RESEARCH
www.commprac.com
ISSN 1462 2815

STUDY OF SHATA DHAUTA GHRITA PROPERTIES: EFFECT OF WASHING ON COW GHEE

Madhuri D. Deshmukh [1]* and Moreshwar P. Patil [2]

[1, 2] Department of Pharmaceutics MET's Institute of Pharmacy, Adgaon, Nashik, Maharashtra, India.
*Corresponding Author Email: deshmukh0072@gmail.com

DOI: 10.5281/zenodo.10241992

Abstract

Ayurveda since ancient times, the science of health care and medicine has been regarded as extremely important. Ghrita is one among the chaturvidha sneha explained in Ayurveda and widely used as Ahara and Aoushada. Shatadhauta ghrita (SDG) is an example of Dhauta samskara and one such unique preparation. SDG (Shata -100 times, Dhauta - washed) is made by washing cow ghee along with water 100 times. This method turns ghee into a soft, cooling, nourishing, silky cream. To investigate the impact of washing of Cow ghee on the formulation of Shata Dhauta Ghrita. SDG was prepared using cow's ghee as per standard Ayurvedic classical texts and subjected to study organoleptic properties (color- white, odour- odourless taste- tasteless texture-Smooth oily and homogenous , weight-increased chemical properties (acid value, Iodine value, Saponification value, Peroxide value, Copper content, RM value and P value) physical properties (Moisture content, pH, refractive index, Viscosity (cp) type of emulsion) analyses as per the standard pharmacopoeial procedures in different no of washing like 10,20,40,60,80,100,120,140.The present research work is successfully investigate the impact of washing on changes of organoleptic properties ,Physical and chemical properties of Shata Dhauta Ghrita formulation.

Keywords: Dhauta Samskara, Shatadhauta Ghrita, Ayurveda.

1. INTRODUCTION

Samskara is the Ayurvedic name for the process of altering a substance's natural qualities. This is accomplished through a variety of techniques such as dilution, heating, cleaning, churning, storing in a certain area, maturing, flavouring, impregnation, preservation, and container. [1,2] Ghee is derived from the ancient Sanskrit word "ghr," which means light or to make luminous. It is commonly manufactured from cow, buffalo, or mixed milk. [3]. It has been shown in both in vitro and in vivo studies to have beneficial effects on health, including anti-obesity, anti-carcinogenic, anti-atherogenic, anti-diabetic, anti-mutagenic, anti-hypertensive, immunomodulatory, apoptotic, osteosynthetic, and in the treatment of various skin disorders. [4]. Due to its amazing ability to enter the deepest tissues, ghee is regarded as the ideal substrate for the creation of Ayurvedic treatments that specifically target specific body organs [5]. Ghee can help transport medications without altering them because to its exceptional ability to swiftly reach every organ and tissue; this is known in Ayurveda as the "yogawahi" action [6, 7]. Ghrta or ghrita are preparations made by boiling ghrita with required liquid medium (Swaeasa/kasaya) and a fine paste (kalka) of medications indicated in the formulation composition [32]. The synthetic topical can cause significant skin damage as well as a variety of adverse effects and toxicity [8].

Clarified butterfat that has been washed 100 times is known as shatadhauta ghrita (SDG; shata = 100; dhauta = washed). Traditional texts cite it as a therapy for burns, chicken pox, leprosy, wounds, and other skin illnesses, as well as a vehicle for drugs to be applied topically. [1, 9,10]. It is an Ayurvedic formulation that combines Dhauta, Jala, and Agni samskara. It includes Bhajana Samskara is prepared in a copper

vessel. Toya samskara entails the usage of water, but dhauta samskara entails washing [1,11].

Shatadhauta ghrita has been studied for its various properties like wound healing [12], management of diabetic foot ulcer [13], pitta yoni [1, 14], fissure [15], incorporation of curcuma amada for the development of anti-inflammatory formulations [16].

Shatadhauta ghrita involves the splitting of fats i.e. ghee in presence of water to produce free fatty acids and glycerol. Because of the pressing force used during fomentation, the molecular size of fat granules decreases as the washing progresses (the surface was non-granular and smooth).To check effect on no of washing of on cow ghee on SDG formulation.

2. EXPERIMENT

2.1. Material and Methods

2.1.1. Procurement of İngredients: Cow ghee was obtained from the local vendor, Nashik. Other of Analysis was procuring Avi Chemicals in Mumbai.

2.1.2. Preparation of Shatadhauta Ghrita

It was prepared by washing required amount of Ghrita (50g) with double amount of cold water (100 ml) till the Ghrita fully immersed in water. Copper vessel is used to facilitate the breakdown of fats. It was rubbed with copper glass. Then water was decanted and again fresh water was poured. This process was repeated for 100 times. (Parihar, *et al*. 2016) [18, 25]. After each 10, 20,40,60,80,100,120,140 washing check Organoleptic Physical and Chemical Properties of SDG. Preparation of SDG shown in fig .no 1.

Figure1: Preparation Method of Shatadhauta Ghrita

3. CHARACTERIZATION OF SHATA- DHAUTA- GHRITA COMPARED TO COW GHEE [28,29]

3.1. Organoleptic Characters

The organoleptic characters of Shata-Dhauta-Ghrita like appearance, color, odour and texture were observed.

3.2. Effect of Washing on weight of ghee (SDG Formulation): SDG was prepared by using traditional Ayurvedic method .After each after each 10, 20,40,60,80,100,120,140 washing check Changes of weight of SDG.

3.3. Evaluation of Chemical Properties of Ghee and SDG

3.3.1. Acid Value: The acid value is the number which expresses in milligrams the amount of KOH necessary to neutralize the free acids present in 1 g of the substance. The obtained Acid value of Shata- Dhauta- Ghrita is shown in Table No.8.7. Where, n = the number of ml of 0.1 M KOH required; w = the weight, in g, of the substance.

$$\text{Acid value} = 5.61\ n/w$$

3.3.2. Saponification Value: The saponification value is the number of milligrams of potassium hydroxide necessary to neutralize the free acids and to saponify the esters present in 1 g of 2g of Shata- Dhauta- Ghrita was weighed accurately and added to 25 ml of 0.5 M ethanolic potassium hydroxide in round bottom flask and the mixture was refluxed using porcelain pieces for 30 min. The mixture was then immediately titrated with 0.5 M HCl using phenolphthalein indicator (a ml). Blank titration was also carried out omitting Shata- Dhauta- Ghrita using 0.5 M HCl (b ml). The Saponification value was calculated from the expression.

$$\text{Saponification value} = 28.05\ (b\ a)/w$$

Where, w = weight of the substance in grams.

3.3.3. Iodine Value

Iodine value is defined as the weight of iodine in gram that reacts with 100g of material.

$$\text{Iodine Value} = (B\text{-}S)\ N/W$$

Where

B=Volume in ml of standard sodium thiosulphate solution required for the blank.

S=Volume in ml of standard sodium thiosulphate solution required for the Sample.

N= Normality of sodium thiosulphate solution.

W=Weight of the substance in gram.

3.3.4. Rechert Meissl

Reichert Meissl value is the number of ml of 0.1N NaOH solution required to neutralize the steam volatile, water soluble fatty acids distilled from oil or fat under precise specified conditions. It is the measure of water soluble steam volatile fatty acids chiefly butyric and cupric acids present in oil or fat.

Reichert-Meissl value = (A – B) × N × 11

A = NaOH solution required for the test (mL),

B = NaOH solution required for blank (mL), and

N = normality of NaOH solution

3.3.5. Polenske Value

Polenske value is the number of ml of 0.1N aqueous NaOH solution required to neutralize the steam volatile, water insoluble fatty acid distilled from oil or fat under precise specified conditions. PV is measure of steam volatile and water insoluble fatty acids mainly caprylic, capric and lauric acids present in fat.

Polenske Value 10x V x N

Where,

V = sodium hydroxide solution (mL) required for the test;

N=Normality of the standard sodium hydroxide solution.

3.3.6. Determination of Peroxide Value

Accurately weighed about 5 g of the sample of ghrita formulation being examined was added into a 250 ml stoppered conical flask. The 30 ml of a mixture of glacial acetic acid (18 ml) and chloroform (12 ml) was added and shaken until it dissolved. The 0.5 ml of saturated potassium iodide solution was added and allowed to stand for exactly 1 minute with occasional shaking. The 30 ml of water was added and titrated gradually with 0.01M sodium thiosulphate with continuous and vigorous shaking until the yellow colour almost disappeared. The 0.5 ml of starch solution was added. The titration was continued with vigorous shaking until the blue colour just disappeared and number of ml of 0.01M sodium thiosulphate required was noted (a). The experiment was repeated omitting the sample and the number of ml of 0.01M sodium thiosulphate required was noted (b).

$$\textbf{Peroxide value} = \frac{10\,(a\text{-}b)}{\text{Weight of } \textit{ghrita}}$$

3.3.7. Copper Content

Atomic absorption spectrometry (AAS) is an analytical technique used to measure the concentrations of metallic elements in food or pharmaceutical products. This technique makes use of the wavelengths of light specifically absorbed by an element. Preparation of Standard solutions of Copper: 0.1 g of copper metal was dissolved in lowest possible 1:1 mixture of concentrated nitric acid and double distilled water, the remaining volume was adjusted using double distilled water up to 1000 ml to obtain stock solution of 100 µg/ml. and calculate conc. of copper present in SDG formulation.

3.4. Evaluation of Chemical properties of Ghee and SDG.

3.4.1. pH

The pH of various formulations was measured by using Digital pH meter. To determined pH, 1 g of each cream formulation were transferred into 100 mL beaker and measured by using the digital pH meter.

3.4.2. Viscosity

The measurement of viscosity of prepared cream was carried out with Brookfield Viscometer (model LV-DV-II, Helipath-spindle type S-96) at 20 rpm for 30 sec.

3.4.3. Determination of Refractive Index

Abbe's Refractometer (ALMICRO) was used to determine refractive index of sample of ghrita formulation. It was cleaned and prisms were wiped out with alcohol and then with acetone and dried. The instrument was kept near proper light source for maximum refraction of light on to the prisms. The Abbe's Refractometer for better accuracy calibrated with water at temperature below 25 ^{o}C for the D-line of sodium. (Water has Refractive Index 1.3009 at 0 ^{o}C). Water was placed in prisms with the help of a capillary tube and the instrument was adjusted to get boundary of crosswire in the field of view. The eye piece was adjusted and the knob was turned to get the boundary of cross wire at the centre of the cross wire. The dial reading was taken as observed refractive index of water (n_{obw}). The prisms were opened, cleaned and dried with alcohol using cotton swab

3.4.4. Type of Emulsion: The type of emulsion of SDG formulation was studied by using dye test, in this test soluble dyes make the color according to their phase solubility, water soluble dyes make the color according to the water phase while oil soluble dyes make the color according to the oil phase. Take a small amount of sample emulsion and mix it with either water soluble dye such as amaranth or an oil soluble dye such as scarlet blue. Observing the result of a small sample mounted on a compound microscope takes in both case.

4. RESULT AND DISCUSSION

In 'Shata-Dhauta' (Shata-one hundred, Dhauta-washing) process, trituration of ghee with aqueous phase was eventually results in formation of w/o type of emulsion as lipid (cow ghee) phase is major. Further washings with trituration (associated with pressure) reduce the particle size of fat granules. Eventually with successive washings, aqueous phase dominates over lipid phase and results in the phase inversion i.e. formation of o/w emulsion. The washings of ghee for one hundred times could led to the formation of a complex system of emulsion i.e. w/o/w and ghee also change its chemical and physical properties. The fat splitting mechanism shown in fig no.2.

Triglyceride + Water → Glycerine (Glycerol) + Fatty Acid

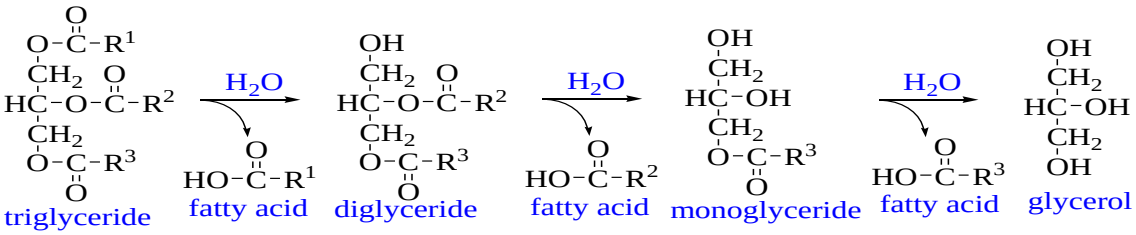

Figure 2: Fat Splitting Process

4.1. Organoleptic Evaluation

The organoleptic characters of Shata-Dhauta-Ghrita like appearance, color, odour and texture were observed. It shown table no.1. The yellow coloring arrives from beta carotene in the cow's butterfat. Due to heating and again washing with water the pigment may leak out into water and change in the colour, odour and texture takes place after washing (Dhauta).

Table 1: Organoleptic Evaluation of Effect on no of Washing on SDG Formulation

No of Washing	Colour	Odour	Texture
0	Golden yellow	Characteristic ghee like	Oily Granular
10	Colour slight changed	Ghee like	Oily Granular
20	Cremish Yellow	Ghee like	Oily yellow cream
40	Slightely Cremish	Slightly change	Oily Homogenous cream
60	Pale yellow	Slightly Change	Oily Homogenous cream
80	Pale yellow	Odorless	Oily Homogenous Cream
100	Light Pale yellow	Odorless	Homogenous cream
120	White	Odorless	Homogenous cream
140	White	Odorless	Homogenous cream

4.2. Effect of Washing on the Weight of Ghee (SDG formulation)

The Cow ghee was washing along with water in different interval of washing like 10, 20,40,60,80,100,120,140 and analysed effect on washing on the weight of ghee. It shown in table no.2. and Fig. no.3. There was 5 g increase in weight during 10th dhauta. The reason may be the passage of water globules in fat molecules by forming the emulsion.Up to 60 washing weight of SDG was increases and after 80 washing it is decreases because washing out fat pigment.

Table 2: Effect of Washing on the Weight of Ghee

Washing time	Initial weight	Volume of water for washing	Final weight
0	50	50	50
10	50	50	55 ±0.288
20	55 ±0.288	50	58.3±0.35
40	58.3±0.35	50	68.3±0.40
60	68.3±0.40	50	72.97±0.66
80	72.97±0.66	50	69.23±0.503
100	69.23±0.503	50	67.11±0.577
120	67.11±0.577	50	65.84±0.223
140	65.84±0.223	50	65.78±0.240

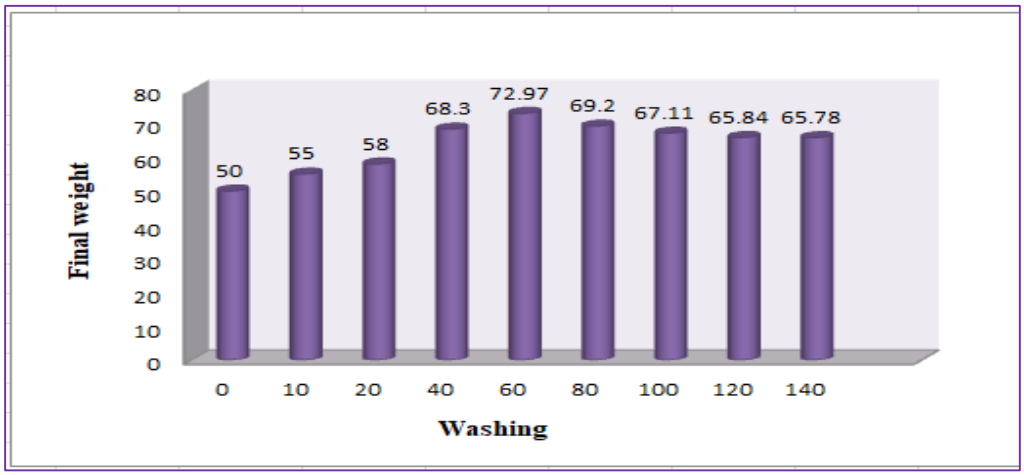

Figure 3: Graphical Represent of Effect of Washing on the Weight of Ghee

4.3. Chemical Characterization

4.3.1. Acid Value

The acid value is the measure of free fatty acids. As oil and fats start to rancidify on storage, triglycerides are converted into fatty acids and glycerol, causing an increase in acid value. The less acid value denotes the less chance of decomposition of ghrita formulation thus increasing life span and therapeutic value. Increasing no of washing of ghee along with water it shown decreasing acid value it shown in fig no.4 and table no 3.

Table 3: Effect of no of Washing on Acid Value of Ghee

Washing	Acid value
0	0.88±0.0057
10	0.807±0.0065
20	0.7573±0.0031
40	0.693±0.0722
60	0.452±0.002
80	0.119±0.058
100	0.091167±0.0015
120	0.09±0.00102
140	0.0877±0.023

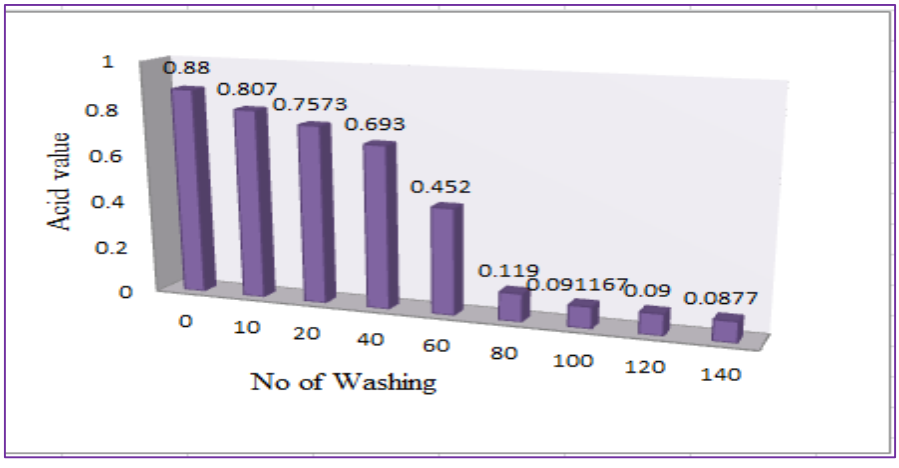

Figure 4: Graphical Represent of Effect of Washing on the Acid Value of Ghee

4.3.2 Saponification Value

The saponification value gives an indication of the number of fatty acids and their average molecular weight in the ghrita formulations. More the fatty matter content or more the carboxylic functional group per unit mass, there will be more chances of rancidity factor and less will be the shelf life and therapeutic value. Increasing no of washing of ghee along with water it shown decreasing Saponification values it shown in fig no.5. and table no.4

Table 4: Effect of no of Washing on Saponification Value of Ghee

Washing	Saponification Value
0	233.61±0.565
10	145.11±0.161
20	97.29±0.294
40	75.4033±0.0221
60	55.3433±0.024
80	27.538±0.036
100	24.979±0.04
120	24.86±0.0088
140	23.91167±0.0058

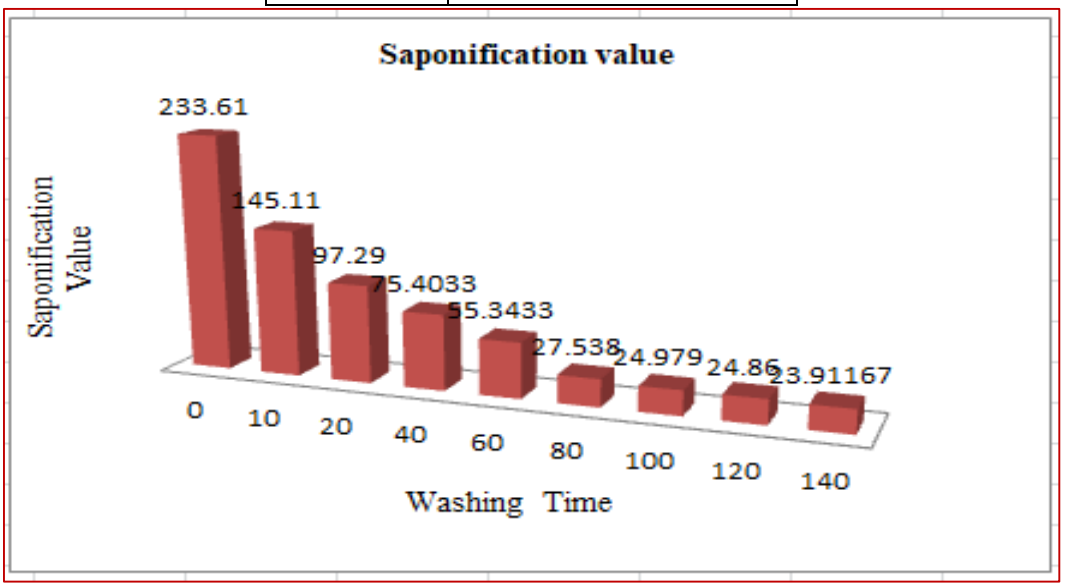

Figure 5: Graphical Represent of Effect of Washing on the Saponification Value of Ghee

4.3.3 Iodine Value

The iodine value indicates quantity of iodine absorbed at unsaturation which signifies the degree of unsaturation of the ghrita formulations. The formulation with higher iodine value is more reactive and susceptible to the oxidation. . Increasing no of washing of ghee along with water it shown decreasing Iodine values it shown in fig no.6 and table no-5

Table 5: Effect of no of Washing on Iodine Value of Ghee

Washing	Iodine value
0	24.26±0.0075
10	17.825±0.0159
20	8.917±0.0068
40	6.426±0.03416
60	4.4776±0.00284
80	3.286±0.0243
100	2.555±0.016
120	2.5240.0092±
140	2.42±0.0447

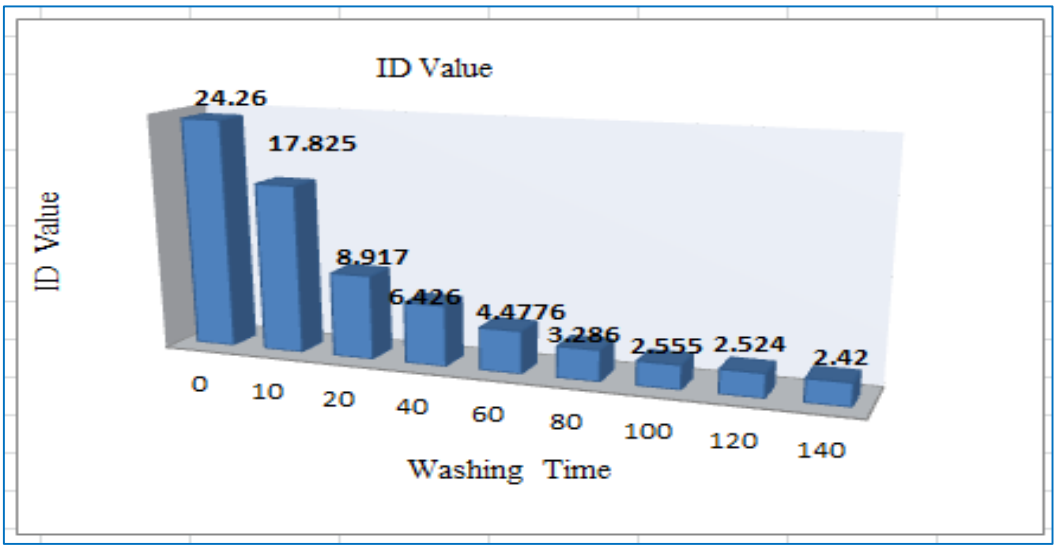

Figure 6: Graphical Represent of Effect of Washing on the Iodine Value of Ghee

4.3.4. Rechert Meissl (RM)

The Rechert Meissl and Polenske values are important indices and principally used for determination of quality of fats of gheebased formulations. The fats of cow ghee can be distinguished from other fats by the presence of glyceryl esters of relatively low molecular weight fatty acids, especially butyric as well as caproic acids. Shown in table no-6 fig.no.7

Table 6: Effect of no of Washing on Iodine Value of Ghee

Washing	RM value
0	21.5±0.1785
10	14.61±0.153
20	7.2986±0.084
40	3.533±0.037
60	1.276±0.0186
80	0.83±0.0186
100	0.588±0.0141
120	0.223±0.0334
140	0.1821±0.1742

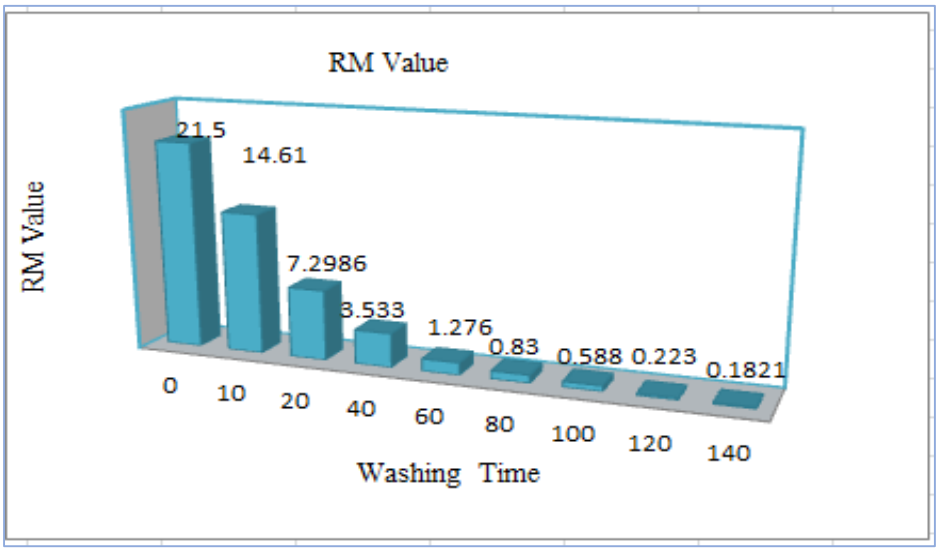

Figure 7: Graphical Represent of Effect of Washing on the RM of Ghee

4.3.5. Polenske value (P value)

The Polenske value measures the steam volatile and water insoluble fatty acids, chiefly caprylic, capric and lauric acids present in ghrita formulations. It shown in table.no 7.and fig.no.8.

Table 7: Effect of no of Washing on Polenske Value of Ghee

Washing	P value
0	1.23±0.01549
10	1.0566±0.026
20	0.974±0.007
40	0.756±0.006
60	0.375±0.125
80	0.1896±0.004
100	0.1186±0.0033
120	0.096±0.0013
140	0.0863±0.0016

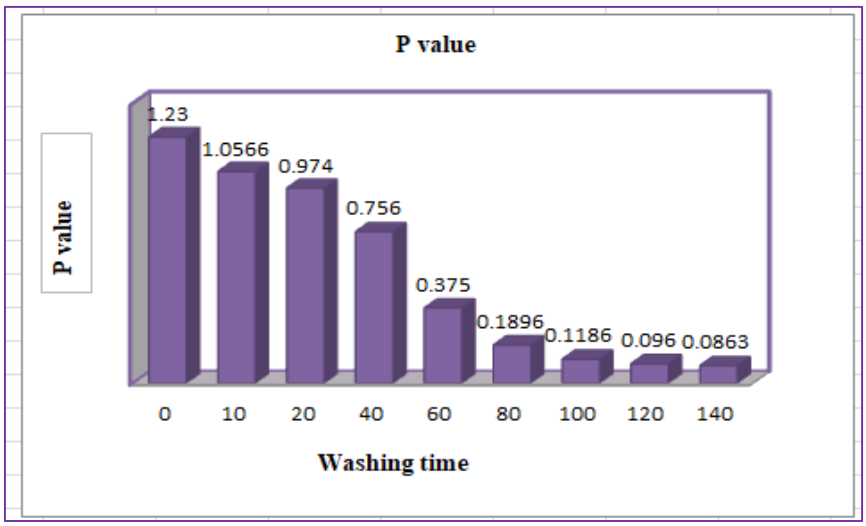

Figure 8: Graphical Represent of Effect of Washing on the Polenske Value of Ghee

4.3.6. Peroxide Value

Usually lipid peroxidation is assumed as a major deteriorative change commonly found in fats and the extent of lipid peroxidation depends upon different attributes viz. unsaturation level, packaging material and storage conditions [32, 48]. The peroxide value was determined to obtain initial evidence of rancidity in formulations. If no of washing increases peroxide value decreases was shown in table no.8 fig no .9

Table 8: Effect of no of Washing on Polenske Value of Ghee

Washing	Peroxide Value
0	2.653 ±0.0033
10	2.4103 ±0.06.6
20	1.289 ±0.0012
40	1.1365 ±0.012
60	1.071 ±0.0295
80	0.987 ±0.0015
100	0.9133 ±0.0012
120	0.9046 ±0.0049
140	0.84 ±0.0173

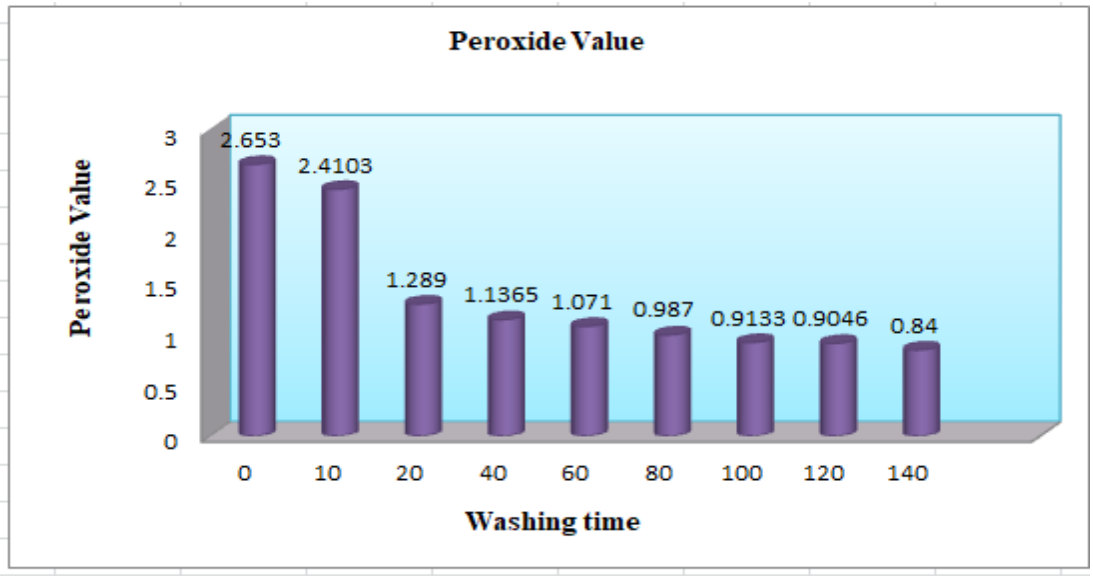

Figure 9: Graphical Represent of Effect of Washing on the Peroxide Value of Ghee

4.3.7. Copper Content

From copper vessel used in Shata-Dhauta ghrita preparation and process in fat splitting seemed to be offering protective effect against rancidity of processed ghrita formulations. The amount of copper found shown in table no 9 and fig.no 10. After successive washing the amount of copper increases.

Table 9: Effect of no of washing on Copper content of ghee

Washing	Copper Content
0	0.0106±0.00066
10	0.117±0.00115
20	0.2363±0.0012
40	0.47266±0.00176
60	0.715±0.0015
80	0.9447±0.0017
100	1.1863±0.234
120	1.213±0.06
140	1.242±0.045

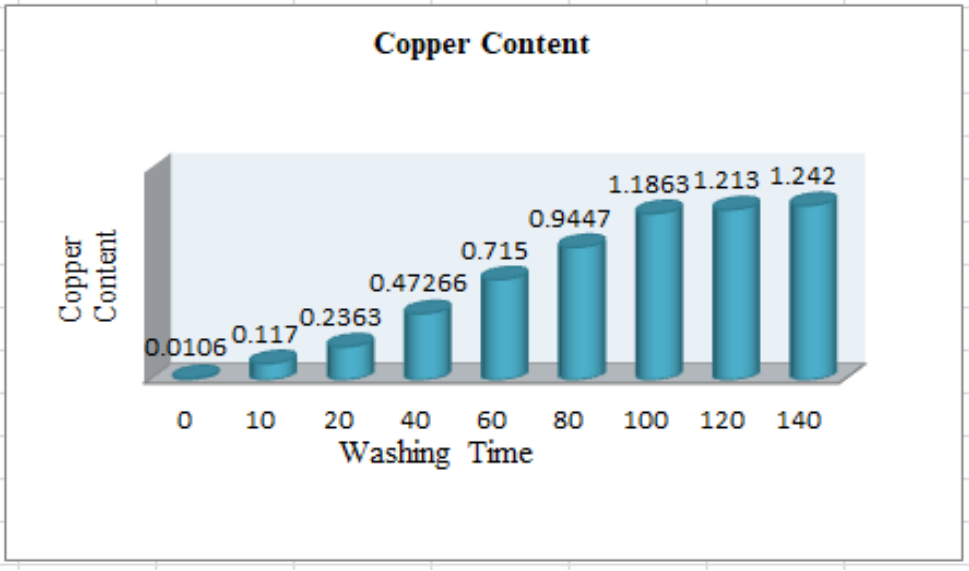

Figure 10: Graphical Represent of Effect of Washing on the Copper Content of Ghee

4.4. Physical Characterization

4.4.1. pH

pH value, measure of hydrogen activity in the formulation conventionally represents the acidity or alkalinity [25]. The pH variations may have impact on flavour, consistency and shelf life of the ghee-based formulations. If the no of washing increases pH value of formulation decreases. It was shown in table no 10 and fig .no.11.

Table 10: Effect of no of Washing on pH of Ghee

Washing	pH
0	4.62 ±0.057
10	4.84 ±0.0088
20	4.9 ±0.035
40	4.95 ±0.035
60	5.82 ±0.008
80	5.9 ±0.0589
100	6.69 ±0.027
120	6.82±0.009
140	6.77 ±0.039

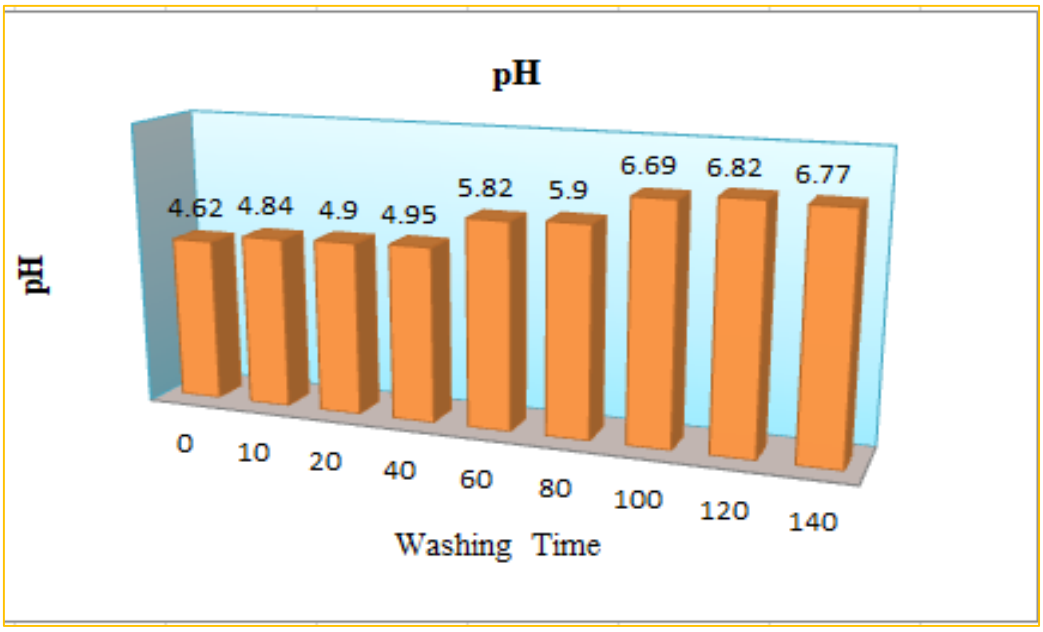

Figure 11: Graphical Represent of Effect of Washing on the pH of Ghee

4.4.2. Viscosity

The viscosity may affect the appearance and the consistency as it measures a resistance of ghrita formulations to the motion under an applied force. According to study washing is increases viscosity also increases the statistical data shown fig no 12 and table no. 11.

Table 11: Effect of no of Washing on Viscosity of Ghee

Washing	Viscosity(CP)
0	6248.667±1.855
10	6540.7±1.201
20	6745±0.577
40	6894.33±1.2018
60	7365±0.333
80	7890.53±0.74
100	9771.66±1.2
120	9852.7±0.90
140	9892.4±1.32

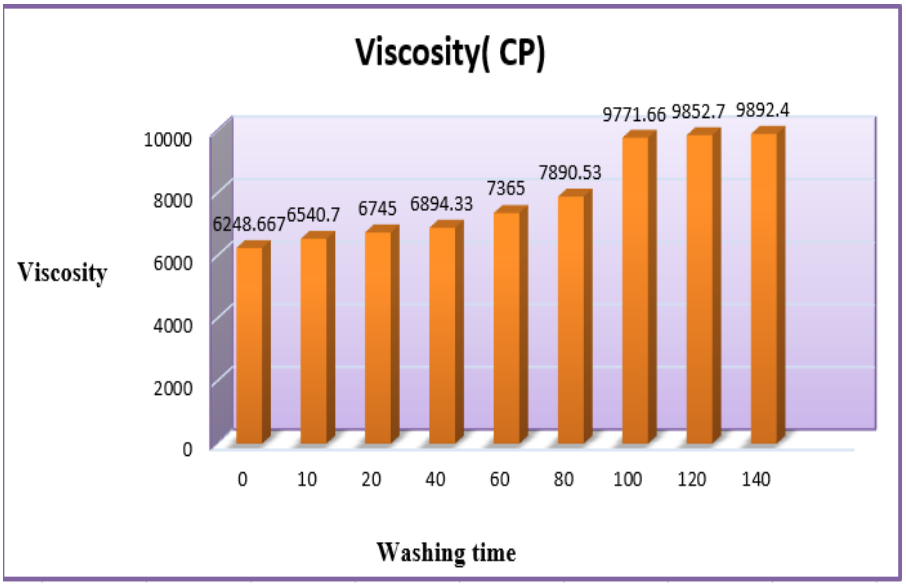

Figure 12: Graphical Represent of Effect of Washing on the Viscosity of Ghee

4.4.3. Refractive Index: The refractive index represents the behaviour of light in the prepared medium, which can be used to determine the concentration of solutes in an aqueous solution. The refractive index increases with decrease in the chain length whereas a double bond elevates the refractive index. Statical data shown in fig no.13 and table no.12.

Table 12: Effect of no of Washing on Viscosity of Ghee

Washing	Refractive index
0	1.447 ±0.0035
10	1.4806 ±0.011
20	1.517 ±0.003
40	1.574 ±0.00305
60	1.592 ±0.00133
80	1.6036 ±0.00317
100	1.611 ±0.0008
120	1.6116 ±0.00251
140	1.624 ±0.00208

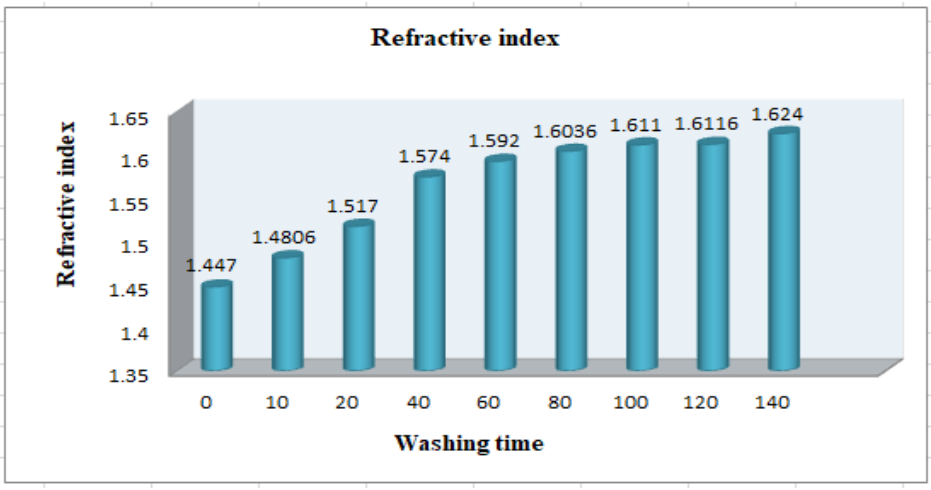

Figure 13: Graphical Represent of Effect of Washing on the Viscosity of Ghee

4.4.4. Identification of Type of Emulsion

Ghrita was triturated along with water and thus formation of water oil (w/o) type of emulsion because water is in dispersed phase and oil is in continuous phase. As the washing continues, due to pressure applied during agitation, particle size of fat granules gets reduced (as per texture it was non granular and smooth). Eventually, successive washings result in o/w type of emulsion. It is possible that it might lead to formation of a complex system like w/o/w emulsion. Data shown in fig.no 14 and table no. 13

Table 13: Effect of no of Washing on Type of Emulsion

Washing time	Type of Emulsion
0	Ghee
10	W/O
20	W/O
40	W/O
60	O/W
80	W/O/W
100	W/O/W
120	WO/W
140	W/O/W

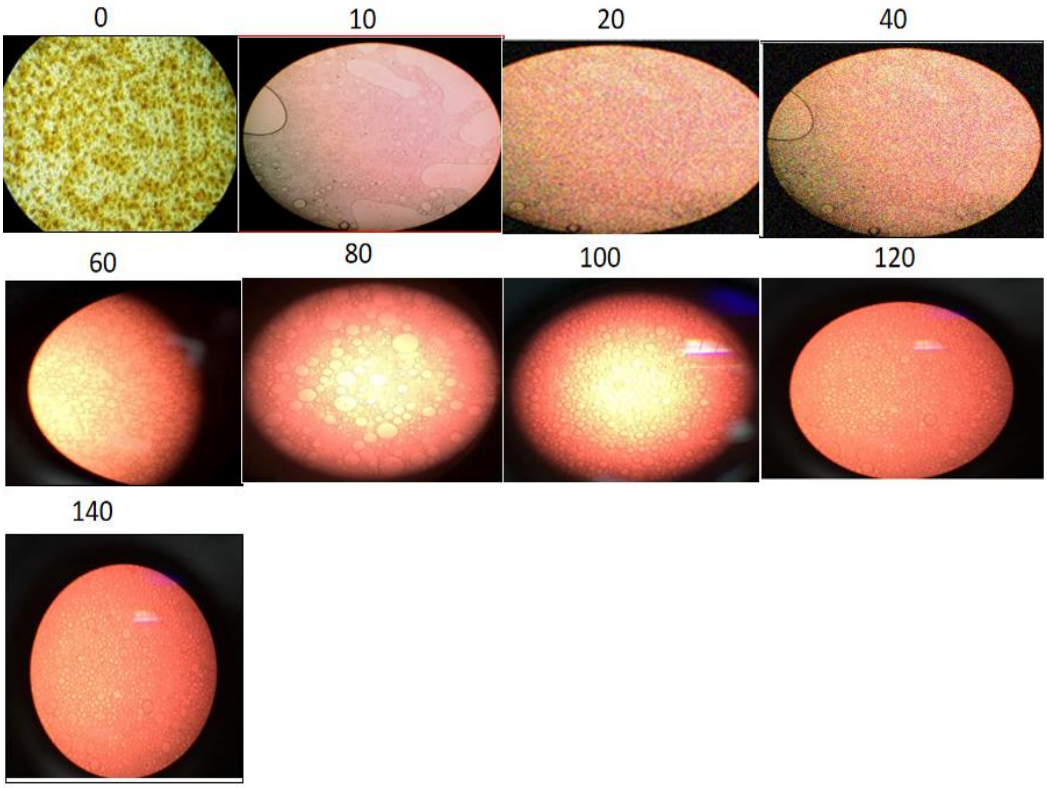

Figure 14: Type of Emulsion

5. CONCLUSION

From the present research work it can be concluded that changes taking place in cow ghee while washing it with hundred times to prepare Shatadhauta ghrita, makes it an elegant and suitable product for topical application. Shata dhauta ghrita is used as natural permeation enhancer in topical product. It is prepared by cow ghee washing with 100 times with water. It is smooth, homogenous, non-oily product, easier to apply and thus improve patient compliance. Chemical analysis and Samskara siddhanta of Ayurveda prove Shatadhauta ghrita is effective and act as good cooling agent, emollient, moisturizer and good scar healer. Shatdhauta ghrita is a popular Ayurvedic medication, but its use in modern medicine has received little attention. The purpose of this study was to look into the utility of Shatdhauta ghrita as a base for topical drug delivery.

Future Prospect

Ayurveda as a prehistoric science of life has a long history and its basic principles may still be compelling today. The call for scientific valuation of Ayurveda has been recognized for a long time. However, the quintessence of any science is a continuous search for new knowledge through research, development and newer applications. It is preferable to conduct ongoing research on the safety, quality, and efficacy of Ayurvedic drugs and procedures. If we combine herbal drugs or phytoconstituent on the Shata dhauta ghrita, it may show an effective herbal formulation

Abbreviations

SDG: Shata dhauta ghrita

RPM: Revolutions per Minute

PPM: parts per million

RM: Reichert Meissl Value

CP: Centipoise

P Value of ghee: Polenske value

NFST: National fellowship of schedule tribal

Acknowledgments

The authors would like to thank the management and Principal Dr. Sanjay Kshirsagar of the MET Institute of Pharmacy in BKC for providing the essential facilities and support. The author would like to express gratitude NFST/RGNF/UGC, Government of India, for providing financial assistance in the form of a fellowship (Award no -2021-NFST-MAH-04091).

Abbreviations

Ethical Approval And Consent Of Participant: Not Applicable

Consent For Publication: Not Applicable

Competing Interest: The Author Declares That They Do Not Have Any Competing İnterests.

Funding: None

Reference

1) BN, A. K., Dixit, S. O., & Patel, S. B. (2016). A Critical Review on Go-Ghrita, Shata Dhouta Ghrita and Sahasra Dhouta Ghrita. *Journal of Ayurveda and Integrated Medical Sciences*, *1*(03), 117-123. https://doi.org/10.21760/jaims.v1i3.4427

2) Gupta, S., Mitra, S., & Prajapati, P. K. (2009). Utility of samskara in pharmaceutics wsr to Sandhana Kalpana. *Indian Journal of Ancient Medicine and Yoga*, *2*(4).

3) Sserunjogi, M. L., Abrahamsen, R. K., & Narvhus, J. (1998). A review paper: current knowledge of ghee and related products. *International Dairy Journal*, *8*(8), 677-688. https://doi.org/10.1016/S0958-6946(98)00106-X

4) Bajaj, K.K. *et al.* (2022) 'Panchgavya: A precious gift to humankind', *Journal of Ayurveda and Integrative Medicine*. The Authors, p. 100525. doi:10.1016/j.jaim.2021.09.003. https://doi.org/10.1016/j.jaim.2021.09.003

5) Wayal, S. R., & Gurav, S. S. (2020). Bhallatakadi ghrita: development and evaluation with reference to murcchana and shata-dhauta process. *Journal of Ayurveda and Integrative Medicine*, *11*(3), 261-269. https://doi.org/10.1016/j.jaim.2020.05.005

6) Sindhuja, S., Prakruthi, M., Manasa, R., & Shivananjappa, M. (2020). Health benefits of ghee (clarified butter)-A review from ayurvedic perspective. *IP J Nutr Metab Health Sci*, *3*(3), 64-72. http://dx.doi.org/10.18231/j.ijnmhs.2020.014

7) Kodituwakku, S. D. (2017, December). Review on Ghee and It's Ayurvedic Uses. In *International Conference on Ayurveda Traditional Medicine and Medicinal Plant* (pp. 1-7).

8) Alnuqaydan, A. M., & Sanderson, B. J. (2016). Toxicity and genotoxicity of beauty products on human skin cells. http://dx.doi.org/10.4172/2161-0495.1000315

9) Sharma, P. V. (2000). Caraka Samhita (Text with English Translation). *Indriya sthana*, 76

10) Babu, B., Ravi, M., Kumar, A. B., Sudheendra, V. H., & Ravishankar, B. (2015). Burn wound healing potential of Plain gritha, Shatadhauta ghrita and Sahasradhauta ghrita on wistar albino rats. https://nopr.niscpr.res.in/handle/123456789/32083

11) Moholkar, S. A., Mukherjee-Kandhare, A. A., Worlikar, P. S., & Tilak, A. (2020). Synergistic Potential of Honey and Shatdhaut Ghrut in the management of Diabetic Foot Ulcer in the Experimental rats. *Research Journal of Pharmacy and Technology*, *13*(3), 1377-1385. http://dx.doi.org/10.5958/0974-360X.2020.00253.X

12) Pallavi, C., (2017). A Clinical Study of Dashmool Bharad Kwath and Tila Taila Yoga Basti in Kashtartava (Dysmenorrhoea) - A Case Study. Aayushi International Interdisciplinary Research Journal (AIIRJ); 4(6):5–11 https://www.aiirjournal.com/uploads/Articles/2017/05/875_08.Dr.%20Pallavi%20Ashok%20Chandanshiv.pdf

13) Matu, M. S., Vijay B.,(2015). Study the role of shatdhaut ghrita in parikartika (fissure in ano). International Ayurvedic Medical Journal; 3(8), 2249-2252 http://www.iamj.in/current_issue/images/upload/2249_2252.pdf

14) Majumdar, S. H., Kulkarni, A. S., Aloorkar, N. H., & Shinde, M. D. Exploring Shat Dhaut Ghrita As Formulation Base For The Topical Delivery Of Curcuma Amada. https://www.academia.edu/download/50431724/36-1470740983_002.pdf

15) Pulate, C. (2021) 'Multidisciplinary Applications Of Tradition And Technology', *Internal Journal of Biology, Pharmacy and allied Sciences*, 10(7), pp. 2418–2429.

16) Ravindra, R. P., & Muslim, P. K. (2013). Comparison of physical characteristics of vanishing Cream base, cow ghee and shata-dhautaghrita as per pharmacopoeial standards. *International Journal of Pharma and Bio Sciences*, *4*(4), 14-21.

17) Pande, V. V., Patel, V. P., Zarekar, N. S., Pandit, S. R., & Pote, A. K. (2020). Investigation of effects of shata dhauta ghrita on the skin permeation of fluconazole loaded topical antifungal nanolipogel. *Journal of Pharmaceutical Sciences and Research*, *12*(7), 948-956.

18) Kohl, E., Steinbauer, J., Landthaler, M., & Szeimies, R. M. (2011). Skin ageing. *Journal of the European academy of dermatology and venereology*, *25*(8), 873-884.

19) Zhang, S., & Duan, E. (2018). Fighting against skin aging: the way from bench to bedside. *Cell transplantation*, *27*(5), 729-738. https://doi.org/10.1177/0963689717725755

20) Baumann, L. (2007). Skin ageing and its treatment. *The Journal of Pathology: A Journal of the Pathological Society of Great Britain and Ireland*, *211*(2), 241-251. https://doi.org/10.1002/path.2098

21) Wadhwa, K., Kadian, V., Puri, V., Bhardwaj, B. Y., Sharma, A., Pahwa, R., ... & Singh, I. (2022). New insights into quercetin nanoformulations for topical delivery. *Phytomedicine Plus*, 100257. https://doi.org/10.1016/j.phyplu.2022.100257

22) Corrêa, R. C., Peralta, R. M., Haminiuk, C. W., Maciel, G. M., Bracht, A., & Ferreira, I. C. (2018). New phytochemicals as potential human anti-aging compounds: Reality, promise, and challenges. *Critical reviews in food science and nutrition*, *58*(6), 942-957. https://doi.org/10.1080/10408398.2016.1233860

23) Parihar, B. K., Thomas, S., Hussain, G., Kadibagil, V. R., & Lakshkar, A. (2016). Pharmaceutical Study of Shatadhauta Ghrita Prepared By Two Different Methods. *Volume*, *5*, 1769-1778.

24) Yadav, A., Gaud, R. S., & Yeole, P. (2016). *Pharmaceutics*. Pragati Books Pvt. Ltd., 33-34

25) Siddhanand, M. A. (2001). Bhaishajyakalpanavidnyan, Chaukumbhe Vidyabhavan, 405-409

26) Bockisch,M.(1998). Analytical Methods. In: Fats and Oils Handbook. AOCS Press, 803-808 https://doi.org/10.1016/B978-0-9818936-0-0.50013-5

27) Odoom, W., & Edusei, V. O. (2015). Evaluation of saponification value, iodine value and insoluble impurities in coconut oils from Jomoro District of the Western Region of Ghana. *Asian Journal of Agriculture and Food Sciences*, *3*(5).

28) Dhurvey Y.R., Kawtikwar P.S., S.D.M. (2012). 'Evaluation of physicochemical properties of cow ghee before and after hydrogenation', International *Journal of ChemTech Research*, 4(1). 185–189.

29) Committee, Ayurvedic Pharmacopoeia (1978). *The Ayurvedic Formulary of India*. Government of India, Ministry of Health and Family Planning, Department of Health. 298-304

30) Nadkarni, A. K., & Km, N. (1955). *Indian materia medica*. 72 https://openlibrary.org/books/OL6173564M/Indian_materia_medica.

31) Committee, *Indian Pharmacopoeia,* (2018). Government of India, Ministry of Health and Family Planning, Department of Health. 142-151

32) Dr. Sudheendra V. H. A handbook of standardization of ayurvedic formulations, chaukhumba orientalia, Varanasi. 201-250

33) Beckett, A. H., & Stenlake, J. B. (Eds.). (1988). *Practical pharmaceutical chemistry: Part II fourth edition* (Vol. 2). A&C Black.

34) Watson, D. G. (2020). *Pharmaceutical analysis E-book: a textbook for pharmacy students and pharmaceutical chemists*. Elsevier Health Sciences.

35) Chen, M. X., Alexander, K. S., & Baki, G. (2016). Formulation and evaluation of antibacterial creams and gels containing metal ions for topical application. *Journal of pharmaceutics*, *2016*.

36) Wayal, S. R., & Gurav, S. S. (2020). Bhallatakadi ghrita: development and evaluation with reference to murcchana and shata-dhauta process. *Journal of Ayurveda and Integrative Medicine*, *11*(3), 261-269. https://doi.org/10.1016/j.jaim.2020.05.005

37) Dr. Minakshi M. N. and Dr. Pallavi B. (2022). Conceptual review of shatadhuta ghrita- an external application. World Journal of Pharmaceutical Science. 11(10):186–91. https://doi.org/10.20959/wjpps202210-23213