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Quality Control Assessment of an Ayurvedic Medicine - *Durvadi Ghrita*

Research Article

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Abstract

Background: *Durvadi Ghrita* is a *Sneha Kalpana* which is claimed to be effective in *Madhumehajanya Timira* (Diabetic Retinopathy). In present study, it has been used for *Nasya*. Objective: Present study was planned to look out on herbal drugs used in the preparation of *Durvadi Ghrita* and standardization of drug by pharmacognostical and physicochemical parameters and HPTLC evaluation. Methods: Identification and authentication of all the raw drug was done by pharmacognostical study i.e. morphological characters, organoleptic characters and powder microscopy. Physicochemical evaluation and HPTLC of final product were done. Results: Pharmacognostical study of all the raw drugs of *Durvadi Ghrita* showed presence of oil globule, prismatic crystals of *Durva.* Lignified branched trichome, pollen grains of *Utpala Kinjalaka.* Trichome, border pitted vessels of *Manjishtha*. Collenchyma cells, border pitted vessel of *Elvaluka.* Lignified fibres, oil globules of *Sita*. Pitted fibres, pitted vessels of *Usheera.* Scalariform vessels, prismatic crystals of *Musta*. Pitted vessels and lignified fibres, crystal fibres of *Chandana*. Lignified cork, and stone cells of *Padmaka* etc. Pharmaceutical evaluation of *Durvadi Ghrita* showed results Specific Gravity 0.9125, Refractive Index 1.47, Acid Value 0.4608, Iodine Value 11.45 and Saponification Value 128.856. High Performance Thin Layer Chromatography, 12 spots were found at 254 nm and five spots were found at 366 nm. Conclusion: Identification and authentication of herbal drug used in the preparation of *Durvadi Ghrita* has been done. Pharmacognostical and physicochemical evaluation of prepared drug has been carried out which can be further useful for standardization of *Durvadi Ghrita* and other clinical researches.

Key Words: *Diabetic Retinopathy, Durvadi Ghrita, Nasya, Standardization.*

Introduction

Durvadi Ghrita is one of the herbal formulations which described in Ayurvedic text *Sahasrayogam- Ghrita Prakarana* (1). This preparation contains polyherbal drugs like *Durva, Utpala Kinjalaka, Manjistha, Elvaluka, Shweta Chandana, Sita, Musta, Usheera, Padmaka* and *Rakta Chandana* are used as *Kalka Dravya*, *Aja Ksheera* and *Tandulodaka* as *Drava Dravya* and *Aja Ghrita* as *Sneha Dravya*. The *Ghrita Paka* was done for three days as per classics (2). It is specially indicated in bleeding disorders. Based on its pharmacological properties, it can be used trans- nasally to arrest bleeding seen in Diabetic retinopathy. Diabetic retinopathy (DR) is an important complication of diabetes mellitus (DM) and the leading cause of visual disturbances in developed countries (3). The pathogenesis of DR includes loss of integrity of capillary walls, micro aneurysms, exudations, pericyte loss, endothelial damage, retinal haemorrhages which lead to visual disturbance initially and turn in to blindness finally. *Durvadi Ghrita* has *Pittasamana*, *Raktastambhana* and *Raktaprasadana* properties, can be used in DR specifically to overcome haemorrhage under *Urdhvanga Raktapitta* spectrum.

Standardization of the drug is very important to assess the quality, purity, safety and efficacy of the drug. Present study, is planned to develop quality parameters of *Durvadi Ghrita* on the basis of pharmacognostical (microscopic) study, physicochemical study and chromatographic evaluation which is useful for future reference. Hence, there is a need of standardization of quality parameters. Therefore, the present study was designed to evaluate the quality parameters of *Durvadi Ghrita*.

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Materials and Methods

Collection of drugs

Most of the raw drugs for *Durvadi Ghrita* were procured from the Pharmacy of Gujarat Ayurved University, Jamnagar. *Aja Kshreera* and *Aja Ghrita* were procured from local milk man of Jamnagar. *Elvaluka* and *Utpala Patra* were collected from Shri Narayana Aushadha Bhandar, Jamnagar. *Tandulodaka* was prepared in Pharmacy of Gujarat Ayurved University, Jamnagar. According to the guideline of Ayurvedic Pharmacopoeia of India (4), raw drugs were identified and certified and authenticated by individual powder microscopy in Pharmacognosy department I.P.G.T. & R.A., Jamnagar.

Table 1: Contents of modified *Durvadi Ghrita*

Sr. No.	Drug	Botanical Name	Part used	Quantity
1	*Durva*	*Cynodon dactylon* Linn.	Whole plant	1part
2	*Utpala Kinjalaka*	*Nymphaea caerulea* Sav.	Stamens	1 part
3	*Manjistha*	*Rubia cordifolia* Linn.	Whole plant	1 part
4	*Elvaluka*	*Prunus cerasus* L.	Resin	1 part
5	*Sita*	Sugar (Eng. name)	--	1 part
6	*Shweta Chandana*	*Santalum album* Linn.	Stem	1 part
7	*Usheera*	*Vetiveria zizanioides* Linn.	Root	1 part
8	*Musta*	*Cyperus rotundus* Linn.	Root	1 part
9	*Rakta Chandana*	*Pterocarpus santalinus* Linn.	Heart Wood	1 part
10	*Padmakam*	*Prunus puddum* Roxb.	Heart Wood	1 part
11	*Aja Ghrita*	Goat's ghee (Eng. name)	--	4 part
12	*Tandulodaka*	Rice water (Eng. name)	--	4 part
13	*Aja Ksheera*	Goat's milk (Eng. name)	--	4 part

Preparation of drug

After getting all the ingredients of *Durvadi Ghrita*, first of all *Aja Ghrita* was taken in large vessel. *Kalka Dravya* of *Durva, Utpala Kinjalaka, Manjistha, Elvaluka, Shweta Chandana, Sita, Musta, Usheera, Padmaka* and *Rakta Chandana* were made into bolus of *Kalka* form by adding sufficient water. This bolus of *Kalka* was added to *Ghrita* when it got melted. Then *Aja Ksheera* and *Tandulodaka* were added slowly. The ratio of *Kalka: Sneha: Drava: Dravya* is 1/16: 1: 4. Throughout the procedure the temperature of heating source was maintained. So, as to generate only bubble in the mixture. The heating was continued till *Sneha Sidhdha Lakshanas* were observed. The mixture was filtered through four folded fine cotton cloth two times after it got partially cooled, and packed in a sterile jar.

Organoleptic characters

With the help of *Panchagyanendriya* (Examination by the sense Organs), organoleptic parameters like colour, texture, odour, touch and taste of the finished products were observed and recorded (5).

Powder microscopy

For *Durvadi Ghrita*, we used *Kalka Dravyas* like *Durva, Utpala Kinjalaka, Manjistha, Elvaluka, Shweta Chandana, Sita, Musta, Usheera, Padmaka* and *Rakta Chandana.* So, it is difficult to examine and analyse the *Durvadi Ghrita* to find out the cellular level of raw drugs. Thus, for powder microscopy study, pinch of powder of *Kalka Dravya* was taken in glass slide covered by cover slip and then stained with phloroglucinol and hydrochloric acid to observe the lignification of the cell wall (6). The sample was studied under the Carl Zeiss Trinocular microscope attached with camera and microphotographs were also taken (7,8).

Physicochemical parameters

Durvadi Ghrita was analyzed by using various qualitative and quantitative parameters at pharmaceutical chemistry laboratory, IPGT & RA, GAU, Jamnagar. The common parameters mentioned in Ayurvedic Pharmacopeia of India and CCRAS (9) guidelines i.e. specific gravity(10), refractive index(11), acid value(12), iodine value(13), and saponification value(14) were taken.

High Performance Thin Layer Chromatography (HPTLC)

Sample preparation 0.1 ML of ghee was taken and 1 ML of hexane was added. The Solution was prepared used for chromatography. Thereafter pre chromatographic derivatization was done. Alcoholic KOH (base) and thereby heated for 10-15 minutes in CAMAG TLC plate heater. Sample application was done using CAMAG linomat 5. HPTLC of *Durvadi Ghrita* was carried out using the solvent system petroleum Ether: Diaethyl Ether: Acetic Acid (9:1:0.1v/v). HPTLC study was performed for the normal phase separation of components of product. Post chromatographic derivatization was done with vanillin sulphuric acid spray reagents (15).

Observations and Results

Organoleptic Characters

Organoleptic characters like colour, odour, taste, touch and texture of *Durvadi Ghrita* are shown in Table 2.

Table no. 2 - Organoleptic Characteristics of *Durvadi Ghrita*

Sr. No.	Characteristics	Results
1	Colour	Golden Yellow
2	Odour	Ghee smell
3	Taste	Sweet Astringent
4	Touch	Soft
5	Texture	Thick liquid

Microscopic Characters of *Durvadi Ghrita*

Pharmacognostical characters of *Durvadi Ghrita* were observed under the microscope were silica deposits, oil globule, prismatic crystals and epidermal cells with stomata of *Durva.* Lignified branched trichome, simple fibres, pollen grains and simple starch

grains with hilum of *Utpala Kinjalaka.* Trichome, border pitted vessels, starch grain, acicular crystals colouring matters of *Manjishtha*. Collenchyma cells, cork cells in surface view and border pitted vessel of *Elvaluka.* Lignified fibres, oil globules, rhomboidal crystals, border pitted vessels of *Shita*. Pitted fibres, pitted vessels, group of fibres of *Usheera.* Scalariform vessels, prismatic crystals, oil globules and silica deposits of *Musta*. Pitted vessels and lignified fibres, crystal fibres, pitted vessels and lignified fibres with oil of *Chandana*. Lignified cork, simple fibres, crystal fibres and stone cells of *Padmaka*. Details of which are depicted in plate no: 1.

Physicochemical analysis

Result of physicochemical analysis of *Durvadi Ghrita*; specific gravity, refractive index value, acid value, iodine value and saponification value are shown in Table 3.

Table 3: Physico-chemical parameters:

No.	Parameters	Result
1	Specific Gravity	0.9125% w/w
2	Refractive Index	1.4700% w/w
3	Acid value	0.4608% w/w
4	Iodine value	11.458% w/w
5	Saponification Value	128.856% w/w

High performance thin layer chromatography (HPTLC):

The colour and R_f value of resolved sports of HPTLC were noted. HPTLC Results of *Durvadi Ghrita* showed 12 spots at 254 nm and 5 spots at 366 nm. Detailed results are shown in the table 4. (Plate no. 2)

Table 4: R_f values obtained by HPTLC

Sample	Detection Condition	No. of spots	Rf value
Durvadi Ghrita	254 nm	12	0.02, 0.05, 0.32, 0.37, 0.42, 0.46, 0.50, 0.55, 0.59, 0.82, 0.84, 0.90
	366nm	5	0.02, 0.32, 0.37, 0.48, 0.90

Plate 1. Powder microscopic photographs of *Durvadi Ghrita*

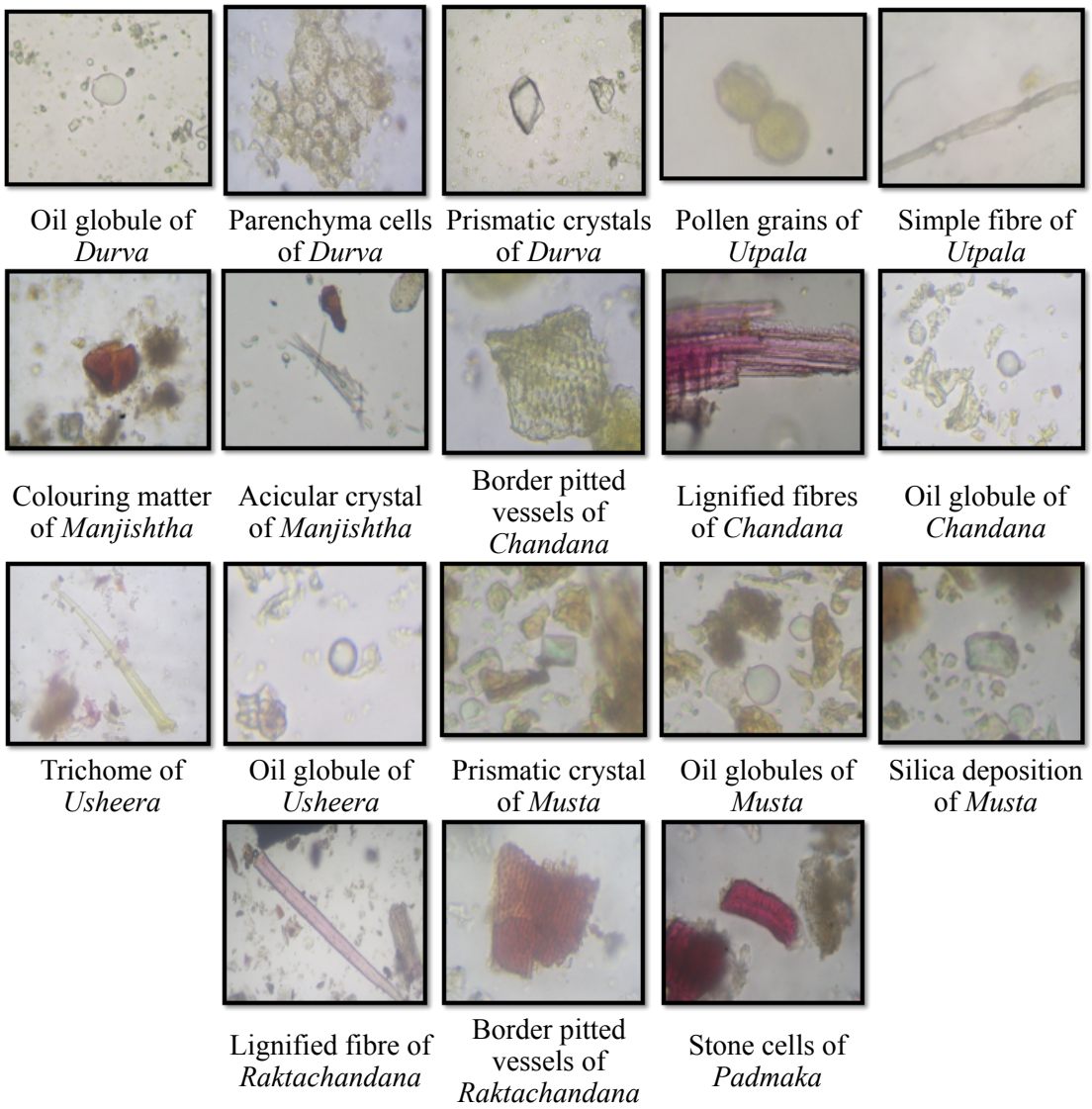
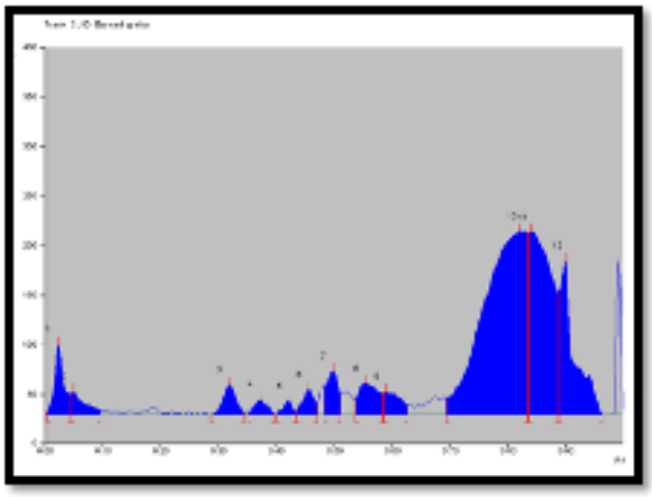
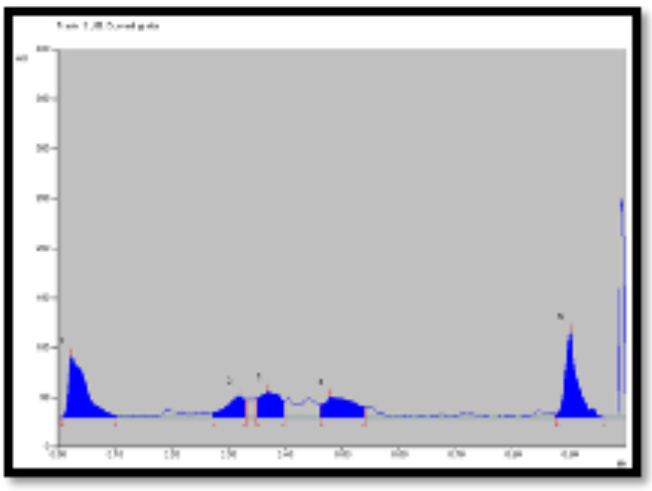


Plate 2. HPTLC of methanolic extract of *Durvadi Ghrita* observed under short UV Light:



Peak display at 254 nm



Peak display at 366 nm

Discussion

In the present study, possible and suitable techniques were taken for the quality evaluation of *Durvadi Ghrita*. Organoleptic characters like colour, odour, taste, touch and texture of *Durvadi Ghrita* are according to the raw drugs, used to prepare the medicated *Ghrita*. The *Durvadi Ghrita* is golden yellow colour, sweet astringent, soft and viscous liquid with characteristic odour. Authentication of used drugs was done by histological and morphological examination. This can prevent misuse of drug adulteration. The pharmacognostical evaluation showed microscopic characters of all the content which were used in drug preparation. This can prove the purity and quality of finished product.

Specific gravity indicates the presence of solute content in the solvent; the value (0.9125) for the drug was appropriate for this medicated ghee (16). Refractive index indicates the density of sample as compared to air and liquid media and the value found to be 1.4700 of medicated *Ghrita* was within the limit (17). The value is a measure of the amount of fatty acids in the *Ghrita* which have been liberated by hydrolysis from the glycerides due to the action of moisture, temperature and/or lipolytic enzyme lipase. It is responsible for rancidity of product; this helps to decide the shelf life of the *Ghrita*; acid value for *Durvadi Ghrita* was found to be 0.4608 thus indicating the good stability of the finished product. Iodine value are used to determine the amount of unsaturation in ghee; higher the iodine value, the more unsaturations are present in the ghee. The degree of unsaturation higher will be the possibility of absorption and atmospheric oxidation leading to rancidity (18). The iodine value of *Durvadi Ghrita* was 11.458 found to be fair enough which indicates the less rancidity of this formulation. The saponification value allows for comparison of the average fatty acid chain length. The long chain fatty acids found in fats have a low saponification value because they have a relatively fewer number of carboxylic functional groups per unit mass of the fat as compared to short chain fatty acids.

In HPTLC of *Durvadi Ghrita*, twelve major spots were observed at 254 nm and five major spots were observed at 366 nm [Table 4, plate 2] indicating its possible compounds of the matrix which may be responsible for its therapeutic activity. These findings could be helpful in identification and authentication of the drug.

Conclusion

Present study reveals that quality of *Durvadi Ghrita* as per pharmacognostical and physico chemical parameters, which helps in justifying the quality of formulation and meets the maximum quality and purity standards of the drug. Chromatographic study results suggest presence of active herbal drug in the lipid formulation. On the basis of observations and experimental results, this study may be used as reference standard in the further quality control research work and clinical studies.

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