

Formulation, Evaluation and Comparison of the Polyherbal Shampoo with the Commercial Shampoos

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Abstract

The present study investigated the effect of Liquid Polyherbal shampoo prepared using herbal extracts with a prominence on safety and efficacy. Polyherbal shampoos prepared using the natural extracts like Pomegranate peel, Reetha, Amla & Hibiscus in different concentrations. Physico-chemical properties and its stability evaluated for manufactured Polyherbal shampoos. Statistical software used to optimize the formulations and to understand main & interaction effects on the shampoo properties along with comparison with commercial products. Prepared Liquid Polyherbal shampoos pH values are nearer to the skin pH. The shampoos containing higher level of Pomegranate content revealed higher detergency and anti-dandruff effect, Amla as well as Reetha showed significant effect on the foaming capacity and cleaning action and similar to commercial formulations. Higher quantity of Hibiscus concentration shown better conditioning behaviour. The manufactured shampoos were stable, unvarying, thicker and comparable as that of commercially marketed shampoos. Based on the Design expert statistical evaluation of results, liquid shampoo formulations containing 1%-3% w/w of Amla, 6%-10% w/w of Pomegranate, 10%-15% w/w Reetha and 11.25%–15% w/w of Hibiscus Polyherbal shampoos exhibited superior performance compared with marketed formulations. LS6, LS16, LS17 & LS20 preparations were identified as the greatest and optimum formulations based on physico-chemical related properties. The identified Polyherbal shampoos have an excellent cleansing, detergency, anti-dandruff effect, conditioning and foaming ability and is ideal for normal hair and has favourable pH with stable organoleptic features. Selected formulations stability results found satisfactory up to 6 months.

Keywords: Amla, Hibiscus, Pomegranate, Polyherbal Shampoo, Reetha

1. Introduction

Now people interested in the Liquid Polyherbal shampoos other than commercially available synthetic shampoos¹. Different types of Polyherbal shampoos prepared using various types of plant/natural extracts and its combinations.

Dandruff is clinical condition which is seen more often including children in now days. Pityrosporum (Malassezia) species cause Dandruff and its Pityrosporum ovale play a role in the manifestation of seborrheic dermatitis². Anti-dandruff shampoos contain

fungistatic ingredients for control of Dandruff³. Anti-dandruff shampoos containing natural components more acceptable in the market due to its less side effects and safety. Currently in the market different types of nutritional and Anti-dandruff shampoo's available with vitamin, amino acids and proteins⁴.

In continuation to our previous research work, the article covers preparation of Liquid Polyherbal anti-dandruff shampoos using available herbal extracts in combination^{5,6}. Herbal extracts like Amla, Reetha, Pomegranate Peel and Hibiscus used for preparation of Liquid Polyherbal shampoo.

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Presence of Polyphenols in the Pomegranate peels powder it reduces hair loss, dandruff and darkens the hair colour⁴. Pomegranate extracts stimulating hair growth and strengthens hair follicles by improving blood flow to the scalp. The presence of high saponin content in Reetha provide hair cleansing property and used as foaming agent⁷. *Hibiscus rosa-sinensis* (Hibiscus) of Malvaceae family helps to boost hair growth, conditioning behaviour and strength the hair. Hibiscus can help to nourish hair and to reduce split ends⁸. Acidic nature presence in amla promotes healthier hair growth, tighten hair follicles and prevent hair fall. It shown better detergency property and shininess of the hair⁹. prepared poly herbal powder shampoos were comparatively assessed for organoleptic, dirt removal, detergent ability, cleaning action, frothing or foaming ability, anti-dandruff activity, pH and conditioning performance¹⁰.

2. Materials and Methods

Amla, Reetha, Pomegranate and Hibiscus extracts were obtained from the Visakhapatnam local bazaar and other components used in this work are either Laboratory testing or medicinal grade. The polyherbal shampoo preparation was done in two parts. Part-A is preparation of powder pre-formulation and Part-B is making the powder pre-formulation into a liquid Polyherbal shampoo.

2.1 Preparation of Powder Poly Herbal Pre-Formulations (Part-A)

Suitable quantity of herbal extracts were individually weighed and screened through 60# mesh. The amount of Amla, Reetha, Pomegranate and Hibiscus were added in altered propositions based on DoE and mixed to get an even mixture of pre-formulation & are evaluated. The evaluation range of assorted propositions of pre-formulations are between 2% to 10% w/w of Pomegranate peel extract, 5% to 15% w/w of Reetha; 1% to 3% w/w of Amla and 7.5% to 15% w/w of Hibiscus concentrations.

2.2 Preparation of Liquid Poly Herbal Shampoos (Part-B)

The powdered pre-formulations were mixed with 1% Guar gum solution to attain better consistency and 10 to 20% w/w solid content in the liquid poly herbal shampoos.

Design-Expert® software was exploited for statistical evaluation and identification of optimized formulations considering physicochemical properties (Part-2). Central composite response surface DoE was selected to augment the composition related aspects and also to analyse key and interaction effects on the properties of polyherbal shampoo. The Alpha value of 1 (face centred) was chosen in the central composite design gives 3 levels of each factor to study the effects in Design Expert® (Software Ver.:12.0.4, Make: Stat-Ease Inc.) for analysis. The ANOVA was used to calculate influence of independent variables on the responses. The study parameter was considered statistically significant if the *P-value* is less than 0.05. The details of the 21 experimental trials and the respective responses were observed and tabulated below (Table 1 & 2).

Table 1. Central Composite response surface design was selected to study three concentration levels of herbal extract used for formulating of Polyherbal shampoo

Factors: Formulation Variable		Level		
		-1	0	+1
A	Amla (% w/w)	1	2	3
B	Reetha (% w/w)	5	10	15
C	Pomegranate (% w/w)	2	6	10
D	Hibiscus (% w/w)	7.5	11.25	15
Response		Target-range		
Y1	Detergency (%)	80 -98		
Y2	Cleaning (%)	5.0 - 7.0		
Y3	Foaming (%)	35 - 50		
Y4	pH	3 - 4		

Table 2. Details of experimental plan

Formulation	A:	B:	C:	D:
	Amla	Reetha	Pomegranate	Hibiscus
	% m/m	% m/m	% m/m	% m/m
LS1	2	10	6	11.25
LS2	2	10	6	11.25
LS3	2	5	6	11.25
LS4	2	10	6	11.25
LS5	1	5	2	7.5
LS6	2	10	6	15
LS7	2	10	10	11.25

LS8	3	5	10	15
LS9	3	5	2	15
LS10	2	10	6	11.25
LS11	2	10	6	11.25
LS12	2	15	6	11.25
LS13	1	5	10	7.5
LS14	1	15	2	15
LS15	3	15	2	7.5
LS16	1	15	10	15
LS17	3	10	6	11.25
LS18	1	10	6	11.25
LS19	2	10	2	11.25
LS20	3	15	10	7.5
LS21	2	10	6	7.5

LS – Liquid poly herbal shampoo.

2.3 Evaluation of Poly Herbal Shampoos

The polyherbal shampoo evaluation was done in two parts. Part-1 evaluation is for general powder characteristics (i.e. with the key ingredients used for formulation) and part-2 evaluation is for physico-chemical properties of liquid shampoo.

After preparation of the Polyherbal pre-formulations as per experimental plan; the evaluation of Polyherbal pre-formulations was performed for powder characteristics (i.e. Power blend earlier mixing with Guar gum 1% solution) for organoleptic and general powder characteristics evaluation. Along with the poly herbal pre-formulations powder characteristics; Liquid Polyherbal shampoos physico-chemical properties were evaluated and the details are given below.

2.3.1 Physical Appearance/Visual Inspection

As Part of pre-formulation studies, Poly herbal shampoos organoleptic evaluation was performed (colour, odour and texture). By visual observation Colour was identified and by touch sensation texture was evaluated. A team of 5 members were evaluated odour sensitivity¹¹.

After reconstitution with Guar gum 1% solution Liquid Poly Herbal shampoos were also evaluated for colour, odour and texture similar to Polyherbal pre-formulations.

2.3.2 Poly Herbal Pre-Formulation Powder Characteristics (Part-1)

The powder characteristics of poly herbal pre-formulation contain assessment of the parameters that effect on the parameters corresponding to its appearance, angle of repose, Bulk and tapped densities of the preparations. Features assessed for pre-formulation were of powder characteristics, particle size, angle of repose and density.

2.3.3 Physico-Chemical Evaluation of Liquid Poly Herbal Shampoos (Part – 2)

2.3.3.1 Determination of pH and physical appearance

The pH for shampoo with 10% solutions in distilled water was measured at 25°C with a pH meter, and its physical appearance was noted¹².

2.3.3.2 Dirt dispersion

About two droplets of liquid shampoo were added to test tube containing 10 ml of distilled water. One droplet of Indian ink dye was inserted and the contents in the test tube mixed and forth horizontally by closing the lid for ten times. The amount of colour (i.e. ink) in the frothing was estimated (None, Light, Moderate, or Heavy)¹³.

2.3.3.3 Cleaning action

A combination of five grams of soil and 0.5 grams of acacia and about 0.5 grams of hair crumple was mixed with 5 ml of water. The soiled hair is washed with the water having one gram of shampoo then the hair was dried. The weight of hair assumed as test weight¹⁴. The hair crumple cleaned with water and with no shampoo, was considered as control. The following equation was considered to calculate% of cleaning action:

$$\text{Cleaning percentage (CP)} = 100(1-T/C) \quad (1)$$

Where,

C is the mass of hair without shampoo;

T is the mass of hair after shampoo.

2.3.3.4 Detergency

The detergency ability of the samples was done by Thompson method. Momentarily, a swatch of hair is cleaned with five percent sodium lauryl Sulfate solution, after drying divided into three grams. The divided portions were slurred in 10% artificial sebum in n-hexane

solution for 15 minutes at 25°C. Then hair was taken out, the solvent was removed by evaporation at 25°C and determined the content of sebum. As a further process, each sample of hair was separated into two equal parts, 1st part was cleaned with 0.1 ml of the 10% test shampoo and the 2nd part was considered as the (-)ve control. Once dried, the adhered sebum was removed with 20 ml n-hexane and checked the weight. The percent DP was evaluated using the equation 2¹⁵

$$\% \text{ detergency power (DP)} = 100 (1 - T/C) \quad (2)$$

Where,

C = mass of sebum for control sample, and

T = mass of sebum for test sample.

2.3.3.5 Foaming ability and foam stability

Foaming ability estimated using Cylinder shake method. Fifty ml of one percent polyherbal liquid shampoo solutions were poured in a measuring jar (250 ml) and lid closed with hand and mixed the contents to and forth horizontally for ten times. The volume of the frothing after mixing was recorded. The frothing measured immediately after mixing and the volume of foam at each minute interval up to four minutes, to evaluate the stability¹⁶.

2.3.3.6 Estimation of conditioning performance

Each 10 cm and weight about 5 gm hair tress was fragmented in to different samples for evaluation of conditioning performance. One of the sample hairs without cleaning will be used as control sample. Remaining three samples were cleaned using prepared Polyherbal shampoos along with commercial market shampoos with uniform cleaning procedure. For each step, the individual sample hair mixed with 15 grams of water in a flask for two minutes and further washed using fifty milliliters of water. After completion of cleaning, samples were dried in air at approximately 25°C. Same procedure was repeated for ten times. The dried samples were evaluated for smoothness and softness using blind touch test by using selected twenty Participants¹⁷. The Participants are blind folded to rate the samples for providing score from 1 to 4. (4- Excellent; 3- Good; 2- Satisfactory & 1- Poor).

2.3.3.7 Evaluation of in-vitro anti-dandruff activity

Anti-dandruff activity was evaluated by *Candida albicans* using cup & plate technique. Sabouraud's agar slants

maintained & inoculated with 0.2 ml of seventy two hours old suspension of *Candida albicans* poured into Petri dish. The 0.05 ml of a solution of sample dissolved in DMSO (10 ml of liquid shampoo in 100 ml of DMSO) filled in the punched cups (10 mm diameter). Filled placed for diffusion at 40°C for one hour and incubated at 30°C for forty eight hours. After completion of 48 hours duration, zone inhibition in mm was measured¹⁸⁻²⁰.

2.3.3.8 Viscosity estimation

Viscosity estimation for Prepared liquid Poly herbal shampoos were performed by using Brookfield Viscometer (LVDV Prime-I spindle). The measurement of viscosity was performed at room temperature (30±2°C).

2.3.3.9 Evaluation of stability of liquid poly herbal shampoo

Stability studies of liquid polyherbal shampoos were performed in line with the regulatory guidelines. The prepared formulations were stored at room temperature (30±2°C) for duration of six months. After completion of six months interval, samples were tested for their physical appearance, pH, viscosity, detergency effect, cleaning action and foam stability.

3. Results and Discussion

With the help of herbal extracts, Polyherbal powdered pre-formulated by mixing different constituents in specific proportions. Selected plant materials are rich in polyphenol compounds such as a flavonoid, phenolic and saponin. They have found to exhibit cleansing and surfactant properties. These herbal extracts also provided viscosity to the formulations. Physicochemical properties of the poly herbal powdered shampoos were statistically evaluated using in-silico tools (statistical Software) to select optimized formulations. The formulations were analysed during the initial month of preparations and after the sixth month time duration, to check their stability and found results similar to the initial analysis.

3.1 Physical Appearance/Visual Inspection

The parameters corresponding to texture, colour and odour are evaluated for organoleptic properties. Colour was evaluated by vision and texture was evaluated by touch sensation. The odour was evaluated by a group

Table 3. Organoleptic evaluation

Sl. No.	Organoleptic evaluation	Result
1.	Colour	Light Brown
2.	Odour	Mild pleasant
3.	Texture	Slippery

of six super smellers by disorganized sampling. All the formulations exhibited acceptable organoleptic characteristics as shown in Table 3.

As shown in Table 3, the formulations of good characteristics in physical properties. Pre-formulations have optimum acceptable odour, light brown colour and a smooth texture.

After reconstitution of liquid Polyherbal shampoos shown acceptable odour, light brown colour suspension and smooth slippery texture.

3.2 Poly Herbal Pre-formulation Powder Characteristics (Part-1)

Pre-formulation Powder Characteristics comprise assessment of the parameters that effects on the appearance, flow properties, sedimentation during the shelf life. The characteristic parameters assessed were of size of particles, angle of repose and weight by volume (i.e. density). Selected one of the pre-formulations i.e. LS17 for general powder characteristics evaluation and results were adequate and acceptable and are given in Table 4-7.

The key ingredients for poly herbal pre-formulations have an optimum particle size and good flow

Table 4. Powder characteristics

Sl. No.	Powder characteristics	Result
1.	Particles Distribution /PSD (in microns)	20 – 30
2.	Angle of repose	32.2
3.	Untapped Bulk Density (g/ml) [BD]	0.321
4.	Tapped Bulk Density (g/ml) [TBD]	0.385

Table 5. Angle of repose calculations for poly herbal pre-formulation

Repli- cate	Meth- od	Height of cone (cm)	Radi- us of cone (cm)	tan θ = (h/r)	Av- erage tan θ	θ = tan ⁻¹ (h/r)	Flow prop- erty
1	Funnel	2.4	3.9	0.615			
2	Meth- od	2.5	3.8	0.658	0.629	32.2	Good
3		2.4	3.9	0.615			

Table 6. Tapped density calculations for poly herbal pre-formulation

Replicate	Tapped volume	Mass of the powder	Tapped density (g/ml)	Avg. Tapped density (g/ml)
1.	52	20.1	0.387	
2.	54	20.5	0.380	0.385
3.	52	20.3	0.390	

Table 7. Bulk density calculation for poly herbal pre-formulation

Replicate	Bulk volume (ml)	Mass of the powder (g)	Bulk density (g/ml)	Avg. Bulk density (g/ml)
1.	63	20.1	0.319	
2.	65	20.5	0.315	0.321
3.	62	20.3	0.327	

characteristics indicating an optimal formulation can be attained. For the shampoo; Particle size is an important aspect, as it has a direct impact on the aesthetic values of the product which connects with person in use. For flow related characteristics; angle of repose and density was found to be adequate, which has to be considered while formulating an ideal shampoo.

3.3 Physicochemical Evaluation (Part -2)

Physico-chemical evaluation of liquid poly herbal shampoos formulated was tested for dirt dispersion, detergency, cleaning, foaming and pH. The data provided in Table 8.

Table 8. Assessment of liquid poly herbal shampoos for pH, dirt dispersion, cleaning action and detergency

Formulations	A: Amla %	B: Reetha %	C: Pomegranate %	D: Hibiscus %	Detergency %	Cleaning %	Foaming %	pH	Dirt dispersion
LS1	2	10	6	11.25	83	5.43	37.6	3.54	Light
LS2	2	10	6	11.25	84	6.1	36.9	3.61	Light
LS3	2	5	6	11.25	74	5.84	35.2	3.24	Light
LS4	2	10	6	11.25	84	5.92	38.9	3.25	Light

LS5	1	5	2	7.5	76	4.12	36.1	3.10	Light
LS6	2	10	6	15	93	6.41	44.1	3.81	None
LS7	2	10	10	11.25	88	5.32	39.2	3.37	Light
LS8	3	5	10	15	78	6.1	36.7	3.42	Light
LS9	3	5	2	15	75	5.97	34.2	3.29	Light
LS10	2	10	6	11.25	82	4.89	38.1	3.08	Light
LS11	2	10	6	11.25	85	5.07	37.4	3.40	Light
LS12	2	15	6	11.25	86	5.32	39.8	3.76	Light
LS13	1	5	10	7.5	81	4.62	40.6	3.53	Light
LS14	1	15	2	15	90	4.83	42.8	3.04	Light
LS15	3	15	2	7.5	91	5.36	41.9	3.47	Light
LS16	1	15	10	15	97	6.28	45.2	3.62	None
LS17	3	10	6	11.25	98	6.43	46.1	3.71	None
LS18	1	10	6	11.25	81	4.92	38.4	3.66	Light
LS19	2	10	2	11.25	83	5.22	39.3	3.28	Light
LS20	3	15	10	7.5	95	6.63	48.4	3.79	None
LS21	2	10	6	7.5	86	5.44	36.5	3.85	Light
MF1	--	--	--	--	94	5.87	44.3	4.29	None
MF2	--	--	--	--	95	6.12	45.5	4.04	None

LS - Liquid poly herbal shampoo; MF - Marketed Formulation.

3.3.1 Determination of pH

The liquid polyherbal shampoos pH was determined in 10% distilled water at 25°C. As depicted in data Table 8, the entire liquid poly herbal shampoo formulations were of acidulous and were in the range of 3.04- 4.29, and is close to the dermal pH. A graphical bar chart representation was presented in Figure 1.

The pH value has a significant effect and can improve and enhance hair quality, reduce irritation to the eyes and

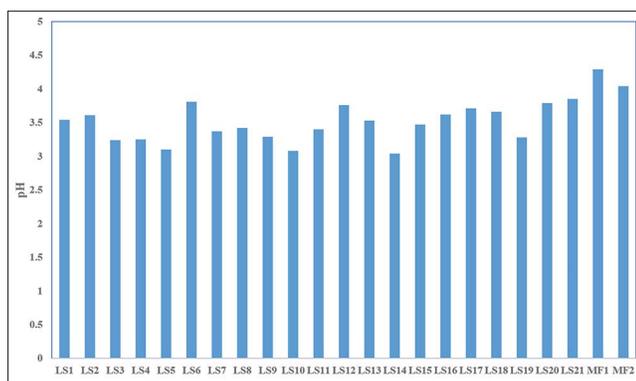


Figure 1. pH profile of liquid poly herbal shampoos.

equilibrates the environmental conditions of scalp. The present development is to endorse poly herbal shampoos with milder pH are the possibilities to decrease the harm to the hair²¹. All formulations are having an acidic pH more correctly mild acidic pH which will prevent swelling and promotes tightening of the scales, which finally results in an increase in glow of hairs. Thus formulations were found to be acceptable with regard to pH²², Amla and Pomegranate shown impact on pH variations.

3.3.2 Dirt Dispersion

Dirt dispersion is considered as key criteria for assessment of cleaning action of liquid polyherbal shampoos. The good qualities of Shampoos selected that rinse away the ink from the foam are considered. It involves the removal of dirt that resides in the froth is complex to wash away and re-adhered on hair. So the capability of shampoo to rinse away the ink is directly related to cleansing action²³. All the tested formulations of poly herbal shampoo contacted with the ink in the aqueous layer, proving their reasonable washing capability. All shampoo formulations showed light and none dispersion of ink in the water portion which shows their actual effectiveness. Amongst the formulations, Amla, Pomegranate and Hibiscus plays major role in the cleansing action.

3.3.3 Detergent Ability

Detergent-Ability of poly herbal shampoos was evaluated by Thompson method. The higher the detergent ability value showed greater the foam. The formulations of poly herbal shampoos portray the detergency between 74% & 98%. Based on the result, it was established that the role of Pomegranate and Reetha are vital in the detergent ability. The bar-chart was presented in Figure 2.

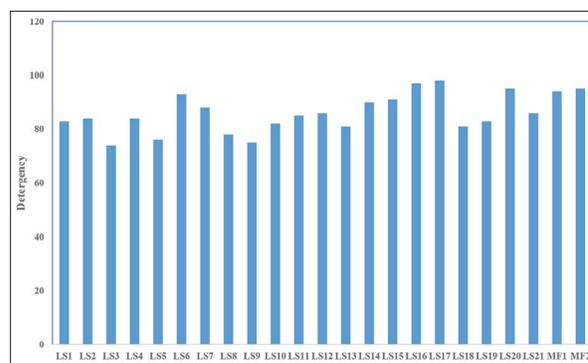


Figure 2. Detergency profile of liquid poly herbal shampoos.

Detergency is an important aspect along with this Soil/sebum removal is the primary aim of a shampoo. The formulations of polyherbal shampoos are containing higher amount of Pomegranate and Reetha shown higher detergency in comparison. The shampoos showed higher or same detergency in comparison with commercial formulations.

3.3.4 Cleaning Action

The polyherbal shampoos containing higher concentration of Amla, Pomegranate and Hibiscus showed higher cleaning action than other formulations comparatively. The poly herbal shampoos containing higher amount of Amla, Pomegranate and Hibiscus showed almost similar cleaning action as that of commercial formulations. The results of cleaning studies indicate that the liquid poly herbal formulations have significant cleaning ability and it was resulted in between 4.12% - 6.63% w/w. Graphical bar-chart representation presented in Figure 3.

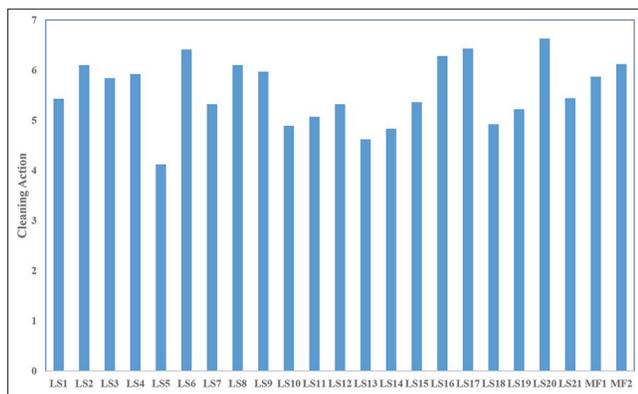


Figure 3. Cleaning action profile of liquid poly herbal shampoos.

As revealed in the results, there is a remarkable variation in the amount of soil removed by the poly herbal formulations²⁴.

3.3.5 Evaluation of Frothing Ability and Froth Stability

Frothing ability is an significant aspect while considering aesthetic value of shampoo. Foaming ability of liquid poly herbal shampoos was evaluated using Cylinder shake technique. Formulated shampoos generated frothing volume above up to 46.1 mL; whereas commercially available shampoos generated frothing volume of

44–45 mL. All ingredients especially Reetha and Pomegranate plays an important role in the foaming ability²⁵. The foam generated by liquid poly herbal shampoos were denser, small, uniform, compact and stable, similar to that of commercial herbal shampoos. Graphical bar-chart representation of foaming ability profile was presented in Figure 4.

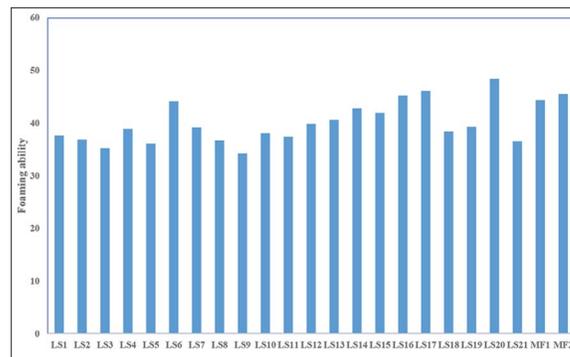


Figure 4. Foaming ability profile of liquid poly herbal shampoos.

3.3.6 Evaluation of Liquid Poly Herbal Shampoo on Conditioning Performance

An Indian woman hair tress was employed to assess the conditioning ability of liquid poly herbal shampoos i.e. for softness and smoothness. This was done by a test known as BLIND-TOUCH, involving 12 participants. The conditioning performance results of poly herbal shampoos were presented in Figure 5, it was concluded that hibiscus play vital role in conditioning the hairs and higher concentration delivers maximum conditioning feel, than marketed formulations. Graphical bar-chart representation of foaming ability profile was presented in Figure 5.

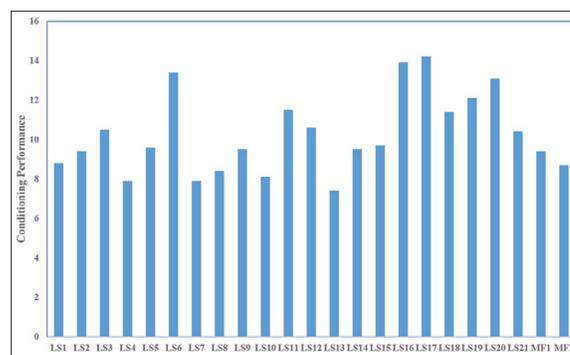


Figure 5. Conditioning performance profile of liquid poly herbal shampoos.

The conditioning outcome is superior to that of commercial formulations. The results noticeably proved that the manufactured shampoos have superior performance in conditioning effect and thus, will have smooth after wash hairs. Hibiscus concentration plays a major role in conditioning performance.

3.3.7 In-Vitro Anti-Dandruff Activity

Cup-Plate method adopted for evaluation of *In-vitro* anti-dandruff activity. Pomegranate Peel extract concentration plays a major role in the anti-dandruff activity. About 6 – 10% Pomegranate extract concentration showed better anti-dandruff activity.

All the constituted liquid Polyherbal shampoos are shown similar viscosity results and good consistency.

3.4 Statistical Evaluation on Physico-Chemical Properties of Liquid Poly Herbal Shampoos

The present study evaluated using DOE (Design Expert) software for statistical design to predict the cleaning, detergency and foaming responses of Liquid Polyherbal shampoo preparations. The resulting contour plots and its polynomial equations aid in predicting the values of selected independent variables for formulation of optimum Liquid polyherbal shampoo formulations with desired properties. The experimental results for Cleaning action, Foaming ability, Detergency ability and pH are presented in Table 8.

3.4.1 Statistical Evaluation to Identify Key-Factors for Detergency

The outcome of ANOVA values are provided in Table 9.

Table 9. ANOVA values for detergency

Source	Sum of Squares	df	Mean Square	F-value	p-value	Comment
Model	620.10	2	310.05	16.93	< 0.0001	significant
B-Reetha	562.50	1	562.50	30.71	< 0.0001	significant
C-Pomegranate	57.60	1	57.60	3.14	0.0931	not significant
Residual	329.71	18	18.32	--	--	--
Lack of Fit	324.51	14	23.18	17.83	0.0065	significant
Pure Error	5.20	4	1.30	--	--	--
Cor Total	949.81	20	--	--	--	--

Based on Table 9 data, the key important factors for detergent action are B (REETHA) and C (POMEGRANATE). The effect of B (%) and C (%) on detergent action was given in Figure 6.

3.4.2 Statistical Evaluation to Identify Key Factors for Cleaning

The outcome of ANOVA is provided in Table 10.

As shown in Table 10, the significant factors affecting cleaning were A (Amla), C (Pomegranate) and D (Hibiscus). The interactions of parameters having impact on ‘cleaning’ are presented in contour plots in Figure 7 & 8.

3.4.3 Statistical Evaluation to Identify Key Factors for Frothing

Outcome of ANOVA is provided in Table 11.

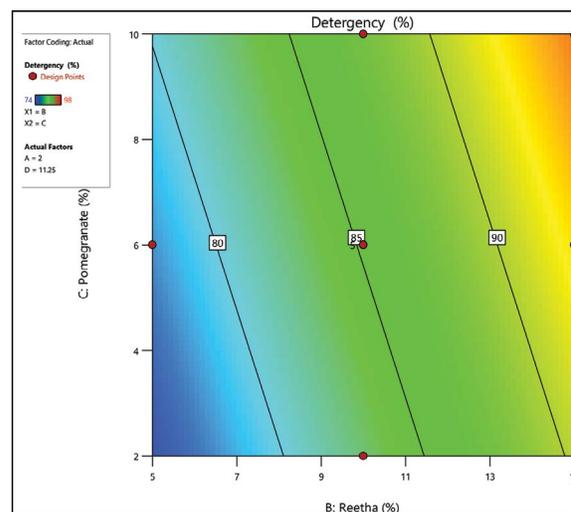


Figure 6. Effect of Reetha (%) and Pomegranate (%) on detergency.

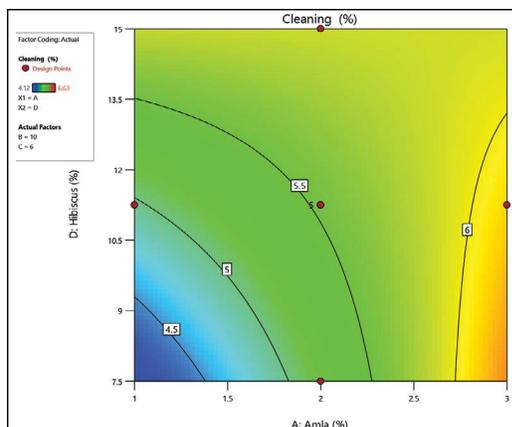


Figure 7. Effect of Amla (%) and Hibiscus (%) on cleaning.

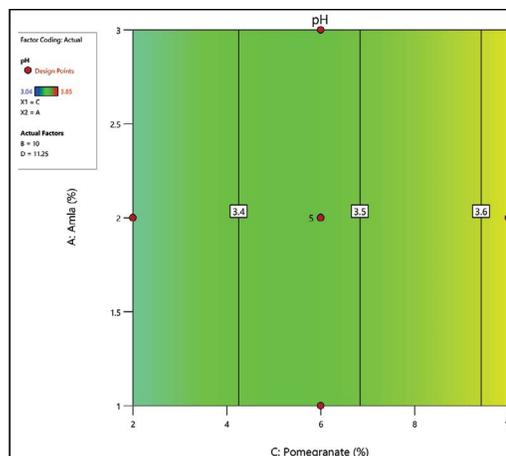


Figure 10. Effect of Pomegranate (%) and Amla (%) on pH.

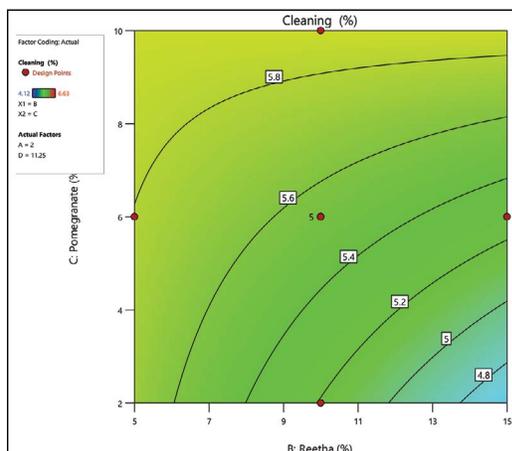


Figure 8. Effect of Reetha (%) and Pomegranate (%) on Cleaning.

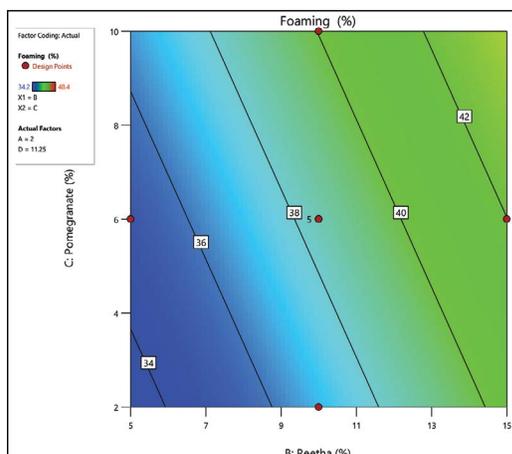


Figure 9. Effect of Reetha (%) and Pomegranate (%) on foaming.

Table 10. Outcome of ANOVA on cleaning

Source	Sum of Squares	df	Mean Square	F- value	p- value	Comment
Model	6.97	6	1.16	8.09	0.0007	significant
A-Amla	3.27	1	3.27	22.78	0.0003	significant
B-Reetha	0.1352	1	0.1352	0.9415	0.3484	not significant
C-Pomegranate	1.19	1	1.19	8.29	0.0121	significant
D-Hibiscus	1.17	1	1.17	8.14	0.0128	significant
AD	0.4774	1	0.4774	3.32	0.0897	Not significant
BC	0.5460	1	0.5460	3.80	0.0715	Not significant
Residual	2.01	14	0.1436	--	--	--
Lack of Fit	0.9138	10	0.0914	0.3333	0.9277	not significant
Pure Error	1.10	4	0.2742	--	--	--
Cor Total	8.98	20	--	--	--	--

3.4.4 Statistical Evaluation to Identify Key Factors for pH

The ANOVA (Analysis of Variance) results are presented in Table 12.

Table 11. Outcome of ANOVA for frothing

Source	Sum of Squares	df	Mean Square	F-value	p-value	Comment
Model	186.35	4	46.59	7.40	0.0014	significant
A-Amla	1.76	1	1.76	0.2802	0.6038	not significant
B-Reetha	124.61	1	124.61	19.80	0.0004	significant
C-Pomegranate	24.96	1	24.96	3.97	0.0638	significant
A ²	35.01	1	35.01	5.56	0.0314	significant
Residual	100.71	16	6.29	--	--	--
Lack of Fit	98.41	12	8.20	14.21	0.0102	significant
Pure-Error	2.31	4	0.5770	--	--	--
Corr-Total	287.07	20	--	--	--	--

The key frothing related factors affecting are B (Reetha), & C (Pomegranate). The interactions of parameters having impact on foaming are presented in contour plots in Figure 9.

Table 12. Outcome of ANOVA for pH

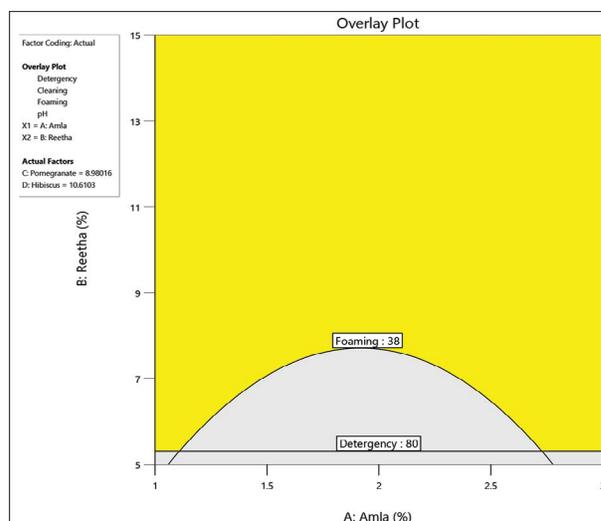
Source	Sum of Squares	df	Mean Square	F-value	p-value	Comment
Model	0.2403	1	0.2403	4.51	0.0470	significant
C-Pomegranate	0.2402	1	0.2402	4.51	0.0470	significant
Residual	1.01	19	0.0532	--	--	--
Lack of Fit	0.8258	15	0.0551	1.19	0.4808	not significant
Pure Error	0.1857	4	0.0464	--	--	--
Cor Total	1.25	20	--	--	--	--

Key factors affecting pH is C (Pomegranate). The interactions of pH contour plot presented in Figure 10.

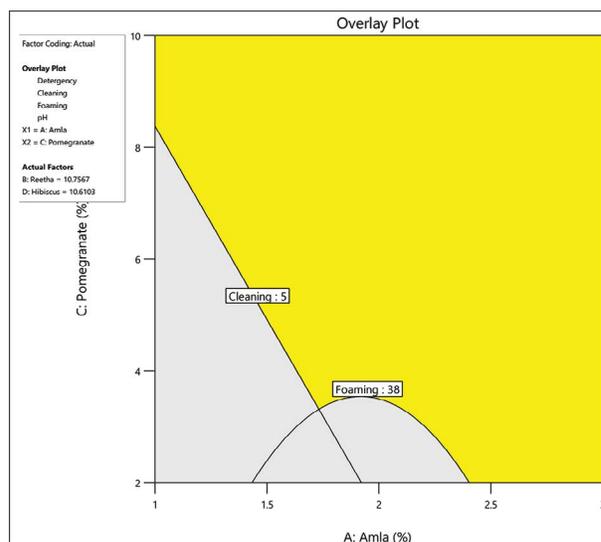
From the evaluation of statistical data, greater concentration of Reetha and Pomegranate have a substantial impact on detergency.

Amla, Pomegranate, and Hibiscus are shown significant impact on cleansing action. The ingredients Reetha and Pomegranate have a significant impact on foaming ability. Amla and Pomegranate have a significant impact on pH.

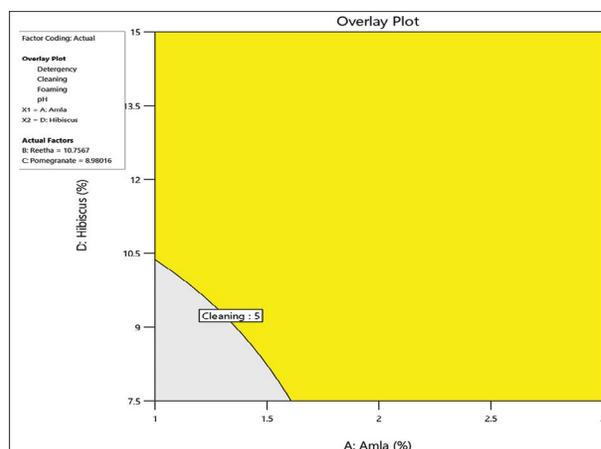
The statistical evaluations are useful to select optimal and acceptable ranges of herbal concentrations of



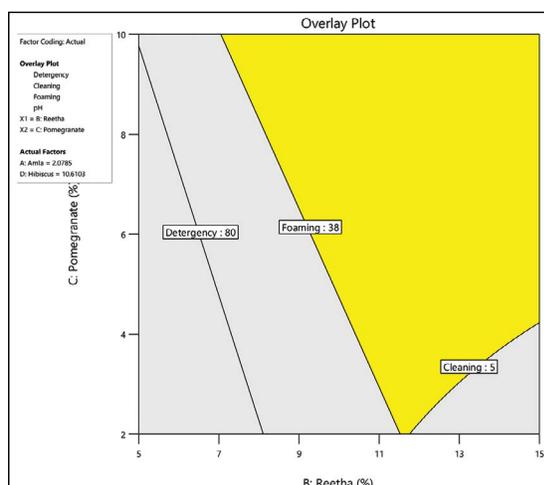
(a)



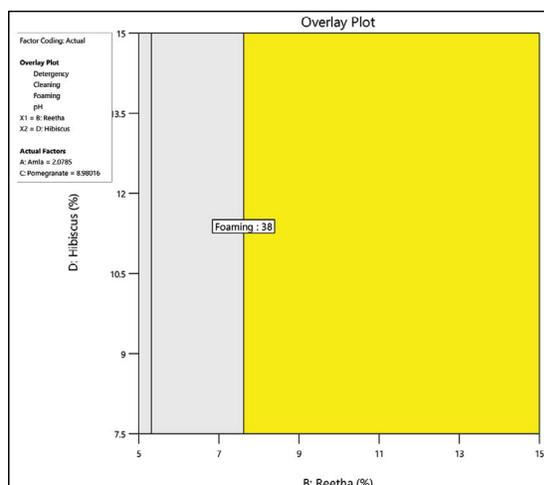
(b)



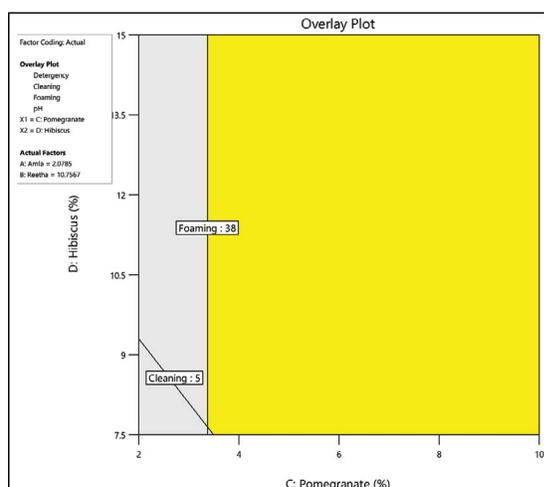
(c)



(d)



(e)



(f)

Figure 11. Overlay plot – effect of formulation variables on responses.

formulation variables. Figure 11 illustrates the overlay plot of responses. The Yellow is h coloured region specifies the acceptance criteria of an ideal liquid polyherbal shampoo formulation with the optimal % range of each of the ingredient.

4. Conclusion

Central composite statistical evaluation found suitable for identification of optimized Liquid polyherbal formulations considering the individual and combination interactions between the herbal extracts; on the performance of shampoos. Polyherbal shampoo formulations containing 1% - 3% w/w of Amla, 6% - 10% w/w of Pomegranate, 10% - 15% w/w of Reetha and 11.25% - 15% w/w of Hibiscus depicts better performance in comparison to commercial marketed formulations. The formulations LS6, LS16, LS17 & LS20 were identified as the better formulations based on their physicochemical properties and further confirmation by statistical evaluation. The optimized Liquid polyherbal shampoos have an excellent cleansing, detergency, conditioning, foaming effects, anti-dandruff activity and are best for regular hair and has optimal pH and stable organoleptic properties, compared with commercial marketed formulations. The physicochemical properties were found stable in optimized formulations up to six months of stability, indicating the formulations were suitably stable.

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