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PHARMACOGNOSTICAL AND PHARMACEUTICAL ANALYSIS OF *PANCHATIKTAGHRITA GUGGLU* AN AYURVEDIC POLYHERBAL FORMULATION

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ABSTRACT

Background: *Panchatikta Ghrita Guggulu* is a polyherbal formulation mentioned in *Ashtanga Sangraha* containing various Ayurvedic medicinal drugs and specially indicated for the treatment of *Asthi-Sandhi-Majjagata Vata*. For assurance of quality of herbal compounds pharmacognostical and pharmaceutical analysis should be done. **Methods:** *Panchatikta Ghrita Guggulu* was subjected to microscopic evaluation for Pharmacognostical study, physiochemical analysis like hardness, weight variation, loss on drying, ash value, acid insoluble extract, pH value, water soluble extract, alcohol soluble extract, high performance thin layer chromatography (HPTLC). **Results:** Pharmacognostical study showed the presence of certain identifying characters of all of the ingredients of *Panchatikta Ghrita Guggulu* that is *Nimba, Guduchi, Patola, Kantakari, Vasa, Gugglu, Patha, Vidanga, Devadaru, Gaja Pipalli, Yavakshra, Sarjikshara, Shunthi, Haridra, Mishi, Chavya, Kushtha, Tejovati, Maricha, Kutaja, Yavani, Chitraka, Katurohini, Bhallataka, Vacha, Pipallimoola, Rasna, Manjishtha, Ativisha, Goghrita*. In pharmaceutical study, preliminary physiochemical analysis showed that hardness of the *Vati* was 4.05 Kg/cm2, ash value 5.83% w/w, loss on drying 9.89% w/w, water soluble extract 23.9% w/w, alcohol soluble extract 32.7% w/w and HPTLC showed 8 spots in 254nm and 5 spots in 366nm. **Conclusions:** Present work was carried out to standardize the polyherbal formulation *Panchatikta Ghrita Guggulu* in terms of its identity, quality and purity. Pharmacognostical and physico-chemical observations revealed the specific characters of all active constituents in the preparation were present in it.

KEYWORDS: *Panchatikta Ghrita Gugglu*, Pharmacognosy, Pharmaceutics.

INTRODUCTION

Panchatikta Ghrita Guggulu, a polyherbal formulation contains various herbal drugs (Table 1) that is *Nimba, Guduchi, Patola, Kantakari, Vasa, Gugglu, Patha, Vidanga, Devadaru, Gaja Pipalli, Yavakshra, Sarjikshara, Shunthi, Haridra, Mishi, Chavya, Kushtha, Tejovati, Maricha, Kutaja, Yavani, Chitraka, Katurohini, Bhallataka, Vacha, Pipallimoola, Rasna, Manjishtha, Ativisha, Goghrita. Panchatikta Ghrita Guggulu* is mainly indicated for the treatment of *Sandhi-Asthi-Majjagata Vata* in a classical text of Ayurveda like *Ashtanga Sangraha Chikitsa Sthana Vatavyadhi Chikitsa*.[1] It is also indicated for the treatment of *Kushtha, Nadi Vrana, Arbuda, Bhagandara, Gndamala, Urdhvajatrugata Roga, Gulma, Arsha, Prameha, Yakshma, Aruchi, Shwasa, Pinasa, Kasa, Shosha, Hridaya Roga, Pandu, Mada Vidradhi* and *Vatarakta*. Ingredients of *Panchatikta Guggulu* are having *Katu, Tikta, Kashaya Rasa, Laghu, Ushna* and *Ruksha Guna, Ushna Virya* and *Katu Vipaka*. Thus, *Panchatikta Ghrita Guggulu* mainly pacify *Kapha* and *Vata Dosha*. In the case of internal administration of hrebomineral drug, it should be safe, effective and free from adulteration, with appropriate quantity and ingredients. It is difficult to identify herbal drug in dry or powdered form.

This condition leads to increase in adulteration. So, it is a need of time to set proper parameters for standardization of herbal drugs. Pharmacognostical studies reveals plant identification and sets parameters for standardization which can be done in the case of herbal traditional medicine. Generally, physiochemical analytical study of drugs help to interpret the pharmacokinetics and pharmacodynamics involved. With the help of physiochemical analytical studies, it is possible to standardize the drug and differentiate the adulterants.

High performance liquid chromatography (HPLC) and thin-layer chromatography (TLC) are the conventional methods used in the analysis of secondary metabolites originating from plants. It is necessity of time in the field of Ayurveda to go for quality control of the raw drugs as well as final products using modern parameters which provides credibility to Ayurvedic medicines and also help in the globalization of Ayurveda.

Objectives of these studies are to evaluate the authenticity of *Panchatikta Ghrita Guggulu* through various pharmacognostical procedures and to develop the pharmacognostical and phyto-chemical profile of *Panchatikta Ghrita Guggulu.*

METHODS

Collection identification and authentication of raw drugs

The raw materials were purchased from local market of Jamnagar, Gujarat. All the raw drugs were identified and authenticated in the Pharmacogonosy laboratory, Institute for Post Graduate Teaching and Research in Ayurveda, Gujarat Ayurved University, Jamnagar, Gujarat, India.

Preparation of Drug

All raw drugs were purchased from local market of Jamnagar, Gujarat. Murchana of Goghrita was done and kept on side. Decoction was prepared from coarse powder of *Nimba, Guduchi, Vasa, Patola, Kantakari* with 8 part of water. It was boiled until water was reduced up to 4th part. Then decoction was filtered. Fine powder of all Kalka Dravyas i.e. *Patha, Vidanga, Devadaru, Gaja Pipalli, Yavakshra, Sarjikshara, Shunthi, Haridra, Mishi, Chavya, Kushtha, Tejovati, Maricha, Kutaja, Yavani, Chitraka, Katurohini, Bhallataka,Vacha, Pipallimoola, Rasna, Manjishtha, Ativisha* was made and then paste was made by adding water in it. After this, Kalka and Kwatha Dravyas were added to Murchita Ghrita and paka was done on Mandagni (Medium flame). *Shuddha Guggulu* was also added to this mixture in Ghrita and was boiled. When whole aqueous part was evapourated the mixture was filtered through cloth. Whole product allowed to cool at room temperature. Then *Vati* of 250 mg was prepared and stored in bottles under hygienic condition.

Pharmacognostical study

The pharmacognostical study was divided in to organoleptic study and microscopic study of the finished product.

Organoleptic study

The genuinity of the polyherbal formulation can be fined with organoleptic characters of the given sample. Organoleptic parameters comprise taste, colour, odour and touch of *Panchatikta Ghrita Guggulu* which was scientifically studied as per the standard references.[2]

Microscopic study

Panchatikta Ghrita Guggulu was powdered and dissolved with water and microscopy of the sample was done without stain and after staining with Phloroglucinol + HCl.

Microphotographs of *Punarnava Guggulu* were also taken under Corl-zeisstrinocular microscope.[3]

Physico-chemical analysis

With the help of various standard physico-chemical parameters, *Panchatikta Ghrita Guggulu* was analyzed. The common parameters mentioned for *Guggulu Kaplana* in Ayurvedic Pharmacopia of India, and CCRAS, guidelines are loss on drying, hardness, total ash value, acid insoluble ash, pH value, water soluble extract, methanol soluble extra total ash and water and alcohol soluble extractives.[4],[5]

High performance thin layer chromatography[6]

High Performance Thin Layer Chromatography (HPTLC) is a powerful analytical method suitable for the separation and quantitative determination of a considerable number of compounds even from complicated matrix. HPTLC is used for identification of active constituents, identification and determination of impurities and quantitative analysis of active constituents. Principle of HPTLC remains the same as of TLC i.e. adsorption. One or more compounds can be spotted in a thin layer of adsorbent coated on a chromatographic figure. The mobile phase solvent flows through because of capillary action against gravitational force. The component with more affinity towards stationary phase travels faster. Thus, the components are separated on a thin layer chromatographic figure based on the affinity of the components towards the stationary phase.

Steps involved in HPTLC were as follows

- Sample and standard preparation
- Selection of chromatographic layer
- Layer pre-washing
- Layer pre-conditioning
- Application of sample
- Chromatographic development
- Detection of spots
- Scanning and documentation.

Methanol extract of *Panchatikta Ghrita Guggulu* were spotted on pre-coated silica gel GF CO254 aluminum figure as 5 mm bands, 5 mm apart and 1 cm from the edge of the figures, by means of camag, linomate V sample applicator fitted with a 100 µL. Hamilton syringe was

used as the mobile phase. After development, densitometry scanning was performed with a camage TLC scanner III reflectance absorbance mode at 254 nm and 366 nm under control of win cats software (V 1.2.1 manufactured by camage Switzerland).The slit dimensions were 6.00 x 0.45 mm and the scanning speed was 20 mm per second.[7]

RESULTS

Organoleptic characters of *Panchatikta Ghrita Gugglu*

Organoleptic characters contents of *Panchatikta Ghrita Gugglu* like colour, taste, touch, odour were recorded. The colour of *Panchatikta Ghrita Gugglu* was black. *Panchatikta Ghrita Gugglu* has aromatic smell, taste was Bitter and felt hard on touch which is shown in Table 2.

Table 1: Ingredient of *Panchatikta Ghrita Gugglu.*

Drug	Latin Name	Proportion	Parts used
Nimba	*Azadirachta indica* A.Juss	480 gm	Stem bark
Amruta	*Tinospora cordifolia* (Willd.) Miers	480 gm	Stem
Patola	*Trichosanthes dioica Roxb.*	480 gm	Leaf
Kantakari	*Solanum surattense Burm. L.*	480 gm	Wlole plant
Vasa	*Adhatoda vesica Nees*	480 gm	Root
Guggulu	*Comiphora wightii (Arn.) Bhand.*	240 gm	Oleoresin
Patha	*Cissampelos Pereira Linn.*	12 gm	Root
Vidanga	*Embelia ribes Burm. L.*	12 gm	Fruit
Devadaru	*Cedrus deodara (Roxb.) Loud.*	12 gm	Heart wood
Gaja pippali	*Piper sylvaticum Roxb.*	12 gm	Fruit
Yavakshara	*Hordeum vulgare Linn.*	12 gm	Whole plant
Sarjikshara	-	12 gm	-
Shunthi	*Zingiber officinale Roxb.*	12 gm	Rhizome
Haridra	*Curucuma longa Linn.*	12 gm	Rhizome
Misi	*Foeniculum vulgare Mill.*	12 gm	Fruit
Chavya	*Piper retrofractum Vahl.*	12 gm	Stem
Kustha	*Saussurea lappa C.B. Clarke*	12 gm	Root
Tejovati	*Zanthoxylum armatum DC*	12 gm	Fruit
Maricha	*Piper nigrum Linn.*	12 gm	Fruit
Kutaja	*Holarrhena antidysenterica (Roth.) A.DC.*	12 gm	Stem bark
Yavani	*Trichyspermum ammi (Linn.) Sprague ex Turril.*	12 gm	Fruit
Chitraka	*Plumbago zeylanical Linn.*	12 gm	Root
Katurohini	*Picrorhiza kurroa Royle Ex Benth.*	12 gm	Rhizome
Bhallataka	*Semicarpus anacardium Linn.*	12 gm	Fruit
Vacha	*Acorus calamus Linn.*	12 gm	Rhizome
Pippali moola	*Piper longum Linn.*	12 gm	Stem
Rasna	*Pluchea lanceolata Oliver & Hiem.*	12 gm	Root/leaf
Manjishtha	*Rubia cordifolia Linn.*	12 gm	Root

Ativisha	*Aconitum heterophyllum Wall. Ex Royle*	12 gm	Root
Visha	*Aconitum palmatum D.Don*	12 gm	Root
Yavani	*Trachyspermum ammi (Linn.) Sprague ex Turril.*	12 gm	Fruit
Go Ghrita	-	768 gm	-

Table 2: Organoleptic characters of *Panchatikta Ghrita Gugglu.*

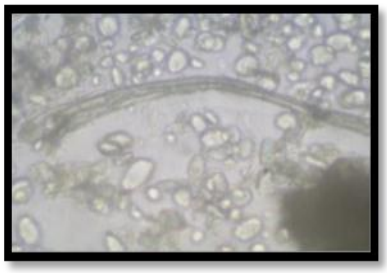
Drug	Colour	Odour	Taste	Consistency
Panchatikta Ghrita Gugglu	Black	Aromatic	Bitter	Hard, *Vati*

Table 3: Physicochemical parameters of *Panchatikta Ghrita Gugglu.*

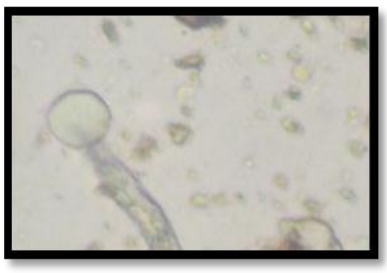
Parameters/ Sample	*Panchatikta Ghrita Gugglu*
Loss on drying	9.89%w/w
Ash value	5.83%at room temp.
Water soluble extractive	23.9% w/w
Methanol soluble extractive	32.7% w/w
pH value (5% aqueous)	2.0
Weight variation of Gugglu	Average wt. 0.342 gm Highest wt.0.380 gm Lowest wt. 0.290 gm

Table 4: HPTLC results for methanolic extract of *Panchatikta Ghrita Gugglu.*

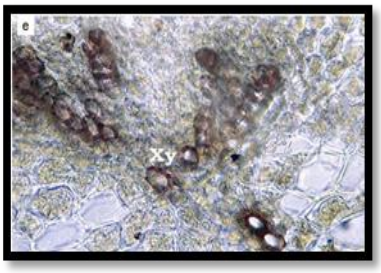
HPTLC	254 nm		366nm	
	No. of spots	Rf value	No. of spots	Rf value
	08	0.05, 0.19, 0.33, 0.35, 0.46, 0.81, 0.86, 0.93	05	0.02,0.36,0.41,0.87,0.94



Fibers of *Yavani*

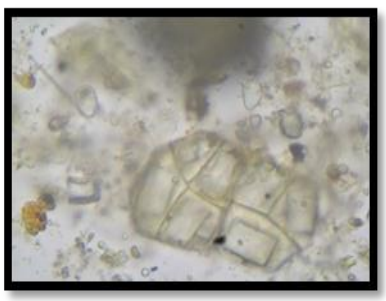


Oil content of *Yavani*

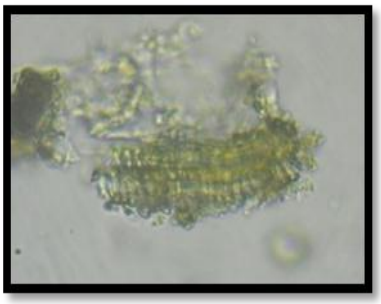


Xylem of *Vatsnabha*

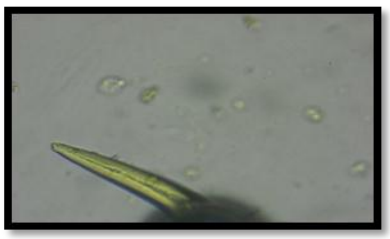
Fibers of *Nimba*



Collenchyma cells of *Guduchi*

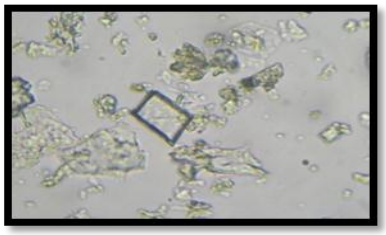


Spiral vessels of *Patola*



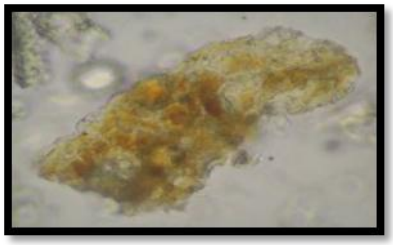
Trichomes of *Patola*

Epidermal cell of *Patola*



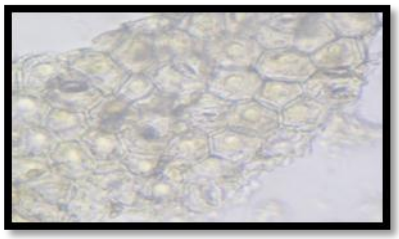
Prismatic crystals of *Kantakari*

Stellete trichome of *Kantakari*



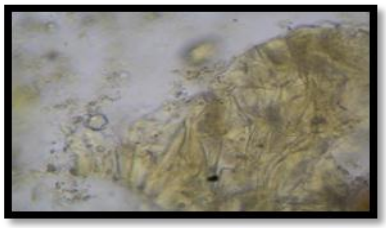
Epicarp cells of *Ativisha*

Lignified fibres of *Devadaru*

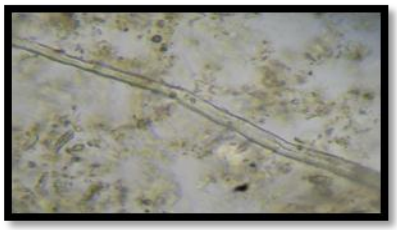


Stomata of *Vasa*

Trichome of *Vasa*

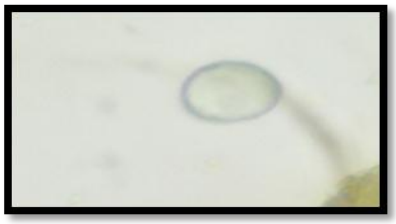


Epidermal cells of *Yavakshara*



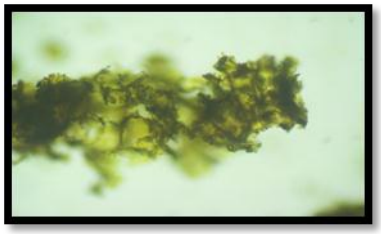
Fibres of *Shunthi*

Starch grains of *Shunthi*

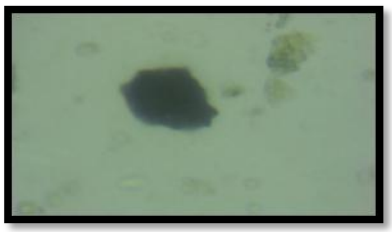


Oil globules of *Haridra*

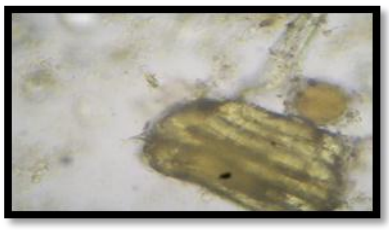
Annular vessels of *Haridra*



Cork view surface of *Haridra*

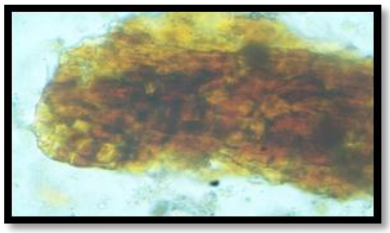


Tannin content of *Kushtha*

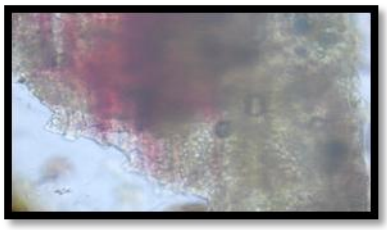


Stone cells of *Maricha*

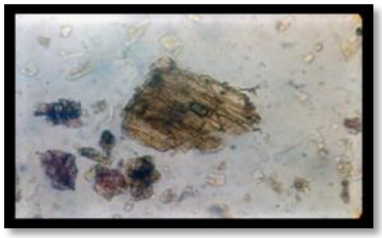
Border pitted vessels of *Chitraka*



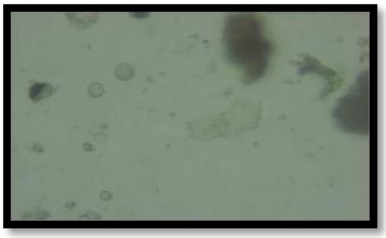
Cork cells with tannins of *Chitraka*



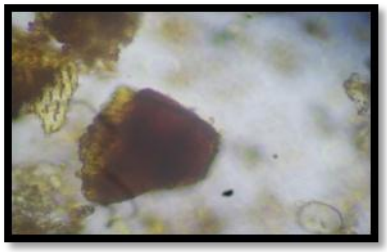
Lignified fibres of *Kutaja*



Fragments of fiber of *Katuki*

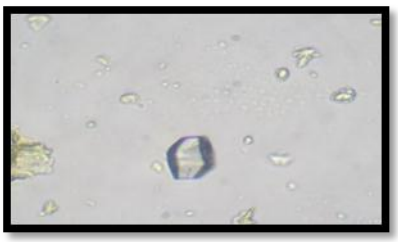


Starch grains of *Vacha*

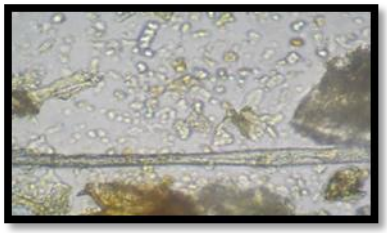


Oleoresin of *Pippali*

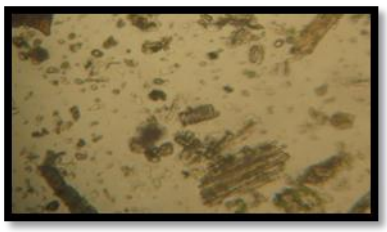
Stone cell of *Rasna*



Calcium oxalate crystals of *Chavya*

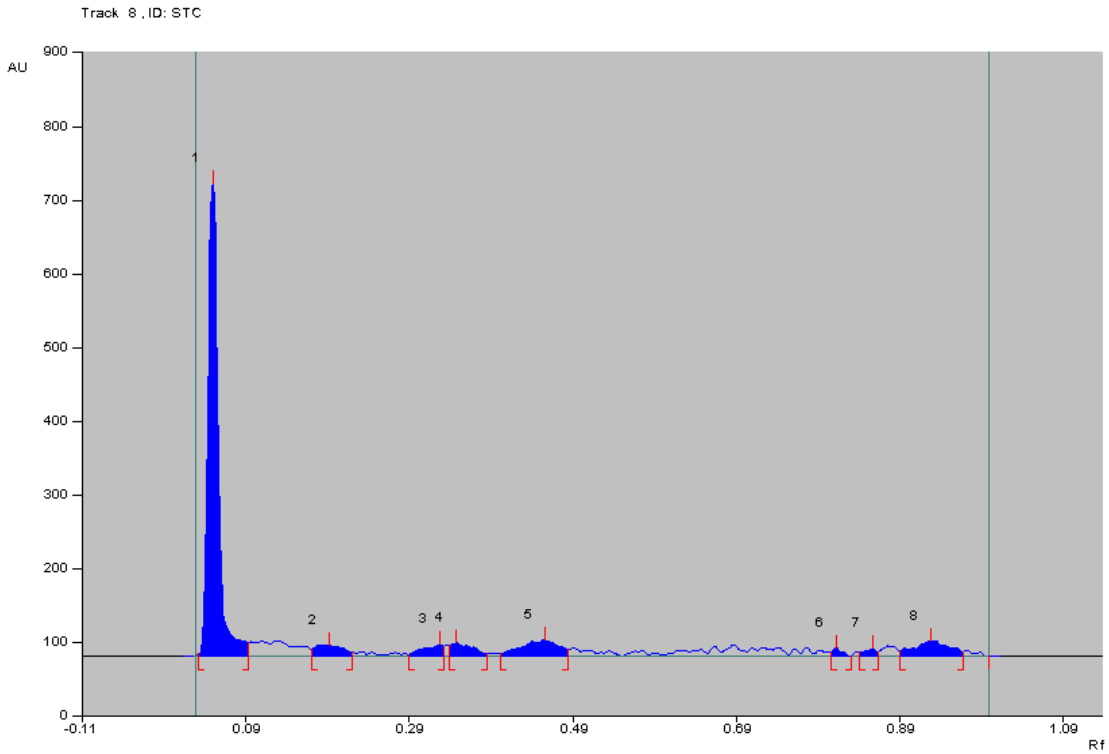


Fibre of *Chavya*



Rod shape crystal of *Manjishtha*

Figure 1: Microphotographs of Panchatikta Ghrita Gugglu.



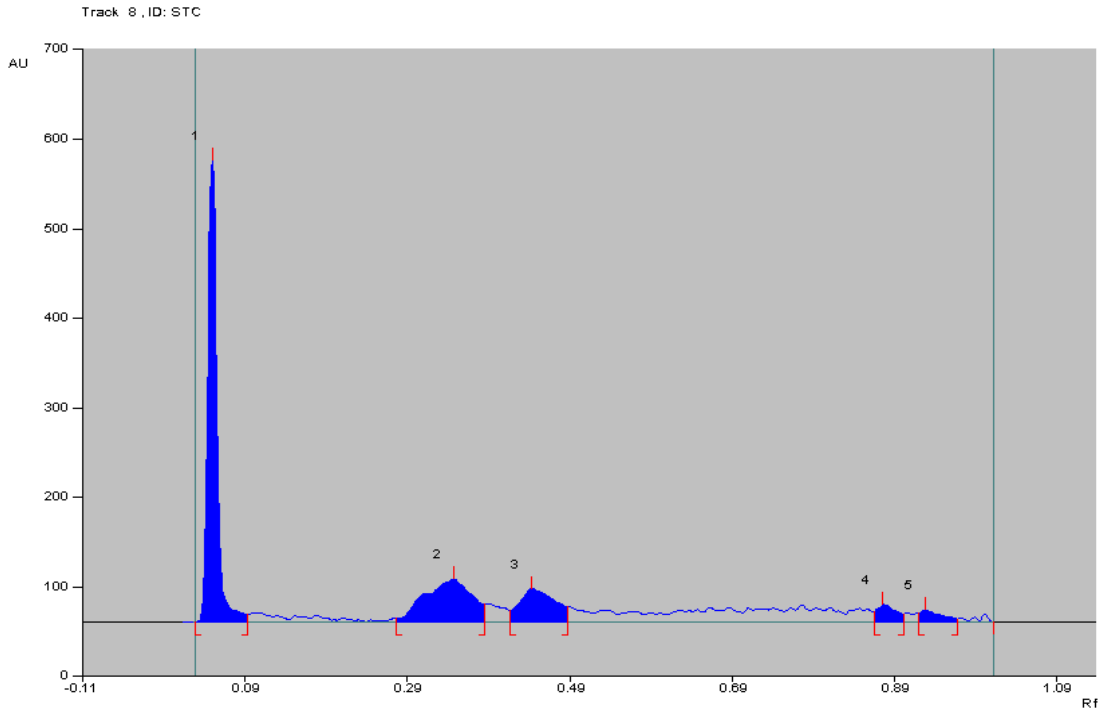


Figure 2: Densitogram of *Panchatikta Ghrita Gugglu* at 254nm and 366nm (A): Peak display at 245 nm (B): Peak display at 366nm.

Microscopic studies of *Panchatikta Ghrita Gugglu*

Identifying characters of ingredients of Panchatikta Ghrita Gugglu under the microscope were Fibres (1) and Oil content (2) of *Yavani*, Xylem of *Vatsnabha* (3), Fibres of *Nimba* (4), Collenchyma cells of *Guduchi* (5), Spiral vessels (6), Trichomes (7) and Epidermal cells(8) of *Patola*, Prismatic crystals (9) and Stellate trichome (10) of *Kantakari*, Epicarp cells of *Ativisha* (11), Lignified fibers of *Devadaru* (12), Stomata (13) and Trichome (14) of *Vasa*, Epidermal cells of *Yavakshara* (15), Fibers (16) and Starch grains (17) of *Shunthi*, Oil globules of *Haridra* (18), Annnular cells (19) and Cork view surface (20) of *Haridra*, Tannin content of *Kushtha* (21), Stone cells of *Maricha* (22), Border pitted vessles (23) and Cork cells (24) of *Chitraka*, Lignified fibres of *Kutaja* (25), Fragments of fibre of *Katuki* (26), Starch grains of *Vacha* (27), Oleoresin of *Pipalli* (28), Stone cells of *Rasna* (29), Calcium oxalate crystals (30) and Fibres of (31) *Chavya*, Rod shape crystal of *Manjishtha* (32).

All these are showed in Figure 1 (1 to 32)

Physicochemical analysis of *Panchatikta Ghrita Gugglu*

Physico-chemical analysis of *Panchatikta Ghrita Guggulu* revealed the loss on drying 9.89% w/w, The ash value was 5.83%w/w, loss on drying 9.89%w/w, water soluble extract 23.9%w/w, alcohol soluble extract 32.7%w/w and pH value was 2.0. (Table 3).

High performance thin layer chromatography of *Panchatikta Ghrita Gugglu*

On performing HPTLC, the chromatogram of *Panchatikta Ghrita Guggulu* showed 08 peaks with maximum Rf values 0.05, 0.19, 0.33, 0.35, 0.46, 0.81, 0.86 and 0.93 at short wave UV 254nm; while at long wave UV 366 nm, the chromatogram showed 5 spots with maximum Rf values 0.02,0.36,0.41,0.87,0.94 (Table 4).

DISCUSSION

Pharmacognostical part of the study of *Panchatikta Ghrita Gugglu* was the step towards identification of all raw material present in the finished product. The presence of all contents of raw drugs in the final product showed the genuinity of the final product. Hence, *Panchatikta Ghrita Gugglu* is herbo-mineral drug, identification of mineral parts of *Panchatikta Ghrita Gugglu* cannot be evaluated through pharmacognosy. All the pharmaceutical parameters were done to analyse the values permissible for the *Panchatikta Ghrita Gugglu*. All the parameters tested under the pharmaceutical study are as per the API.[8] The physic chemical parameters showed that percentage of alcohol soluble extract was more than water soluble extract which indicates the presence of flavonoids, tannins and anthocyanidins in the drug. While alcohol soluble extract value denotes the presence of tannins, resins and alkaloids in the drug. Ash value of the final product is 5.83% w/w shows the presence of inorganic material which cannot be identified through pharmacognosy.

CONCLUSION

The Pharmacognostical and physic chemical analysis of *Panchatikta Ghrita Gugglu* confirmed the purity and genuinety of the drug. Published information is not available on Pharmacognostical and physic-chemical profiles of *Panchatikta Ghrita Gugglu*. Information acquired from this study may be beneficial for further research work and can be used as a reference standard for quality control researches.

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