



PHARMACOGNOSTICAL AND PHYSICOCHEMICAL ANALYSIS OF TRIPHALADI YOGA: AN AYURVEDIC POLY- HERBAL FORMULATION

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ABSTRACT

In Ayurvedic classical texts administration of potent psycho-physical rejuvenator formulations comes under *Rasayana chikitsa* which frees one of diseases, improves quality of life and delays senile degenerative changes. Eleven among the most commonly used anti-oxidant, adaptogenic potent, well established rejuvenator herbs viz., *Hareetaki* (*Terminalia chebula* Retz), *Vibheetaki* (*Terminalia bellerica* Roxb) *Amalaki* (*Embllica officinalis* Linn), *Guduchi* (*Tinospora cordifolia* Miers), *Gokshura* (*Tribulus terrestris* Linn), *Yashtimadhu* (*Glycyrrhiza glabra* Linn), *Tulasi* (*Ocimum sanctum* Linn.), *Haridra* (*Curcuma longa* Linn), *Daruharidra* (*Berberis aristata* DC), *Shunthi* (*Zingiber officinale* Rosc), *Punarnava* (*Boerhavia diffusa* L) were administered along with unequal quantity of ghee and honey as *Anupana* (vehicle) to delay senile changes and reverse cataract formation in the eyes. For the first time the herbal compound in the powder form was subjected to Pharmacognostical evaluation, physico-chemical screening, phyto-chemical analysis and HPTLC studies. The presence of Scalariform vessels, Acicular crystals, Border pitted vessel, Starch grains were the characteristic features observed in the microscopy. The Phyto-chemical analysis indicated presence of alkaloids, tannins, saponins, flavonoids and glycosides.

Keywords: *Triphaladi yoga*, Pharmacognosy, Physicochemical, HPTLC.

INTRODUCTION

An estimated 200 million people worldwide have cataracts.¹ Data presented by WHO shows that one-third of the world's 45 million blind and half of the world's 1.5 million blind children live in South-East Asia region. The blind persons in this Region are among the poorest in the world and the weakest in the society (most of them women), often leading a miserable life and are disenfranchised.² In India alone, 4 million people turn blind due to cataract every year. Data available from three national level surveys in India was used to project the estimates of cataract blindness in the country. Extrapolating the observations to the entire country it was estimated that in 2001, there were 7.75 million individuals whose blindness could be attributed to cataract and this would increase to 8.25 million by 2020³.

The lens of the eye is formed of water and special proteins that are initially clear. Cataract is the clouding that develops in the crystalline lens of the eye or in its capsule, varying in degree from slight to complete opacity and obstructing the passage of light. Early in the development of age-related cataract the power of the lens may be increased, causing myopia, and the gradual yellowing and opacification of the lens may reduce the perception of blue colours. Cataracts typically progress slowly to cause vision loss and are potentially blinding if untreated. The condition usually affects both the eyes, but almost always one eye is affected earlier than the other. Cataracts develop when proteins of the lens begin to cluster and become denser. The formation of these protein bundles makes it difficult for light to pass through the eye to the retina and creates a discolored or opaque area in the lens. Yellowing of the lenses due to

disintegration of proteins is another way to develop cataract. A senile cataract, occurring in the elderly, is characterized by an initial opacity in the lens, subsequent swelling of the lens and final shrinkage with complete loss of transparency. Moreover, with time the cataract cortex liquefies to form a milky white fluid in a Morgagnian cataract, which can cause severe inflammation if the lens capsule ruptures and leaks. If untreated, the cataract can cause phacomorphic glaucoma. Very advanced cataracts with weak zonules are liable to dislocation anteriorly or posteriorly. Such spontaneous posterior dislocations (akin to the historical surgical procedure of couching) in ancient times were regarded as a blessing from the heavens, because some perception of light was restored in the cataractous patients.

The only available treatment of cataract is surgery. However due to its high cost, post operative complications and uneven availability around the world, propels researchers to find out other modalities of treatments for cataract, like use of antioxidants, vitamins, and others to breakdown pathophysiology of cataract.

Data available in India shows that all cataract surgeries are not sight-restoring surgeries as nearly 40-50% surgeries are done in individuals with a vision > 20/200 in the better eye.⁴ Clearly, the tremendous burden of blindness due to cataract remains a major challenge for eye care professionals with public health perspective. Surgical treatment of cataract imposes great economic burden on the state and the backlog is perhaps too big to be handled by surgery alone. Alternatively, preventive ophthalmology offers another approach to tackle the problem by seeking to identify factors that might modify the onset or progression of cataract. If such

a factor is identified which simply delays the onset of cataract by a period of 10 years, the number of cataract surgeries would drastically decrease by 45% or more⁵

So it becomes important to find successful medical management to prevent the onset or delay the progress of cataract. As like all *svabhavabalapravritta vyadhis* (Diseases which occur due to natural changes or ageing process), cataract also needs treatment that can help in delaying the ageing process. Hence Rasayana drugs were chosen for the study.

Triphalaadi yoga is a combination of eleven dried herbs in powder form administered in cataract patients to delay senile changes in the lens. The dose administered is 6gms of the powder mixed with 6gms honey and 3 gms of cow's ghee twice daily in the morning and before bedtime after food.

The eleven drugs chosen for this study is an anubhoota yoga from the Shalakya Tantra Department of I.P.G.T& R.A, Jamnagar and has the solid backing of previous researches for their most important individual indactivities viz. Adaptogenic properties of Guduchi, Hareetaki, Amalaki⁶, Anti-AGE properties (Anti- Advanced glycation end products) of Shunthi, Tulasi⁷, Immunomodulatory and Rejuvenating aspects of Guduchi, Gokshura and Amalaki⁸, Adaptogenic, anti- inflammatory, anti – hypercholestraemic action of Haridra and Daru haridra etc.⁹

MATERIALS AND METHODS

Collection/Procurement of the drug:

Ingredients viz. fruits of *Haritaki* (*Terminalia chebula* Retz), *Vibheetaki* (*Terminalia bellerica*) *Amalaki* (*Emblica officinalis* Garten), roots and rhizomes of *Yashtimadhu* (*Glycyrrhiza glabra* Linn), fruits of *Gokshura* (*Tribulus terrestris* Linn), stem of Guduchi (*Tinospora cordifolia* Miers), rhizome of *Haridra* (*Curcuma longa* Linn), *Daruharidra* (*Berberis aristata* DC), rhizome of *Shunthi* (*Zingiber officinale* Rose), whole plant of *Punarnava* (*Boerhavia diffusa* L) were procured from the pharmacy, and churna was prepared (Table: 1) in the Pharmacy, I.P.G.T. & R.A., Jamnagar, India. The whole plant / Panchanga of *Tulasi* (*Ocimum sanctum* Linn) was procured from Sadvaidyashala, K.R.Puram, Bangalore. Their identities were confirmed by correlating their morphological and microscopical characters with those given in the literature.

Preparation of test drug:

Equal quantities of the obtained fruits, stems, roots/rhizomes, whole plant were shade dried and made in to fine powder separately with the help of mechanical grinder, sieved through 60# and mixed together mechanically to get a homogenous mixture sieved again and stored in airtight containers.

Organoleptic Evaluation:

Various parameters such as colour, odour, taste, touch and texture of the finished product (*churna*) were observed and recorded⁹.

Microscopic Evaluation:

Sample drug was dissolved in small amount of distilled water for a while and then mounted in glycerin. Microscopic examination was carried out with and without staining¹⁰. By powder microscopy, observed the characters, determined the chemical nature of the cell wall along with the form and chemical nature of the cell contents. Microphotographs were taken by using Carl Zeiss binocular microscope attached with camera.¹¹

Physico-chemical Constants:

In physical evaluation foreign matter, moisture content, ash values viz., total ash, acid insoluble ash and extractive values viz., alcohol soluble extractive value, water soluble extractive value as well as pH value etc was determined.

Phyto-chemical Analysis:

Preliminary tests were carried out on methanolic extract for the presence or absence of phytoconstituents like alkaloids, tannins & phenolic compounds, flavonoids, saponins and anthraquinon glycosides¹².

High Performance Thin Layer Chromatography (HPTLC):

HPTLC was performed as per the guidelines provided by API¹³. Methanolic extract of drug sample was used for spotting. HPTLC was performed using Toluene + Ethyl acetate (7:3) solvent system and observed under visible light after derivatisation with vanilline sulfuric acid followed by heating the plate at 110°C. The colour and R_f values of the resolved spots were noted. (Table: 5)

TABLE: 1 - INGREDIENTS OF TRIPHALADI YOGA

Sr No	Name of Ingredients	Parts used	Ratio
1	Haritaki	Pericarp	1 part
2	Vibheetaki	Pericarp	1 part
3	Amalaki	Pericarp	1 part
4	Yashtimadhu	Roots & stolons	1 part
5	Gokshura	Fruits	1 part
6	Guduchi	Stem	1 part
7	Haridra	Rhizomes	1 part
8	Daruharidra	stem	1 part
9	Tulasi	Whole plant	1 part
10	Punrnava	Whole plant	1 part
11	Shunthi	Rhizomes	1 part

TABLE: 2 - ORGANOLEPTIC CHARACTERS

Sr no	Parameters	Results
1	Colour	Golden yellow
2	Odour	Aromatic
3	Taste	Bitter, Astringent
4	Touch	Smooth
5	Texture	Soft

TABLE 3: SHOWING PHYSICOCHEMICAL CONSTANTS

Sr No	Parameters	Results
1	pH	4.73
2	Loss on drying	3.99%
3	Particle size 60 mesh 60-85 mesh 85-120 mesh Above 120 mesh	66.11% 19.16% 1.66% 3.73%
5	Water soluble Extract	26.19%
6	Alcohol soluble extract	26.79%
7	Total Ash	8.5%

TABLE 4: SHOWING RESULTS OF PHYTOCHEMICAL ANALYSIS

Sr No	Components	Results
1	Alkaloids	+
2	Tannin & Phenolic compounds	+
3	Flavonoid	+
4	Saponin Glycosides	+
5	Anthroquinon glycosides	+

+ Present

TABLE 5: Results of TLC study of Triphaladi yoga

No of spots	Rf values Triphaladi yoga		After spray
	254 nm	366 nm	
1	0.35	0.23	0.35
2	0.73	--	0.73

Photo Plate.1. Microphotographs.

Cont..... with HPTLC.

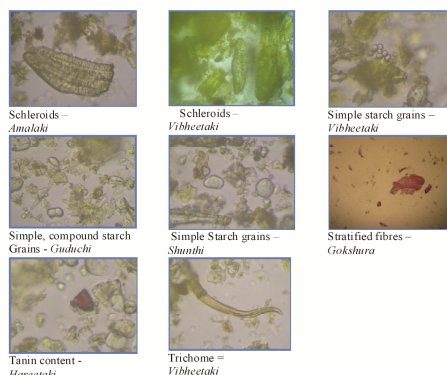
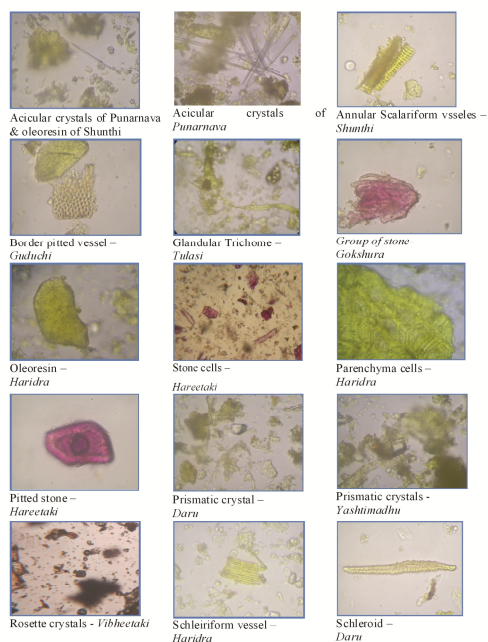


Photo Plate: 2 - Densitograms of Methanolic extract of *Triphaladi yoga*

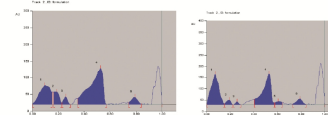
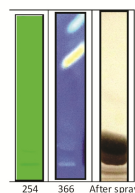


Photo Plate: 3 - HPTLC Plates of Methanolic extract of *Triphaladi yoga*



RESULTS AND DISCUSSION:

Organoleptic Characters:

Triphaladi yoga churna was characterized as soft in texture, smooth in touch, golden yellow in colour, aromatic in odour, bitter and astringent in taste. (Table: 2)

Microscopical Characters:

The diagnostic characters of microscopic analysis of *Triphaladi yoga* showed the presence of tannin content from *Hareetaki*, simple starch grains and trichomes (type) of *Vibheetaki*, schleroids of *Amalaki*, *Vibheetaki* and *Daru haridra*, Annular scalariform vessels and simple starch of *Shunthi*, border pitted vessels and simple and compound starch grains of *Guduchi*, glandular trichomes of *Tulasi*, groups of stones and stratified fibers of *Gokshura*, oleo-resin, pitted stones and scalariform vessels belongs to *Haridra*, parenchyma cells and prismatic cells from *Yashtimadhu* and acicular crystals belongs to *Punarnava*. (Photo Plate: 1)

Physico-chemical Parameters:

Physio-chemical parameters of *Triphaladi yoga* are tabulated in Table: 3. Loss on drying at 110°C is one of the major factors responsible for the deterioration of the drugs and formulations. Low moisture content is always desirable for higher stability of drugs. The results of loss on drying at 110°C of churna showed the lower limits than prescribed in API. A high ash value is indicative of contamination, substitution, adulteration or carelessness in preparing the formulation. The results of ash value revealed that the preparation has lower value than mentioned in API. Water soluble and alcohol soluble extractive value plays an important role in evaluation of crude drugs. Less extractive value indicates addition of exhausted material, adulteration or incorrect processing during drying or storage or formulating.

The extractive values of preparation were observed almost equal in both water as well as alcohol.

Phyto-chemical analysis:

Preliminary qualitative analysis showed the presence of alkaloids, tannin & phenolic compounds, flavonoids, saponin glycosides, anthraquinone glycosides, indicating the active compounds were not disturbed during the preparation and storage. (Table: 4)

High Performance Thin Layer Chromatography:

The HPTLC profiles of methanolic extract of the formulation are super - imposable indicating the presence of all the constituents as per the ingredients. Two spots at R_f 0.35, 0.73 were observed in 254nm UV light spectrum. (Photo plate: 3)

CONCLUSION:


After analysis of *Triphaladi yoga* by different parameters such as foreign matter, moisture content, total ash, acid insoluble ash, water and alcohol soluble extractives and HPTLC densitograms shows good co-relation between them. The study of microscopic characters of present formulation shows the presence of diagnostic identifying characters of ingredients which are used. So it can be concluded that these parameters can be used for the evaluation of *Triphaladi yoga*. Purity and potency of the materials and formulations following the procedures given could be performed in QC/QA laboratory of pharmaceutical house. The present study can serve as the reference for the future works on *Triphaladi yoga Rasayana* in churna form.

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