

Development and Evaluation of Polyherbal Hair Oil Fortified with Milk of Indigenous Cow

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Abstract

Hair, an accessory part of the body is important for overall appeal of the body. Ailments related with hair and scalp is chronic dermatological condition, for which several chemical based products are available in the market but with adverse side effects. In the present study a polyherbal hair oil fortified with milk of indigenous cow was developed, standardized using physico-chemical parameters such as Refractive index, weight per ml, iodine value, acid value, peroxide value, saponification value and HPTLC. Efficacy of Hair Oil was carried out by repeated open application test - (ROAT) on adult human volunteers for four weeks to determine if the Hair oil is efficient in controlling Dandruff and itching of scalp and thus improving overall hair quality in human subjects. Parameters used for efficacy with respect to Hair were dryness, roughness and texture of hair and with that of Scalp were dandruff and itching of scalp. At the end of the fourth week, more than 80% participants got Excellent results for quality of hair and scalp. Based on the results obtained from the Study, it can be concluded that use of polyherbal hair oil fortified with cow milk produces an evident clinical improvement in hair and scalp ailments. The product is suitable for micro industries.

Keywords: Hair; Scalp; Polyherbal; Oil.

Introduction

Hair is a string of dead keratin cells [1]. It is an accessory part of the body derived from ectoderm of the skin. It is important for overall appeal of the body [2]. They also can be termed as a mirror of nutritional imbalance of the body. Ailments related with hair and scalp is chronic dermatological condition, its etiology is not properly understood but its unpredictability of the condition together with its visible nature can result in stressful and psychological inferiority for the patients [3]. Fashion pro Hair strengthening, highlighting, coloring, use of chemical based creams lotions result in hair loss, premature graying, Minoxidil, a common chemical based treatment for hair ailments show side effect after prolong use [4]. Herbal hair care oils are one of the most well recognized hair treatments. Ayurvedic hair oils not only moisturize scalp, reverse dry scalp and dry hair conditions, but provide numerous essential nutrients required to maintain normal functions of sebaceous glands and promote natural

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hair growth. Hair oils have been traditionally used to treat irritated stressed scalp, reduce effect of aging on hair shape and growth, combat seborrhoea and alopecia. Herbal hair care oils work through nutritional support of natural skin restorative and hair growth processes. Regular continuous application is the best way to achieve significant improvement. In various ailment of hair, preference is being given to herbal hair oils [5]. They not only support hair growth and improve elegance of hair but also prevent hair fall [6]. Hair oil also provides essential moisture to the scalp depicting in beautiful hair [7].

Many herbs and oils like coconut oil, sesame oil show excellent effect on hair and scalp ailments. The present study aims at formulation, standardization, and efficacy study of polyherbal hair oil based on kshirpak vidhi of its preparation. Use of indigenous cows milk enhances the extraction of the herbs along with minimizing the rancidity of the oils. The use of herbs like *Amla*, *Hirda*, *Baheda*, *Bhrungraj*, *Jatamansi*, *Kapoorkachari* as an oil base extracts are found to be very effective. *Emblica officinalis*, *Gaertn* (Fruit), *Terminalia chebula Retz* (Fruit), *Terminalia bellirica* (*Gaertn*) *Roxb* (Fruit) belonging to family (Euphorbiaceae) contains tannins and minerals, providing nutrition to hair (8), *Amla* is rich in vitamin C, tannins and minerals such as phosphorus, iron and calcium which provides nutrition to hair and also causes darkening of hair (9), *Eclipta alba* Hassk (Whole plant) is well known in Ayurved for hair treatments. *Nardostachys jatamansi* (Dried rhizome), *Hedychium spicatum* (Rhizome) are known for their soothing effect and mental peace.

The present study aims to standardize the product according to Ayurvedic Pharmacopoeia of India, and study the efficacy of the product.

Materials and Methods

The dried rhizome of *Nardostachys jatamansi* DC., *Hedychium spicatum*, dried whole plant of *Eclipta alba* Hassk, dried fruits of *Emblica officinalis* Gaertn, *Terminalia chebula* Retz, *Terminalia bellirica* (Gaertn) *Roxb*, fresh fruits of *Citrus lemoni* and dried root and rootstock of *Arnebia euchroma* (Royle) Johnston var were purchased from local market and authenticated by Dr. S.K. Padoley, Head, Dept. of Botany, Porwal college of Science, Kamptee, R.T.M. Nagpur

University, Nagpur. Sesame oil and Coconut oil were purchased from locally from *Ghani*. Milk of indigenous cow (Gir) was provided by *Goshala* of Go-Vigyan Anusandhan Kendra, Dewalapar, Nagpur.

Formulation of Hair Oil

The herbal materials used for the formulation were checked as per the Ayurvedic Pharmacopoeia of India. There are various methods available for the preparation of hair oils direct boiling method, paste method and cloth method (10). All the herbal materials except *Citrus lemoni* were dried in shade, crushed and passed through the sieve number 80. Measured quantity of Sesame oil and Coconut oil were taken in a pan and heated on a low flame. Pieces of *Citrus lemoni* were added to it. Measured quantity of milk of indigenous cow (Gir) was added in pan. *Arnebia euchroma* (Royle) Johnston var was added to it to get naturally red coloured hair oil. Rounded dough mentioned as *Kulk* in Ayurveda were prepared by adding minimum quantity of water in weighed quantity of *Eclipta alba* Hassk and *Triphala* powder prepared by mixing equal quantity of *Emblica officinalis* Gaertn, *Terminalia chebula* Retz. and *Terminalia bellirica* (Gaertn) *Roxb*. Dough were added in boiling oil. Completion of heating process was tested by deeping cotton weaks and holding it under the flame for smooth burning. After satisfactorily heating, the oil was filtered through a muslin cloth and allowed to cool. Weighed quantity of *Nardostachys jatamansi* DC. and *Hedychium spicatum* were mixed in a piece of cotton cloth which was tied, dipped in the oil and left for seven days. Finally the oil was filtered to get finished product. Composition of the formulated polyherbal hair oil is summarized in Table 1.

Table 1: Composition of polyherbal hair oil fortified with cow milk

S. No.	Botanical name/English name of ingredients	Hindi names	Quantity
1.	Sesame oil	Til Tail	200 g
2.	Coconut oil	Nariyal tail	1800 g
3.	<i>Arnebia euchroma</i> (Royle) Johnston var (Dried root and rootstock)	Ratanjot	25 g
4.	<i>Nardostachys jatamansi</i> DC. (Dried rhizome)	Jatamansi	25 g
5.	<i>Hedychium spicatum</i> (Rhizome)	Kapoorkachari	25 g
6.	<i>Eclipta alba</i> Hassk (Whole plant)	Bhrungraj	25g
7.	Mixture of	Trifala	75 g
	1. <i>Emblica officinalis</i> Gaertn (Fruit)	Mixture of 3 herbs in equal parts :	
	2. <i>Terminalia chebula</i> Retz (Fruit)		
	3. <i>Terminalia bellirica</i> (Gaertn) <i>Roxb</i> (Fruit)		
8.	<i>Citrus lemoni</i> (Fruit)	Nimboo	6
9.	Milk of indigenous cow	Godugdha	2 L



Ingredients of hair oil



Preparation of hair oil



Filtration of hair oil

Standardization of Developed Formulation (DF)

The formulated hair oil was prepared in three batches and tested for following physico-chemical parameters as per methods mentioned in Ayurvedic Pharmacopoeia of India published by Government of India.

Refractive Index

The refractive index was measured at 25° C by placing a drop of DF in the prism of Abbe's refractometer.

Weight /ml

Weight per ml of the oil was measured at 25° C using dry pycnometer.

Saponification Value

The saponification value is the number of mg of potassium hydroxide required to neutralize the fatty acids, resulting from the complete hydrolysis of 1 g of the oil or fat, when determined by the following method:

35 to 40 g of potassium hydroxide was dissolved in 20 ml water, sufficient alcohol was added to make 1,000 ml. and allowed to stand overnight. The clear liquor was poured off. 2 g of the DF was weighed in a tared 250 ml flask, 25 ml of the alcoholic solution of potassium hydroxide was added. The flask was attached to a reflux condenser and boiled on a water-bath for one hour, with frequently rotating the contents of the flask. It was cooled, 1 ml of solution of phenolphthalein was added and the excess of alkali was titrated with 0.5 N hydrochloric acid. The number of ml required (a) was noted. The experiment was repeated with the same quantities of the same reagents in the manner omitting the sample. The number of ml required (b) was noted. The

saponification value was calculated using the following formula:—

$$\text{Saponification Value} = \frac{(b-a) \times 0.02805 \times 1.000}{W}$$

Where 'W' is the weight in g of the sample taken.

Iodine Value

It was determined by using Iodine Monochloride Method

Acid Value

The acid value is the number of mg of *potassium hydroxide* required to neutralize the free acids in 1 g of the substance, when determined by the following method:

10 g of the oil was weighed into a 250 ml flask, 50 ml of a mixture of equal volumes of alcohol and solvent ether were added to it, which was neutralized after the addition of 1 ml of solution of phenolphthalein. The flask was heated gently on a water-bath and titrated with 0.1 N potassium hydroxide, shaking constantly until a pink colour with persistence for fifteen seconds was obtained. The number of ml required was noted. The acid value was calculated from the following formula:

$$\text{Acid Value} = \frac{a \times 0.00561 \times 1000}{W}$$

Where 'a' is the number of ml of 0.1 N potassium hydroxide required and 'W' is the weight in g of the sample taken.

Rancidity Test

The rancidity test was carried out as per Kreis test.

Mineral Oil

The test for detection of mineral oil was carried out as per Holde's test.

High Performance thin Layer Chromatography

2 g of formulated Hair oil was taken in a flask and 20 mL methanol was added to it. The flask was placed in rotator shaker for 3 hours at 40°C, then cooled and alcoholics layer was separated to get methanolic extract. It was concentrated to about 5 ml. 10 µl of the methanolic extract of hair oil along with same quantity of methanolic extracts of all the ingredients was applied on Silica gel "G" plate (F 254) and developed to a distance of 8 cm using Toluene: Methanol: Ethyl acetate (6: 0.5: 3) as mobile phase.

Evaluation of Efficacy

Efficacy of Hair Oil was carried out by repeated open application test - (ROAT) on adult human volunteers for four weeks to determine if the Hair oil is efficient in controlling Dandruff and itching of scalp and thus improving overall hair quality in human subjects.

Selection of Volunteers for Study

A total of 07 volunteers (2 male and 5 female) of age group 18-45 years satisfying following eligibility criteria were enrolled in the study.

Inclusion Criteria

Healthy male and female volunteers having at least one problem related hair /Scalp ailments such as dry hair, rough hair, damaged hair, dandruff & itching of scalp were selected and asked to refrain themselves from the use of any other means of Dandruff control or hair treatment other than the test product. Written Informed Consent was received from the volunteers after informing details of the treatment procedures, benefits and potential risks, if any.

Exclusion Criteria

Exclusion criteria were individuals under Hair/Scalp therapy at least one month prior to the study and using Specific anti dandruff products, individuals who were medically compromised, individuals who had undergone pharmacological treatments, individuals with past history of contact dermatitis or who were using

any other branded Hair products.

Design of Efficacy Study

The volunteers were enrolled for the efficacy study after satisfying inclusion criteria. Case history of subjects with regard to Hair/Scalp related problems were obtained and their demographic medical histories were recorded. All the subjects were provided with the product and instructed to massage gently their Scalp and hairs with the allocated Hair oil for 10 minutes both in the morning and before going to bed in the night. Subjects were instructed to refrain from any other Hair/Scalp products. The volunteers were examined for hair quality and scalp quality at baseline and after hair wash on 2nd, 3rd, and 4th week of the study with the help of magnifying lens.

Assessment of Efficacy

Four point scale (i.e. bad, average, good & excellent) for hair & scalp related ailment was the basis for overall efficacy assessment of therapeutic effect. Volunteers underwent for follow up for four weeks on the basis of following criteria:

Hair: dryness, roughness and texture of hair

Scalp: Dandruff and itching of scalp

Results and Discussion

Standardization of developed formulation (DF)

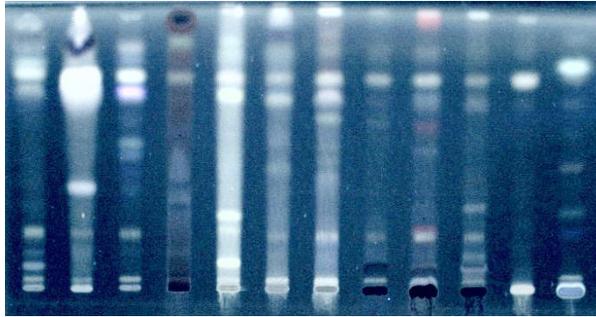
Based on data generated from analysis of three batches of hair oil, range of values for each parameter was obtained and summarized in Table 2.

HPTLC Studies

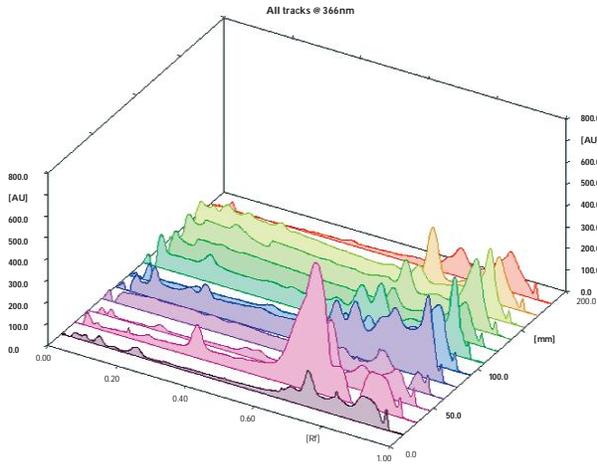
HPTLC of methanolic extract on Silica gel "G" Toluene:Methanol:Ethyl acetate (6:0.5:3) shows 3 major spots at 254nm at Rf.0.11, 1.84 and 1.93 (all black). On spraying with 5% Methanolic Sulphuric acid reagent and heating the plate at 105°C for ten minutes; shows under U.V. 366 at Rf.0.04 (yellowish), 0.07(blue), 0.11(faint blue), 0.22(yellow), 0.67(faint blue), 0.72(pink), 0.81, 0.89, 0.95 .

Table 2: Standards Developed for DF

S.no.	Parameters	Obtained range
1.	Refractive index at 25 °	1.450 to 1.460
2.	Weight /mL at 25 °	0.90 to 0.92
3.	Saponification value	210 to 230
4.	Iodine value	10.00 to 12.00
5.	Acid value	Not more than 4
6.	Rancidity test	Negative
7.	Mineral oil	Absent



Chromatogram: Derivatized at 366 nm



Graph for Derivatized plate at 366 nm

Efficacy of DF

Results obtained for evaluation of hair quality and scalp quality are as summarized in table 3 and 4 respectively. Total of 07 Participants were enrolled in the present study out of which 02 were male and 05 were Female. Among 7 participants, 06 participants enrolled in the study were complaining about dry/rough or damaged hair and 05 were complaining for Dandruff and itching of scalp. At the end of the study there were total of 07 participant with no drop offs.

At the end of the fourth week, out of total 07 participants, 05 participants got Excellent results for Hair quality (Dryness, Roughness, texture of hair) and 02 participants got Good results for Hair quality.

At the end of the fourth week, out of total 07 participants, 04 participants got Excellent results for Scalp quality (Dandruff and itching of Scalp) and 03 participants got Good results for Scalp quality.

In all the participants, any kind of complications or adverse effects was neither reported by participants nor observed by the investigator. It is obvious that addition of herbs in cosmetics does not have any side effect on skin (11, 12, 13). Results obtained for evaluation of hair quality and scalp quality are as summarized in Table 3 and 4 respectively.

Table 3: Observation sheet, (Assessment results for Hair quality)

Participant no.	Age/Sex of participant	Week of observation	Assessment results for Dryness, Roughness, texture of hair			
			Bad	Average	Good	Excellent
1	24/F	0 th	■			
		2 nd			■	
		3 rd				■
		4 th				■
2	30/F	0 th	■			
		2 nd		■		
		3 rd				■
		4 th				■
3	26/M	0 th		■		
		2 nd		■		
		3 rd			■	
		4 th			■	
4	24/M	0 th		■		
		2 nd		■		
		3 rd			■	
		4 th			■	
5	19/F	0 th	■			
		2 nd			■	
		3 rd				■
		4 th				■
6	25/F	0 th	■			
		2 nd			■	
		3 rd			■	
		4 th				■
7	42/F	0 th			■	
		2 nd				■
		3 rd				■
		4 th				■

■ Indicates positive results

Table 3: Observation sheet
(Assessment results for Scalp quality)

Participant no.	Age/Sex of participant	Week of observation	Assessment results for Dandruff and itching of Scalp			
			Bad	Average	Good	Excellent
1	24/F	0 th	■			
		2 nd		■		
		3 rd			■	
		4 th				■
2	30/F	0 th	■			
		2 nd			■	
		3 rd			■	
		4 th				■
3	26/M	0 th			■	
		2 nd			■	
		3 rd			■	
		4 th			■	
4	24/M	0 th			■	
		2 nd			■	
		3 rd			■	
		4 th				■
5	19/F	0 th	■			
		2 nd		■		
		3 rd		■		
		4 th			■	
6	25/F	0 th	■			
		2 nd		■		
		3 rd			■	
		4 th			■	
7	42/F	0 th			■	
		2 nd			■	
		3 rd			■	
		4 th				■

■ Indicates positive results

Conclusion

Based on the results obtained from the Study, it can be concluded that use of polyherbal hair oil fortified with cow milk produces an evident clinical improvement in hair and scalp ailments. The demand for medicines that modify hair augmentation and look has led to a multibillion dollar industry [14]. The product is suitable for micro industries.

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