Research Article

DEVLOPMENT AND EVALUATION OF ORGANIC LEAVE-IN-HAIR CONDITIONER

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Abstract

Hair products like hair styling gels, straight irons, curling irons along with other similar tools cause the hair shaft to become damaged and brittle, dry, and causing split ends. In this work we have revealed the application of organic, natural UV-protective substances such as grape seed oil and marshmallow root extract (Resha Khatmi).and grape seed oil which acts great on dry hairs and helps in easy detangling as well as softening of the hair as well as has Anti- UV effect. Grapeseed oil helps strengthens weak or brittle hair and contains tocopherol (vitamin E) which has great anti-oxidant property and protects against UV damage, and De-Frizz the hair. Hydrolyzed Keratin Protein which increases cystine content is used which smoothens and moisturizes the hairs. This work also comprised of extraction of marshmallow root and evaluation of formulation.

Keywords- Grape *seed Oil, Hydrolysed Keratin Protein, Marshmallow Root Extract*

1. INTRODUCTION

There are number of product present in the market for hair and are used by both men & women, there are products such as shampoos, hair straighteners, shaving products & depilatories. Hair consists of central medulla surrounded by cortex. Hard keratin makes up the cortex, whereas soft keratin makes up the center medulla. The cuticle is formed by the very thin overlapping flat scales that cover the outside of hair. **[1]**

Keratinization of germinative cells leads in a transformed epithelial structure that is identified as hair. **[1]**

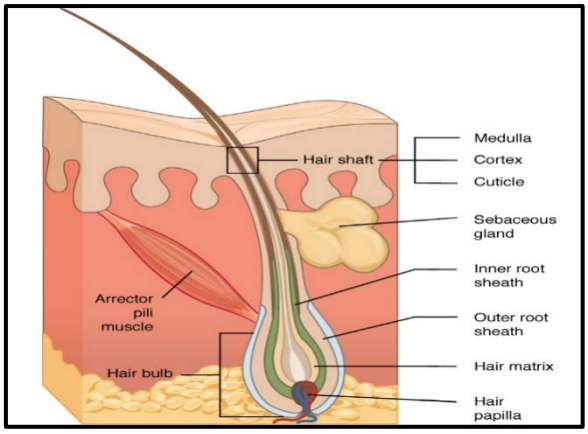

Figure 1. Structure of hair

The hair shaft of mammals is divided into three main categories as mentioned

- **Cuticle:** The Cuticle of the hair is naturally coated with oily substances named as sebum that haves the property to protect the hair from drying. A healthy cuticle must be seen smooth & flat. **[2]**

- **Cortex:** The Cortex of the hair consists of long stands of keratin, held together by disulfide & hydrogen bonds. **[2]**

- **Medulla**: The most sensitive section of the hair shaft is the medulla. **[2]**

Hair conditioners are the cosmetic products designed to improve the texture, manageability & overall health of our hair. They work by adding moisture nourishing ingredients & protective layer to our hair shaft. Different types of hair conditioners are available in the market such as Rinse out conditioners, leave in conditioners, deep conditioners, and conditioning serums. **[3]**

Leave in hair conditioners are applied to wash and dried hair and it remain on it till the next shampoo. The property of leave in hair conditioner is to smoothen down the cuticle and protect the hairs from weathering. They are convenient and popular. They contain cationic detergents, water, lipids, thickeners and are available in thick/thin formulations.it has short lived effect. **[4]**

Leave in hair conditioners are the product used for curly and unmanageable hair, blowing hair, and hair that has been damaged by chemical reactions.as it contain mineral and vegetable oil, petrolatum, and silicone. They maintain the moisture and give hair a glossy finish and fine hair is not a good fit for them **[4]**

1.1 Grape seed oil (Vitis Vinifera):

In response to their nutritional qualities the berries of Vitis Vinifera L ssp sativa grapes have attracted attention from all around the world.it has the biological properties such as antioxidant, anti-inflammatory. Grape seed oil is a vegetable oil derived from the seeds of grapes.it contains. Grape seed oil contains 8% - 20% of oil (dry basis). Vitamin E has the beneficial effects of grape seed oil because of its high antioxidant activity. This oil is non-greasy in texture. It helps to protects against UV damage, smooth’s and blocks split ends and act as a moisturizer. **[5]**

1.2 Marshmallow root extract (Khatmi):

Khatmi consists of the root of Althea officinalis Linn. (Family Malvaceae) a perennial, uniformly downy herb, occurring in Kashmir region. This root contains lauric acid, which is the fatty chain which protects your roots and follicles, giving them a better chance to flourish. Additionally rich in mucilage, a viscous material composed of proteins and polysaccharides, is marshmallow root. Because of its texture, marshmallow root works well as a detangler, making brushing your hair easier **[6]**

1.3 Argan oil:

Argan oil is a plant oil obtained from the kernels of the Argan tree (Argania spinosa L.), which includes tocopherols (Vit E), essential fatty acid and anti-oxidants that nourish hairs. Argan oil has rich source of (Vit E), which provides a fatty layer to hair that may prevent dryness and can help to reduce fizziness and boost shine. **[7]**

1.4 Hydrolysed keratin protein:
In basic terms, hydrolyzed keratin is a big protein molecule that is extracted from sheep wool and cow hoof and undergoes a chemical breakdown to enable it to pass through the hair cuticle.
Weak hair strand gaps will be filled up by the hydrolyzed keratin. **[8]**

1.5 Cocoa butter:

Cocoa butter derived from Theobroma cacao (Sterculiceae) has a property particularly soothing after windburn or sunburn. It helps in smoothening of hair and heals dry and damaged hair. Combined with vitamin E hair is less prone to dullness **[9]**

1.6 BTMS-50 (Behentrimonium Methosulphate):

It works as an emulsifying agent that was added in the formulation to combine water and oil together. It is also natural plant-based emulsifier derived from natural oil called rapeseed oil. It is one of the best emulsifier any leave in conditioner could ever have those conditions hair and preparing hare care products. **[10]**

Figure 2. Product Image

Sr. No	Brand of Product	Main compound	Overall Application
1.	Cantu	Grape seed oil+ almond oil+ Mango butter	To seal moisture in the hair, reconstructing, smoothing, curl enhancing.
2.	Curl up	Argan oil+ Flax seed oil extract.	Help to reduce frizz, define and add shine to curly hair.
3.	RIZVAN	Argan oil+ Marshmallow root+ Burdock root oil+ aloevera etc.	Hydrates hair, reduces frizz, add shine, repair damage.
4.	Moraccanoil All-in-One leave in conditioner.	Sunflower seed oil+ Argan oil+ quinoa and barley extracts.	Nourishes hair, protect against heat damage, and detangles with ease.
5.	EDEN	Hydrolyzed keratin protein+ Coconut+ Shea.	Repair your hair, adds hydration.
6.	TRUE by made Beautiful Nourishing Leave-in-conditioner.	Shea+ Honey+ Coconut &mango+ Hydrolyzed keratin protein.	Detangle and moisturises the hairs
7.	Crème of Nature	Argan oil+ Hydrolyzed keratin protein+ Hydrolyzed wheat protein.	Adds shine, protect hairs, improve hair strength, add moisture and shine.

Table 1. Comparative Market Product

MATERIALS:

Raw Material Vendor list

Protein Keratin Liquid Hydrolysed (BRM Chemicals)

- Behentrimonium Methosulphate Btms-50 (BRM Chemicals)
- Grape seed Oil (BRM Chemicals)
- Marshmallow Roots (Jio Mart)
- Lavender Oil (Wellness Forever)
- Argan Oil (Wellness Forever)

3. EXPERIMENTAL

3.1 Development of Conditioner

Table 2. Product Formulae

Sr.No	Component	Quantity
1	Grapeseed oil.	3ml
2	Argan oil	1ml
3	Marshmallow Root Extract	2 ml
4	Hydrolyzed Keratin Protein	2ml
5	BTMS-50	4g
6	Cocoa butter	1g
7	Cetyl Alcohol	1g
8	Glycerine.	2ml
9	Water	Q. S
10	Sodium Benzoate	0.1g
11	Citric acid	Q. S
12	Fragrance	2-3Drops

Measure out aqueous phase ingredients like water, glycerine, marshmallow root extract. Cover the container using aluminium foil to avoid evaporation (If required) Measure out oil-phase ingredients like Argan oil, Grapeseed oil, Cocoa butter, Btms-50, Cetyl alcohol. Gently melt all the ingredients using water bath at temperature 60 degrees till 20 minutes approx. till it completely melts. Once two phases have completely melted (no solids remain) then transfer into a glass bowel. Slowly blend both the phases together using electronic blender. Blend until conditioner has thickened and emulsified. Cool the mixture, creamy-glossy consistency.

Now after cooling add hydrolyzed keratin protein, sodium benzoate as a preservative and lavender oil as an essential oil. Completely blend the conditioner and adjust the pH using citric acid. Store the conditioner in a squeeze bottle and perform evaluation.

3.2 Viscosity Determination:

The viscosity of prepared formulation was analysed by the Brookfield Viscometer LVDVE with helipath, using spindle number 96 at 10 rpm. **[11]** Prepared formulation is taken. A level indicator was used to set the instrument's base level. After being cleaned, the spindle was fitted to the device. After that, the spindle was turned inside the gel until the viscometer displayed a consistent reading. Three times through the process, determine the average viscosity value. **[12]**

Figure 3. Brookfield Viscometer & its Spindles

3.3 Spread ability:

Spread ability is measured in terms of the amount of time, measured in seconds that the slides need to move off a formulation placed in between them when a specific load is applied. If the time taken for separation two slides is less better is the spread ability. It is calculated by using following formula:

$$S=M\times L/T \quad (1)$$

Where,

M = weight tied to upper slide

L = length of glass slide

T = time taken to separate the slide

Spreading 0.5 g of the gel on a premarket glass plate allowed researchers to measure the gel's spreadability. After then, another glass plate was set on top of it.The suitable gram of weight was placed in the balance. Time required by the glass plate to slip off was determined. Spread ability of the gel formulation was calculated using the given formula. **[13]**

3.4 pH determination:

The pH of formulation was determined by using digital pH meter.1gm of formulation was taken and dispersed in 10 ml of distilled water and kept aside for two hours. **[14]**

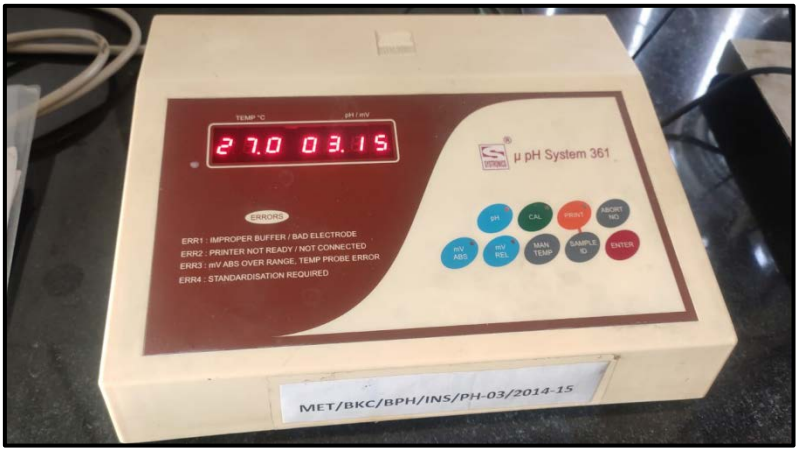

Figure 4. Formulation pH by Digital pH Meter

3.5 Homogeneity:

Following the placement of the leave in conditioner into the container, the created formulation was examined visually to ensure homogeneity.

3.6 Melting Point

Using Thiele's tube, the melting point of the formulation was found to be between 90 and 98°C

- Introduce small amount of formulation into Capillary Tube.
- Place this in the middle of the thermometer in the heating bath
- Maintain an eye on the temperature where melting starts.
- Note the Melting Point. **[15]**

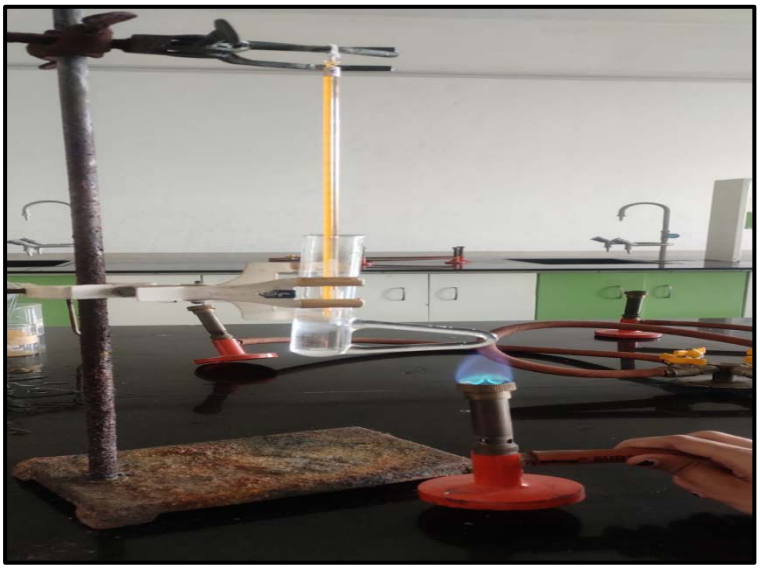

Figure 5. Melting Point Assembly.

3.7 Skin Irritation Test:

Acute dermal irritation test was performed as per OECD guidelines: 6 female/male (either sex) wistar rats were used for experiment. The animals were allowed tp acclimatize for 7 days after procurement. Temperature (22+20) and humidity (45-55%) was controlled. The animals were kept 12hrs in light/dark cycle. On the day before administration of the test formulation, fur was removed from the dorsal/flank area of the test animals by using hair removal cream.

Over the exposed dorsal/flank skin area, the test formulation was applied uniformly, covering at least 10% of the body's surface area. During the course of a 24-hour exposure period, the test formulation was kept in contact with the skin using non-irritating tape and a porous gauze dressing. The test site will be further covered in a suitable manner to retain the gauze dressing and test formulation and ensured that the animals cannot ingest the test chemical. During the 24-hour exposure period animals were caged individually in order to avoid oral ingestion of the test chemical by other animals in the cage. At the end of the exposure period, residual test chemical was removed using water. A period of 48 hours was allowed between the testing of each animal, though this will depend on the onset, duration, and severity of toxic signs. Animals were observed as soon as possible after dosing, at least once in the first thirty minutes and then again every few hours over the next twenty-four hours. During the first two to six hours following the start of the exposure period and every day after that, extra care was given to the animals.

Table 3. Skin Irritation Test Experimental Design

Test group	
3 animals	3

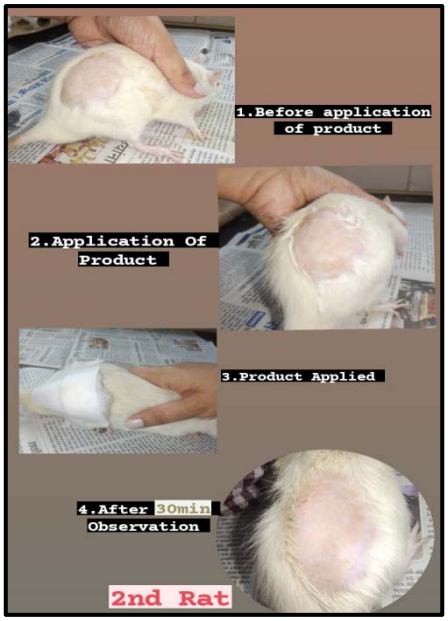

Figure 6. Observation on Rat 1

Figure 7. Observation on Rat 2

Figure 8. Observation on Rat 3

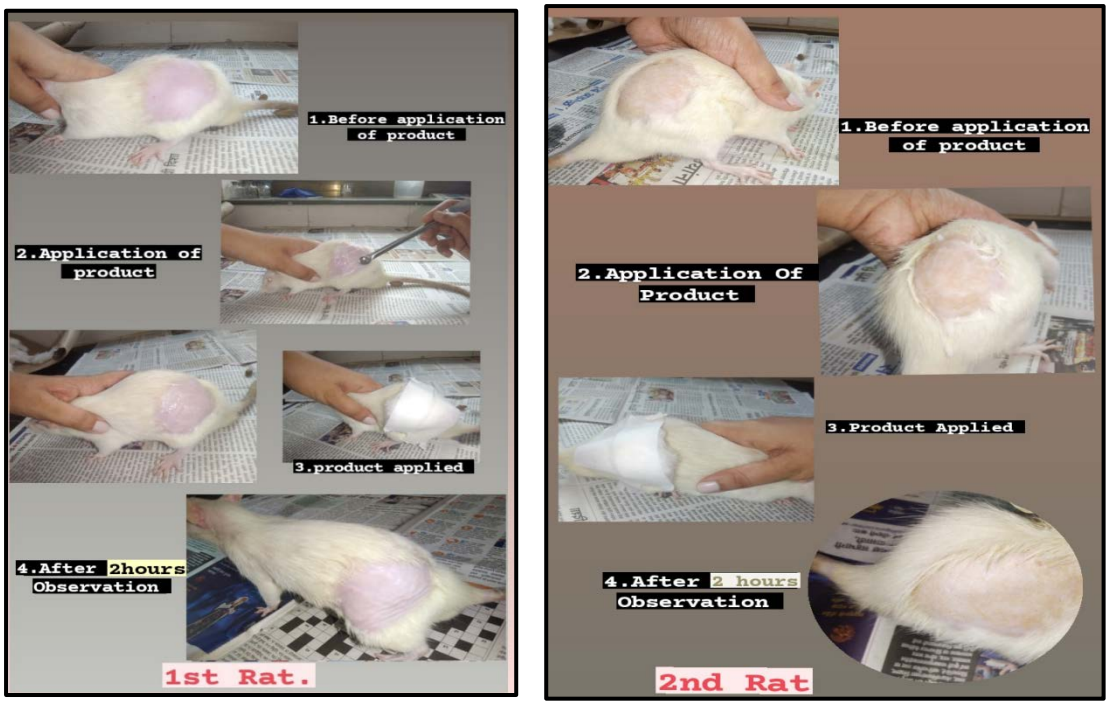

Figure 9. Skin Irritation Observation after 2 Hours.

After 1 Day and 4th Days

Observation:

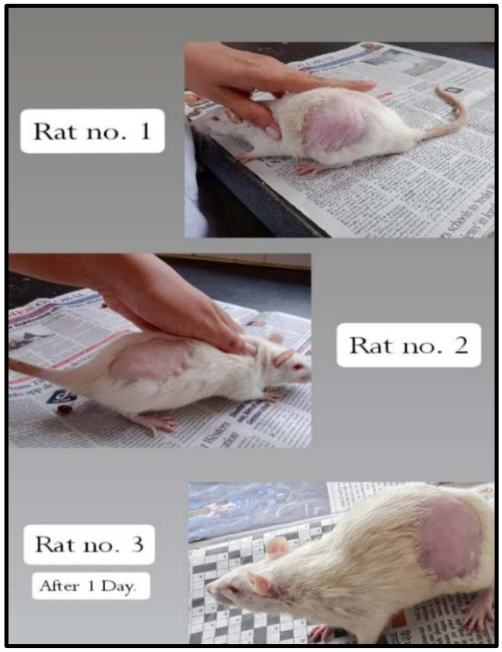

Figure 10. Observation after 1 day

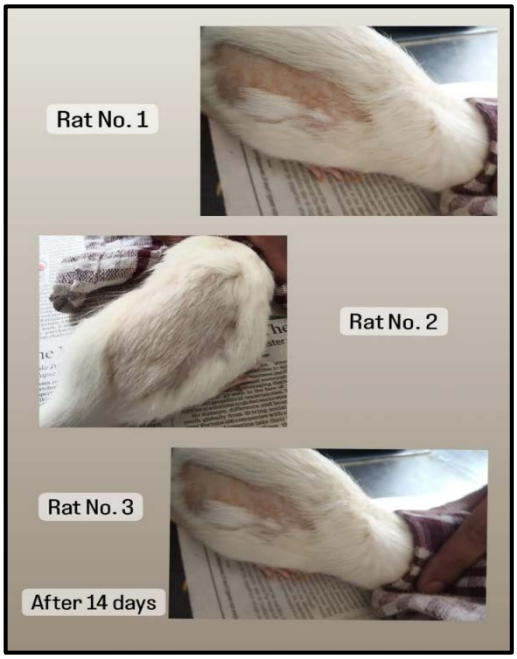

Figure 11. Observation after 14 days

3.8 DPPH Assay:

The antioxidant activity of grapeseed oil and marshmallow root extract was determined by a methodology using the 2, 2, -diphenyl-1-picrylhydrazyl (DPPH). It is a stable free radical with purple colour which turns yellow when scavenged. This method is used to measure the Total antioxidant capacity (TAC). The % radical scavenging activity or % inhibition of the sample can be calculated using the formula:

% Inhibition= (A0-At)/A0 *100

Where, A0 - Absorbance of control. At - Absorbance of sample. **[16]**

1. Prepare stock solution of DPPH by dissolving 4 mg DPPH in 100ml methanol. Take its absorbance at 517 nm. Dilute the solution by adding 1 ml of above stock solution in 10 ml methanol. Take its absorbance (A0)

2. Make 100 ppm stock solution of sample and std ascorbic acid Make dilutions from this as 20, 40, 60, 80 ppm take 1 ml DPPH stock & 3 ml of above diluted sample solution in another test tube and repeat the process for all dilutions. Similarly, for std ascorbic acid (1ml DPPH+ 3ml diluted ascorbic acid solutions) Keep dilutions aside for 30 min after mixing. Take absorbance of all samples at 517 nm (At) **[16]**

4. RESULT AND DISCUSSION:

Table 4. Result and Discussion

Parameters	Result
Viscosity Determination	2000+-25000cps
pH Determination	Values of formulation lies in acidic and value found to be 3.3
Spread ability	Spread ability was found to be 14.2
Homogeneity	The formulation was found to be homogenous and no visible particles were seen
Melting point	The Melting point was observed to be 90-98°C
Skin irritation Test	There was no irritation on skin of rats

Table 5. DPPH Assay Absorbance of Control

Control	Absorbance	Mean (A0)
DPPH Stock Solution	0.434	0.422
	0.411	

Table 6. Characterization of Product

Parameters	Result
Color	Whitish

Odour	Pleasant
Texture	Smooth
Physical State	O/W Emulsion
Solubility in Water	Slightly Insoluble
pH	2.83
Viscosity	20000+-25000cps
Spread ability	14.2
Melting point	90-98 degree Celsius
Specific Gravity	0.99-1.30
Skin Irritation Test	No Irritation.

5. SUMMARY AND CONCLUSION:

The formulation containing Marshmallow extract and Grapeseed oil along with Hydrolyzed Keratin Protein was found to have good properties with appropriate pH, Spread ability, Viscosity, Melting point, and Compatibility with each other. Skin irritation not observed in preclinical study. The formulation was also found to be stable and having good physical stability as well as no phase separation was seen. Formulation softens and smoothen the Hair shafts. Adds luster and Vitality. No greasy appearance was found. Moreover, it was found to be stable and effective for hair management.

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