

# Formulation of Herbal Hair Gel - A Hair Growth Stimulant

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**Abstract:** Hair loss is a disorder in which the hair falls out from skin areas where they are usually present, such as the scalp and the body. This loss interferes with the many useful biological functions of the hair, including sun protection (mainly to the scalp) and dispersal of sweat gland products. It is a controversial issue as there is no general agreement about what are the main factors that cause loss of hair. It is a universal problem having affected both sexes of all races to different extents for as long as humankind has existed. To overcome baldness, various methods are carried out, one of which is the innovation of synthetic hair growth formula and herbal products.

**Keywords:** hair fall, herbal gel, hair growth, herbal formulation, gender baldness

## 1. Introduction

Hair loss is a disorder in which the hair falls out from skin areas where they are usually present, such as the scalp and the body. This loss interferes with the many useful biological functions of the hair, including sun protection (mainly to the scalp) and dispersal of sweat gland products. As hair cover to the scalp has psychological importance in our society, patients with hair loss suffer tremendously. The most common hair disorder is termed as alopecia which is frequently used to express the patterned loss of scalp hair in genetically vulnerable men and women. In mammals, hair plays a vital role in thermal insulation and for social and sexual communication, both visually and as a means for dispersing scents secreted by skin glands. Humans are relatively hairless compared to other mammals.

Though hair loss (alopecia) is not a debilitating or life-threatening sickness, the very thought of becoming bald can lead to emotional stress and traumatic experience for those who suffer from premature or excessive hair loss. It is a controversial issue as there is no general agreement about what are the main factors that cause loss of hair. It is a universal problem having affected both sexes of all races to different extents for as long as humankind has existed (Libeccoetal., 2004).

Angiogenesis (through endogenous substances), androgen antagonism, vasodilation through potassium channel opening 5 - alpha reductase inhibition and modulation of hair cycle are the major non - surgical therapeutic strategies for hair growth promotion. Minoxidil (useful in both male and female pattern baldness) and Finasteride (useful in male pattern baldness) are two US FDA - approved synthetic drugs finding concomitant use for treatment of androgenic alopecia, but their side effects have reduced their usage

(Prince *et al.*, 1999) The side effects associated with the use of these synthetic compounds include erythema, scaling, pruritus, gynaecomastia, dermatitis, itching or skin rash.

To overcome baldness, various methods are carried out, one of which is the innovation of synthetic hair growth formula and herbal products. The use of these types of material has induced the production of multiple drugs, so in the market today there are many hair growth drugs.

A gel is a preparation that is easier to formulate than other semisolid preparations and has good dispersion. The gel is also easy to wash and not sticky and does not cause irritation and allergies. This sums up the rationale for the formulation an Herbal hair Gel – a Hair Growth Stimulant.

## 2. Materials and Methods

### Collection and Preparation of plant extraction

The plant sample was collected from Forest Genetic Resources Tree Park, Kolapakkam. The fresh whole plant of *Merramia tridentate* was washed with distilled water and dried under shade at room temperature. It was then coarsely grounded and passed through sieve no 40 and stored in an airtight container for further use. Aqueous extraction of the coarsely powdered plant materials of *Merramia tridentate* was sterilized at 15 lb. pressure and the extract was filtered through Whatman filter paper (No.1), preserved in airtight containers and kept at 4°C until further use.

### Preliminary phytochemical analysis

Preliminary analysis was carried out on the plant extract of *Merramia tridentate* to identify the useful constituents like alkaloids, flavonoids, saponins, tannins, phenols and terpenoids using standard methods.

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### Preparation of formulation

Three different herbal hair gel formulations were prepared by simple gel formulation preparation method with carbopol gel base. Carbopol 2gms and measured quantity of extracts was dispersed in 80 ml of distilled water and mixed by stirring continuously in a magnetic stirrer at 800 rpm for 1 hour with all the ingredients under continuous stirring. The mixture was neutralized by adding triethanolamine. Mixing was continued until a transparent gel was formed.

The aqueous extract of *Merramia tridentate* in various concentrations as shown in Table 1 were incorporated in the carbopol base gel and prepared herbal hair gel formulations.

**Table 1**

Formulation	HGF 1	HGF 2	HGF 3
Herbal Extract (%)	15	30	45
Carbopol 934 (gm)	2	2	2
PVP (mg)	5	5	5
Methyl paraben sodium (mg)	75	75	75
Glycerine (ml)	3	3	3
PEG (ml)	6.25	6.25	6.25
Triethanolamine (ml)	1.5	1.5	1.5

### Animal study

Healthy C57 / BL6 mice 25 - 30 gms of either sex were used for hair growth promoting activity. The experimental protocol was approved by the Ethical Committee.

### Treatment for hair growth activity in vivo

Fifteen C57/BL6 mice were divided into five groups of 3 animals in each group. Hairs from 3 cm<sup>2</sup> area at the dorsal portion of all the mice were shaved using electric shavers and applied with marketed hair remover to completely remove hair. Group 1 served as a negative control was applied with simple gel where there was no drug treatment. Group 2 was topically applied with 2 % minoxidil over the shaved area as positive control. Group 3 was topically applied with 15 % w/w gel formulation of HGF 1. Group 4 was topically applied with 30 % w/w gel formulation of HGF 2. Group 5 was topically applied with 45 % w/w gel formulation of HGF 3 respectively. All the gel and standard drug were applied once in a day. The treatment was continued for 30 days and hair growth pattern was observed and tabulated. Skin biopsies were taken on the 30<sup>th</sup> day for follicular observation. Increase in thickness and presence of the follicles in the subcutis layer were taken as evidence for transition of follicles from telogen to anagen phase of hair growth.

### Qualitative hair growth study

Qualitative hair growth analysis was undertaken by visual observation of two parameters: hair growth initiation time (i. e., minimum time to initiate hair growth on denuded skin region) and hair growth completion time (i. e., minimum time taken to complete cover the denuded skin region with new hair).

### Quantitative hair growth study

Hair was plucked randomly using sterile forceps from the shaved dorsal area of mice on 10<sup>th</sup>, 20<sup>th</sup> and 30<sup>th</sup> day of treatment. Hair length was measured and the results were recorded as mean length  $\pm$  SEM of 25 hairs.

### Histological studies

One mouse from each group was authenticated after 30 days of treatment. Skin biopsies were obtained from the shaved portion and preserved in 10 % formalin. Sections of tissues were implanted in paraffin wax and sectioned into a thickness of 10  $\mu$ m. The sectioned tissues were stained with haematoxylin and eosin and the follicular phases of hairs were examined under microscope with an ocular micrometre and also the number of hair follicles per mm area of skin, and percentage ratio of hair follicles in different cyclic phases, like anagen (growth phase) and telogen (resting phase) was determined microscopically.

### Hair follicle counting

Digital photomicrographs were taken from representative areas of slides at a fixed magnification of 100X. All images were cropped in a fixed area with a width of 1500  $\mu$ m and then counted hair follicles in deep subcutis manually.

### Statistical analysis

Statistical analysis of the data was carried out by one way ANOVA in respect of the test and control groups and followed by Dunnett's test. Differences between data were considered highly significant  $P < 0.05$ . The data are reported as mean  $\pm$  SEM.

## 3. Results

### Primary dermal irritation study

The total scores for skin irritation in terms of erythema and oedema was calculated after 12, 24, 48 and 72 hours according to OECD scoring system. Results revealed that the developed herbal gel formulation did not cause any erythema or oedema on the intact rabbit skin when observed for 72 hours. The Primary Dermal Irritation Index (PDII) of the formulation was zero; therefore, according to OECD guidelines the formulation can be classified as non - irritant to the mice skin.

### Preliminary phytochemical analysis

Preliminary phytochemical studies of *Merramia tridentate* reveal the presence of alkaloids, carbohydrates, cardiac glycosides, tannins, saponins, phenols and flavonoids (Table 1).

### Hair growth promoting activity of HGF gel formulation in telogenic C57/BL6 mice

To measure the hair growth promoting activity of HGF gel formulation *in vivo* telogenic C57/BL6 mice were showed one day before topical application of gel formulations. The skin colour of mice in the telogen phase was pink and became dark along with anagen initiation. Since the active growth of hair follicles and black pigmentation occur in C57/ BL6 mice during the anagen phase, the hair growth activity of HGF gel formulation was evaluated by observing the skin colour.

More blacken skin areas were observed in HGF 1, HGF 2 and HGF 3 gel formulation treated groups at 10 days, compared to the control and 2 % minoxidil groups. At 30 days, dorsal skin hairs were fully recovered in HGF 3 gel formulation treated groups as Minoxidol done. These results

suggest that HGF 3 gel formulation induces early telogen to anagen conversion of hair follicles (Figure 1).

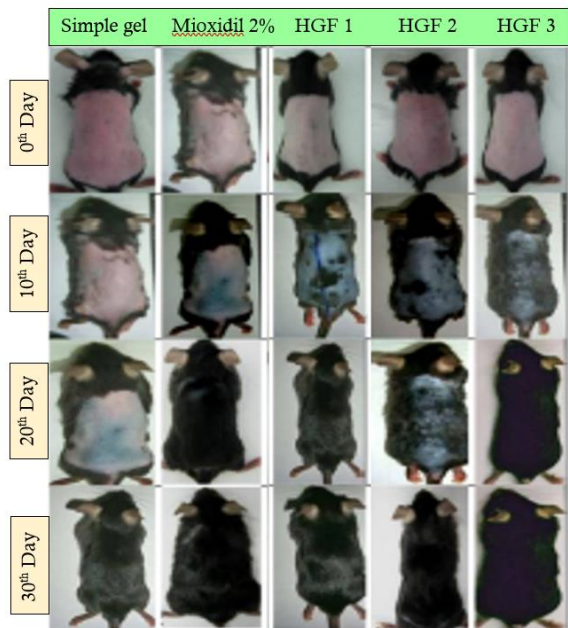


Figure 2: Effect of HGF gel formulation on hair growth in mice model

Table 5: Effect of HGF Gel Formulation on Hair Length

Treatments	Mean length of hair in mm		
	10 <sup>th</sup> Day	20 <sup>th</sup> Day	30 <sup>th</sup> Day
Negative control – simple gel	0.0 ± 0.0	0.3 ± 0.0	1.2 ± 0.20
Positive control - 2 % minoxidil	0.6 ± 0.21	1.4 ± 0.24	2.4 ± 0.20 ***
HGF 1	0.1 ± 0.0	1.4 ± 0.24	1.6 ± 0.24
HGF 2	0.6 ± 0.20	1.6 ± 0.24	2.8 ± 0.20 ***
HGF 3	0.8 ± 0.20	2.6 ± 0.24	3.6 ± 0.24 ***

\*\*\* P < 0.05 significant when compared to control

**Histological studies**

A considerable difference in cyclic phases of hair growth was observed in groups treated with minoxidil, HGF gel formulations. An increase in the number and size of hair follicles has been designated as an indicator for the transition of hair growth from the telogen to anagen phase. To examine the progression of hair follicles in the hair cycle, hematoxylin - eosin staining was performed, since an increase in size and number of hair follicles can be observed in the deep subcutis. The photomicrographs obtained indicated that control (simple gel) treated animals had less percentage of anagenic hair follicles (47 %) while the HGF 3 (73.6 %), minoxidil (71 %) and HGF 2 (69.4 %) treated animals showed maximum percentage of anagenic hair follicles and higher follicle density (Figure 3). HGF 1 gel formulations treated group showed the anagenic follicles percentage of 55.1 respectively and were significant as compared to control but was not as much significant as HGF 3, HGF 2 and minoxidil treated groups (Table 6).

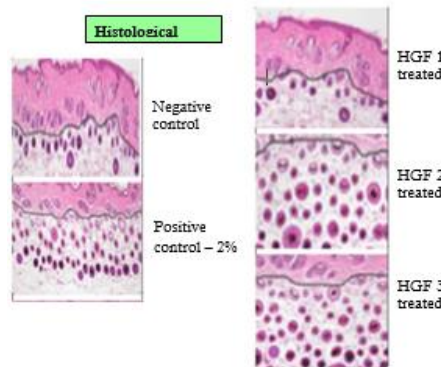


Figure 3: Hair follicle growth in transverse section of the dorsal skin of C57/BL6 micetreated with HGF gel formulation

Table 6: Effect of HGF Gel Formulation on percent of hair follicles after 30 days

Treatments	Telogen	Anagen	T / A Ratio	Percent hair follicles > 0.5 mm in length
Negative control - simple gel	48.3 ± 0.94	47.0 ± 0.72	1.03	28.4 ± 0.92
Positive control - 2 % minoxidil	25.2 ± 0.17	69.4 ± 0.77	0.36	46.3 ± 1.03***
HGF 1	26.7 ± 0.81	65.1 ± 0.32	0.41	39.6 ± 0.69
HGF 2	23.1 ± 0.36	71.0 ± 0.43	0.33	45.9 ± 1.11***
HGF 3	22.6 ± 0.44	73.6 ± 0.57	0.31	46.9 ± 0.91***

\*\*\* P < 0.05 significant when compared to control

**4. Conclusion**

In conclusion, the effect of HGF gel formulation on the qualitative hair growth and length was found to be more significant as compared to standard and control group treated animals. The quantitative effect of HGF 3 gel formulation definitely promotes hair growth by inducing hair follicles in the anagen phase. The percentage of anagen induction HGF 3 gel formulation and minoxidil were comparable. On the basis of similarities observed between the minoxidil and the HGF 3 studies, it is expected that HGF 3 gel formulation showed the best hair growth activity compared with minoxidil. In future this gel formulation will be tested on human volunteers for its hair growth activity.

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