

**FORMULATION AND EVALUATION OF HYDRATING GEL USING
SALVIA HISPANICA SEED OIL****¹*Vedanti V. Shete and ²Bharati Ganu**¹Student, ²Assistant Professor

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ABSTRACT

Salvia Hispanica L. is annual herbaceous plant, native of Southern Mexico and Northern Guatemala belonging to family Lamiaceae. It is widely cultivated for its oils that are known for health promoting characteristics due to high content of omega-3 α -linolenic acid (ω -3 ALA) that protects from inflammation and other polyunsaturated fatty acids (PUFA's). Chia seed oil is obtained from seed through different extraction methods which result in varying quality parameters of oil. In the further study, gel base was formulated and active i.e. Salvia Hispanica oil was incorporated. After incorporation of active in base, observations of pH, assessment of skin moisture, washability, spreadability were performed to see the stability and effectiveness of

gel. Physical observation of gel's organoleptic properties like pH, viscosity, colour and odour were found to be stable for 30 days of storage.

KEYWORDS: Salvia Hispanica seed, Salvia Hispanica seed oil, Chia seed oil, hydration, α -linolenic acid.

INTRODUCTION

Dry skin is a common condition that is attributed to lack of water in the stratum corneum. Dry skin is caused by many different things like environmental causes and certain health conditions.^[1] Dry skin treatment and prevention's pathway is proposed as a platform for optimal skin care. This approach includes therapeutic treatment concepts, that does not address specific conditions like eczema and psoriasis, and is limited to prevention and

treatment of dry skin. Dry skin which is also called as xerosis is major problem due to the environmental changes. Hydration to the skin is much needed in such case.^[2]

Of all beneficial properties claimed for cosmetic creams/gels, hydrating is possibly the most widely used. If water lost more rapidly from the stratum corneum then it is received from the lower layer of epidermis, the skin becomes dehydrated and loses its flexibility.^[3]

In humans one of the major skin problems is dry skin. It is decreased in the water content of stratum corneum. There are two basic types of dry skin, the first one is due to prolonged exposure to low humidity and air movement and second is due to physical and chemical changes to the skin due to process such as aging, continual decreasing etc.

The function of the epidermis is to produce and maintain an efficient barrier called stratum corneum (horny layer). Skin is continuously losing moisture to the atmosphere by diffusion of water vapour through the stratum corneum and sweat gland (below the thermal threshold of sweating). The keratinized cells, NMF and intracellular lipid keep this transepidermal water loss (TEWL) to an acceptable minimum.^[4]

It has been seen that water keeps the horny layer supple and ensures neutralization of acids and alkalis. Water content 10-20% in stratum corneum is said to be required to keep it soft and pliable. Unfortunately, the water content of keratinocytes in the basal layer (70%) decreases slowly in the stratum granulosum (65%), and then more rapidly (to about 35%) in lower layer of stratum corneum. As the stratum corneum matures, its water level drops more slowly until it reaches desquamating layers (about 15-20%).

Alteration of the barrier can cause an increase in TEWL to the extent that water is lost and it can be replaced by the underlying tissues which can then result in the horny layer drying out, precipitating dry skin conditions.

Dry skin feels rough and itchy. Closer observation reveals the presence of fine wrinkles and at more advanced stage, larger scales. The surface of severe dry skin is cracked and is reddened and as a result of the dilated peripheral blood capillaries in the dermis. Environmental factors, sunlight and aging plus numerous skin diseases and dietary deficiencies, all produce dry skin. The environmental factors include low relative humidity caused by central heating, wind, cold weather and repeated contact with water, surfactant and solvents. Thus many conditions can cause symptoms of dry skin.^[5]

Gels are type of base that produce uniform external appearance and give moist feeling. With development of new material and technology aqueous and oil gels having more function than providing moisture have been developed. Aqueous gel contains more moisture so are used for supplying moisture and cooling effect. Oily gels supply oil to skin. As they give moist feeling these gels are liked more and are becoming popular.

Skin care products are promoted vigorously in the cosmetic industry with claims of tremendous benefit for good and healthy looking skin. There exists in markets an elementary understanding of what a proper skin care regime should consist of.^[6]

Basically market product are based on different skin type i.e.; oily, dry, combination or sensitive and then divide treatment into basic steps of exfoliation, treatment, hydration and protection.

Salvia Hispanica L. is annual herbaceous plant, native of Southern Mexico & Northern Guatemala belonging to family Lamiaceae. Seeds are composed of total dietary fiber from 47.1- 59.8% and contain upto 40% of oil with high content of unsaturated fatty acid of which linolenic acid represents upto 68%. Also they are good source of protein, dietary fibre, vitamin, minerals and antioxidants. This oil can be used for all skin types those who have sensitive, inflamed and dehydrated skin would get benefit from chia seed oil and it is also used for irritated and oil-prone skin. There are different skin benefits of chia seed oil for skin like, it moisturizes skin, improves dryness and itchiness, increases skin hydration, have antioxidant property.^[7]

MATERIALS AND METHODS

Salvia Hispanica seed oil, Carbopol 940, Glycerin, Triethanolamine, Methyl paraben, Distilled water.

Collection of Salvia Hispanica oil

Salvia Hispanica oil was procured from Moksha Lifestyle Products, Delhi.

Extraction of Salvia Hispanica seed oil

Chia seed oil is obtained from seeds of Salvia Hispanica through cold pressing method.

Seeds were pressed at 25-30°C using a Komet screw press with a 5-mm restriction dye and screw speed of 20 rpm. The screw press was first run for 15 min without seed material, but

with heating by means of an electrical resistance-heating ring attached around the press barrel to raise screw press barrel temperature to desired temperature ($25 \pm 2^\circ\text{C}$). Running temperature was checked with digital thermometer inserted in restriction dye. The meal obtained by pressing (Mp) was ground with a laboratory mill at 489 μm . Meals were homogenized and stored in plastic vessels at 5°C from both extraction methods (Ms and Mp) until use.^[8]

Determination of acid value

Different testings for *Salvia Hispanica* oil were done which includes acid value, it is a number which expresses in milligrams the amount of potassium hydroxide necessary to neutralize the free fatty acid present in 1gm of substance. Here 10 gms of oil was dissolved in 50ml of mixture of equal volume of ethanol (95%) and ether previously neutralized with 0.1 M potassium hydroxide to phenolphthalein solution. 1ml of phenolphthalein was added and titrated with 0.1 M potassium hydroxide until the solution remains faintly pink after shaking for 30 seconds and then acid value was calculated.

Determination of saponification value

Saponification value is number of milligrams of potassium hydroxide necessary to neutralize the free fatty acids and to saponify the esters present in 1gm of the substance. Here 2gm of oil was weighed into 200ml flask of borosilicate glass fitted with reflux condenser. 25ml of 0.5M ethanolic potassium hydroxide as added and little pumice powder was also added and boiled under reflux on water bath for 30 minutes. 1ml of phenolphthalein solution was added and titrated immediately with 0.5 M hydrochloric acid (a ml) and the operation was repeated omitting the oil (b ml)

Also specific gravity was checked using hydrometer, refractive index was checked using refractometer and solubility of oil was also checked which is shown in table 1.

Table 1: Physiochemical testing of Saliva Hispanica oil.

Sr.no.	Name of test	Standard range	Obtained value
1	Acid value	0.70- 2.05 mg KOH/g	0.849
2	Sap value	193- 222mg KOH/g	195.0025
3	Specific gravity at 20°C	0.890 to 0.988	0.895 to 0.945
4	Refractive index at 20°C	1.4811 to 1.4832	1.460 to 1.505
5	Solubility 1. In water 2. In alcohol	Insoluble Soluble	Insoluble Soluble

Gas Chromatography testing

Gas chromatography of given oil sample was analysed according to the procedure given in AOAC 969.33 and the results were found as followed in (table no. 2).

As per the graphical analysis (fig. 1) provided by Qualichem laboratories it was found that polyunsaturated fatty acid was present in very high percentage (72.43%). Omega 3 was found to be (54.95%) followed by omega-6 (17.48%). Monosaturated fat was also present (6.99%) with (19.38%) saturated fat which is shown in (table no.2).^[9]

Table 2: Gas Chromatography results.

Name of Sample & Description : CHIA SEED OIL			
Manufactured by : -			
Supplied by : -			
Customer Reference : DT.09/04/2022			
Date of Manufacture : -	Batch No : -		
Date of Expiry : -	Batch Size : -	Quantity submitted : APP.50 G	
Date of receipt : 9-Apr-22	Start Date of Analysis : 12-Apr-22	Completion Date of Analysis : 12-Apr-22	

Not Sampled By Qualichem		Report no : M/85/22-23	
No.	Test		Result
1	DESCRIPTION Pale yellow coloured liquid filled in a plastic container.		-
2	TRANS FAT Method : AOAC 969.33 (Detection limit : 0.01-1g/100g)	g/100g	0.1
3	SATURATED FAT Method : AOAC 969.33	g/100g	19.38
4	MONOUNSATURATED FAT Method : AOAC 969.33	g/100g	6.99
5	POLYUNSATURATED FAT Method : AOAC 969.33	g/100g	72.43
6	OMEGA 3 Method : AOAC 969.33 (Detection Limit : 0.01 g/100g)	g/100g	54.95
7	OMEGA 6 Method : AOAC 969.33	g/100g	17.48

Environmental Condition : Maintained as per the requirements of the samples and the test methods.			

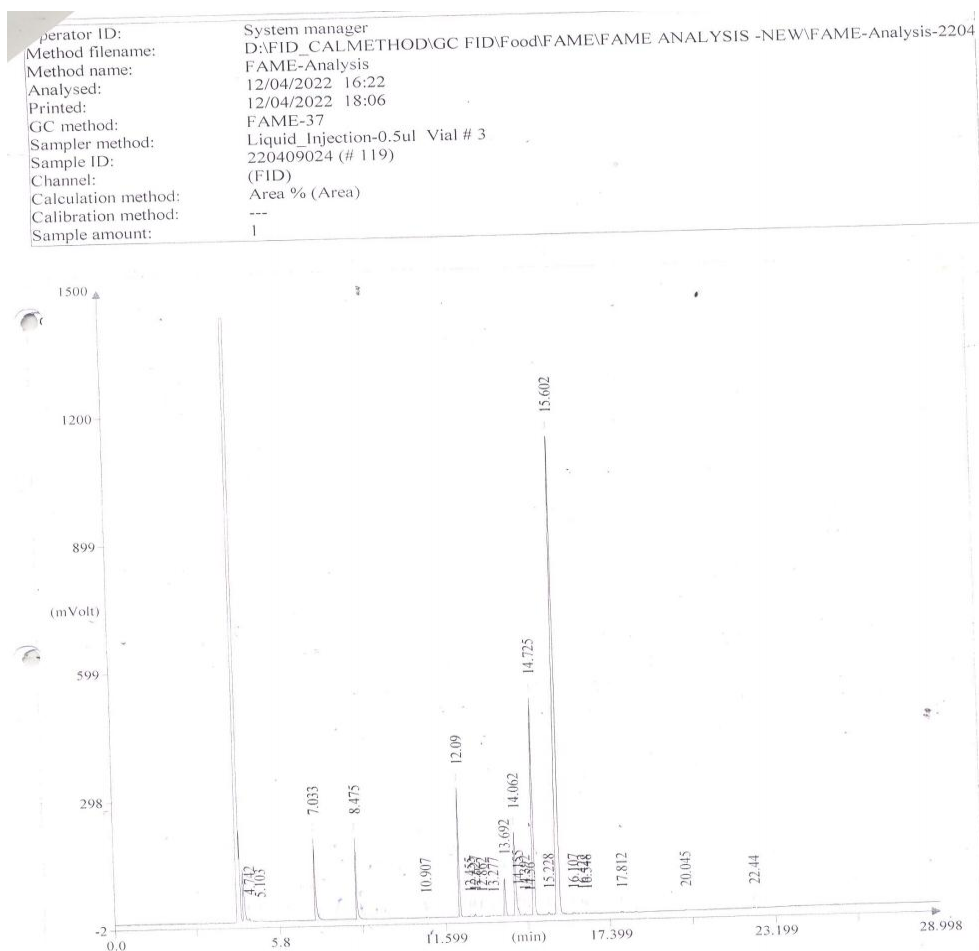


Fig. 1: Gas Chromatography graph.

Fatty Acid Profile testing

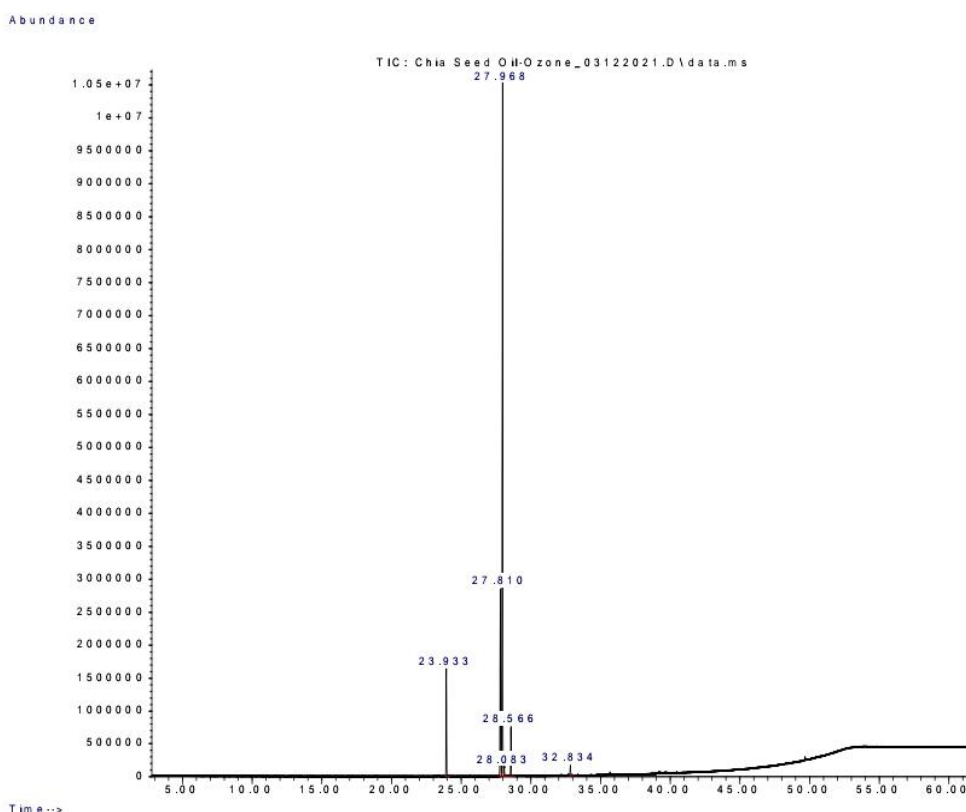
Fatty Acid Profile of given oil sample was analysed according to the procedure and the results was found as followed in (table no. 2).

As per the graphical analysis provided by moksha (fig 11) it was found that palmitic acid, linoleic acid, linolenic acid, oleic acid, stearic acid and arachidic acid was found.(Table no.3).^[10]

Table 3: Fatty Acid Profile results.

Product Name: Chia Seed Oil

PK	RT	Area %	Library/ID	CAS
1	23.9327	08.5667	Palmitic Acid	000112-39-0
2	27.8097	16.5794	Linoleic Acid	000112-63-0
3	27.9684	68.7785	Linolenic Acid	000301-00-8
4	28.0828	00.9369	Oleic Acid	000112-62-9
5	28.5656	04.0643	Stearic Acid	000112-61-8
6	32.8336	01.0742	Arachidic Acid	000506-44-5

Product Name: Chia Seed Oil**Fig. 2: Fatty Acid Profile graph.****Formulation and development of hydrating gel**

For the formulation of hydrating gel 3 base formulations were prepared and one was selected from them which gives satisfactory transparency and consistency. The formulation of selected base was shown in Table. In the formulation of hydrating gel, Carbopol 940 was allowed to swell by adding half amount of water and then slowly glycerin was added with methyl paraben with remaining amount of water. Lastly Triethanolamine was added as thickening agent in the base. After the observation of stiffer form or viscosity of three bases, formulation (T3) was selected as base for hydrating gel which is shown in table 4.

Table 4: Formulation of gel base.

Sr. no.	Ingredients	Quantity for 100 gm			Uses
		Trial 1	Trial 2	Trial 3	
1	Carbopol 940	1gm	1.5gm	2gm	As a gelling agent
2	Glycerin	10ml	8ml	6ml	As a humectant
3	Triethanolamine	0.9ml	0.9ml	0.6ml	As emulsifier, thickening agent
4	Methyl paraben	0.5gm	0.5gm	0.5gm	Preservative
5	Water	Upto 100 ml	Upto 100 ml	Upto 100 ml	As a continuous phase and an external phase

Incorporation of active in hydrating gel

Five different concentrations of active were added in the selected gel base. 2%, 3%, 4%, 5%, and 6% concentration of Salvia Hispanica oil were used. This was shown in Table 5.

Table 5: Formulation of gel with active.

Sr. no.	Ingredients	Quantity for 100% F ₁	Quantity for 100% F ₂	Quantity for 100% F ₃	Quantity for 100% F ₄	Quantity for 100% F ₅
1	Carbopol 940	2%	2%	2%	2%	2%
2	Glycerin	6%	6%	6%	6%	6%
3	Triethanolamine	0.6 %	0.6 %	0.6 %	0.6 %	0.6 %
4	Methyl paraben	0.5%	0.5%	0.5%	0.5%	0.5%
5	Water	85.4%	85.4%	85.4%	85.4%	85.4%
6	Salvia Hispanica oil	2%	3%	4%	5%	6%

Formulation of gel with hyaluronic acid

On another side a gel was formulated with same base (T3) adding hyaluronic acid of about 2% to compare the moisture content with Salvia Hispanica hydrating gel using Skin moisture meter.

Table 6: Formulation of gel with hyaluronic acid.

Sr. no.	Ingredients	Quantity for 100% B ₁
1	Carbopol 940	2%
2	Glycerin	6%
3	Triethanolamine	0.6 %
4	Methyl paraben	0.5%
5	Water	85.4%
6	Hyaluronic acid	1-2%

Evaluation of the hydrating gel

1) Observation of the organoleptic properties

Here, the properties like colour, odour and viscosity were visually observed.

2) Determination of Skin moisture content

To measure the moisture content of skin ten subjects of each dry skin, normal skin and oily skin were taken. The test was performed on inner side of arm where four squares of 2cm each were made giving 1cm gap in between using a scale and a marker. Different formulations were applied into each square, the amount taken was 0.2 gm approx. which should fill each square. Then each formulation F₃, F₄, and F₅ were compared with a gel base with 2% of hyaluronic acid. This was performed using digital skin moisture meter. After every 15

minutes of applying the gel the readings were taken till 2 hours. Then the moisture content of F_3, F_4, F_5 was found to be similar as hydration given by all the concentration of active (*Salvia Hispanica* oil) was similar on dry skin, normal skin and oily skin, so the minimum concentration i.e. 4% of *Salvia hispanica* oil in F_3 formulation was selected for studies of accelerated stability test, spreadability and washability. The readings are further given in Table 7,8 and 9 respectively.



Fig. 3: Reading of skin after applying hydrating gel for different formulations by skin moisture meter.

Table 7: Observation of moisture meter on dry skin for 2 hrs after every 15 min.

- Dry skin reading- 11.4

Formulation/time ↓ →	15min	30min	45min	60min	75min	90min	105 min	120min
B_1	53.2	53.6	54.4	56.2	56.9	57	58.5	68.9
F_3	48.2	48.6	49	49.8	50.6	51.2	52.2	62.9
F_4	52.4	52.9	53.6	55	55.9	56.7	57.4	58
F_5	52.1	52.4	53.1	53.5	54.2	54.9	55.4	56

Table 8- Observation of moisture meter on normal skin for 2 hrs after every 15 min.

- Normal skin reading- 24.3

Formulation/time ↓ →	15min	30min	45min	60min	75min	90min	105 min	120min
B_1	63.2	63.6	64.4	66.2	66.9	67	68.5	68.9
F_3	58.2	58.6	59	59.8	60.6	61.2	62.2	62.9
F_4	62.4	62.9	63.6	65	65.9	66.7	67.4	68
F_5	62.1	62.4	63.1	63.5	64.2	64.9	65.4	66

Table 9: Observation of moisture meter on oily skin for 2 hrs after every 15 min.

- **Oily skin reading- 32.1**

Formulation/time ↓ →	15min	30min	45min	60min	75min	90min	105 min	120min
B ₁	73.2	73.6	74.4	76.2	76.9	77	78.5	78.9
F ₃	68.2	68.6	69	69.8	70.6	71.2	72.2	72.9
F ₄	72.4	72.9	73.6	75	75.9	76.7	77.4	78
F ₅	72.1	72.4	73.1	73.5	74.2	74.9	75.4	76

3) Determination of pH

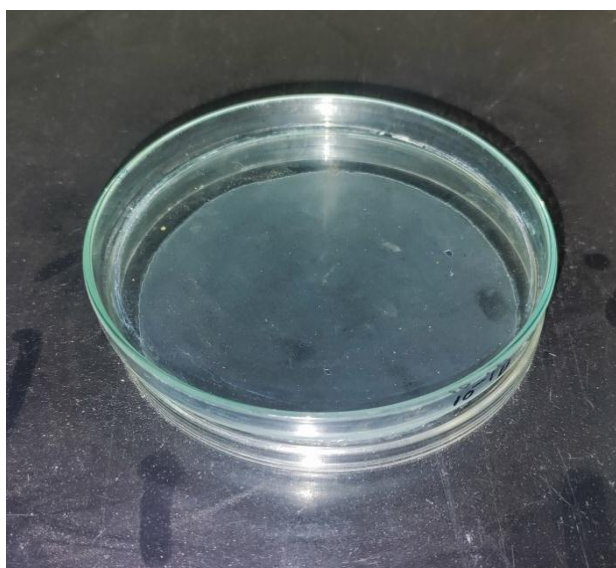
In case of gel, the pH was directly measured in the sample using pH metre. The pH metre was equipped with glass electrode which determines the pH of the resulting solution at 27±2°C. pH of hydrating gel with *Salvia Hispanica* oil was found to be **6.1**.

4) Determination of Washability

Formulations were applied on the skin and then ease and extend of washing with water were checked manually. The washability of gel was found to be good.

5) Determination of Spreadability

Spreadability indicates the extent of area to which gel readily spreads on application to skin. About 1gm of gel was weighed at the centre of the glass plate (10x10cm) and carefully another glass plate was placed over it. 2kg weight was placed on the plate (avoid sliding of the plates). The diameter of gel in cms was measured after 30mins. The diameter of gel was found to be **9.2 cm**.^[11]

**Fig. 4: Spreadability of hydrating gel.**

So, after performing skin moisture meter test F_3 was selected as it was giving similar hydration as F_4 and F_5 and the further tests were performed for final product i.e F_3 . Test included determination of pH, spreadability test, washability test. And then accelerated stability test was performed for F_3 .

RESULT AND DISCUSSION

Chia seed contains omega-3 fatty acid and α -linolenic acid that have strong hydrating effect to maintain healthy and fresh looking skin. The hydrating materials help to balance natural hydrating mechanism and keeps the skin in healthy condition. Hydration is much needed to skin in case to keep it soft and supple.

Standardization of chia seed oil was carried out, acid value, saponification value, specific gravity and refractive index were determined. From the result, it was found that the values of the test of chia seed oil was in the standard range so it was found to be pure and could be used in cosmetic preparation.

Gas chromatography was performed and the result showed that 72.43 % of polyunsaturated fat was prominent followed by 54.95% omega 3 fatty acid and 17.48% omega 6 fatty acid. The result also showed 6.99% of monounsaturated fat, 19.38% saturated fat and trans fat was found to be 0.1%. the peak was formed at the highest concentration of fatty acid. Ali et al. found that on an average chia oil contains about 64% omega-3 and 19% omega-6 fatty acids and we found that it contains about 54.93% omega 3 and 17.48% omega 6 comparatively in our result.^[12]

Fatty acid profile was performed and the result showed that palmitic acid, linoleic acid, linolenic acid, oleic acid, stearic acid and arachidic acid was found. Bartosz Kulcznski found palmitoleic acid, margaric acid, oleic acid, eicosenoic acid, linoleic acid, linolenic acid, eicosadienoic acid, myristic acid, palmitic acid, stearic acid, arachidic acid, behenic acid, lignoceric acid.^[13]

Three different base formulations of gel were developed and out of which F_3 formulation was selected as final formulation, it was observed after 24 hours of observation that gel formed with uniform consistency, had good transparency without any tacky film.

The recommended percentage of active was 1-5% as per (Jeong et al. 2010). Jeong et al. prepared topical formulation of 4% chia oil and applied for 8 weeks. It was found that the

formulation with chia seed oil applied on skin improved the skin hydration, lichen simplex chronicus and prurigo nodularis. Healthy volunteers suffering from xerotic pruritus also revealed improvement in skin hydration followed by trans epidermal water loss and capacitance of skin.^[14] As indicated by Jeong et al. in present study different percentage of active i.e. chia seed oil was added in base formulation i.e. 4%, 5%, and 6% and these formulations were found to be stable and also gave good effect for hydration so the least concentration was selected i.e 4%(F₃) for further studied for performance evaluation.

Spreadability was determined to check the extent of area to which gel is spread. Here glass plates were used to check the spreadability and some weight was kept on it for around half an hour. The gel passed the spreadability test which was found to be 9.2 cm.

Washability of gel was checked manually after applying different formulations on skin and ease and extent of washing with water was checked. The gel passed the washability test.

Digital skin moisture meter (SK-3) was used to determine the moisturizing efficacy of gel with different concentrations. Ten subjects were taken for every skin type i.e. dry skin, normal skin and oily skin and the average was found out for final result. These formulations were compared with hyaluronic base (B₁) where the formulation showed result closer to B₁. According to the standard interpretation of moisture meter, more than 50% indicated moisturized skin. The test was passed by all the given samples. Three formulations i.e. F₁, F₂, and F₃ were selected which gave similar result after comparing it with hyaluronic acid base so the least concentration was selected i.e. F₃ containing 4% of chia seed oil.^[15]

The sample with 4% chia seed oil were evaluated for accelerated stability testing at different conditions and following changes were observed for month in which stability was checked twice in a week. The colour, odour and pH of the product were found to be stable. Graph was formed of change in pH of hydrating gel against no. of days. The formulation of hydrating gel was found to be effective in terms of all functional parameters.

CONCLUSION

Herbal formulation are considered safer than synthetic formulation as synthetic formulation are associated with side effects such as contact allergy, local irritation, scaling, photosensitivity, itching, pruritus, redness, skin peeling etc.

Although various topical herbal formulations for hydrating/ moisturizing are available in the market like creams, lotions, but the present study was done in gel based formulation because gels are generally more stable and active and water-based gel being less irritant is more preferred over creams and lotions.

A hydrating gel containing chia seed oil (*Salvia Hispanica L*, Lamiaceae) are reported to have very beneficial effect on hydration due to high concentration of α - linolenic acid also contains omega 3, omega 6 of different chemical constituent.

The aim of formulating chia seed oil hydrating gel was found successful with good results. The hydrating gel showed a good spreadability. The formulation showed a good hydrating effect on human skin without causing skin irritation. Accelerated stability studies were also done for the formulation. The formulation was found to show good stability result and was found to be stable at room and in refrigerator temperature. It was found to be non-irritant. The study revealed that the formulation is capable of give hydrating effect to skin. So it can be concluded that **Salvia Hispanica oil will help to prevent dry skin.**

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