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Preparation and Evaluation of Classical Ayurvedic Formulation Pathyadikada

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ABSTRACT

Objective: The objective of this study was to prepare and evaluate **Pathyadikada**.

Methods: The selected formulation **Pathyadikada** was prepared as per the methodology described in Ayurvedic texts. The prepared and marketed formulations were analyzed for their physico-chemical constants using established methods in accordance with WHO guidelines, the Indian Pharmacopoeia, and standard protocols from the Ayurvedic Pharmacopoeia of India, Herbal Pharmacopoeia, and other relevant sources.

Results: A profile consisting of distinguishing parameters was developed to provide means of correct identification. Some important organoleptic features of **Pathyadikada** laboratory and market formulations like color, odour, taste, appearance, touch, etc. were documented. Physico-chemical constants were determined and documented. The formulations were tested for the presence of heavy metals, microbial content, pesticide residues and aflatoxin, which were also found within the limits of WHO specifications. Preliminary phytochemical studies of **Pathyadikada** revealed the presence of terpenoids and phenolics as chief constituents. High performance thin-layer chromatography (HPTLC) method was developed and reported for the quantification of **Berberine** and **Quercetin** as marker constituents to ensure identity and quality of **Pathyadikada**.

Conclusion: The studies carried out on **Pathyadikada** can serve as a valuable tool and provide suitable standards for its identification. Phytochemical screening and the quantitative HPTLC assay method developed for quantification of ascorbic acid and gallic acid in **Pathyadikada** can serve for the routine quality control of commercial samples and help in justifying the traditional claims endowed upon this formulation in a scientifically accepted manner.

Keywords: Pathyadikada, Ayurvedic Formulation, Phytochemical Evaluation, Berberine and Quercetin.

INTRODUCTION

Our traditional medicine has a long history. It is purely based on the practices derived from the theories, skills, knowledge, beliefs and experiences indigenous to different cultures. Ayurveda is a widely accepted holistic tradition that reaches far beyond the sphere for wellbeing through healing and the prevention of disease. It has a amazing capacity to help us sync up with our inner nature, develop strengths and offer real support wherever it is needed-so that we can better maintain balance in the face of ill conditions. Ayurveda is a gift of life that can help each of us to stay fit and celebrate our sense of wellness.

Pathyadikadacontains Terminalia chebula, Terminalia bellirica, Embelicaofficnalis, Curcuma longa andTinosporacardiofolia. Pathyadikadha is indicated in the treatment shiroroga (Headache; also called shirahshula or shiro tapa), Migraine; Headaches due to eyestrain. It reduces the intensity and frequency of migraine attacks.

Pathyadikadha is also therapeutically used in Trigeminal neuralgia (Trigeminal neuralgia is a disorder of the trigeminal nerve, the fifth cranial nerve, that causes episodes of sharp, stabbing pain in the cheek, lips, gums, or chin on one side of the face), Sinusitis, Pain in Teeth, Night blindness, etc. Since this medicine contains Triphala, it also relieves constipation.

The aim of the present study was to explore**Pathyadikada**formulations appearing in Ayurvedic traditional medicine for establishing the scientific data pertaining to quality assessment.

MATERIALS AND METHODS

Materials

Procurement of Selected Formulations and Crude Drugs

The ingredients of PathyadiKada were procured from SohamAyurvedRasashala, Solapur, Maharashtra, The ingredients purchased for PathyadiKada (Kashyam) were fruits of Haritaki (Terminalia Cheubula), fruits of Bhibitak (Terminalia belerica), Fruits of Amalaki (Embelica officinalis), roots of Chirayita (Swartiachirata) Rhizomes of Haridra (Curcuma longa), stem of Guduchi (Tinosporacordifolia) and water for decoction.

Reagents and Chemicals

Chemicals, solvents and reagents used for the study were of analytical grade.

Methods

In order to develop the methods to assess the quality control parameters of the selected formulations, selected formulations were subjected to determination of physico-chemical constants using reported methods as per WHO guidelines (WHO, 1998) and Indian Pharmacopoeia, 1996.

The selected formulation**Pathyadikada**was prepared as per the methodology described in Ayurvedic texts.

Pathyadikada

Pathyadikada is polyherbalAyurvedic decoction containing Pathya and other medicinal herbs. This Kashaya has an antioxidant adaptogenic and laxative properties. It is a useful medicine for treating headaches as it is tridodhnashak, anti-inflammatory, purgative and pain relieving. The decoction of medicinal herbs is also known as Kwath, Quath or Kada.

Pathyadi**kada / kwatha** is a non-prescription ayurvedic medicine which is mainly useful in various types of headaches, migraine, ear, nose, throats disorder, eye disease, blurred vision etc.

Method of Preparation ofPathyadikada

The laboratory formulation of**Pathyadikada**was prepared as per the Sharangdhar Samhita.

Ingredients ofPathyadikada

The ingredients purchased for**Pathyadikada**were fruits ofHaritaki, Bhibitak, Amalaki, Chirayita, Haridra, Guduchi(Table 1).

Table 2.1. Ingredients of Pathyadikada

Sr. No.	Ingredients	Botanical name	Part used	Quantity
1	Haritaki	Terminalia Cheubula	Fruit	30 gm
2	Bhibitak	Terminalia belerica	Fruit	30 gm
3	Amalaki	Embelica officinalis	Fruit	30 gm
4	Chirayita	Swartiachirata	Root	30 gm
5	Haridra	Curcuma longa	Rhizome	30 gm
6	Guduchi	Tinosporacordifolia	Stem	30 gm
7	Jal	---	---	Q.S.

Preparation of Pathyadikada

Patyadikada was prepared by a method described in Sharangdhrasamhita. 30 gm each of Haritaki, Bhibitak, Amalaki, Chirayita, Haridra, Guduchiwere boiled with 02 L of water in an iron pot and boiled until reduced to 200ml of decoction or kwath. The kwath was filtered. Then, the mixture was cooled and stored in tightly closed container.

Standardization of Pathyadikada Laboratory and Marketed Formulation

The Laboratory and Marketed formulations of Pathyadikadawere standardized as per WHO guidelines under the various headings using standard protocol obtained from AyurvedicPharamacopoeia of India, Herbal Pharmacopoeia, Herbal compendium, reference books and official agency guidelines listed under table 2.

Table 2. Parameters for Pathyadikada

Sr. No.	Test
1	Description
2	Loss on drying at 105°C
3	Total – ash
4	Acid – insoluble ash

5	Total Solid content (%w/w)
6	pH value
7	Specific gravity at 25°C (g/ml)
8	Viscosity (millipoise)
9	Refractive Index
10	Total Phenolic Content
11	Identifications, TLC/HPTLC-with marker (wherever possible)
12	**Test for heavy metals** Lead Cadmium Mercury Arsenic
13	**Microbial contamination** Total bacterial count Total fungal count
14	**Test for specific Pathogen** E. coli Salmonella spp. S.aureus Pseudomonas aeruginosa
15	**Pesticide residue** Organochlorine pesticides Organophosphorus pesticides Pyrethroids
16	Test for Aflatoxins (B1,B2,G1,G2)

5.3.7 Determination of Phenolic Content: (AP part – I, 2001, API part – I-1999, AFI part – I -2003)

The total phenolic content of the formulations was determined by the Folin-Ciocalteau method with some modifications. Five grams of KALF and KAMF was mixed in 50 ml water and then filtered with Whatman No.1 paper. 0.5 ml of the filtrate was added to 2.5 ml of 0.2 N FolinCiocalteau reagent and placed for 5 minutes. Two ml of 75 g/L of Na_2CO_3 was then added and the total volume made upto 25 ml using distilled water. The above solution was then kept for incubation at room temperature for 2 hours. Absorbance was measured at 760 nm using 1 cm cuvette in a Perkin-Elmer UV-VIS lambda 25 spectrophotometer. Tannic acid (0 - 800 mg/L) was used to produce standard calibration curve. The total phenolic content was expressed in mg of Tannic acid equivalents (TAE) / g of extract.

Considering wide therapeutic applications of **Pathyadikada / kwatha**, and presence of the marker constituents in the selected formulations, to ensure identity and quality of **Pathyadikada / kwatha** a simple, sensitive, specific and reproducible HPTLC method was developed for the quantification of markers in the **Pathyadikada / kwatha** formulations.

Standardization of Finished Product

The prepared sample was standardized by employing various parameters such as organoleptic, physicochemical, qualitative, and quantitative analysis. The data were also compared with commercially available product. The organoleptic characteristics of both prepared and the commercially available product of PathyadiKada were analyzed on the basis of observational criteria (appearance, color, taste, and odor). The physicochemical parameters such as loss on drying, extractive value in different solvent, pH, and acid value were determined as per the guidelines mentioned in Ayurvedic Pharmacopeia of India. The phytochemical screening was performed using different qualitative assay methods described previously. Total reducing sugar content was determined using a titration method as described previously. Quantitative estimation of total polyphenolic content was performed using spectroscopic method and was expressed in equivalent of Gallic acid.

Table 3. Organoleptic evaluation of PathyadiKada(PKLF/PKMF)

Sl. No.	Organoleptic Parameters	Laboratory Sample (PKLF)	Marketed sample (PKMF)
1	Color	Blackish-brown	Brown
2	Odor	Characteristic	Characteristic
3	Taste	Bitter/Acrid	Bitter/Acrid
4	Appearance	Liquid	Liquid
5	Touch	Soft	Soft

Table 4. Physicochemical evaluation of PathyadiKada(PKLF/PKMF)

Sr. No.	Physicochemical Parameters	Laboratory Sample (PKLF)	Marketed sample (PKMF)
1	Loss on drying 110 °C (% w/w)	84.99	36.31
2	Total ash value (% w/w)	00.19	00.47
3	Acid-insoluble ash (% w/w)	< 0.01	< 0.01
4	Total Solid content (%w/w)	18.72	17.95
5	pH value (10% aqueous solution)	03.97	4.28
6	Specific gravity at 25°C (g/ml)	1.075	1.078
7	Viscosity (millipoise)	1.065	1.015
8	Refractive index at room temperature	1.424	1.858
9	Total Phenolic content (mg/100g)	1640.62	192.96

Table 5. Qualitative phytochemical screening of PathyadiKada(PKLF/PKMF)

Sr. No.	Phytoconstituent Screening Test	Laboratory Sample (PKLF)			Marketed sample (PKMF)		
		Aq. Ext.	Me. Ext.	Chl. Ext.	Aq. Ext.	Me. Ext.	Chl. Ext.
1	**Carbohydrates**						
	Molisch's reagent	+	+	+	+	+	−
	Fehling test	+	+	−	+	+	−
	Reducing sugar test	+	+	−	+	+	−
2	**Alkaloids**						
	Dragendorff's test	+	+	+	+	+	+
	Mayer's test	+	+	+	+	+	+
	Wagner's test	+	+	+	+	+	−
3	**Glycosides**						
	Borntrager's test	+	+	−	+	+	−
	Legal test	+	+	−	+	+	+
4	**Phenolic compounds& tannin**						
	Ferric chloride test	+	+	+	+	+	+
5	**Flavanoids**						
	Shinoda	+	+	−	+	+	−
	Lead acetate test	+	+	−	+	+	−
6	**Proteins & amino acids**						
	Millon's reagent	−	+	−	+	+	−
	Ninhydrin reagent	−	+	−	−	+	−
7	**Saponins**						
	Foam test	+	+	−	+	+	−
	Sodium bicarbonate test	+	+	−	+	+	−
8	**Triterpenoids**						
	Salkowski test	+	+	+	+	+	+
	Liebermann-Burchard's test	−	+	+	+	+	+

+ (positive) represents presence of the Phytoconstituent
− (negative) represents absence of the Phytoconstituent
Aq.: Aqueous; Me.: Methanolic; Chl.: Chloroform; and Ext.: Extract

Table 6. Heavy metal contamination of PKLF and PKMF

Sr. No.	Heavy Metal	Observation (ppm)		Permissible limits (ppm) As per (API)
		PKLF	PKMF	
1	Mercury	<0.1	<0.1	1
2	Cadmium	<1.0	<1.0	0.3
3	Lead	<1.0	<1.0	10
4	Arsenic	<1.0	<1.0	3

Table 7. Total bacterial count and Total fungal count of PKLF and PKMF

Sr. NO.	Microbial contamination	Observation (cfu/g)		Permissible limits As per (API)
		PKLF	PKMF	
1	Total bacterial count	15×10^2	24×10^2	NMT 10^7cfu/g
2	Total fungal count	2×10^1	12×10^1	NMT 10^4cfu/g

Table 8. Test for specific Pathogen of PKLF and PKMF

Sr. No.	Pathogen	Observation (ppm)		Permissible limits (ppm) As per (API)
		PKLF	PKMF	
1	Escherichia coli	Absent	Absent	NMT 10^2cfu/g
2	Salmonella spp.	Absent	Absent	Absence Per Gram
3	Pseudomonas aeruginosa	Absent	Absent	Absence Per Gram
4	Staphylococcus aureus	Absent	Absent	Absence Per Gram

Table 9. Test for Pesticide residue of PKLF and PKMF

Sr. No.	Pesticide	Observation (ppm)	
		PKLF	PKMF
1	Organochlorine pesticides	Not Detected	Not Detected
2	Organophosphorus pesticides	Not Detected	Not Detected
3	Pyrethroids	Not Detected	Not Detected

Table 10. Estimation of Aflatoxins of PKLF and PKMF

Sr. No.	Aflatoxins	Observation (ppm)		Permissible Limit (ppm) As per (API)
		PKLF	PKMF	
1	B1	<10	<10	0.5
2	G1	<10	<10	0.5
3	B2	<10	<10	0.1
4	G2	<10	<10	0.1

DISCUSSION

Some important organoleptic features of PathyadiKada laboratory and market formulations like color, odour, taste, appearance, touch, etc were documented.

Determination of physico-chemical constants indicated a Loss on drying of 34.99 % & 36.11%, total ash value 00.19 and 00.47 %w/w, acid insoluble ash was <0.01, pH, Viscosity, Refractive index, specific gravity and Phenolic content 164.62 and 192.96 respectively were documented for Laboratory formulation and marketed formulation The Patyadikada Laboratory formulation and marketed formulation were tested for the presence of heavy metals, Microbial content, pesticide residues and aflatoxin which were also found which arewithinthelimitsofWHOspecifications.

Preliminary phytochemical studies of PatyadiKadha revealed the presence of carbohydrates, alkaloids, phenolic compounds and tannins, flavonoids, steroids and terpenoids as chief constituents.

The phytochemical studies showed presence of terpenoidal moieties in the PatyadiKadawhile performing co-TLC studies with some of similar compounds available the Rf of one of the components corresponded with that of Berberine and Quercetin thus revealed its presence. Therefore, simple, sensitive, specific and reproducible HPTLC method was developed for thequantification of Berberine and Quercetin as marker constituents to ensure identity and quality of PatyadiKadha.

CONCLUSION

Thus, from the present study, it can be concluded that the pharmacognostical studies carried out on PatyadiKada can serve as a valuable tool and provide suitable standards for the identification of Ayurvedic formulations. Phytochemical screening and the quantitative HPTLC assay method developed for quantification of Berberine and Quercetin in PatyadiKada can serve for the routine quality control of commercial samples and help in justifying the traditional claims endowed upon the formulations in a scientifically acceptable manner.

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