Formulation and Characterization of Herbal cream containing Fenugreek seed extracts.

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Abstract- The aim of present study was formulation of a stable W/O emulsion containing fenugreek seeds extract using olive oil as a base. Extract of Fenugreek seeds was obtained by using methanol as a solvent and the extract of fenugreek seeds, was entrapped in the inner aqueous phase of W/O emulsion. The formulation was prepared in internal aqueous phase (W/O emulsion) containing the extract of fenugreek seeds. The herbal formulation showed good spreadibilty, good consistency, homogeneity, pH, non greasy and no evidence of phase separation. The formulation, was stable at all the accelerated conditions regarding color, liquifaction and phase separation. The extract-containing cream substantially increased skin elasticity, hydration and decreased the skin melanin.

Index Terms- Fenugreek seed extract, cream, emulsion, formulation.

I. Introduction

Emulsions are a class of disperse systems consisting of two immiscible liquids. The liquid droplets (the disperse phase) are dispersed in a liquid medium (the continuous phase). To disperse two immiscible liquids, one needs a third component, namely, the emulsifier which is a surface active agent. Emulsifiers reduce interfacial tension between two immiscible liquids. The choice of the emulsifier is crucial in the formation of the emulsion and its long-term stability. In cosmetics, emulsions are the delivery vehicle for many hair and skin conditioning agents. Oil in Water (O/W) emulsions are the most commonly used cosmetic delivery systems which supply moisture to skin and improve the skin condition by forming the occlusive barrier on the skin. Anionic and non-ionic emulsions are used to deliver various oils and waxes which provide miniaturization, smoothness and softness to hair and skin. Emulsions formed with cationic emulsifiers are themselves effective conditioning agents since their positive charge is attracted to the negative sites on the hair, thus allowing them to resist rinse off.

In recent years, scientists are looking towards the cosmetic products utilizing natural resources. Fenugreek (Trigonella foenum graecum) is an annual herb that belongs to the family Leguminosae widely grown in Pakistan, India, Egypt, and Middle Eastern countries. Due to its strong flavor and aroma, the leaves and seeds of fenugreek are widely consumed in Indo-Pak subcontinent as well as in other oriental countries as a spice in food preparations, and as an ingredient in traditional medicine. It is rich source of calcium, iron, β-carotene and other vitamins. Fenugreek seeds extract is usually used to treat severe skin inflammation, chapped lips and skin aging. The seeds are used as a traditional remedy for the treatment of diabetes and hypercholesterolemia in Indian and Chinese medicines.

Olive oil is obtained from the olive (the fruit of *Olea europaea*; family Oleaceae), a traditional tree crop of the Mediterranean Basin. The oil is commonly used in cooking, cosmetics, pharmaceuticals, and soaps, and as a fuel for traditional oil lamps. Olive oil has a long history of being used as a home remedy for skincare. Egyptians used it alongside beeswax as a cleanser, moisturizer, and antibacterial agent. It can be used to relief of seborrheic dermatitis, acne, psoriasis or atopic dermatitis. The aim of our study is to prepare the emulsion of olive oil and aqueous fenugreek extract and compare the efficacy of this formulation with base i.e. olive oil emulsion in water and marketed product.

II. MATERIALS AND METHODS

Material: Fenugreek seeds and Olive oil were purchased from local market of Matunga, Mumbai. Span 80, Tween 80, Guar gum (additive) were ordered from TCI Chemicals(India)Pvt. Ltd.

Methods:

Preparation of plant extracts:

Dried and fresh batches of fenugreek seeds were purchased from local market, Matunga, Mumbai. Seeds were washed with distilled water to get rid of extraneous matter. Fenugreek seeds (10 g) were dried in oven at 50°C. After drying, they were ground into a fine powder in a mixer. Methanol (250 ml) was used for extraction by Soxhlet extraction method for six hours. The extracts were filtered. The residue was re-extracted twice under the same condition to ensure complete extraction. The extracts were combined, filtered and evaporated to dryness under reduced pressure at 65°C by a rotary evaporator. Extract was stored at dry place. 10 ml of the extract was measured into a pre-weighed aluminum dish and kept in an oven at 85°C for 24 hours then followed by placing in desiccator for 14 hours. The weight difference was calculated and percentage yield as well as weight expressed in mg/10 ml was determined.

Emulsion preparation

Oil phase that consisted of olive oil and calculated quantity of Span 80 (HLB No. 4.5) and Tween 80 (HLB No. 15) was prepared. At the same time, aqueous phase was prepared by adding the fenugreek seeds extract and natural polymer in water. After that, oil phase was added to the aqueous phase drop by

drop at 2500 rpm stirring to get the O/W emulsion based emulsion and further stirred for 30 minutes.

Preparation of base: The procedure was continued exactly as described above for preparation of cream without using the fenugreek seeds extract. This is the emulsion of olive oil and water.

Standardization of cream parameters:

The effect of various parameters like surfactant content, oil content, fenugreek seeds extract, HLB number of surfactant blend and additive content on emulsion stability was studied by trial and error method. A series of surfactant blends of different HLB numbers were prepared by the proper mixing of individual emulsifiers. Initially the surfactant concentration was kept at 0.5% (w/w) level and oil content was varied from 5 to 20% (w/w) level. As these emulsions were found physically unstable, procedure was repeated by increasing emulsifier concentration from 0.5 to 5% (w/w).

Characterization of emulsions and stability tests:

Rheological properties of an emulsion are important. These properties decide the physical stability, skin feel, ease of application on skin and its overall performance. The factors like oil content, amount of additive are the controlling factors of viscosity of an emulsion. The viscosity of emulsion and base was measured at 20±0.1ºC over a shear rate-range of 0 to 600 s^{-1}, using a rotational viscometer RV-2.

Stability tests were performed under different conditions for the emulsion and base. These tests were performed on samples kept at 8 ± 0.1^{O}C (in refrigerator), 25± 0.1^{O}C (in incubator), 40 ± 0.1^{O}C (in incubator) and 40 ± 0.1^{O}C (in incubator with 75% relative humidity).

Dermatological tests and product evaluation on skin:

Evaluation Test:

pH: 10% w/v suspension of the cream was prepared with water and the pH was measured using pH meter.

Homogeneity: The formulations were tested for the homogeneity by visual appearance and by touch.

Appearance: The appearance of the cream was judged by its color, pearlscence and roughness and graded.

Dilution method: Fixed amount of cream was diluted with water and mineral oil separately

Filter paper method: A streak of the cream was applied on the filter paper

Ash: Product was taken in a flat –bottom platinum dish and ashing was done at 600 ˚C in muffle furnace

feel: Emolliency, slipperiness and amount of residue left after the application of fixed amount of cream was checked.

Type of smear: After application of cream, the type of film or smear formed on the skin were checked.

Sixteen human volunteers having ages between 25 and 35 years were selected. Prior to the tests, a cosmetic expert examined the volunteers for any serious skin disease or damage especially on cheeks and forearms. The volunteers were instructed to apply the emulsion for 12 weeks and to come after every week for the dermatological tests.

Skin Irritation Tests were carried out by applying 0.5 g of emulsion and the base daily for seven days on 4cm^2 area of forearms and cheek. The skin was observed for any visual change such as redness *(erythema)* and swelling *(oedema)* after every 24 hours for seven days. Evaluations were done by using the "Primary Skin Irritation Index" which is the composite score obtained by summing individual scores of erythema and oedema which can range from 0 to 8 representing the degree of skin irritation.

The study of moisturizing property of formulated emulsions was carried out using a CM-825PC Corneometer against an aqueous marketed cream. The base line values were taken on 2 cm × 2 cm area on forearms. Test area was then treated with 0.1 g of formulated emulsion and control. Humidity conditions were set at 25 ±0.1°C and 48.5±0.1°C.

III. RESULTS AND DISCUSSION

The pH of human skin typically ranges from4.5 to 6.0 and 5.5 is considered to be average

pH of the skin. Therefore, the formulations intended for application to skin should have pH close to this range The pH values of the formulation remained constant with slight decrease in values.

Days	Temperature	Parameters					
		pH	Homogenity	Spreadability	After feel	Colour	Type of smear
0	RT	5.92	**	**	E	NC	NG
	40^{0}C	5.91	**	**	E	NC	NG
5	RT	5.91	**	**	E	NC	NG
	40^{0}C	5.90	**	**	E	NC	NG
10	RT	5.89	**	**	E	NC	NG
	40^{0}C	5.86	**	**	E	NC	NG
15	RT	5.87	**	**	E	NC	NG
	40^{0}C	5.83	**	**	E	NC	NG
20	RT	5.82	**	**	E	NC	NG
	40^{0}C	5.80	**	**	E	NC	NG
25	RT	5.76	**	**	E	NC	NG
	40^{0}C	5.77	**	**	E	NC	NG
30	RT	5.69	**	**	E	NC	NG

	40^0C	5.66	**	**	E	NC	NG
45	RT	5.61	**	**	E	NC	NG
	40^0C	5.62	**	**	E	NC	NG
60	RT	5.55	**	*	E	NC	NG
	40^0C	5.52	**	**	E	NC	NG

Table no. 01:Stability Testing
**: Good, *: Satisfactory, E: Emollient, NC: No Change, NG: Non greasy.

These formulations had almost constant pH, homogeneous, pearlescent, emollient, non-greasy after the application. There was no significant colour change observed in the formulation at the end of 3 months indicating the good stability of the emulsion. The stable formulations were safe in respect to skin irritation and allergic sensitization. The prepared herbal face cream is intended for cosmeceutical use rather than as mere cosmetic. It contains fenugreek seed extract which act as an anti-oxidant agent.

The yields of the extracts obtained by the soxhelt method were calculated as percent by weight of the fenugreek seed. According to the chemical composition and polar nature of phenolic compounds, fenugreek contains a relatively high percentage yield in polar solvents like water, methanol etc. while lower in nonpolar solvents. The yield was found to be 55.6 in mg/10 ml and in 23.5% yield.

The optimized parameters of the cosmetic creams showing six months storage stability at 28±2°C are shown in below:

	Oil (w/w)	Surfactant blend (w/w)	Additive (w/w)	Aqueous phase (w/w)	Surfactant blend ratio Span 80 : tween 80	HLB no. of Surfactant blend
Formulation	24%	4%	0.8%	71.2%	71.43:28.57	7.5
Base	24%	4%	0.8%	71.2%	76.19:23.81	7

Table no.02 : optimized parameters of the cosmetic cream and base

The surfactant molecules from oil phase and water phase get associated in a particular way at the O/W interface. Sufficient emulsifier concentration is necessary to produce the tightly packed monolayer of surfactant molecules at the interface which acts as a mechanical barrier to flocculation and coalescence of oil droplets thereby stabilizing the emulsion. All stable creams obtained were having close matching of HLB no. of surfactant blend and oil phase. Thus, preparation of surfactant blend with identical HLB no. that with oil phase is found to be one of the most important criteria for obtaining stable creams. The use of natural polymers as additives has enhanced texture, viscosity of external phase, rheological behavior and storage stability of creams against various phenomena such as coalescence, flocculation, layer separation, etc.

The formulated cream was found to be stable for 60 days without separation of layers at different temperatures. No phase separation was observed for the emulsion indicating the formation of tightly packed monolayer of surfactant molecules at the interface which acts as a mechanical barrier to flocculation and coalescence of oil droplets thereby stabilizing the emulsion. No phase separation even on centrifugation was seen in any of the samples kept under different storage conditions, i.e. 8^OC, 25^OC, 40^OC and 40^OC (75% RH) up to 60th day of observation. This indicated that the emulsions were stable at all the storage conditions for 60 days.

Spreadability denotes the extent of area to which the gel readily spreads on application to skin or the affected part. The bioavailability efficiency of a cream also depends on its spreading value.

Formulation	**Ingredients**	**Concentrations**	**Spreadability**
1	Aqueous phase	71.2	10.3
	Oil phase	24	
	Surfactant	4	
	Span 80:tween 80	76.19:23.81	
	additive	0.8	

2	Aqueous phase	72.2	9.35
	Oil phase	23	
	Surfactant	4	
	Span 80:tween 80	72.44:27.56	
	additive	0.8	
3	Aqueous phase	73.2	8.29
	Oil phase	22	
	Surfactant	4	
	Span 80:tween 80	71:29	
	additive	0.8	

Table No. 03 Spredability in different formulations

The application of base resulted in slight increase in water content of skin at the end of three hours while the application of formulation showed significant increase in water content of skin at the end of three hours. The significant increase in moisture after application of formulation may be due to the fact that fenugreek extract contains mucilaginous polysaccharide that is a natural effective ingredient for improving skin hydration, possibly through a humectant mechanism. The results shows that the emulsion of olive oil and fenugreek extract can be used as effective moisturizer. On the other hand, marketed products give short term relief to dry skin.

IV. CONCLUSION

The emulsion of aqueous extract of fenugreek seeds and olive oil can be prepared. The rheological parameters and storage stability of each formulated cream were found to be identical with marketed product. The formulation was found to be superior to the marketed product for long term effect and spreadability. Due to no adverse effect of the formulation on skin and good moisturizing properties, the formulation can be used as a skin care product and an effective moisturizer.

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