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Formulation and Evaluation of Herbal Hair Dye: An Ecofriendly Process

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Abstract:

The increase in environmental and health hazards in the manufacture of dyes and its use throughout world is a major concern. This work was made possible while investigating the alternative to the synthetic and semi synthetic dyes. The composition of 100% herbal hair dye consist of 30% aqueous extract of *Cymphomandra betacea* containing 35% flavanoid and 10% tannins, 30% aqueous extract of *Tagetes erecta* containing 20% carotenoid and 40% *Aloe vera* gel, as natural mordant, containing 0.3% polysaccharide and 98.5% water. The present study is also directed to synergistic hair dye compositions containing aqueous extract of *Cymphomandra betacea* and aqueous extract of *Tagetes erecta* blended with *Aloe vera* gel, a natural mordant. Further, this study is directed to a method for coloring hair, by applying an effective amount of the herbal hair dye composition with distilled water as an acceptable carrier. The herbal hair dye shows permanent dying to the applied regions of human hair without causing any hair damage or hair loss or skin irritation when compared to the synthetic and semi synthetic dyes. The active constituent also prevents the hair from damage caused by photoreaction and pollution. This formulation proves to be a vital alternative for the synthetic and semi synthetic dyes.

Key words: *Cymphomandra betaceae, Tagetes erecta, Aloe vera, synthetic dye and flavanoids.*

1. Introduction

In comparison to natural hair dyes, synthetic hair dyes are reported to cause skin and other skin related diseases. The manufacturing process is hazardous to health of the people involved in the process and its applications leads to environmental pollution and also causes potential side effects to the consumers of the product. The fear of side effects from the synthetic dyes has limited its use by health conscious customers throughout the world and has to overcome various regulatory barriers before it reaches its destination.

A dye can generally be described as a colored substance that has an affinity to the fiber, fur or hair. The dye is generally applied as aqueous solution, and may require a mordant to improve the fastness of the dye on the fiber, fur or hair. Natural dyes also referred as mordant dyes. Different mordant will give different hue color with the same dye. A mordant is thus an agent which allows a reaction to occur between the dye and the fiber, hair or fur [1].

Hair dyes include dyes modifiers, antioxidants, alkalizers, soaps, ammonia, wetting agents, fragrance, and a variety of other chemicals used in small amounts that impart special qualities to hair such as softening the texture or give a desired action to the dye. The chemicals that are normally used in the dye are amino compounds (4-amino-2-hydroxytoluene and m-Aminophenol). Metal oxides, such as titanium dioxide and iron oxide, are also often used as colorants in the process. Colorants are classified as being temporary or permanent. In temporary coloring the color can be washed from hair easily. Permanent coloring of hair involves addition of aromatic diamine or hydric phenols or polycompounds such as para phenylenediamine in the formulation. Continuous usage of such compounds containing dye on natural hair causes so many side effects such as skin irritation, erythrema, loss or damage of hair and skin cancer. Other chemicals used in hair dyes act as modifiers, which stabilize the dye pigments or otherwise act to modify the shade.

Antioxidants protect the dye from oxidizing with air. Most commonly used is sodium sulfite. Alkalizers are added to alter the pH of the dye formula, because the dye works best in a highly alkaline milieu. Ammonium hydroxide is a common alkalizer. Apart from these basic chemicals, many other chemicals are used to impart special qualities to a manufacturer's formulala. In order to color human hair by oxidative dye technology, the hair is generally treated with a mixture

of oxidative hair coloring agents and an oxidizing agent. Hydrogen peroxide is the most commonly used oxidizing agent. However, in addition to oxidizing the oxidative coloring agents, hydrogen peroxide treatment of the hair can also solubilise and decolorize the colored melanin component in the hair, which can lead to undesirable hair qualities, such as brittleness and hair damage.

Composition of herbal dyes and hair coloring mordant can be used to deliver a variety of hair colors to the hair. However, substantial improvement is needed in the areas of color saturation, color development, precise initial color consistency, improved wash fastness, improved hair conditioning without causing hair damage and skin irritation. Because of the manufacturing hazards, environmental pollution, its side and toxic effects there is a vital need for an alternative to the existing black dye. These limitations of the chemically derived dye can only be overcome by replacing the constituents in the composition, by non toxic ingredients derived from herbal resources[2]. The black dye produced from herbal resources may be used in wide variety of context including hair color products. At this juncture, there is enormous need for a method to increase the yield of such dyes from herbal products.

2.Materials and Methods:

2.1. Plant source:

(a). *Cymphomandra betacea*: Commonly known as Tamarillo or Tree tomato belongs to Solanaceae family. The fruit contains flavanoids, pectin, gums and mucilage. It is also rich in vitamins, minerals and antioxidants.

(b). *Tagetes erecta:* Fresh and dried flowers of *Tagetes erecta* belongs to Compositae family. The flower consists of carotenoids consisting of leutin, zeaxanthin, neoxanthin plus violaxanthin, β-carotene, lycopene, α-cryptoxanthin, phytoene and phytofluene. It is commonly known as Marigold flower. Flowers are edible and also used as coloring agent and condiment. A yellow dye obtained from the flower can be used as a saffron substitute for coloring and flavoring foods[3]. This probably refers to the use of the flowers as an edible dye. This flower is medicinally used.

(c). *Aloe vera :* Aloes is the dried juice of the leaves of *Aloe vera* belonging to the family Liliaceae. Anthraquinones (aloin, aloe-emodin), resins, tannins and polysaccharides are the major chemical constituents; Aloe vera gel consists primarily of water and polysaccharides (pectins, hemicelluloses, and glucomannan, acemannan, and mannose derivatives). It also contains amino acids, lipids, sterols (lupeol, campesterol) and enzymes. Clear gel has a dramatic ability to heal wounds, ulcers and burns, application of a protective coat on the affected area speed up the rate of healing[4].

2.2. Extraction and formulation of dye:

The extraction and formulation method of dyes from the above plants are stated in flow chart-1. The method of obtaining dye from plant material is environmentally friendly since water is used as solvent.

2.3. Standard hair dyes used for the study:

(a).Standard I :

Synthetic hair dye containing para-phenylenediamine marketed as Permanent hair dye (Natural black).

(b). Standard II

Semi-synthetic hair dye containing Amla, Bhrinraj, Methi, Henna, Hibuscus and Para-phenylenediamine, a marketed as Indica herbal hair color.

2.4 sample hair dyes used for the study:

(a). Sample –I

- 10%Aqueous extract of *Cymphomandra betaceae* (Fruits) containing 35% flavanoid and 10% tannins.
- 20% Aqueous extract of *Tagetes erecta* (Flower) containing 20% carotenoid.
- 70% *Aloe vera* gel Leaf) containing 0.3% polysaccharide and 98.5% water.

Step 1: Collection of raw materials from medicinal plant garden and authenticated by the Botanist.

↓

Step 2: Evaluation of purity and quality of raw materials by physical, chemical, analytical and microscopical techniques.

↓

Step 3 : 100 g of the dried *Tagetes erecta* powder was treated with 100 ml of distilled water and was heated for one hour at 100^0C. The extract was filtered to obtain a yellowish brown dye solution. The optical density was recorded. The extracted dye solution was evaporated to $1/10^{th}$ volume under controlled temperature. The concentrated extract was subjected to freeze drying and the final yield was found to be 33 g.

↓

Step 4 : Fully ripped *Cymphomandra betacea* fruits were cleaned thoroughly.
The fruits were cut in to small pieces and 100g of the sliced fruit was cooked with 500ml of distilled water for 2hrs by maintaining the temperature at 60˚C.After 2hrs the whole mass was filtered to obtain a ruby-red colored aqueous extract. 500ml of the obtained aqueous extract was concentrated for 1hr at 60˚C to obtain 100ml of aqueous extract. The concentrated extract was subjected to freeze drying and the final yield was found to be 36 g.

↓

Step 5 : Fresh leaves of *Aloe vera* were collected washed thoroughly and the outer green surface (pericyclic fibers) was peeled off and the inner "fillers", white mass was collected by cooping. 100 g of the collected material was crushed to a semi-solid consistency which was subjected to filtration. The filtrate was subjected to evaporation to $1/10^{TH}$ of its volume under controlled temperature (60˚C) and the final yield was found to be 45 g.

↓

Step 6 : Quantitative determination of the active constituents namely flavanoids, tannins, carotenoid and polysaccharides by using HPTLC/ HPLC technique.

↓

Step Step 7 : Formulation of natural dye with mordant: 30g aqueous extract of *Cymphomandra betacea* containing 35 % flavanoid and 10% tannins , 30g Aqueous extract of *Tagetes erecta* containing 20% carotenoid and 40g *Aloe vera* gel, as natural mordant, containing 0.3% polysaccharide and 98.5% water were mixed together.

Flow Chart: 1

Table 1: Comparison of coloring effect of herbal hair dye with marketed brands on human hair

Name of the hair dye with constituent	Coloring effect & fastness property	Duration of exposure	Physical appearance
1. Natural hair dyed marigold flower dye + Aloe vera juice	4/5 to 5	30minutes	No damage
2. Semi synthetic hair dye Indigo herbal hair color. (Henna, Amla, Bringraj, Methi, Hibiscus, resorcinol, Paraphenylene diamine and H_2O_2)	4/5 to 5	15minutes	Cortex damaged
3. Synthetic hair dye Godrej permanent hair dye (Paraphenylene diamine)	4/5 to 5	15minutes	Cortex damaged

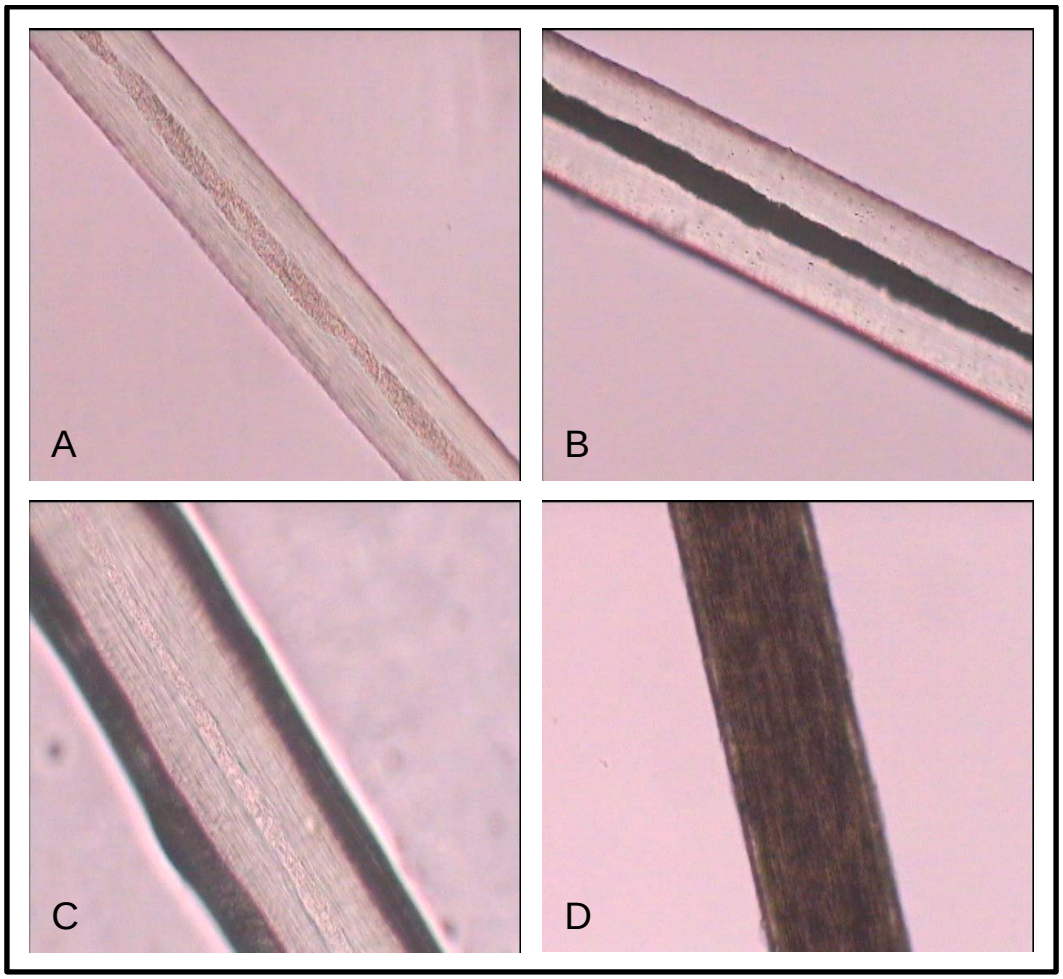

Figure 1: Coloring effect of herbal hair dye with natural mordant

(A) - Microscopic structure of human grey hair; (B) - Microscopic features of medulla region of human grey hair stained with herbal hair dye for 30 mnts; (C) - Microscopic features of cortex region of human grey hair stained with herbal hair dye after 30 mnts; (D) - Microscopic features of human grey hair treated with herbal hair dye after 3 treatments.

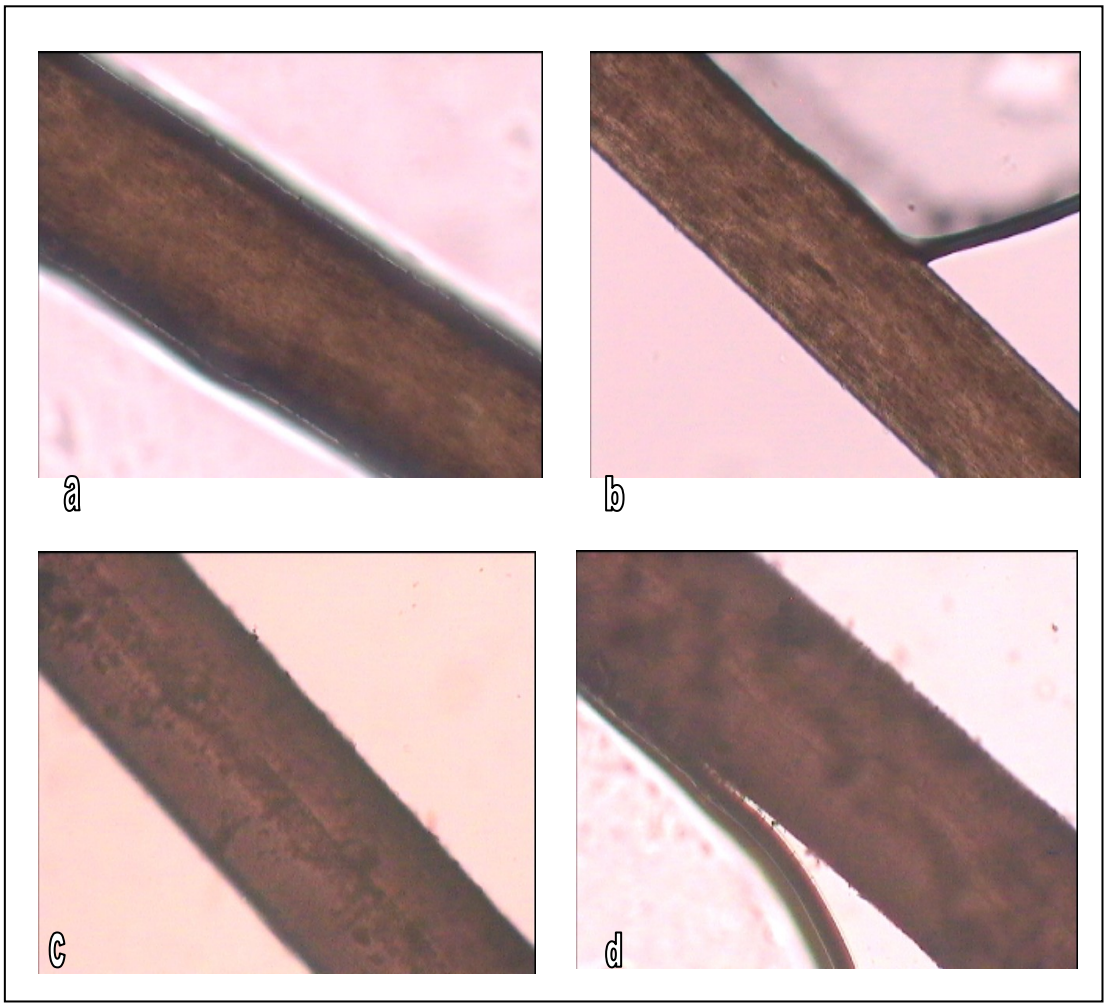

Figure 2: Comparison of coloring effect of herbal hair dye with marketed brands on human hair

(a) - Microscopic structure of human grey hair treated with synthetic hair dye for 15 mnts; **(b)** - Microscopic features of grey hair treated with synthetic hair dye showing damaged cortex after 2 treatments; **(c)** - Microscopic structure of human grey hair treated with semi synthetic hair dye for 15 mnts; **(d)** - Microscopic features of grey hair treated with semi synthetic hair dye showing damaged cortex after 2 treatments.

(b). Sample –II

- 30%Aqueous extract of *Cymphomandra betaceae* (Fruits) containing 35% flavanoid and 10% tannins.
- 30% Aqueous extract of *Tagetes erecta* (Flower) containing 20% carotenoid.
- 40% *Aloe vera* gel Leaf) containing 0.3% polysaccharide and 98.5% water.

(c). Sample –III

- 40%Aqueous extract of *Cymphomandra betaceae* (Fruits) containing 35% flavanoid and 10% tannins
- 40% Aqueous extract of *Tagetes erecta* (Flower) containing 20% carotenoid
- 20% Gel of *Aloe vera* (Leaf) containing 0.3% polysaccharide and 98.5% water.

2.5 Collection of hair sample:

Human grey hair was collected from male and female volunteers.

2.6. Physichemical evaluation of the extracts and dye:

The extracts and the formulated dye was subjected to various physiochemical analysis namely Microbial content,moisture content ,pesticide residue,heavy metals,ash value , extractive value and qualitative and quantitative phytochemical analysi [5].

2.7. Study on the dying effect:

The formulated dye was applied over grey hair sample and the fastness property and dying effect was observed using system microscope [6].The effect was recorded and the results are shown in Table 1 and Figure 1 &2.

2.8. Skin irritation study:

Draize modified scoring technique was used to evaluate the skin irritation at 24hrs for seven days, after the application of the dyes the skin irritation studies were performed on healthy rabbits [7]. The dorsal surface ($50Cm^2$) of the rabbits was cleared and the hair was removed by shaving. The skin was cleansed with rectified spirit. Natural dye was applied over the skin. The skin was observed for skin irritation, erythrema formation and edema formation for 6 days.

3. Results and Discussion:

3.1. *Aloe vera* gel

Microbial content:

The test for Salmonella spp and Staphylococcus spp. in Aloe vera gel was negative and the acceptable maximum limits of microorganisms are as follows. For external use: Aerobic bacteria—not more than 102/ml; Fungi—not more than 102/ml; enterobacteria and Gram-negative bacteria—not more than 101/ml; Staphylococcus spp.—0/ml.

Moisture Content:

98.5% water.

Pesticide residues:

Established in accordance with WHO guidelines on quality control methods for medicinal plants and guidelines on predicting dietary intake of pesticide residues and the pesticide residual content was within the limit.

Heavy metals:

Lead levels were found to be 0.5 mg/kg and no traces of cadmium, Permited lead and cadmium levels are not more than 10 and 0.3mg/kg, respectively.

Total ash:

4%, Accordance with WHO guideline requirements

Alcohol-soluble residue:

8 %, Accordance with WHO guideline requirements

Water-soluble extracts:

43%, Accordance with WHO guidelines requirements

Chemical assays:

Carbohydrates 0.3%, water 98.5% Polysaccharide composition analysis by gas–liquid chromatography.

3.2.Aqueous extract of *Tagetes erecta*

Chemical assays:

20% carotenoid was estimated by HPLC analyses.

Tagetes erecta extract was saponified and analyzed for carotenoid composition. HPLC analyses were performed on two normal-phase columns (β-Cyclobond and silica) and on a C_{30} reversed-phase column. The extract contained 93% utilizable pigments (detected at 450 nm), consisting of all-trans and cis isomers of zeaxanthin (5%), all-trans and cis isomers of lutein, and lutein esters (88%). All were identified by chromatographic retention, UV−visible spectra, and positive ion electrospray mass spectrometry in comparison to authentic standards. This compositional determination is important for the application of marigold extract in cosmetic colorant because it contains more biologically useful lutein compounds[8].

3.3. Aqueous extract of *Cymphomandra betacea*

Chemical assays:

35% flavanoid and 10% tannins by Reverse phase HPLC and GC-MS technique [8]. Reversed phase high performance liquid chromatography was used for the analysis of the plant extracts. Gas chromatography–mass spectrometry method was also used for identification of polyphenolic compounds after silylation. The most abundant poly phenolic compound was Ellagitannins, ferulic acid and (+)-Catechin and (−)-epicatechin were the main flavonoids identified. This compositional determination is important for the application of tree tomato extract in food and cosmetic industry as colorant anti

oxidant and antimicrobial agent, because it contains more biologically useful Ellagitannins and Catechin compounds.

3.4. Study on the effect of successive application of selected combination of herbal dye with natural mordant.

For dying optimized conditions used were: wave length: 410nm, dye extraction time:30min, medium of extraction: aqueous, dye material concentration:10%, dying time: 30 minutes and pH of dying solution: 7.0.

Data pertaining to fastness properties and mordant concentration of sample-I had good coloring effect (3/5 to 4),where as it was very poor when subjected to three washing with shampoo.

The fastness property and dying effect was observed to be very good even after three washings with shampoo. (4/5 to 5), in Sample-II. The fastness property and dying effect was observed to be good (4/5 to 5) even after three washing with shampoo in Sample-III but the sample stained the skin.

3.5. Comparative evaluation of the formulation with marketed brands

The coloring effect, fastness and effect on the skin structure of the selected herbal hair dye composition Sample-II was compared with Standard-I, Synthetic hair dye containing para-phenylenediamine and Standard-II Amla, Bhrinraj, Methi, Henna, Hibuscus and para-phenylenediamine. The above three were applied to human hair samples, after 30min the hair was washed thrice with shampoo and observed microscopically.

The fastness property and dying effect was observed to be good even after washing (4/5 to5) in Standard-I synthetic hair dye containing para-phenylenediamine but it stained the skin and damaged the hair. Rupture of cortex region of the hair was observed.

The effect was also observed to be good in Standard-II, containing Amla, Bhrinraj, Methi, Henna, Hibuscus and para-phenylenediamine but treatment time was prolonged to 1 hour and it also damaged the hair. Slight rapture of cortex region of the human hair was observed.

The fastness property and dying effect was observed to be very good even after washing (4/5 to5) in Sample-II and it did not stain the skin or damaged the hair. The results are shown in Table 1 , Figure I A, I B, I C & I D and Figure 2 A, 2 B, 2 C & 2 D.

3.6. Skin sensitivity test

The herbal dye samples were subjected to skin irritation study since the herbal hair dyes should not produce any skin sensitization or irritation when applied on hair. Draize modified scoring technique was used to calculate the skin irritation at 24hrs for seven days, after the application of the dyes the skin irritation studies were performed on healthy rabbits. The dorsal surface ($50cm^2$) of the rabbits was cleared and the hair was removed by shaving. The skin was cleansed with rectified spirit. Herbal hair dye samples was applied over the skin. The skin was observed for skin irritation, erythrema formation and edema formation for 6 days and the result showed no skin irritation or erythema or edema on the skin treated with sample dye I and II when compared sample III.

4. Summary and Conclusion:

The present study is directed to a herbal hair dye composition for dying human hair comprising a mixture of plant extract and plant essential oils which include 30% aqueous extract of *Cymphomandra betaceae*, 30% Aqueous extract of *Tagetes erecta* and 40% *Aloe vera* gel, as natural mordant. Efficacy data shows that this embodiment is highly effective, exhibited fast dying effect without causing hair damage and skin irritation at this composition . A mixture of plant extracts containing higher percentage (>30%) aqueous extract of *Cymphomandra betaceae*, (>30) aqueous extract of *Tagetes erecta* (>40%) *Aloe vera* gel, exhibited fast dying property but causing hair damage and skin irritation, which is not a desirable effect for a novel herbal hair dye.

The herbal hair dye composition consists of 30 % aqueous extract of *Cymphomandra betaceae* containing 35% flavanoid and 10% tannins 30% Aqueous extract of *Tagetes erecta* containing 20% carotenoid and 40% *Aloe vera* gel, as natural mordant, containing 0.3% polysaccharide and 98.5% water. The plant extracts mentioned above should contain the specified percentage of active constituents for producing a prolonged and effective coloring of human hair.

Efficacy data shows that this composition is highly effective, exhibited fast and prolonged coloring effect on human hair. A mixture of plant extracts containing lower percentage of active constituents, i.e. <35% flavanoid, <10% tannins, <20% carotenoid and <0.3% polysaccharide and < 98.5% water fails to produce the fast and prolonged coloring effect.

The herbal hair dye formulation also relates to synergistic compositions comprising of carotenoids, flavonoids and tannins which contributes to the coloring effect on human hair and protects human hair and skin from photodamage. The fruits of *Cyphomadra betacea* with high content of carotenoids, vitamins, flavonoids and anthocyanins is a good source of raw material for natural dye [9] . Aloe vera gel containing polysaccharide and water is effective as natural mordant and also nourishes the dehydrated aged skin and hair and photodamge [10].

The herbal dye extracts and natural mordant extracts were subjected to freeze drying process. The substances can be stored at room temperature without refrigeration. Preservation is possible because the greatly reduced water content inhibits the action of micro organism and enzymes.

The coloring activities of the herbal hair dye were comparable with that of the commercially available synthetic and semi synthetic hair dye.

The significances of the formulated herbal hair dye :

- The active constituents present in the hair dye are non toxic and devoid of any side effects.
- The mordant used is of natural origin and is derived from the *Aloe vera* gel with which the active dye is mixed to facilitate its application and enhance penetration of the dye in to cortex region of the human hair.
- It is prepared from100% water soluble plant ingredients; hence it is free from any obnoxious odor.
- The raw materials used and the final product is totally biodegradable.
- The extracted pigments entirely impart its color to the hair and do not react with it.
- The pigment penetrates deep in to medulla region with out rupturing the cortex.
- The formulation contains 100% water soluble herbal extracts.
- The solvent and carrier used in the whole preparation is only distilled water.
- Very simple to use and a highly economical hair dying process.
- The product is stable at room temperature.
- The composition and mode of preparation is environmental friendly.

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