Pharmacogn. Res., 2024; 16(2):294-299.
https://www.phcogres.com

Original Article

The Herbal Hue: Formulation and Evaluation of Herbal Hair Dye

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ABSTRACT

Background: The plant henna has a natural coloring pigment that is used to dye skin, fingernails, hair, and textiles like leather, silk, and wool. It may also be utilized for impermanent body art. An active ingredient in henna plants identified as "lawsone" is an orange-reddish dye that combines to the protein keratin of human skin to color it. **Materials and Methods:** Herbal hair dye formulation is prepared to get a darker black natural hair dye that is free from any harmful chemicals. The main constituent used is henna which gives an orangish colour to hair. Different other natural compounds were mixed to get a darker color as well as to protect the hair. The formulation also prevents hair from premature greying, hair fall, cleaning of dandruff, etc. The formulation promotes hair growth and helps to maintain a healthy environment for hair to grow. The main four constituents used for coloring were Henna (50%), Indigo (30%) Coffee (4%) and Amla (4%), which also promote hair growth. Other constituents such as Shikakai (2%), Reetha (2%), Brahmi (2%), Red Sandalwood (2%), Hibiscus (2%), Bhringraj (2%) were mixed to get natural black color to hair. **Results:** The herbal hair dye pack was formulated, evaluated and its application on the blonde hair was carried out. The two formulations A and B when applied to blonde hair string the color obtained are similar to natural black hair color. **Conclusion:** However, further studies on the application of formulated hair dye packs on different shades of human hair are required to determine its full range of potential advantages.

Keywords: Herbal hair dye, Henna, Phytochemical evaluation, Molisch test, Hair dye formulation.

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Received: 13-10-2023;
Revised: 25-11-2023;
Accepted: 18-12-2023.

INTRODUCTION

Hair coloring or dying are methods of altering the color of one's hair. The main motivation for this is to recolor grey and white hair or to bring back the original color of hair that has been discolored by sun exposure or cosmetics. Herbal dyes have all of the benefits of natural substances. These formulations, in addition to working as a hair color, function as a hair growth stimulant and nourishment due to the right amalgamation of herbs.

Henna

The plant henna has a natural coloring pigment used to dye skin, fingernails, hair, and textiles like leather, silk, and wool. It may also be used for impermanent body art. An active ingredient in henna plants identified as "lawsone" is an orange-reddish dye that attaches to the protein keratin of human skin to color it. Henna, scientifically identified as *Lawsonia inermis*, has a long-established history of application owing to its natural dyeing properties. The chemical composition of henna comprises a cluster of organic compounds referred to as lawsone or hennotannic acid, which is accountable for the dyeing effect. The henna plant's leaves are the main source of these chemical constituents.

The process of dyeing hair with henna involves a chemical reaction between lawsone and hair proteins. Henna powder is typically mixed with a mildly acidic liquid, like lemon juice or tea, to create a paste. This acidic environment helps activate the dye molecules in henna.[1] When applied to the hair, the lawsone molecules in the henna paste penetrate the hair shaft by permeating the outer protective layer, known as the cuticle.[2] Once inside the hair shaft, the lawsone attaches to proteins, specifically keratin, present in the hair. This binding forms a durable and impermanent dye complex.[3] After lawsone binds to proteins, an oxidation process begins. Exposure to air triggers a chemical reaction which enhances the color of the dye. This oxidation typically takes several hours to fully develop.[4] The mechanism by which henna stains hair distinguishes it from chemical hair dyes, which penetrate the hair cortex and permanently alter its color. Henna acts as a surface-level dye that coats the hair shaft, resulting in a semi-permanent color that gradually fades as the hair grows.[5] It is important to note that the effectiveness and color outcomes of henna can vary based on factors such as the quality of the henna powder, the natural

DOI: 10.5530/pres.16.2.37

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color of the hair, and individual differences in hair chemistry.[6] The henna shade ranges from intense crimson to pale orange, varying according to the nature of the leaves and the henna paste combination applied.[7]

Variation of Henna

There are typically three types of henna available.

Natural Henna

When natural henna (*Lawsonia inermis*) is first applied, it leaves skin with a profound brown-red stain that lasts for around a week. It is derived from the Lawsonia plant's leaves.[8]

Neutral Henna

Neutral henna, also known as *Cassia obovata*, is not the same as pure henna powder. Actually, it's a different plant altogether called *Senna italica*. Natural and neutral henna plants have different properties and molecular consistency, despite their appearance being comparable. In their research findings, Irwin suggested that neutral henna does not directly alter hair's color.[9]

Black Henna

Indigo, natural henna, and other unlisted chemicals or colors like para-phenylenediamine (PPD) are the ingredients of black henna. Although the primary purpose of this dye is to color fabric whereas some manufacturers are using it to make henna tattoo cones and hair dye.[10]

MATERIALS AND METHODS

Ten essential herbs have been chosen for the manufacture of the Herbal Hair Dye Pack as depicted in Table 1 (Formulation's Composition), powdered versions of all herbs purchased from the local market-authorised retailers. To create a homogenous composition, all the herbs were then blended consistently.[11-14] The blonde hair used in the study were collected from a local barber's shop.

Evaluation: Herbal hair dye

The developed herbal hair dye formulations were examined for several parameters, including organoleptic, physicochemical, phytoconstituents, and rheological features.[15-19]

Organoleptic Evaluation

Organoleptic aspects for a variety of sensory attributes, such as colour, odour, etc., were meticulously recorded. As shown in Table 2, the organoleptic and morphological characteristics of the raw medications and powders, such as colour, odour, texture, and appearance, were independently examined.[20]

Phytochemical Evaluation

Phytochemical testing was done on the formulations of herbal hair dye in order to determine which phytoconstituents were present, including sugars, lipids, carbohydrates, and alkaloids. Several phytoconstituents were detected in the aqueous extract of the resulting herbal hair dye using standard operating procedures and established methodologies. Table 3 highlights the phytochemical screening outcomes.[21]

Molisch's Test

Materials Required

- Herbal hair dye formulations as a sample,
- Distilled water,
- α-Naphthol,
- Absolute Ethanol,
- Sulphuric Acid.

The Molisch test is a chemical method for detecting the presence of carbohydrates in a material. Test solution preparation: Take 1 g of the sample to be tested and dissolve it in a test tube containing 2 mL distilled water test. Pretreatment or extraction procedures according to the specific sample can be performed. Molisch Reagent Preparation: Prepare the Molisch reagent by adding 1 g of α-Naphthol to 100 mL of ethanol. Mix thoroughly to ensure complete dissolution. Perform the Test: Add a few drops of the prepared test solution to a clean test tube. Carefully add 2-3 mL of the Molisch reagent to the test tube and mix gently by swirling. Add strong sulfuric acid slowly and gently down the edges of the test tube to produce a layer over the mixture. Be careful not to combine the layers. Examine the test tube to see if a violet or purple ring forms at the intersection of the two layers. The formation of a violet or purple ring indicates the presence of carbohydrates in the sample. The color's intensity might change based on the proportion of carbohydrates.[22,23]

Volatile Oil Test

Material required

- Herbal hair dye formulations as a sample,
- Sudan (III) reagent,
- Absolute ethanol,
- Distilled Water.

The volatile oil test is a widely used method for analyzing and identifying essential oils. It involves the utilization of an alcoholic solution of Sudan III as a reagent to detect the presence of volatile oils. When Sudan III is added to a sample, it undergoes a characteristic color change or staining in the presence of volatile oils. This reaction is due to Sudan III affinity for the lipid-based

constituents found in essential oils. The intensity of the resulting color provides valuable information about the quantity and quality of volatile oils in the sample. The volatile oil test is a simple and rapid qualitative assessment tool used to determine the presence of essential oils in various natural products, including plants and herbs.[24]

The procedure for conducting the volatile oil test involves the Preparation of Sudan III Solution: Prepare a solution of Sudan III dye in ethanol or a suitable alcohol solvent at a specified concentration.[24] Test Sample Preparation: Obtain the sample suspected to contain volatile oils, such as a plant extract, herbal material, or any substance believed to contain essential oils. Place a small portion of the test sample in a clean test tube or on a spotting plate. Add a few drops of the prepared alcoholic Sudan III solution to the sample. Observation: Observe the sample for any changes in color or staining effects that occur upon the addition of Sudan III solution. The presence of volatile oils typically results in the development of a distinct color, often ranging from orange-red to reddish-brown.[24,25] Interpretation: To ascertain if volatile oils are present in the sample or not, compare the staining or color seen with established standards or references.

Table 1: The formulation's composition.

Sl. No.	Ingredient	Botanical Name	Uses	Formulation A (g)	Formulation B (g)
1	Henna	*Lawsonia inermis*	Hair dyes, hair care products.	25	25
2	Indigo powder	*Indigofera tinctoria*	Improves hair colour. Soothes hair scalp and follicles.	5	15
3	Amla	*Phyllanthus emblicia*	A great conditioner for the hair, gives soft and shiny look.	5	2
4	Coffee	*Coffea arabica*	Stimulate hair follicles. Improves blood circulation to scalp.	5	2
5	Shikakai	*Acacia concinna*	Natural foaming agent gently cleanses the scalp.	2	1
6	Reetha	*Saipindous mukorossi*	Used as a cleanser and remove lice from hair.	2	1
7	Hibiscus	*Hibiscus Rosa-sinensis*	Add volume, treat dandruff, prevent split ends.	1	1
8	Bhringraj	*Eclipta prostrata*	Increases blood circulation to the scalp and roots.	1	1
9	Red Sandalwood	*Pterocarpus santalinus*	Keeps the scalp hydrated, reduce dandruff formation.	2	1
10	Brahmi	*Bacopa monnieri*	Reduce inflammation and hair loss by treating dandruff.	2	1

Table 2: Organoleptic Evaluation.

Sl. No.	Parameter	Formulation A	Formulation B
1	Colour	Brownish Green	Greenish Brown
2	Odour	Characteristic	Characteristic
3	Texture	Fine	Fine
4	Appearance	Powder	Powder

Table 3: Summary of Phytochemical evaluation.

Sl. No.	Parameter	Formulation A	Formulation B
1	Molisch's test	Carbohydrates was present.	Carbohydrates was present.
2	Mayer's test	Alkaloids was present.	Alkaloids was present.
3	Volatile oil test	Volatile oil was present.	Volatile oil was present.

Table 4: Rheological evaluation of herbal hair dye.

Sl. No.	Parameters	Formulation A	Formulation B
1	Bulk density	0.5 g/cm^3	0.5 g/cm^3
2	Tapped Density	1.25 g/cm^3	1.1 g/cm^3
3	Angle of Repose	41.250	38.99
4	%Carr's index	60%	54.54%
5	Housner's ratio	2.5	2.2

Table 5: Patch Test of Herbal hair dye.

Sl. No.	Parameter	Formulation A	Formulation B
1	Swelling	Negative	Negative
2	Redness	Negative	Negative
3	Irritation	Negative	Negative

Table 6: Stability Testing of Herbal Hair Dye.

Sl. No.	Parameters	Formulation A		Formulation B	
		Room Temperature (20°C)	35°C	Room Temperature (20°C)	35°C
1	Color	No change	No change	No change	No change
2	Odor	No change	No change	No change	No change
3	pH	6.7	6.8	6.8	6.9
4	Texture	Fine	Fine	Fine	Fine
5	Smoothness	Smooth	Smooth	Smooth	Smooth

Figure 1: Blonde human hair.

Figure 2: Colour of Formulation B in blonde human hair.

Figure 3: Colour of Formulation A in blonde human hair.

Mayer's Test (For Alkaloid)

Material Required

- Herbal hair dye formulations as a sample,
- Potassium Iodide,
- Mercuric Chloride,
- Distilled Water.

Procedure

Weigh 5 g of Potassium Iodide (KI) and 1.358 g of mercuric chloride using a precise balance. Dissolve the weighted potassium iodide in 60 mL of distilled water in a clean container. Stir the solution in order to completely dissolve the KI. Add the mercuric chloride to the KI solution while stirring continuously. Continue stirring until the mercuric chloride is completely dissolved. Once the solution is homogeneous, take a small amount of finely powdered sample and place it in a test tube. Add a few drops of Mayer's reagent to the test tube using a dropper or pipette. Observe the color change in the test tube. The presence of alkaloids in the sample will result in the formation of a creamy precipitate or turbidity within a few minutes. Compare the test tube with a control tube containing distilled water and Mayer's reagent. The control tube should remain clear, indicating the

absence of alkaloids. A summary of Phytochemical evaluation has been tabulated in Table 3.

Rheological Evaluation

For the internal formulation, physical parameters including angle of repose, bulk or untapped density, tapped density, Housner's Ratio, and Carr's index were measured and calculated. The formula for computing bulk Density (D) is D=M/V, where M is the mass of the particles and V is the total volume, they fill. This is measured with a graduated cylinder. 100 g of the weighted formulation were added to the cylinder using a funnel. Following the recording of the starting volume, the sample was extensively tapped. The bulk density value was obtained by comparing the starting volume with the volume identified after tapping. This value was then utilized to calculate the tapped density. The angle of repose measures the powder's flow properties since it affects how cohesive the different particles are with one another. The fixed funnel cone method is utilized to determine the Height (H) above the paper that is put on a level surface. The pack was slowly poured into the funnel until the peak formed. In this case, an is the angle of repose, and tan is equal to either H/R or arc tan H/R. R stands for the radius of the conical heap. Housner's Ratio is connected to the powder flow and is influenced by the antiparticle friction.

Housner's Ratio is calculated as D/D' where D' is the tapped density and D is the bulk density. Carr's index is a measure of a powder's compressibility. Table 4 presents the results collected for the rheological assessment of herbal hair coloring.

Patch Test

This often entails applying a little amount of the hair dye's aqueous solution behind the ear or on the inner elbow in an area measuring 1 cm^2 and letting it dry. Any indications of annoyance or a feeling of ill health are documented. The designated area received measured and sparing applications of the prepared hair pack for a predetermined amount of time. Any sign of redness, irritation and swelling were monitored and recorded for up to 24 hr. Table 5 lists the outcomes of tests for irritability symptoms.

Stability Test

The created formulation was subjected to a month's worth of storage at various temperatures in order to verify its stability. The formulations of packed glass vials were examined for physical characteristics like smoothness, texture, odour, pH, and colour as shown in Table 6, while being stored at room temperature 35°C.

RESULTS AND DISCUSSION

Four different mixtures were prepared by taking 10 g of powder (Henna, Indigo, Formulations A and B) and making a slurry of it. Keep that slurry for 24 hr. Now take a Blonde hair (Figure 1) and apply paste on it. Wash it after one hour. The observed colour of the henna is reddish-orange while the colour of indigo is blue-black. The produced herbal hair dye is made up entirely of beneficial natural components. Due to the ideal herbal combination in this composition, it also functions as a conditioner, anti-dandruff agent, and hair growth stimulator in addition to being a hair dye. The inert nature of the hair dye pack was revealed by a patch test. It is simple to store and stable at temperatures 20 and 35°C. The formulation B (Figure 2) shows better hair colour than formulation A (Figure 3), which also promotes hair growth, strength and decreases hair damage. When Henna (50%) is mixed with Indigo (30%) along with Amla (4%), Coffee (4%), Shikakai (2%), Reetha (2%), Brahmi (2%), Red Sandalwood (2%), Hibiscus (2%), Bhringraj (2%) the observed hair color become darker after washing. It is devoid of the harmful effects of ammonia-based chemical colors because it is a formulation made from natural herbs. However, consistent use of it results in thick, silky, and beautifully colored hair.

CONCLUSION

In this study, the herbal hair dye pack was formulated, evaluated and its application on the blonde hair was carried out. The color obtained from the pack is close to natural hair black color. Also, the flow property of formulation B is better than the first formulation A so it is easy to make a slurry and apply on hair. However, further studies on the application of formulated hair dye packs on different human hair of different shades are required to determine its full range of potential advantages.

ACKNOWLEDGEMENT

The authors are grateful to Shri Guru Ram Rai University, Patel Nagar, Dehradun for providing the necessary facilities for this research work.

CONFLICT OF INTEREST

The authors declare that they have no conflicts of interest.

FUNDING

The author(s) received no financial support for this research.

ABBREVIATIONS

mL: Millilitre; **g:** Gram; **hr:** Hours; **KI:** Potassium Iodide.

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Cite this article: Patni P, Gahtori A, Bhandari P, Dimri T, Ahmad M. The Herbal Hue: Formulation and Evaluation of Herbal Hair Dye. Pharmacog Res. 2024;16(2):294-9.