



Antifungal Activity of Hair Dyes Gel from *Henna* Leaves and *Clove* Flower

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ABSTRACT

Henna leaves and clove flowers contain natural dyes that can be used as hair dye. Apart from that, it can also overcome dandruff problems because these plants contain antifungal activity. This study aims to determine the physical properties and antifungal activity of hair dyes gel extract from henna leaves and clove flowers. Research methods include preparing raw materials for henna leaves and clove flowers, making extracts of henna leaves and clove flowers, hair dyes gel formulation, physical properties testing, and antifungal activity testing. The hair dyes gel consists of 4 formulas, with a ratio of henna leaves and clove flowers in F0 without extract, F1 (10%: 2%), F2 (20%: 4%), and F3 (30%: 6%). Test parameters include physical properties tests which include organoleptic tests, homogeneity tests, pH tests, adhesion tests, coloring effectiveness tests, and antifungal activity tests against *Candida albicans*. The results of the physical properties test of the hair dyes gel showed that the combination of henna leaves extract and clove flowers can be used as a natural hair dye. The results of the hair dyes gel inhibition test at F0 was 12.77 mm, F1 was 25.97 mm, F2 was 29.80 mm, and F3 was 32.68 mm. The gel formula that provides the best color was F3, which provides the most striking color, namely dark brown, and offers antifungal activity of 32.68 mm, with a very strong category.



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ABSTRAK

Daun pacar kuku dan bunga cengkeh adalah tanaman yang mengandung zat warna alami yang dapat digunakan sebagai pewarna rambut. Selain itu juga dapat mengatasi masalah ketombe karena kedua tanaman ini mengandung aktivitas antifungi. Penelitian ini bertujuan untuk mengetahui sifat fisik dan aktivitas antifungi gel pewarna rambut alami ekstrak daun pacar kuku dan bunga cengkeh. Metode penelitian meliputi persiapan bahan baku daun pacar kuku dan bunga cengkeh, pembuatan ekstrak daun pacar kuku dan bunga cengkeh, formulasi gel pewarna rambut, uji sifat fisik dan uji aktivitas antifungi gel pewarna rambut. Sediaan gel pewarna rambut terdiri dari 4 formula, dengan perbandingan daun pacar kuku dan bunga cengkeh pada F0 tanpa ekstrak, F1 (10%:2%), F2 (20%:4%), dan F3 (30%:6%). Parameter pengujian diantaranya uji sifat fisik yang meliputi uji organoleptik, uji homogenitas, uji pH, uji daya lekat, uji efektivitas pewarnaan, serta uji aktivitas antifungi terhadap *Candida albicans*. Hasil uji sifat fisik gel pewarna rambut yang diteliti menunjukkan kombinasi ekstrak daun pacar kuku dan bunga cengkeh dapat digunakan sebagai pewarna rambut alami. Hasil uji daya hambat gel pewarna rambut pada F0 sebesar 12,77 mm, F1 sebesar 25,97 mm, F2 sebesar 29,80 mm, dan F3 sebesar 32,68 mm. Formula gel yang memberikan warna paling baik adalah F3, yang memberikan warna paling mencolok yaitu coklat pekat dan memberikan aktivitas antifungi sebesar 32,68 mm, dengan kategori sangat kuat.

Kata Kunci: Pewarna rambut; Pacar kuku; Cengkeh; Antifungi

1. Introduction

Hair dye is a type of cosmetic that is popular with the public, to affect hair, either darkening, reducing, or changing hair color. As you get older, your hair will also turn white. One effort made to restore hair color is by using hair dye [1]. Hair dye are used to color hair, either restoring it or changing it to another color [2]. Using synthetic hair dye can cause side effects such as allergies to the scalp, irritation, and damage to the tissue in the hair. So the use of natural dyes as basic ingredients for hair dye is very necessary [3]. Hair dye is not only for coloring hair but can also nourish hair [4]. Hair problems experienced by humans today include dandruff, which is a condition where the horny layer occurs excessively from the scalp [5]. Dandruff appears in the form of itching caused by a fungal infection [6]. One of the fungi that causes dandruff problems in hair is *Candida albicans* [7], [8], [9].

Indonesia's biodiversity can be used as cosmetics [10]. Traditional beauty care is a cultural manifestation that has been passed down from generation to generation and has become part of Indonesian culture [10]. One of the plants that contains natural dyes is henna (*Lawsonia inermis* L.) leaves [11]. Henna leaves contain Lawsone dye which can be extracted and used as a dye [12]. Henna leaf extract also has astringent properties which can reduce wounds on the skin [13]. Henna leaf extract has antifungal activity against *Candida albicans* [14],[15]. Another natural ingredient used to strengthen hair color is clove flowers (*Syzygium aromaticum* L). Cloves are used as a natural dye for hair because they contain tannin which is a natural dye [16]. Clove powder can provide a darker, sharper, and clearer brownish color to gray hair. Apart from that, cloves can act as an antifungal, and conditioner, and prevent hair loss [17]. Cloves have antifungal activity against *Candida albicans*[18].

Based on this background, the author made an innovation by utilizing henna leaf extract into a natural hair coloring gel preparation combined with clove flowers to obtain

stronger color stability. Apart from that, the active ingredient in the natural hair coloring gel preparation, a combination of henna leaf extract and clove flowers, is expected to have antifungal activity against the *Candida albicans* fungus that causes dandruff in the hair. This research aimed to determine the physical quality and antifungal activity of natural hair coloring gel from extracts of henna leaves (*Lawsonia inermis*) and clove flowers (*Syzygium aromaticum* L.) on the growth of the fungus *Candida albicans*.

2. Method

Materials

The materials used include henna leaves, clove flowers, distilled water, 96% ethanol, H₂SO₄, glycerin, BaCl₂, nipasol, nipagin, CMC-Na, filter paper, shampoo, hair wig, tissue, Potato Dextrose Agar (PDA), fungal culture *Candida albicans*, NaCl, and Ketoconazole 2%.

Procedure

The research procedure includes several stages, namely preparation of raw materials and making extracts of henna leaves and clove flowers, formulation of hair coloring gel, physical quality tests, and antifungal activity tests of hair coloring gel extracts of henna leaves and clove flowers.

Preparation of raw materials

Samples of henna leaves were obtained from Banmati village, Sukoharjo. Select henna leaves that are perfectly green in color. Dried clove flowers were obtained from Ir Market. Soekarno Sukoharjo. The clove flowers taken are dark brown, dry, not rotten, and blackish. Each sample was washed with running water and sorted. The samples were dried in an oven at a temperature of 40-50 °C for ± 48 hours. The dried samples were ground with a grinder to obtain simplicia powder and then sieved using an 80 mesh sieve [19], [20].

Making extracts of henna leaves and clove flowers

Extraction was carried out using the maceration method. Each henna leaf powder and clove flower powder were prepared in a ratio of 1:10, 100 grams of powder, and 1000 ml of 96% ethanol. Then the simplicia powder was soaked using 96% ethanol solvent under closed conditions. Maceration was carried out for 5 days. The maceration results are filtered to separate the solvent from the filtrate. The filtrate obtained was then evaporated over a water bath at a temperature of 70 °C until a thick extract was obtained [19], [20].

Formulation of hair coloring gel

Making hair coloring gel is done in several stages. First, weigh each ingredient according to the formula in table 1. Second, develop the gelling agent (CMC-Na) with hot water, and stir until homogeneous or a gel mass is formed. The gelling agent is used for each formula. Third, add henna leaf extract and clove flowers. Fourth, add nipagin and nipasol, as well as glycerin, then mix until homogeneous. Fifth, the finished hair coloring gel preparation is put into a clear pouch that has been sterilized.

Table 1. Formulation of Hair Dye Gel of Henna Leaf Extract (*Lawsonia inermis*) and Clove Flower (*Syzygium aromaticum* L.)

Material	Function	Formulas (%)			
		F0	F1	F2	F3
Henna leaf extract	Active substance	-	10	20	30
Clove flower extract	Active substance	-	2	4	6
CMC-Na	Gelling Agent	3	3	3	3
Nipagin	Preservative	0,2	0,2	0,2	0,2
Nipasol	Preservative	0,2	0,2	0,2	0,2
Glycerin	Humectant	10	10	10	10
Aquadest add	Solvent	Add up to 10 mL	Add up to 10 mL	Add up to 10 mL	Add up to 10 mL

Information: Hair dyes gel formula with a ratio of henna leaves extract and clove flowers extract: F0 (0%:0%), F1 (10%:2%), F2 (20%:4%), and F3 (30%:6%).

Physical Quality Test of Hair Dye Gel

There are several physical quality tests for hair coloring gel with extracts from henna leaves and clove flowers. Organoleptic tests refer to research [21]. The pH test refers to research [22]. Homogeneity test, referring to research [23]. The adhesion test refers to research [24]. The coloring effectiveness test refers to research [25]. pH measurement test is carried out to determine whether the pH of the gel matches the pH of the skin, namely between 4.5 and 6.5. Homogeneity is indicated by the absence of coarse grains in the preparation. The adhesion power of semisolid preparations is more than 1 second.

Test the Antifungal Activity of Hair Dye Gel

The antifungal activity test of hair coloring gel extract from henna leaves and clove flowers against *Candida albicans* was carried out using the well method, using PDA (Potato Dextrose Agar) media. The test materials included hair coloring gel, extracts of henna leaves and clove flowers in the formulas F0, F1, F2, and F3, as well as Ketoconazole 2% as a positive control and distilled water as a negative control. Observations were made after a 3x24 hour incubation period. The clear area indicates the sensitivity of the fungus to the test material, which is expressed by the diameter of the inhibition zone. The diameter of the inhibition zone is categorized as to the strength of its antifungal power based on the classification of fungal growth inhibition response, namely <10 mm (weak), 10-15 (medium), 16-20 mm (strong), >20 mm (very strong) [26] [27], [28].

3. Results and Discussions

The research began with making thick extracts of henna leaves and clove flowers. Based on the research results, the results of a thick extract from 100 grams of simplicia henna leaf powder resulted in 12.035 grams of extract so the yield obtained was 12.035%. Meanwhile, 100 grams of clove flower simplicia powder resulted in a thick extract of 13.481 grams of extract, so the yield obtained was 13.481%. Hair coloring gel is made in

4 formulations. Physical quality tests on hair coloring gel include organoleptic tests, homogeneity tests, pH tests, adhesion tests, coloring effectiveness tests, and *Candida albicans* antifungal activity tests.

Table 2. Physical Quality of Hair Dye Gel of Henna Leaf Extract (*Lawsonia inermis*) and Clove Flower (*Syzygium aromaticum* L.)

F	Organoleptic (color, shape, smell, skin feel)	pH	Homogeneity	Adhesion (seconds)	Staining Effectiveness
F0	Transparent, semi-solid, odorless, non-sticky	5,22	Homogeneous	5,14	-
F1	Orange brown, semi-solid, typical of cloves, not sticky	5,57	Homogeneous	5,57	Blonde Brown
F2	Brown, semi-solid, typical of cloves, not sticky	5,75	Homogeneous	5,89	Brown
F3	Dark brown, semi-solid, typical of cloves, not sticky	6,02	Homogeneous	6,58	Dark Brown

Information: Hair dyes gel formula with a ratio of henna leaves extract and clove flowers extract: F0 (0%:0%), F1 (10%:2%), F2 (20%:4%), and F3 (30%:6%).

The organoleptic test aims to determine the color, shape, smell, and taste of the skin of the hair coloring gel [29]. Based on table 2, the results of the organoleptic test for the hair coloring gel formulation of henna leaf extract and clove flower extract, there is a slight difference, because the henna leaf extract and clove flower extract used have different concentration ratios. The organoleptic test results showed that F1 produced an orange-brown gel, semi-solid, had a distinctive clove odor, and was not sticky to the skin. F2 produces a gel that is brown in color, semi-solid, has a distinctive clove odor, and is not sticky on the skin. F3 produces a gel that is dark brown in color, semi-solid, has a distinctive clove odor, and is not sticky to the skin. The color in F3 is the most concentrated because it uses a larger ratio of extracts than other formulas. Meanwhile, F0 shows transparent, semi-solid, odorless, non-sticky results on the skin. F0 is a base formula without added extracts.

The pH test aims to determine the safety of a preparation, especially topical preparations. Ideally, topical preparations have a pH value that is the same as the skin's pH so that irritation does not occur on the skin surface. Gel preparations that are too acidic for the skin's pH or pH < 4.5 can irritate the skin, whereas if gel preparations are too alkaline or pH > 6.5 it can cause dry skin [30]. From the research results in table 1, it shows that the hair coloring gel of henna leaf extract and clove flowers at F0, F1, F2, and F3 have a pH of 5.22 respectively; 5.57; 5.75 and 6.02. These results show that the higher the concentration of extract in the preparation, the more the pH value increases [31]. This is different from previous research conducted by [32], which stated that the greater the amount of extract used, the lower it would be. Overall, the pH of the preparation obtained is still safe for hair and meets the requirements for a good pH value, namely 4.5-6.5.

Homogeneity testing aims to determine whether the ingredients in the formulation are mixed evenly or not. Homogeneous is indicated by the absence of coarse grains in the preparation [33]. Based on the results in table 2., it is known that the homogeneity test results for each formula provide homogeneous results for each preparation, which is known based on the absence of lumps or coarse grains in the gel preparation.

Adhesion testing aims to determine how much the gel is able to stick to the skin or hair within a certain time so that it can function optimally. It is recommended that the adhesive force of semisolid preparations be more than 1 second [34]. Based on table 2., the results of the adhesion test for the hair coloring gel formulation of henna leaf and clove flower extract were 5.14 seconds at F0, 5.57 seconds at F1, 5.89 seconds at F2, and 6.58 seconds at F3. . This shows that the adhesion test results of each gel formula have good adhesion because they comply with the standard value, namely more than 1 second. The longer the gel sticks to the hair, the more of the active substances released will be absorbed by the hair, meaning the ability of the gel to stick to the hair can function optimally [35].

Testing the effectiveness of hair coloring gel was carried out to determine whether the hair coloring gel made could provide a coloring effect on hair. Tests were carried out on hair coloring gel in F1, F2, and F3 using nine hair bundles of more than 30 strands each which had been washed with shampoo, smeared with hair coloring gel marked with formulas F1, F2, F3, and left for 2 hours. Then the hair is washed and observed whether there is a color change in the hair. Based on table 1, it is known that the results of the hair dye effectiveness test can provide color and can stick to the hair. The coloring effectiveness test can be said to be good when the gel preparation is applied to the hair, the color can stick [3]. The F1 produces blonde brown hair. F2 produces a light brown hair color. F3 produces a dark brown hair color. The results of the coloring effectiveness test show that this preparation can be used as a natural hair dye, where the gel formula that gives the best color to hair is F3.

Table 3. Antifungal Activity for Hair Dye Gel of Henna Leaves Extracts (*Lawsonia inermis*) and Clove Flowers (*Syzygium aromaticum* L.)

F	Inhibition Zone Diameter (mm)	Category
Positive control	37,13	very strong
Negative control	0	weak
F0	12,77	medium
F1	25,97	very strong
F2	29,80	very strong
F3	32,68	very strong

Information: Hair dyes gel formula with a ratio of henna leaves extract and clove flowers extract: F0 (0%:0%), F1 (10%:2%), F2 (20%:4%), and F3 (30%:6%).

The results of the antifungal activity test showed that the hair coloring gel with a combination of henna leaf extract and clove flowers could inhibit the growth of the *Candida albicans* fungus. Based on table 3, the antifungal activity of distilled water as a negative control was weak, no antifungal activity was obtained, whereas with Ketoconazole 2% as a positive control, an average inhibitory power was found to be 37.13 mm. The antifungal activity was very strong in the positive control because Ketoconazole was able to inhibit filamentous growth and metabolism in *Candida albicans*, causing damage to cells [36], [37]. The inhibitory power of hair dye gel at F0 is 12.77 mm

in the medium category. Meanwhile, F1 is 25.97 mm, F2 is 29.80 mm, and F3 is 32.68 mm, each in the very strong category. This shows that the hair coloring gel preparation, henna leaf, and clove flower extract, have antifungal activity in inhibiting the growth of the *Candida albicans* fungus. As mentioned in previous research, the addition of *Lawsonia inermis* L [38], [39] and cloves [40], [41] can control the growth of *Candida albicans*.

The antifungal effect produced can be caused by secondary metabolite compounds found in henna leaf extract [42] and cloves [43] namely flavonoids, alkaloids, saponins, tannins, and terpenoids. Flavonoids interfere with the permeability of *Candida albicans* cell membranes by forming complex bonds with cell membrane proteins, thus initiating damage to *Candida albicans* cell membranes [44], [45]. Tannin can inhibit the formation of chitin, which is one of the components that make up *Candida albicans* cell walls [46]. Terpenoids disrupt the balance of ion homeostasis so that *Candida albicans* will experience damage to cell membranes [47]. Alkaloids cause damage to fungal cell walls and cause intercalation of fungal DNA [48]. Saponins inhibit the growth of *Candida albicans* by interacting with membrane sterols [49]. The results of this research show that the higher the extract concentration, the higher the inhibitory power against fungi. The results of this study are in line with previous research, that hair tonic with papaya leaf extract has antifungal activity against the fungus *Candida albicans* [50].

4. Conclusion

The results of tests on the physical properties of hair dyes gel show that the combination of henna leaves and clove flowers can be used as a natural hair dye. The results of the hair dyes gel inhibition test at F0 were 12.77 mm, F1 was 25.97 mm, F2 was 29.80 mm, and F3 was 32.68 mm. Each formula has antifungal activity, but based on the physical properties obtained, the gel formula that provides the best color is F3 which provides the most striking color, namely dark brown, and produces the greatest inhibitory power against *Candida albicans*.

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