Indirect Titration in Aqueous-alcohol Media. Croat. Chem. Acta, 2005, 78:1:99-103

14.M.M. Chakrabarty, Chemistry And Technology of Oils And Fats, Allied Publishers Pvt. Ltd , 2003.

15.Firestone D et all, Determination of the iodine value of oils and fats: summary of collaborative study J AOAC Int. 1994 May-Jun;77(3):674-6.

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