

# FORMULATION AND EVALUATION OF NON-GREASY HERBAL HAIR CREAM

**Ahamed Mueen Mubeen <sup>1\*</sup>, Amitha Susan Achenkunju <sup>2</sup>, Dashlima Begum. K <sup>2</sup>,  
Thangameena. M <sup>2</sup>, Narmadha. K.N <sup>2</sup>**

<sup>1</sup>Department of Pharmacognosy, Swamy Vivekanandha College of Pharmacy, Elayampalayam,  
Tiruchengode, Tamil Nadu-637205

<sup>2</sup>UG Scholar, Swamy Vivekanandha College of Pharmacy, Elayampalayam, Tiruchengode, Tamil Nadu-  
637205.

## **Corresponding Author**

Ahamed Mueen Mubeen\*

Contract No.: +91-9791166468

Email ID: ahmdmueen96@gmail.com

## **ABSTRACT:**

*The aim and objective of this study to formulate and evaluate the herbal based non-greasy herbal hair cream. here, totally five herbal extracts were used hydroalcoholic extracts of Hibiscus rosa sinensis, Withania somnifera, Eclipta alba, Olea Europea and Nigella sativa were used in different composition to get multipurpose effects on scalp and hair such as antimicrobial, prevent hair fall, increased hair growth and follicles, prevent graying of hair, prevent dryness of hair and scalp, protection of hair from UV rays. There are two different formulation were formulated (F1 and F2) and standardization procedure were done with this two formulations. By the results of standardization F2 formulation is better one than F1 formulation, it was selected for further evaluation studies of antimicrobial studies and Sun protection factor determination studies. The F2 formulation significantly has anti bacterial and anti fungal effect, and the F2 formulation has ability to block 50% of UV rays, it was confirmed by SPF value determination. In future in-vivo animal studies can be conducted for F2 formulation.*

**Key word:** Hair cream, antimicrobial, Sun protection factor, Cream evaluation.

## INTRODUCTION

Herbal cosmetics also known as natural cosmetics. Now these days, people become more health conscious than before, so, they prefer best. Today, herbal cosmetics are growing popular not only in developed countries like United States, Canada, United Kingdom, Australia, Germany and France etc. The German and the French herbal cosmetics markets are the most upcoming markets among the European countries. The major problem in analyzing world trade particularly herbal cosmetics is the limited statistical data available on production and trade [1]. In India AYUSH a separate department was established in March 1995 to promote indigenous systems of medicine and homeopathy. This provided a platform and boosted the herbal cosmetic industries too. The overall Indian personal care market has the potential to grow at 15-16% per annum and thereby double to US\$ 8 billion (approx. 40,000 crore) by 2012 [2].

### **Pharmaceutical and therapeutic aspects of cosmetics:**

Cosmetics are such preparations which are being utilized by every age group. The market of cosmetics has existed all over the world and proves economically beneficial. Today the market is flooded with various cosmeceutical preparations including baby preparations, bath preparations, cleansing preparations etc.

The public's interpretation of what constitutes a drug or a cosmetic may differ somewhat from that of regulatory agencies. The ingredients used in cosmetics to a large extent are the same as those employed in drugs. According to US law, cosmetics may contain ingredients that treat or prevent disease or alter the structure or function of the human body. The objective of cosmetics is limited to the enhancement of appearance [3].

## HAIR

Hair is an important part of human body. The problems associated with it includes hair loss, unruly hair, lack of hair volume, conditioning, immature graying, dandruff, thinning of hair, dullness etc. Hair can vary in shape, length, diameter, texture, and color. The cross section of the hair could also be circular, triangular, irregular, or flattened, influencing the curl of the hair. All mammals have hair. Its main purpose is to regulate body temperature. It is also wants to decrease friction, to guard against sunlight, and to act as a way organ. [4]

### **Hair consists of two parts:**

#### **1. Follicle**

The follicle is a club shaped structure in the skin. At the top of the follicle may be a network of blood vessels that provide nutrients to feed the hair and help it grow. This is called the papilla. Each follicle arises from an interaction between epidermis and dermis.

Follicle is divided into three segments:

- **Infundibulum:** Extends from the surface opening of the follicle to the level of opening

of its sebaceous gland

- **Isthmus:** Extends from the infundibulum to the level of insertion of the arrector Pili muscle.
- **Inferior segments:** The base of the bulb is invaginated by a tuft of vascularized loose connective tissue called dermal papilla. The papilla of the hair contains many blood vessels and provides nourishment for hair growth [5].

## 2. Shaft

The hair shaft is divided into three layers:

- **Medulla:** It is the middle of hair. It's going to be fragmented or segmented and continuous or doubled. It is often a hollow tube or crammed with cells. Most hairs don't contain a medulla, while others have medullas that are discontinuous or fragmented. It forms the middle of the hair shaft. Fine hairs especially tend to lack this layer.
- **Cortex:** It is the largest part of hair shaft which provides the color to hair, i.e. melanin (hair pigment).
- **Cuticle:** The cuticle may be a transparent outer layer of the hair's shaft. It's made from scales that overlap each other and protect the inner layers of the hair. The scales point from the proximal end of the hair, which is closer to the scalp and to the distal end [6].

## HAIR DISEASE

**Mycotic condition:** seborrheic dermatitis and dandruff are known to be two ends of single disease spectrum caused by *Malassezia* species .

**Parasitic capitis:** pediculosis capitis is caused by infestation with *Pediculus humanus capitis*.

**Inflammatory conditions:** psoriasis is characterized by discrete erythematous plaques covered by silver-grey scale .

The bacterial agent often responsible for folliculitis is *Staphylococcus aureus* [7].

## CAUSES OF HAIR LOSS

- Reduction in hair follicle function due to male hormones
- Reduction in metabolic function of hair follicles and hair bulb
- Reduction in scalp physiological functions
- Local impairment of the circulation due to tension in the scalp

- Common causes of hair fall such as stress, eating habits, lack of protein, chemical led products / wrong products, heredity, dramatic weight loss.
- Hormonal changes and medical conditions
- Radiation therapy to the head
- Hairstyles and treatments
- Medications and supplements [8]

### **TYPES OF HAIR LOSS:**

Most hair loss can be categorized into three types:

- Noncicatricial (potentially reversible)
- Cicatricial
- Due to hair shaft abnormalities [9]

## **MATERIALS AND METHODS**

### **COLLECTION OF PLANT MATERIAL**

Crude plant powdered material was purchased from Siddha Pharmacy and checked basic quality control parameters. Then powdered material was subjected to extraction process by Soxhlet apparatus.

### **PROCEDURE FOR EXTRACTION**

The finely powdered material undergoes the Soxhlet extraction method by using hydroalcoholic solvent (70% ethanol and 30% water) for a duration of three successive days (72 hrs). The temperature should not exceed more than 100°C. The dark green color hydroalcoholic extract was collected, filtered and the filtrate was subjected to concentration under reduced pressure using rotary vacuum evaporator. The concentrated product was freeze dried and the product was finally used for further experimental studies [10].

The same procedure was followed for the crude drug ashwagandha, bhringraj and hibiscus.

### **PROCEDURE FOR FORMULATIONS**

Oil in water (O/W) type herbal hair cream was fixed for the study. The emulsifier (stearic acid) and the other oil soluble components (bees wax, lanoline, cetyl alcohol, *Nigella sativa* oil, *Olea europea*) were dissolved in the oil phase (part A). The other water-soluble components (extract of *Eclipta Alba*, *Withania Somnifera*, *Hibiscus Rosa Sinensis*) and triethanolamine were

dissolved in the aqueous phase (part B). Both the mixture A and B heated in a water bath until reaches 70°C, aqueous phase was mixed with oil phase with constant stirring. Finally add preservatives and perfume. [11] The formula for the cream is given in table 1.

**Table 1: Composition of cream % w/w**

INGREDIENTS	F1 (%w/w)	F2 (%w/w)
<i>Eclipta Alba</i> extract [12]	1	1
<i>Hibiscus Rosa Sinesis</i> extract [13]	1	1
<i>Withania Somnifera</i> extract [14]	1	1
Stearic acid	5	4.5
White bees wax	2	2.5
Cetyl alcohol	4	3.5
Lanoline	1	1.5
Black cumin oil [15]	1	1
Olive oil [16]	4	4
Methyl paraben	0.02	0.02
Propyl paraben	0.01	0.01
Triethanolamine	2	2.1
Water	upto 100	upto 100
Perfume	Qs	Qs

## EVALUATION OF CREAM [17,18,19]

### Appearance:

The appearance of the cream was judged by its color, pearlescence, roughness and graded.

### pH of the cream:

The pH meter was calibrated using standard buffer solution. About 0.5g of the cream was weighed and dissolved in 50.0ml of distilled water and its pH was measured.

**Viscosity:**

Viscosity of the formulation was determined by Brookfield viscometer at 100 rpm, using spindle no :7

**Dye test:**

The scarlet red dye is mixed with the cream. place a drop of the cream on a microscopic slide covers it with a cover slip, and examine it under a microscope. If the disperse globules appear red and the ground colorless that is o/w type emulsion. The reverse condition occurs in w/o type emulsion i.e. the disperse globules appear colorless in the red ground.

**Acid value:**

Take 10 gm of cream and dissolved in 50 ml mixture of equal volume of alcohol and solvent ether, then the flask was connected to reflux condenser and heated until the content was dissolved completely, then add 1ml of phenolphthalein and it is titrated with 0.1N NaOH until light pink color appears after shaking the flask for 30 seconds.

$$\text{Acid value} = n \times 5.61 / w$$

n = Amount of NaOH required

W = weight of the substance

**Saponification value:**

Introduce about 2g of substance refluxed with 25ml of 0.5 N alcoholic KOH for 30 minutes to this 1ml of phenolphthalein added and titrated immediately, with 0.5 N HCl.

$$\text{Saponification value} = (b-a) \times 28.05 / w$$

a = The volume in ml of titrant

b = The volume in ml of titrant

w = The weight of substance in gram

**Homogeneity:**

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

**Irritancy test:**

Mark an area (1sq.cm) on the left side of the scalp surface. The cream was applied to

the specified area and time was noted. Irritancy, edema, erythema was checked if any for regular intervals up to 24hrs and reported.

### **Rheological studies:**

The formulated cream was found to be Non – Newtonian. Take a fixed quantity 10grams of cream in a 10 ml beaker. Keep it impact for 1 hr. the beaker was inclined to one side see whether the cream is liquefied or not. Beaker is shaken for continuous 5min and checked whether consistency has changed or not. The beaker was again tilted and checked for pourability of the cream.

### **Stability testing:**

Stability of herbal hair cream was determined by centrifugation studies the cream were centrifuged at 3500-13500 rpm at the interval of 10 minutes and further observe for phase.

### **Type of smear:**

It was determined by applying the cream on the skin surface on human volunteer. After application of the cream, the type of film or smear formed on the skin were checked.

### **Antibacterial assay using agar well diffusion method**

The well diffusion method was used to screen the antimicrobial activity. *In vitro* antimicrobial activity was screened by using Muller Hinton Agar (MHA) obtained from Himedia (Mumbai). The MHA plates were prepared by pouring 15 ml of molten media into sterile petriplates. The plates could solidify for 5 minutes and 0.1% inoculums suspension was swabbed uniformly, and the inoculums could dry for 5 minutes. Wells were cut and 20 µl of the different concentration of test drug (25µl, 50µl, 75µl and 100µl) were added. The plates were incubated at 37°C for 24 hours. The antibacterial activity was calculated by measuring the diameter of the inhibition zone formed around the well (NCCLS, 1993). Chloramphenicol disc was used as a positive control.

### **ANTIFUNGAL ACTIVITY BY WELL DIFFUSION METHOD:**

Antifungal activity was measured using methods of well diffusion plates on agar. To test the antifungal activity, the fractions of different concentration of plant extract were dissolved in 70% ethanol. 20 mL of Sabouraud Dextrose Agar was poured into each 15 cm Petri dish. *C. albicans* were grown in sabouraud dextrose broth at 27° C for 48 h. Growth was adjusted to OD (600 nm) of 0.1 by dilution with sabouraud dextrose broth. Then, Wells were cut and 20 µL of

the different concentration of test drug were placed on agar to load 10 and 15  $\mu\text{L}$  of each spice sample (1 mg/mL). 100 units of Fluconazole, obtained from a local pharmacy, were used as a positive control. Inhibition zones were determined after incubation at 27° C for 48 hrs.

### DETERMINATION OF SPF VALUE

1g of Herbal hair cream was weighed, transferred to 100 ml standard flask, diluted to volume with ethanol to get 10 mg/ml of concentration and it was subjected to ultra-sonification for 5 min. After that filtered through Whatman No.1 filter paper and collect the filtrate by rejecting the first 10 ml of filtrate, then 5 ml of aliquot was taken in 50 ml standard flask, diluted to volume with ethanol to get 1 mg/ml of concentration. Subsequently, 5 ml of aliquot was transferred to 25 ml standard flask and the volume completed with ethanol to get 200  $\mu\text{g}$  /ml of concentration. The absorbance values of each aliquot were determined from 290 nm to 320 nm at 5 nm interval, using ethanol as a blank. The readings were taken in triplicate and the determinations were made at each point, followed by application of Mansur equation.

Mansur et al. (1986) developed a very simple mathematical equation which substitutes the in vitro method proposed by Sayre et al. (1979), utilizing UV Spectrophotometry and the following equation.

$$SPF(spectrometry) = CF \times \sum_{290}^{520} EE(\lambda) \times I(\lambda) \times abs(\lambda)$$

Where, CF=Correction factor (10), EE ( $\lambda$ )=Erythmogenic effect of radiation with wavelength  $\lambda$ , Abs ( $\lambda$ )=spectrophotometric absorbance values at wavelength  $\lambda$ . The values of EE×I are constant [20].

### RESULTS AND DISCUSSION

Formulation F1 and F2 was successfully completed with respective procedures and then the formulations F1 and F2 was evaluated for its stability, so the basic standardization parameters was calculated and it was plotted in the table 2



**Table no 2: Physicochemical parameters**

S.No	Parameters	F1	F2
1	Appearance	Pale green	Pale green
2	pH	6.9	6.7
3	Viscosity	27001-27089 cps	25025-25099 cps
4	Dye test	o/w emulsion	o/w emulsion
5	Acid value	3.81	3.87
6	Saponification value	26.8	26.3
7	Homogeneity	Uniform distribution	Uniform distribution
8	Irritancy test	No irritation	No irritation
9	Rheological study	Pseudo plastic flow	Pseudo plastic flow
10	Stability test	No phase separation	Phase separation

Here totally two formulations were prepared (F1 and F2) and evaluated. From the evaluation study F1 formulation more stable than F2 formulation.

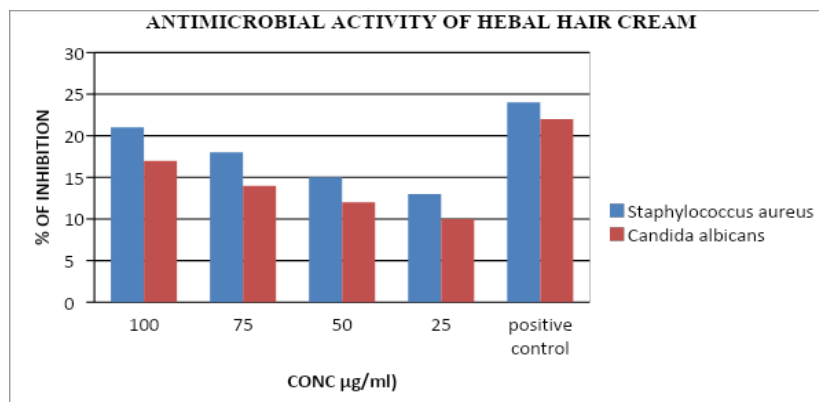
In the present work decided to extract crude powder drug and formulate herbal hair cream. The herbal hair cream was O/W type of emulsion hence; it can be removed easily by water that gives better customer complaints. So, the better formulation F1 choosed for further *in-vitro* studies.

## ANTIMICROBIAL ACTIVITY OF HERBAL HAIR CREAM

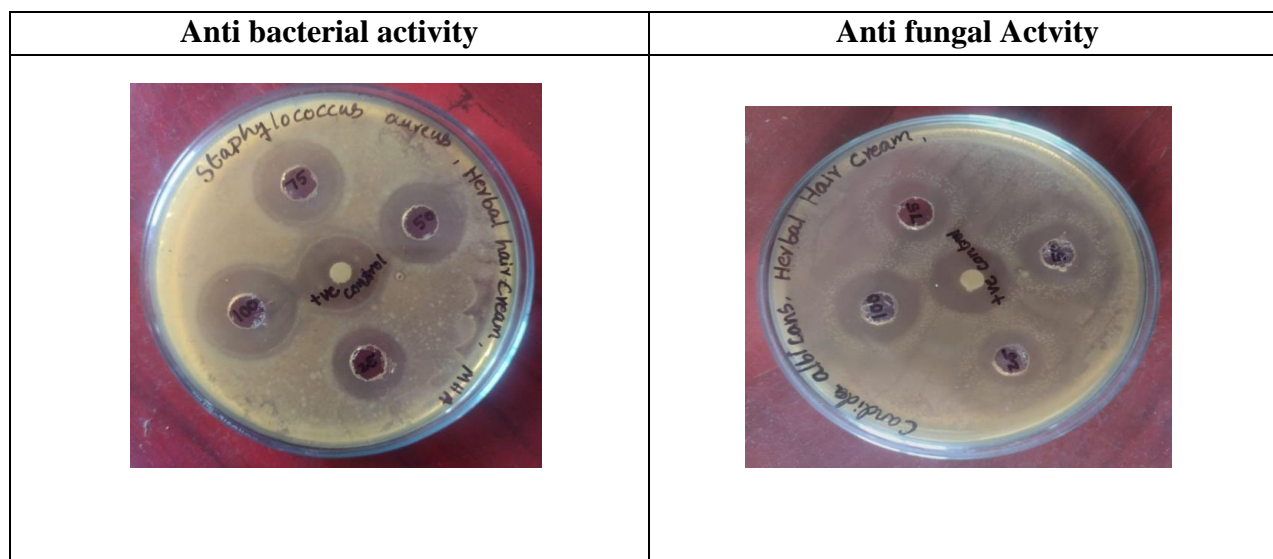
The multipurpose herbal hair cream (F1) was evaluated for *in-vitro* antimicrobial activity here totally two stains were observed for this study that is *Staphylococcus aureus* (bacteria), *Candida albicans* (fungi). This study revealed that multipurpose herbal hair cream exhibited significant antimicrobial activity. The observed results were given in the table 3 and figure 1

**Table no 3: Zone of inhibition**

S. NO	MICROORGANISMS	ZONE OF INHIBITION (MM)				
		100 $\mu$ l	75 $\mu$ l	50 $\mu$ l	25 $\mu$ l	+ VE CONTROL
BACTERIA – GRAM POSITIVE						
1	Staphylococcus aureus	24	18	15	13	24
FUNGAL						
2	Candida albicans	17	14	12	10	22



## PICTORAL REPRESENTATION OF ANTIBACTERIAL AND FUNGAL ACTIVITIES OF HERBAL HAIR CREAM



**Bacteria (*Staphylococcus aureus*):** The zone of inhibition of standard drug (chloramphenicol) and test sample (herbal hair cream) was found to be 24mm and 24mm respectively.

**Fungi (*Candida albicans*):** The zone of inhibition of standard drug (fluconazole) and test sample (herbal hair cream) was found to be 22mm and 17mm respectively.

The *in-vitro* antimicrobial study is so important for reduce the usage of animals in research works. Generally, four methods were used for *in-vitro* antimicrobial assay.

This study performed by using different concentration of sample (25 $\mu$ g, 50 $\mu$ g, 75 $\mu$ g and 100 $\mu$ g) were used for this study and finally the *in-vitro* antimicrobial study shows significant antimicrobial activity against *staphylococcus aureus*. We can observed the maximum zone of inhibition at maximum concentration of the test sample. So, it gives dose dependent activity and zone of inhibition of test sample gives same like chloramphenicol inhibitory activity against *Staphylococcus aureus*. The antifungal activity of herbal hair cream shows good action against *Candida albicans* but lesser than fluconazole. This notable antimicrobial activity of herbal hair cream may be exhibited due to the presence of *Nigella sativa* oil (thymoquinone) and *Eclipta alba*.

## UV PROTECTION ACTIVITY OF HERBAL HAIR CREAM

Multipurpose herbal hair cream was evaluated for *in-vitro* uv protection activity by uv spectrometric method. This study revealed that multipurpose herbal hair cream exhibited significant uv protection activity and the results are plotted in the Table 4, 5 and fig 2

**Table no 4: In-vitro SPF value determination of herbal hair cream formulations measured under different wavelength**

S.No	Wavelength ( $\lambda$ nm)	EE×I (Normalized)	Absorbance	Absorbance× CF×EE×I	SPF= $\sum EE(\lambda) \times I(\lambda) \times 10 \times$ Absorbance
1	290	0.015	0.296	0.0444±0.0011	2.1633
2	295	0.0817	0.258	0.2107±0.0012	
3	300	0.2874	0.231	0.6638±0.0011	
4	305	0.3278	0.211	0.6916±0.0013	
5	310	0.1864	0.196	0.3653±0.0011	
6	315	0.0839	0.186	0.1560±0.0012	
7	320	0.018	0.175	0.0315±0.0012	

**Table no 5: Reference standard value**

SPF	Percent of UV blocked
2	50
4	75
5	80
10	90
15	93
25	96

The sun protection factor of multipurpose herbal hair cream was found to be 2.1633. According to the reference standard value the cream has ability to block 50% of UV radiation

The proposed UV spectrometric method is simple, rapid, use low reagents hence, we used for *invitro* determination of SPF values in many cosmetic formulation in recent years, natural compound or bioactive products have gained considerable attention as UV protective agents due to the presumable safe utilization, ecological issues and minimum side effects besides their antioxidant activity, Plant extract containing wide range of phenolic acids, flavonoids and high molecular weight polyphenols usually covers the full range of UV wavelength. so, the multipurpose herbal hair cream containing four herbal extracts that can be absorb UV radiation and prevent our hair and scalp from UV rays. so, due to the sun protection activity we can control or overcome greyish of hair, burning sensation, to prevent hair follicles from UV rays.

## SUMMARY

The aim and objective of present work was to formulate and evaluate multipurpose herbal hair cream for treat several scalp and hair follicle related problems. It should be stimulating hair growth, hair follicle stimulations and it can able to act against bacterial and fungal related disease. So, our research study completely focused on selections of pharmacologically potent herbal drug, it should be control hair fall and scalp related microbial diseases. The plants are more potent healers because they promote the repair mechanism in the natural way. The knowledge and experience of usage of herbs are being blend with advanced cosmetic technology to develop a safe and effective product.

In the present study, the multipurpose herbal hair cream was prepared using herbal plant extracts such as *Nigella sativa*, *Withaniasomnifera*, *Eclipta alba*, *Hibiscus rosasinensis*, *Olea europea*. The cream type was O/W emulsion, this type of emulsion generally non-greasy, easily removed by water and it is more suitable for transdermal drug delivery system and it can be gives sustained release of drug through the scalp and the active principles can easily penetrate through the scalp. Here totally two formulations were prepared (F1 and F2) with different ratio of oil and aqueous phase. The formulation F1 and F2 evaluated for its physiochemical evaluation such as appearance, pH, viscosity, dye test, acid value, saponification value, homogeneity, irritancy test, rheological studies based on the evaluation study, stability test the F1 formulation is more stable than F2 formulation so, the F1 formulation choosed for *in-vitro* antimicrobial study, it was performed by using well diffusion method totally two different stains *Staphylococcus aureus* (bacteria) and *Candida albicans* (fungus) were used. Both the microbial growth was significantly inhibited by herbal hair cream. Then the cream was subjected to sun protection factor analysis by UV spectrometric method and the result of the study was positively noted. It has ability to block 50% of UV rays from sunlight. From the present study it can be concluded that it has significant antimicrobial activity and UV protection activity so we can continue with further *in-vivo* studies.

## CONCLUSION

This study concluded that, the combining extracts of *Hibiscus rosa sinensis*, *Withania somnifera*, *Eclipta alba*, *Olea Europea* and *Nigella sativa* were used in different composition to get multipurpose effects on scalp and hair such as antimicrobial, prevent hair fall, increased hair growth and follicles, prevent graying of hair, prevent dryness of hair and scalp, protection of hair from UV rays. We know that it is not possible to get efficient effect with single herb, but by taking combination of different extracts can be possible to increase efficiency of formulation. Based on the results, F1 formulation was more stable than F2 formulation; it was confirmed by evaluation study. So, the further *in-vitro* studies were completed with F1 formulation and it shows significant antimicrobial activity and UV protection activity. Further detailed study can be done with *in-vivo* pharmacological evaluation.

## REFERENCE

1. Davinder kumar, Gajendra rajora, Om parkash, Himanshu, Mamtaantil, Virender kumar. *Herbal cosmetics an overview. International journal of advanced scientific research* 2016; 1(4):36-41
2. Dr. Sudhanshu kumarmeher, Dr. Banmali Das, Dr. Purnendu panda, Dr. G.C. Bhuyan, Dr.M.M. Rao. *Uses of Withania somnifera (Linn)Dunal (Ashwagandha) in Ayurveda and its pharmacological evidences. Research Journal of Pharmacology and Pharmacodynamics. Jan – Mar 2016; 8(1): 23-29*

3. Boni E. Elewski, *Clinical Diagnosis of Common Scalp Disorders. The society for investigative dermatology*, 2005; 10(3) :190-193
4. Ameya Lanjewar, Soni Maurya, Devendar Sharma, Anchal Gaur. *Review of hair problem and its solution, Journal of Drug Delivery and Therapeutics*. 2020; 10(3-s): 322-329
5. Katlein Franca, Thiago Saldanha Rodrigues, Jennifer Ledon, Jessica Savas, Anna Chacon. *Comprehensive Overview and Treatment Update on Hair Loss. Journal of Cosmetics, Dermatological Sciences and Applications*. August 2013; 3:1-8
6. Rashmi S. Pal., Nikita Saraswat., Pranay Wal., Ankita Wal., Yogendra Pal. *Preparation & Assessment of Poly-Herbal Anti-Dandruff Formulation. The Open Dermatology*. 2020; 14: 22-27
7. Boni E. Elewski, *Clinical Diagnosis of Common Scalp Disorders, The society for investigative dermatology*, 2005; 10(3) :190-193
8. Soni kk and Soni S, *Eclipta alba (L.) An Ethnomedicinal Herbal Plant, traditionally use in Ayurveda. Journal of Horticulture*. 2017;4(3):2376-0354
9. Hans Wolf, Tobias W. Fischer, Ulrike Blume-Peytavi. *The diagnosis and treatment of hair and scalp diseases. Deutsches Arzteblatt International. DtschArztebl Int* 2016; 113: 377-86
10. Ahamed Mueen Mubeen, Venkata Rathina Kumar, Vasuki K. *Pharmacological potential of Excoecaria agallocha L. on Rheumatoid arthritis. International Journal of Experimental Pharmacology*. 2021; 11 (2): 12-18.
11. Mandeep Singh, Shalini Sharma, Sukhbir Lal Khokra, Ram Kumar Sahu, Rajendra Jangde. *Preparation and Evaluation of Herbal Cosmetics Cream. Pharmacology online*. 2011; 2: 1258- 1264
12. Kakali Datta, VishuwajeetRohil, Anu T Singh, Ashok Mukherjee, Beena Bhat. B.Ramesh, *Eclipta alba extract with potential for reversing chemotherapy-induced alopecia: An experimental study in mice, Ann Natl Acad Med Sci (India)*. 2012; 48(3&4): 43-64
13. Laila Che Rose, L, Syahirah Rusdi, N.N, Asari, A, Effendy Abd Wahid, M, Suhaimi. H. *Potential hair growth of crude extract from Hibiscus rosasinensis Linn. Archives of pharmacy practice*. 2020; 11(4): 13-19
14. Biswajit Auddy, Jayaram Hazra, Achintya Mitra, Bruce Abedon, Shibnath Ghosal. *A Standardized withania somnifera Extract Significantly reduces stress- related parameters in chronically stressed Humans: A Double- Blind, Randomized, placebo-controlled study*. 2008; 11: 51- 57
15. Salih Hamad Mohamad Aljabre, Mohamad Akram Randhawa, Naeem Akhtar, Omar Mohamad Abdulrahman Mohamed Alquarshi, Ali Aldossary. *Anti-dermatophyte activity of ether extract of nigella sativa and its active principle, thymoquinone. Journal of Ethnopharmacology* 2005 ; 101 : 116-119

16. Wantida Chaiyana, Pimporn Leelapornpisid, Rungsinee Phongpradist and Kanokwan Kiattisin. *Enhancement of antioxidant and skin moisturizing effects of olive oil by incorporation into microemulsions, Nanomaterials and Nanotechnology.* 2016; 6:1-8
17. Mandeep Singh, Shalini Sharma, Sukhbir Lal Khokra, Ram Kumar Sahu, Rajendra Jangde. *Preparation and Evaluation of Herbal Cosmetics Cream.* *Pharmacology online.* 2011; 2: 1258- 1264
18. Ravi Shankar, Babita Sharangi, Ramesh Gupta, Kamla Pathak. *Formulation and Characterization of Polyherbal Cream for skin Manifestations.* *JAASP Research paper* 2016; 1: 360-366.
19. Sharmila Dusi, J. Saminathan. *Formulation and evaluation of Aloe vera and Dacus Carota Herbal Cream,* *International Journal of Pharmacy Research & Technology* 2020; 10(1)
20. Elizangela Abreu Dutra, Daniella Almanca Goncalves da Costa e Oliveira, Erika Rosa Maria Kedor-Hackmann, Maria Ines, Rocha Miritello Santoro. *Determination of sun protection factor (SPF) of sunscreen by ultraviolet spectrophotometry.* *Brazilian Journal of Pharmaceutical Science,* 2004;40,381-385