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Analytical profile of *Brahmi Ghrita*: A polyherbal Ayurvedic formulation

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Abstract

Brahmi Ghrita, a polyherbal Ayurvedic formulation is recommended in the management of various psychological disorders like *Unmada, Apasmara* and *Graharogas*. The present study deals with the pharmacognostical identification of ingredients of *Brahmi Ghrita* and its physico-chemical analysis. Pharmacognostical study containing both macroscopic and powder microscopy of raw drug revealed the quality and genuineness of all the constituents of *Brahmi Ghrita*. Organoleptic features of coarse powder made out of the crude drugs were within the standards prescribed. Acid value was 0.16075, saponification value 184.17, Refractive Index value 1.467 at room temperature, Iodine value 26.715, Specific gravity at room temperature was 0.9133. HPTLC was carried out after organizing appropriate solvent system in which maximum 9 spots were distinguished and most of the R_f values were identical in alcoholic extract which shows the presence of certain definite constituents in *Brahmi Ghrita*.

Key words: *Brahmi Ghrita*, HPTLC, pharmacognosy, physico-chemical analysis

Introduction

Brahmi being a *Medhya* drug is recommended for various psychosomatic and psychiatric disorders. Most of the formulations acting on psyche are ghee based. It is well established that, the drugs to have its action on brain should have the capacity to cross the blood–brain barrier and for that purpose ghee is the best drug vehicle. *Brahmi Ghrita* is recommended for the management of *Unmada* (Insanity), *Alakshmi* (Inauspicious), *Apasmara* (Epilepsy), *Papavikaras* (Diseases due to sinful acts),[1] and for *Apasmara*, *Unmada*, *Graha Rogas* (Diseases afflicted by evil spirits).[2]

The objectives of adulteration are 4-fold, namely, to increase the bulk or weight of the substance, to improve its appearance, to give it a false strength, or to rob it of its most valuable constituents. The recognition of such impurities, and the tracing of them to their source, is of prime importance in pursuing a charge of adulteration.[3] The objective of the present study is to ascertain the genuinity of all the ingredients of *Brahmi Ghrita* and presence of components as recommended in Ayurvedic Pharmacopoeia of India (API) –*Brahmi*,[4] *Vacha*,[5] *Shankhapushpi*,[6] *Kushtha*[7] through pharmacognostical and physico-chemical studies.

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Aims and objectives

1) Pharmacognostical study of individual components of *Brahmi Ghrita*.
2) Physico-chemical analysis of *Brahmi Ghrita*.

Materials and Methods

Collection and authentication of raw drugs

Whole plant of *Brahmi* (*Bacopa monnieri* (L.) Pennel) was collected from Foundation for Revitalization of Local Health Traditions (FRLHT), Bangalore in the month of December 2011. Other ingredients of *Brahmi Ghrita* [Table 1] were procured from Pharmacy, Gujarat Ayurved University. All these were identified and authenticated in Pharmacognosy Laboratory, IPGT and RA, Gujarat Ayurved University, Jamnagar. *Ghrita* (Cow's ghee) was procured from Khadi Gramodyoga Bhandar, Jamnagar and *Brahmi Ghrita*[1] was prepared in Pharmacy of Gujarat Ayurved University, Jamnagar.

Method of preparation of *Brahmi Ghrita*

Brahmi Swarasa was extracted by exerting mechanical pressure on fresh *Brahmi*. In a large vessel *Go-Ghrita* was poured, when it liquefies under moderate flame, *Kalka* of *Vacha*, *Kushtha*, *Shankhapushpi* made in *Brahmi Swarasa* was added, followed by

addition of *Brahmi Swarasa*. To get final product, the contents were subjected to heat till up to *Sneha Siddhi Lakshanas* were observed.[8]

Pharmacognostical evaluation of ingredients of *Brahmi Ghrita*

Organoleptic study

Individual powders were subjected for various sensory characters like color, taste odor, etc., and were carefully noted down.[9]

Powder microscopy

The powders of respective parts of [Table 1] *Brahmi*, *Vacha*, *Kushtha*, *Shankhapushpi* were studied separately with and without staining. The microphotographs were taken under Corlzeiss binocular microscope attached with camera.[10,11]

Physico-chemical study

Brahmi Ghrita was analyzed using various standard physicochemical parameters such as Acid value, saponification value, Refractive Index value, iodine value, specific gravity. High Performance Thin Layer Chromatography (HPTLC) was carried out after making appropriate solvent system with methanolic extract of *Brahmi Ghrita*[12] at Pharmaceutical chemistry laboratory, IPGT and RA, Jamnagar.

High Performance Thin Layer Chromatography

Preparation of sample solution

The *Ghrita* sample was adsorbed on silica gel. The mixture was extracted with hexane. Hexane fraction was discarded. The material was extracted with methanol. Process was repeated for 3 times. The methanol layer was collected, filtered, and evaporated off. The dried material was again dissolved in methanol and used for Thin Layer Chromatography (TLC) identification.

Chromatographic conditions

1) Stationary phase: Silica gel GF 254(E. Merck) precoated TLC plates
2) Mobile phase: Dichloromethane: methanol: water (4.5:1.0:0.1 v/v/v)
3) Sample volume: 5 µl
4) Sample for HPTLC: Methanol extract of *Brahmi Ghrita*
5) Spray reagent: Vaniline-sulfuric acid

Instrumental conditions

Camag HPTLC instrument catalog No 0276481(Switzerland) was used for experiment
Application mode: Camag Linomat V
Development chamber: Camag twin trough chamber.
Plates: Precoated silica gel GF254 plates.
Chamber saturation: 30 min.
Development time: 30 min.
Development distance: 7 cm.
Scanner: Camag scanner III.
Detection: Deuterium lamp, Tungsten lamp
Data System: Win cats software

Procedure

Before spotting, the plates were prewashed with methanol. Sample solutions were applied to the plates as sharp bands by means of Camag Linomat V sample applicator. The spots were dried in a current of air. The mobile phase (20 ml) was poured into a twin trough glass chamber whole assembly was left to equilibrate for 30 min and the plate was placed in the chamber. The plate was then developed until the solvent front had travelled at a distance of 80 mm above the base of plate. The plate was then removed from chamber and dried in a current of air. Detection and quantification was performed with Camag TLC scanner 3 at a wavelength of 254 and 366 nm.

Observations and Results

Pharmacognostical analysis

Organoleptic characters were noted down and are depicted in Table 2.

Microscopic characters: Powder microscopy of *Brahmi Ghrita* ingredients was studied and microphotographs were placed at respective figures.

Kushtha: Fiber, prismatic crystals, tannin, cork in surface, annular, and pitted vessels [Figure 1a–d].

Table 1: Ingredients of *Brahmi Ghrita*

Name	Botanical name	Part used	Form	Part
Brahmi	*Bacopa monnieri* (L.)Pennel	Whole plant	Juice	4 l
Vacha	*Acorus calamus* Linn.	Rhizome	Powder	85 g
Kushtha	*Saussurea lappa* C.B.Clarke	Root	Powder	85 g
Shankapushpi	*Convolvulus pluricaulis* Choisy	Whole plant	Powder	85 g
Go-Ghrita (cow's ghee)	-	-	-	1 Kg

Table 2: Organoleptic characters of ingredients of *Brahmi Ghrita*

Observation	*Kushtha*	*Shankhapushpi*	*Brahmi*	*Vacha*
Color	Dark brown	Grayish brown	Greenish grey	Creamish
Odor	astringent	Characteristic	Pungent	Aromatic
Taste	Bitter astringent	Slight astringent	Astringent-Bitter	Bitter ends in sweet

Brahmi: Prismatic crystals, fiber, fragments of annular vessels, stomata, fragments of pallside parenchyma, tannin contents, and fragments of pitted vessels [Figure 2a–d].

Vacha: Tannin, simple starch grains, oleoresins, parenchyma cells with starch grains, fiber with lumen, fragments of annular and pitted vessels [Figure 3a–d].

Shankhapushpi: Simple unicellular trichome, stellate trichome, prismatic crystals, fragments of spiral vessels, lignified fibers, stomata, fragments of pitted vessels, tannin, starch grains in group [Figure 4a–d].

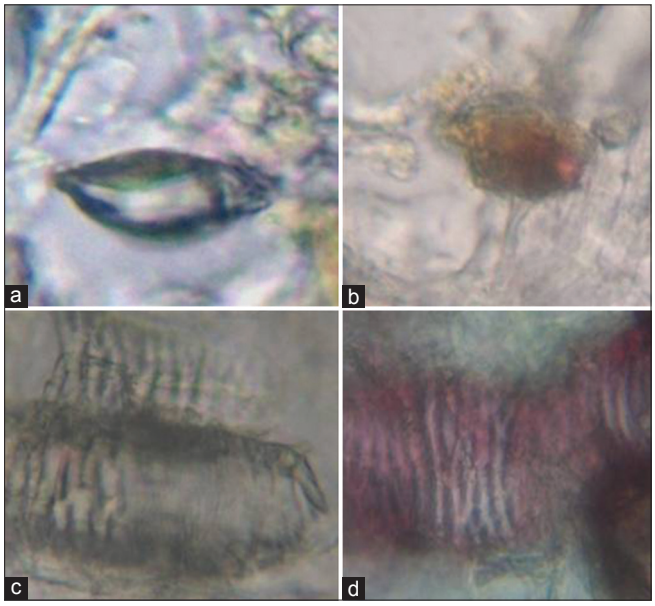

Figure 1: Powder microscopy of *Brahmi Ghrita ingredients Kushtha* (a) Prismatic crystals (b) Tannin (c) Annular vessels (d) Pitted vessels

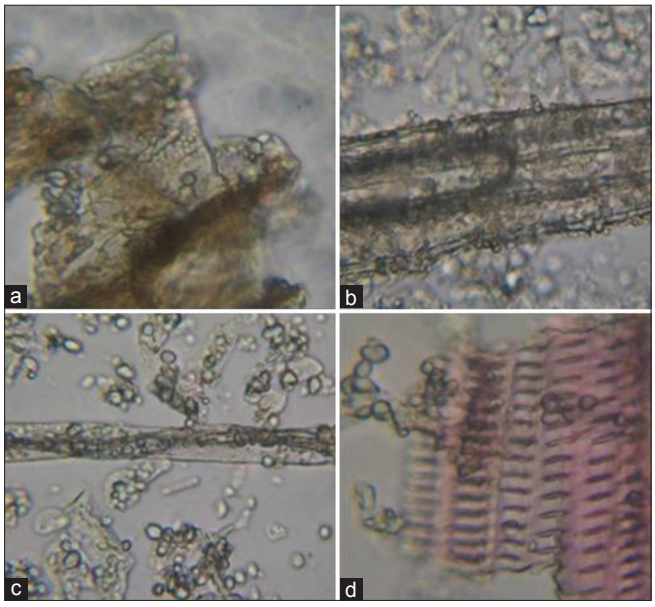

Figure 3: *Vacha* (a) Olioresins (b) Parenchyma cells with starch grains (c) Fiber with lumen (d) Fragments of annular and pitted vessels

Physico-chemical analysis

Brahmi Ghrita was analyzed using various standard physico-chemical parameters such as acid value, saponification value,

Table 3: Physicochemical parameters

Test	Result
Acid value	0.16075
Saponification value	184.17
RI value	1.467
Iodine value	26.715
Specific gravity at room temperature	0.9133
Rancidity test	Negative

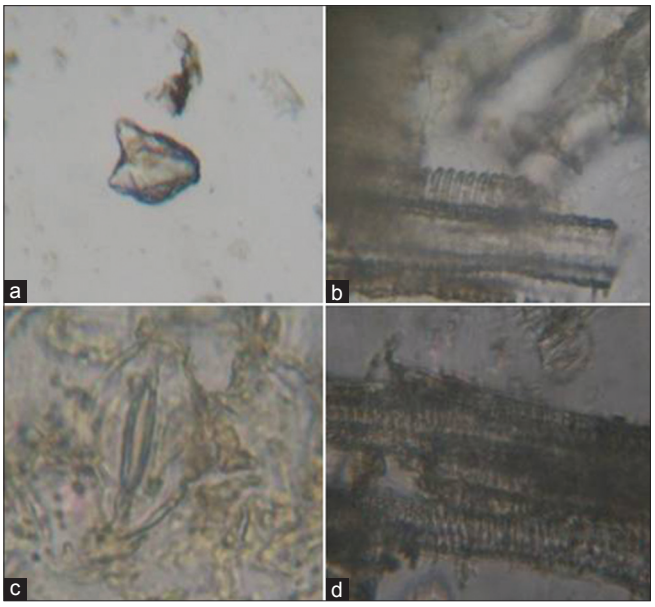

Figure 2: *Brahmi* (a) Prismatic crystals (b) Fragments of annular vessels (c) stomata (d) Fragments of pitted vessels

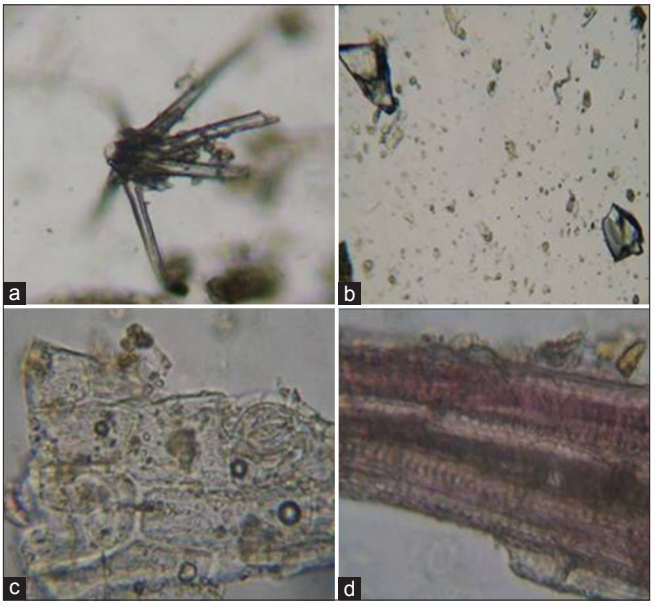

Figure 4: *Shankhapushpi* (a) Stellate trichome, (b) Prismatic crystals (c) Stomata (d) Fragments of pitted vessels

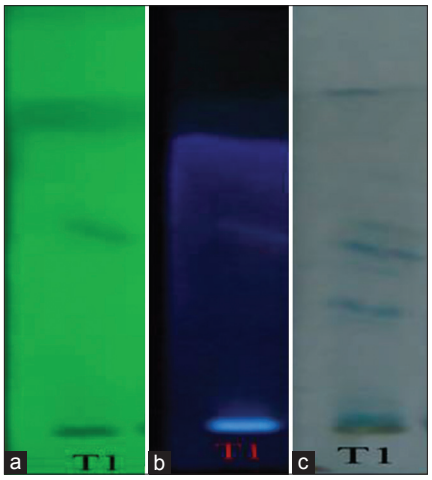

Figure 5: HPTLC figure prints (a) at 254 nm (b) at 366 nm (c) After sprays T1-*Brahmi Ghrita* sample

Table 4: High performance thin layer chromatography

	Spots	Rf values
At 254nm	9	0.03, 0.06, 0.13, 0.20, 0.78, 0.83, 0.86, 0.49, 0.63
At 366 nm	6	0.16, 0.58, 0.61, 0.71, 0.77, 0.81
After spray	8	0.05, 0.29, 0.35, 0.47, 0.52, 0.6, 0.78, 0.86

RI value, iodine value, specific gravity [Table 3].

High Performance Thin Layer Chromatography

On analyzing under densitometer at 254 nm, the chromatogram showed 9 peaks, while at 366 nm the chromatogram showed 6 peaks. And after spray the chromatogram showed 8 peaks. [Table 4], [Figure 5a–c]. Three dimensional (3d) densitogram at 254 nm shows comparative R_f value of sample with standard [Figure 6a–c].

Discussion

Pharmacognostical study reveals authentification of individual raw drugs of *Brahmi Ghrita* and is cross verified.[3-6] The oleoresins, pitted vessels, tannin, prismatic crystals, stomata, fiber are observed in ingredients. All the physico-chemical parameters, acid value, saponification value, RI value, iodine value, specific gravity analyzed were within the normal reference range.[13] In HPTLC one spot was detected at R_f value 0.78, which indicates presence of Bacoside in *Brahmi Ghrita*.[14] All the results show that the prepared *Ghrita* formulation is not rancid (after 10 months of preparation) and the quality of the *Ghrita* is standard.

Conclusion

Pharmacognostical study findings confirm the ingredients present in the *Brahmi Ghrita*. Identified phytochemical components like bacoside support the intended action of the formulation. Under densitometer at 254 nm 9 peaks and under 366 nm 6 peaks were found and after spray, 8 peaks were found. It is inferred that the formulation meets maximum qualitative standards. The results of this study may be used as the reference standard in further research undertakings of its kind.

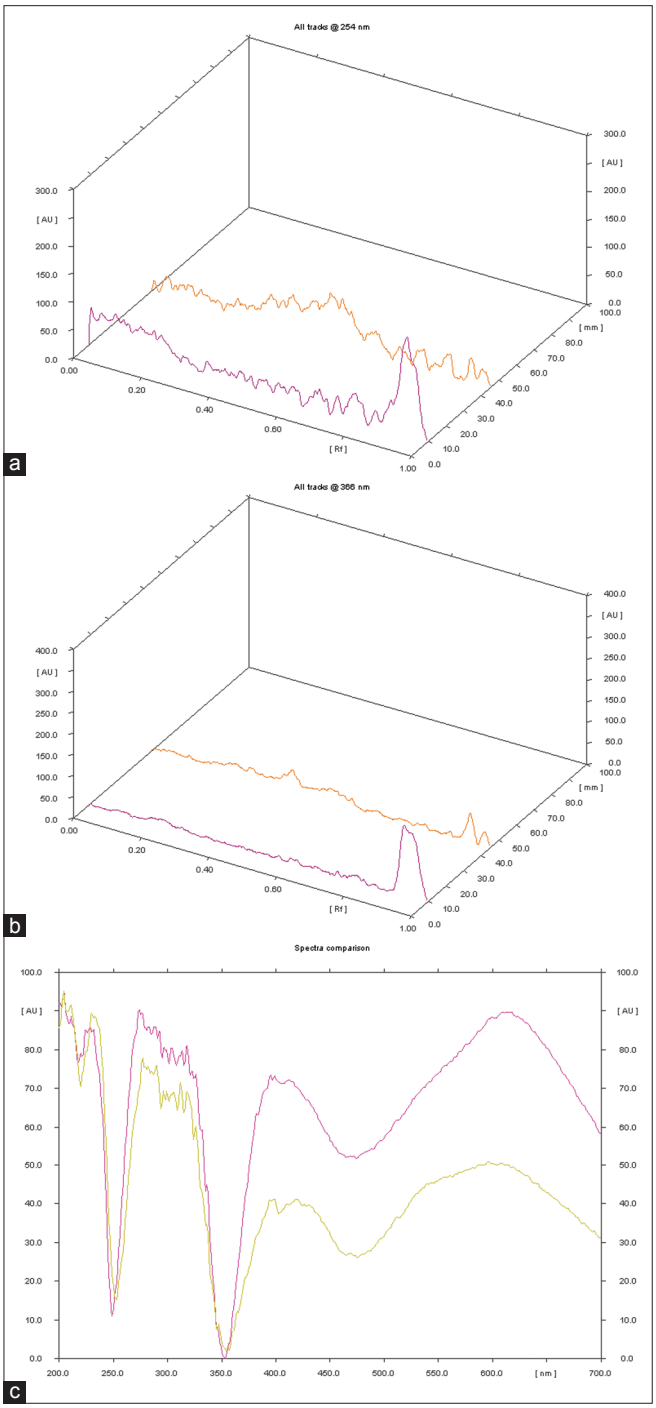

Figure 6: (a) 254 3D (b) 366 nm (c) Spectral comparison at Rf 0.78

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हिन्दी सारांश

ब्राह्मीघृत के घटकों का द्रव्यपरिचय एवं रासायनिक विश्लेषण

ज्योति गुब्बण्णवर, हरिमोहन चन्दोला, हरीशा सी. आर., रेणुका कल्याणी, विनय जे. शुक्ला

प्रस्तुत अध्ययन में ब्राह्मीघृत के घटकों का द्रव्य परिचयात्मक एवं रासायनिक विश्लेषण किया गया है । द्रव्यपरिचयात्मक अध्ययन, ब्राह्मीघृत के घटकों के स्थूल स्वरुप एवं चूर्ण के सूक्ष्मदर्शीय अध्ययन द्वारा उनकी गुणवत्ता एवं शुद्धता को दर्शाता है । सभी घटक द्रव्यों के परिचयात्मक नामरुप लक्षण सामान्य परिधि के अन्तर्गत पाये गए । औषधविलयनतन्त्र तैयार करने के बाद HPTLC किया गया, जिसमें अधिकतम ९ चिन्ह पाए गए । अल्कोहल में घुलनशील तत्वों का Rf मान अधिकाँशतः एक समान पाया गया, जो कि ब्राह्मीघृत में निश्चित तौर पर कुछ प्रभावशाली तत्वों की उपस्थिति को सिद्ध करता है ।