



Testing The Effectiveness of Hair Growth Tonic with Cocoa Bean Extract on Mice for Physical Stability and Activity

Muhammad Alfian^{*1}, Muhammad Nurul Hasanudin², & Mochamad Luqni Maulana³

¹Departement of Pharmaceutical Technology, Faculty of Halal Industry, University of Nahdlatul Ulama Yogyakarta, D.I Yogyakarta

²Departement of Pharmacology, Faculty of Halal Industry, University of Nahdlatul Ulama Yogyakarta, D.I Yogyakarta

³Bachelor Program, Faculty of Halal Industry, University of Nahdlatul Ulama Yogyakarta, D.I Yogyakarta

#This research has been presented at The 3rd International Conference on Contemporary Science and Clinical Pharmacy (ICCCSP 2023), Universitas Andalas, Indonesia, 30-31 October 2023

ABSTRACT: Cocoa is a highly valued plant due to its versatile uses, including being processed into cosmetic products. Hair loss and slow hair growth are common hair-related concerns that can be effectively addressed through the use of hair tonics. Previously, research has shown that cocoa bean can effectively stimulate hair growth when made into a cream. Our research aimed to determine the secondary metabolite content of cocoa beans extract, formulate the extract into a hair growth tonic, and test its physical stability and effectiveness on hair growth in mice. Cocoa beans extracted by maceration method and conducted phytochemical screening. This extract was then used to create three formulations with different extract concentrations of 0.2%, 0.3%, and 0.5%, which were tested for physical stability, including organoleptic properties, homogeneity, pH, and specific gravity. Mice were divided into five treatment groups, hair length growth measurements were taken every week, on the 7th, 14th and 21st days. Six strands of hair were taken from the application area, and their length was measured using a calliper. Results revealed that cocoa seed extract contains flavonoids, alkaloids, tannins, and phenolics. The formula with a 0.3% concentration of cocoa bean extract showed the best physical stability. Hair growth tests on mice showed no significant difference between the 0.3% and 0.5% cocoa bean extract formulas and the positive control in the second week. The 0.3% formula is the best for ensuring good physical stability and promoting hair growth in mice when used twice daily for two weeks.

Keywords: cacao bean; hair tonic; hair growth.

Introduction

Cocoa is one of the leading plantation commodities in Indonesia, cocoa beans are one of the exported commodities but their value is low. Utilizing semi-finished processed products such as simplicia powder which can be used as cosmetic products can increase the selling value. Indonesia is a leading producer of cocoa, but the value of exported cocoa beans is relatively low. However, by using products like simplicia powder, which has cosmetic applications, producers can increase the selling value of cocoa [1]. Cocoa (*Theobroma cacao* L.) is rich in polyphenol compounds that act as antioxidants. The main polyphenols found in cocoa beans are catechin and epicatechin, which are flavonoids [2]. Additionally, the functional compounds in cocoa bean fat, such as stearic

acid, oleic acid, palmitic acid, and vitamin E, are beneficial for skin health. They can promote cell regeneration and stimulate hair growth [3].

The hair is an essential aspect of the body, serving both a cosmetic and protective role for the scalp. Hair loss can lead to baldness (alopecia), and this condition can negatively impact a person's self-esteem, affecting both genders [4]. Hair tonic is a popular cosmetic product known for its easy application and quick absorption into the scalp. It offers a variety of benefits, including promoting hair growth, maintaining a healthy scalp, preventing hair loss, boosting blood circulation, and relieving dandruff and itching. Additionally, it provides a refreshing sensation when applied [5].

Article history

Received: 29 Okt 2023
Accepted: 05 Des 2023
Published: 30 Des 2023

Access this article



***Corresponding Author:** Muhammad Alfian

Departement of Pharmaceutical Technology, Faculty of Halal Industry, University of Nahdlatul Ulama Yogyakarta, Kota Yogyakarta, Daerah Istimewa Yogyakarta, Indonesia, 55162 | Email: muhammadalfian@unu-jogja.ac.id

The integration of cocoa bean extract into the hair tonic product line has the potential to yield positive results in terms of promoting hair growth. This innovative addition presents an opportunity for the company to expand its product offerings and cater to a wider range of consumers. Further research could be conducted to determine the optimal concentration of cocoa bean extract to maximize its effects on hair growth [5].

In order to explore the secondary metabolite compounds present in cocoa bean extract (*Theobroma Cacao L.*), researchers conducted a study on a different form of product made from cacao bean extract cream [6]. The extract was then used to formulate a hair tonic product, which was tested for physical stability over a 4-week period at room temperature (25°C). The tonic was also tested for its effectiveness in promoting hair growth in mice over a 3-week period.

Methods

Equipments and Materials

The equipments used in this research were blender (COSMOS), rotary evaporator (IKA), analytical balance (OHAUS), maceration jar container, water bath (memmerth), pH meter (PHS-3C), viscometer (Lamy Rheology), pycnometer (IWAKI), Whatmann paper no 1 (cytiva), 40 mesh sieve, custom roasting, custom desheller, custom oscilating granulator and glass tools (Herna).

The materials used in this research are, cocoa beans (KWT Pawon Gendis Kulon Progo, Indonesia), 70% ethanol (PT. Bratachem, Indonesia), 96% ethanol (PT. Bratachem, Indonesia), EDTA (PT Alfa Kemika Indonesia), DMDM Hydantoin (SLB Brazil), distilled water (PT. Bratachem, Indonesia), FeCl₃ (Merck, Germany), HCL (Merck, Germany), Dragendorf Reagent (Merck, Germany), white mice (balb/c, 2-3 month) and Minoxidil (Regrou).

Extraction

The cocoa beans used in this process were provided

by the KWT Pawon Gendis Kulon Progo, Yogyakarta, which is a women's farmer group. The beans were carefully cleaned and sun-dried for four days. After drying, they were roasted at 120°C for 20 minutes using a roasting tool. The roasted beans were then separated into cocoa nibs and bean epidermis using a desheller. Next, the cocoa nibs were ground into powder using an oscillating granulator and blender (cosmos). The resulting powder was sifted through a 40 mesh sieve and extracted using the maceration method for 48 hours. To achieve a ratio of 1:3 between the material and solvent, 500 g of the material was soaked in 1500 mL of 70% ethanol. [7]. Maceration was performed at room temperature (30 ± 2 °C) with manual stirring every six hours. The mixed solvent extract was filtered using whatman number 1 filter paper. The filtered extract was evaporated in a rotary vacuum evaporator IKA RV8V at a temperature of 40°C, pressure of 100 mbar, and speed of 100 rpm [8]. Finally, the thickened extract was obtained by further thickening the rotated extract in a water bath memmerth [9].

Phytochemical Screening

In order to determine the tannin content, dissolve 1 g of extract in 2 mL of distilled water and heat the solution. Filter the solution and mix it with 2 drops of FeCl₃. If the resulting colour is blackish-blue or green, then it indicates a positive tannin content [10].

To analyze flavonoids, mix 0.5 mg of extract with 2 mg of Mg and 2-3 drops of concentrated HCl. If the color changes to red-orange, it means that flavonoids are present [11]. For phenolic compound testing, dissolve 50 g of sample extract in 5 mL of distilled water and mix it with 3-4 drops of FeCl₃. A green to black-blue color indicates the presence of phenolic compounds [12].

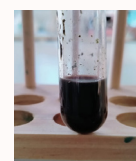
To test for alkaloids, mix 1g of sample extract with 1.5 mL of 2N hydrochloric acid solution, filter it, and then mix it with 3-4 drops of Dregendorf's reagent. If an orange-yellow precipitate appears, it confirms the presence of alkaloid compounds [12].

Table 1. Hair tonic formulation

Material	Function	Formulation 1	Formulation 2	Formulation 3
Cacao bean Ext	Activ Ingredients	0.2 %	0.3 %	0.5 %
Alcohol 70%	Solvent	10%	10%	10%
EDTA	Chelating Agent	0.05%	0.05%	0.05%
DMDM Hydantoin	Preservative	0.4%	0.4%	0.4%
Aquadest	Solvent	Add 100%	Add 100%	Add 100%

Table 2. Results of phytochemical screening

Secondary metabolite	Observation results if positive	Observation results	Conclusion
Tannin	the color of the solution changes to blackish blue	solution changes to blackish blue	+
Flavonoid	the color of the solution changes to yellowish brown	solution changes to yellowish brown	+
Fenol	the color of the solution changes to blackish green	solution changes to blackish green	+
Alkaloid	There is a yellow precipitate	There is a yellow precipitate	+



Cacao Seed Extract Hair tonic Formulation

The process of developing the formulation involved the utilization of lower extract concentration, a methodology that was informed by prior research [6]. The preparation of hair tonic cocoa bean extract involves the use of three different concentrations of cocoa bean extract, namely 0.2%, 0.3%, and 0.5%. The formulation for the extract is carefully measured and designed to yield desirable results .

All ingredients are then weighed by analytical balance OHAUS, then the cacao seed extract is mixed with 70% alcohol and dissolved until homogeneous which is referred to as mixture A. The EDTA is then dissolved first in distilled water to become mixture B, the mixture between A and B is added with Dmdm hydantoin and stirred again until homogeneous.

Evaluation of Physical Stability 3 Formulation of Cacao Seed Extract Hair Tonic Preparations Organoleptic Test

Over the course of 4 weeks, the shape, colour, and odour of the three formulation F1, F2 and F3 hair tonic preparation were visually observed at room temperature storage (25°C) using an organoleptic test [13].

PH Test

The hair tonic F1, F2, F3 was also subjected to a pH test using a PHS-3C pH meter with measurements taken every week [13].

Viscosity Test

To measure the hair tonic F1, F2, F3 viscosity, the preparation was placed in a 100 ml beaker and tested using a Lamy Rheology B-One Plus LR viscometer with an L-4 spindle needle, spinning at a speed of 30 RPM for 30

Table 3. Result of organoleptic test

Formula	Organoleptic test	Week			
		I	II	III	IV
1	Form	Liquid	Liquid	Liquid	Liquid
	Colour	Brown	Brown	Brown	Brown
	Odour	Chocolate	Chocolate	Chocolate	Chocolate
2	Form	Liquid	Liquid	Liquid	Liquid
	Colour	Brown	Brown	Brown	Brown
	Odour	Chocolate	Chocolate	Chocolate	Chocolate
3	Form	Liquid	Liquid	Precipitate	Precipitate
	Color	Brown	Brown	Light Brown	Light Brown
	Odour	Chocolate	Chocolate	Chocolate	Chocolate

seconds. This test was conducted every week for 4 weeks [9].

Specific Gravity Test

Lastly, the specific gravity of the hair tonic F1, F2 and F3 was measured using a IWAKI pycnometer at room temperature. The empty pycnometer (W1) was weighed, filled with distilled water and weighed again (W2). After being emptied, it was then filled with hair tonic and weighed once more (W3). The specific gravity of the hair tonic could be calculated using the given formula :

$$p = \frac{W3-W1}{W2-W1} \times \text{specific gravity of aquadest (g/ml)}$$

with this test also carried out every week for 4 weeks [14].

Hair Growth Activity Test

The procedure was evaluated and approved by the ethic committee of Ahmad Dahlan University at 2015 number 011504040. Male white mice (*Mus musculus*) aged 3-4 months were used for the experiment. The mice were allowed to adapt for 3-4 days after purchase, and then divided into five groups:

1. Normal control group (base of hair tonic)
2. Positive control group (minoxidil 2%)
3. Cacao Seed Extract Hair tonic formulation I (concentration 0.2%)
4. Cacao Seed Extract Hair tonic formulation II (concentration 0.3%)
5. Cacao Seed Extract Hair tonic formulation III (concentration 0.5%)

The normal group received hair tonic base treatment,



Figure 1. Cacao seed extract hair tonic at week 4

Table 4. Result of pH test

Formula	Average of pH \pm SD			
	Week I	Week II	Week III	Week IV
1	6.1 \pm 0.05	6.2 \pm 0.05	5.1 \pm 0.05	5.3 \pm 0.05
2	6.1 \pm 0.05	6.2 \pm 0.05	5.5 \pm 0.05	5.6 \pm 0.1
3	6.1 \pm 0.05	5.5 \pm 0.1	4.1 \pm 0.05	4.1 \pm 0.05

while the positive group received 2% minoxidil hair growth treatment. The backs of the mice were shaved clean of hair before the treatment. During the treatment, the backs of the shaved mice were first smeared with 70% ethanol as an antiseptic, followed by the hair tonic application for each group. The treatment was administered twice a day 2 mL, in the morning and evening, for 21 days. The first day of administration was considered day 0. Hair length growth measurements were taken every week, on the 7th, 14th and 21st days. Six strands of hair were taken from the application area, and their length was measured using a calliper.

SPSS 16.0 used for statistical calculations were performed using normality (Shapiro-Wilk) and homogeneity (Levene) tests on the results of hair length measurements for each treatment group. Data that was distributed normally and homogeneously was tested using ANOVA to determine significant differences between all groups. Data that was not normally distributed and homogeneous was subjected to non-parametric testing using Kruskal Wallis and Mann-Whitney tests with a 95% confidence level between groups [15].

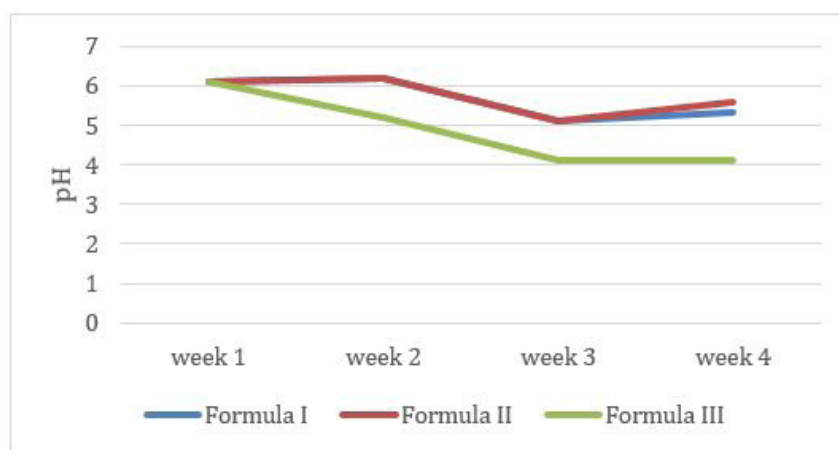
Result and Discussion

Extraction of Cacao Seed

Following the maceration of 500 g of cocoa bean powder with 1,500 mL of ethanol solvent, a thick extract weighing 62.31 g was obtained, with a yield of 12.8%. This yield falls within the optimal range of 10-15% for a good yield, as recommended [16].

Furthermore, the phytochemical screening of the macerated extraction of cocoa beans with 70% ethanol solvent demonstrated the presence of various chemical compounds, including phenols, tannins, flavonoids, alkaloids, and saponins.

Qualitative chemical screening is an examination of a plant's chemical composition to determine the type of compounds it contains. Results from a phytochemical screening indicate that cocoa bean extract contains alkaloids, phenolics, tannins, and flavonoids presented in table 2. These findings are consistent with previous research that examined the chemical composition of cocoa bean extract [2]. Flavonoids, known for their antioxidant properties, can counteract the harmful effects of UV radiation on hair fibers. UV radiation can interfere with hair growth by damaging melanin pigment and protein

**Figure 2.** Graph of pH for 4-week

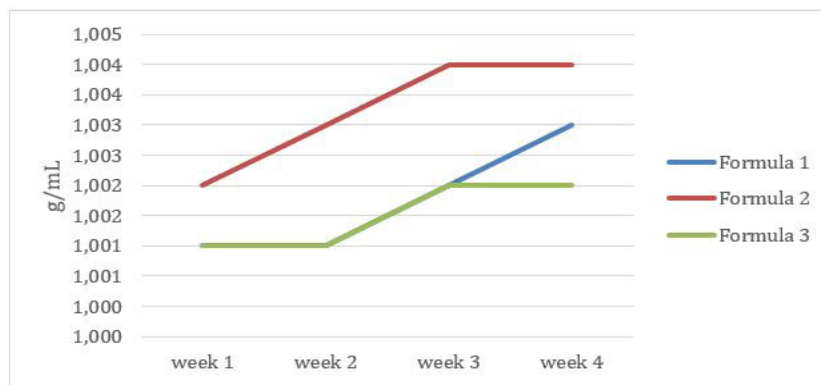


Figure 3. Specific gravity graph for 4 weeks

fractions, leading to oxidative stress and dermal papilla cell death. However, flavonoids in cocoa bean extract can promote hair growth by relaxing the muscles in hair follicle blood vessels, allowing for optimal nutrition. In addition, flavonoids can repair and protect the hair/skin system by disrupting the radical chain process and reducing oxidative damage [17].

Furthermore, the alkaloid, flavonoid, and phenol content in cocoa bean extract can also stimulate hair growth by inhibiting the activity of the 5 α -reductase enzyme. This enzyme is responsible for converting testosterone into dihydrotestosterone, which can lead to hair loss and perifollicular fibrosis through continued miniaturization of hair follicles. The tannin compounds in the extract can also promote hair growth by inhibiting the IFN- γ signal in the JAK/STAT3 pathway, preventing hair follicle dystrophy [18].

Evaluation of Physical Stability of Cacao Seed Extract Hair Tonic Preparations

Organoleptic Test

Organoleptic testing of hair tonic preparations is carried out by observing the physical form, odour and colour of the preparation. The following are the results of the hair tonic organoleptic test :

Organoleptic testing is a method of evaluating the

physical properties of a product for qualitative analysis. It is used to assess the stability and quality of a product, based on its appearance, texture, color, odor, and taste. During week 0, all three formulations were in liquid form and had a distinctive chocolate odor due to the use of cocoa bean extract. They were also brown in color. The stability of all formulas remained intact during the first week. In the second week, formulas 1 and 2 continued to remain stable, but formula 3 experienced slight settling. By the third week, formulas 1 and 2 remained stable, while formula 3 had sediments that were starting to enlarge and float on the surface. As the last week progressed, formula 3 had sediment floating on the surface and its color had faded to a brownish hue. This could be attributed to the large concentration of cocoa extract used, which led to mold formation on the surface. However, hair tonic formulas 1 and 2 exhibited good physical stability throughout the four weeks there was presented in [table 3](#) and [figure 1](#).

PH Test

The pH test aims to determine the degree of acidity or alkalinity of the hair tonic. The average results of the pH test are presented in [table 4](#).

The determination of a product's potential to cause irritation can be assessed by measuring its pH level. The manifestation of scaly or irritated skin can be attributed to

Table 5. Result of specific gravity test

Formula	Average of specific gravity (g/mL) \pm SD			
	Week I	Week II	Week III	Week IV
1	1.001 \pm 0.001	1.001 \pm 0.001	1.002 \pm 0.001	1.003 \pm 0.001
2	1.002 \pm 0.001	1.003 \pm 0.001	1.004 \pm 0.001	1.004 \pm 0.001
3	1.001 \pm 0.001	1.001 \pm 0.002	1.002 \pm 0.001	1.002 \pm 0.001

Table 6. Result of viscosity test

Formula	Viscosity (PaS) ± SD			
	Week I	Week II	Week III	Week IV
1	1.1 ± 0.1	1.2 ± 0.05	1.2 ± 0.1	1.2 ± 0.05
2	1.3 ± 0.05	1.4 ± 0.05	1.5 ± 0.02	1.5 ± 0.05
3	1.5 ± 0.05	1.6 ± 0.1	1.7 ± 0.05	1.7 ± 0.1

pH imbalances in the body [19]. The pH levels of various cacao seed extract hair tonic formulations decreased over a month when kept at room temperature. Among all the formulas, Formula 3 has significant pH decrease during the third week, as depicted in Figure 2. Maintaining an appropriate pH level can enhance blood circulation on the scalp, leading to better absorption of nutrients, oxygen, and minerals by hair follicles. Although Formulas 1 and 2 also observed a decline in pH, their pH levels remained safe for the scalp, falling within the range of 4.5 to 6.5 [19].

Specific Gravity Test

The specific gravity of a preparation is a measure of its density in relation to the density of water. This property can have a significant impact on the viscosity of the product, as a higher specific gravity indicates a higher density, which can lead to a thicker, more viscous product. Therefore, understanding the specific gravity of a preparation is important in predicting its flow and consistency [14]. The present study involved the determination of the specific gravity of hair tonic cacao seed extract, which was carried out on a weekly basis for a total duration of four weeks. The measurement was conducted using a pycnometer, which is a widely accepted method for determining the density of liquid substances.

Figure 3 and Table 5 present the specific gravity calculations for the hair tonic. It was observed that the average specific gravity value of all the formulations increased after being kept at room temperature for one month. These findings indicate that none of the hair tonic formulas meet the required specific gravity standards, which should be less than 1 g/mL. [14].

Viscosity Test

Over the course of four weeks, we conducted weekly viscosity measurements on our hair tonic using a B-One Plus LR Lamy Rheology viscometer. After leaving the formulations at room temperature for one month, the average viscosity value increased. Our findings indicate that the hair tonic's viscosity value will be higher or thicker with a higher concentration of cocoa extract. It's worth noting that the increase in viscosity value remains within the quality standard range set by the Indonesian National Standard (SNI) 16-4955-1998, which calls for a viscosity value of less than 5 PaS there was presented in table 6 and figure 4 [20].

Hair Growth Activity Test

The present study aimed to assess the hair growth activity through a measurement of the length of hair over

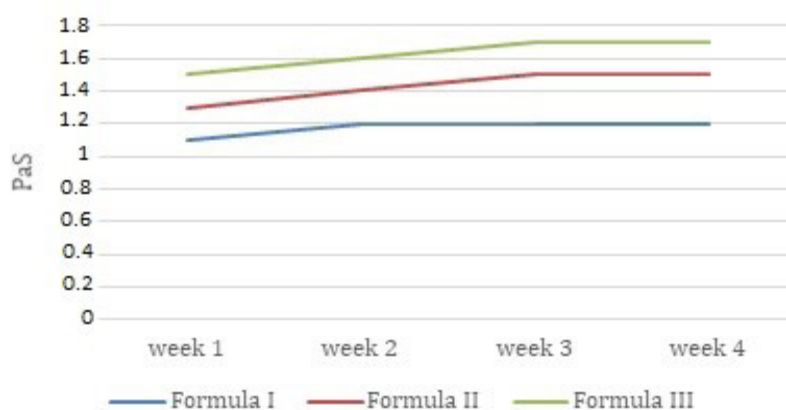


Figure 4. Graph of viscosity for 4 weeks

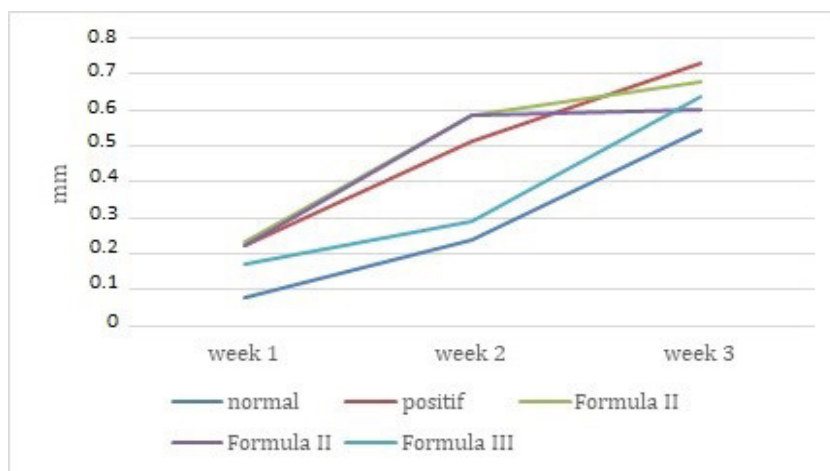


Figure 5. Graph of mouse hair length for 3 weeks

a period of three weeks. The obtained results are illustrated in Figure 5.

Based on the data presented in Figure 5, it appears that the test animals given minoxidil for 3 weeks (the positive group) had the longest hair growth, with an average length of 0.73 cm. On the other hand, the normal group - those given hair tonic base without cacao seed extract - had the shortest hair growth, averaging only 0.546 cm over the same period. To determine whether these differences were statistically significant, the data was analyzed using ANOVA and the results are presented in Table 7.

Statistical analysis was performed on the data in table 7 to determine differences in each treatment group. Results were calculated weekly and in the first week, the average growth rate for the normal, positive, formula 1, formula 2, and formula 3 groups was 0.076mm, 0.223mm, 0.236mm, 0.224mm, and 0.172mm. Analysis was conducted to determine normality of the treatment group data, which showed that it was normally distributed with a significant value > 0.05. Homogeneity variation analysis was then performed, which showed that the data was homogeneously distributed. ANOVA analysis was conducted and showed

no significant differences between each group. Therefore, it was possible to determine the results of giving hair tonic twice a day to each treatment group during the week. In the first week, there was no significant difference in hair growth. In the second week, the average growth rate for the normal, positive, formula 1, formula 2, and formula 3 groups was 0.240mm, 0.513mm, 0.584mm, 0.586mm, and 0.292mm. Analysis was conducted to determine normality of the treatment group data, which showed that it was not normally distributed with a significant value of <0.05. Homogeneity variation analysis was then conducted, which showed that the data was distributed non-homogeneously. Non-parametric test analysis was conducted and showed a significant difference between the normal and positive groups (sig 0.050). This showed that there was a significant difference in hair growth between the normal group and the positive group, namely minoxidil 2%. There was also a significant difference between the normal group and the formula I group (0.017). The study demonstrated a noteworthy contrast in hair growth between the normal group (base) and the Formula I group (2% cocoa bean extract concentration). Additionally, there was a significant

Table 7. Hair growth activity test results

Group	Treatment	Average of Hair Long (mm) ± SD		
		Week I	Week II	Week III
1	Normal (Base)	0.076 ± 0.07	0.240 ± 0.28	0.546 ± 0.15
2	Positif (Minoxidil 2%)	0.223 ± 0.18	0.513 ± 0.2	0.730 ± 0.08
3	FI (Cocoa seed ext 0,2 %)	0.236 ± 0.08	0.584 ± 0.18	0.680 ± 0.15
4	FII (Cocoa seed ext 0,3 %)	0.224 ± 0.09	0.586 ± 0.22	0.6 ± 0.17
5	FIII (Cocoa seed ext 0,5 %)	0.172 ± 0.05	0.292 ± 0.06	0.644 ± 0.08

difference between the normal group and the Formula II group (3% cocoa bean extract concentration) with a significance level of 0.023. These findings suggest that the results of applying hair tonic twice a day from the Formula I and Formula II groups during the second week resulted in a significant improvement in hair growth compared to the normal group. When compared with the positive control group (minoxidil), there were no significant differences between the Formula I and Formula II groups in the second week. Therefore, the Formula I and II groups exhibited the same level of hair growth activity as minoxidil. In the third week, the average growth rate for all groups was 0.546mm (normal), 0.730mm (positive), 0.680mm (Formula 1), 0.6 (Formula 2), and 0.644mm (Formula 3). The data was analyzed for normality and homogeneity variation, which showed that there were no significant differences between each group. Consequently, the study concluded that applying hair tonic twice a day to each treatment group during the third week resulted in no significant difference in hair growth. The findings from previous research on the use of cocoa bean extract cream to stimulate hair growth in rabbits align with the results of our study [20].

Conclusion

Based on the results of phytochemical tests, cocoa bean extract contains secondary metabolites including tannins, phenols, flavonoids and alkaloids which can be beneficial for hair growth. In the physical stability test for 4 weeks at room temperature 25 C, formulas 1 and 2 with a cacao seed extract concentration of 0.2% and 0.3% showed good physical stability except for the specific gravity which did not meet SNI requirements. Based on hair growth activity testing for 3 weeks, formula 2 with a concentration of 0.3% cacao seed extract had the best activity on mice hair growth during 2 weeks of use.

References

- [1]. N. Asyik and A. Ansi, "Proses Pengolahan Sekunder Biji Kakao Menjadi Produk Olahan Kakao Setengah Jadi," *Pros. Semin. Nas. Agribisnis*, vol. 0, no. 0, pp. 43–47, 2018 <http://dx.doi.org/10.37149/11328>.
- [2]. P. Sari, E. Utari, Y. Praptiningsih, and Maryanto, "Karakteristik Kimia-Sensori dan Stabilitas Polifenol Minuman Cokelat-Rempah," *J. Agroteknologi*, vol. 09, no. 01, pp. 54–66, 2015. <https://jurnal.unej.ac.id/index.php/AGT/article/view/3070>.
- [3]. T. Mariyana, A. E. Wibowo, and Y. Farida, "Uji Aktivitas Antiketombe dan Penyubur Rambut Kombinasi Ekstrak Daun Jeruk Kalamansi (*Citrofortunella microcarpa*) dan Ekstrak Biji Kakao (*Theobroma Cacao*) dalam Sediaan Creambath," *J. Sains dan Kesehat.*, vol. 3, no. 5, pp. 694–700, 2021, <https://doi.org/10.25026/jsk.v3i5.579>.
- [4]. N. Sativa, N. Noviyanti, R. A. Pratiwi, and S. Hindun, "Formulasi Dan Uji Aktivitas Tonik Rambut Ekstrak Etanol Daun Bidara (*Ziziphus Nummularia*) Pada Kelinci," *Bul. Penelit. Tanam. Rempah dan Obat*, vol. 32, no. 1, p. 40, 2022, <http://doi.org/10.21082/bullitro.v32n1.2021.40-51N>.
- [5]. Syilfiana Anwar and Fitrianti Darusman, "Hair Tonic dengan Kandungan Senyawa yang Memiliki Aktivitas Penumbuh Rambut dari Berbagai Bahan Herbal," *Bandung Conf. Ser. Pharm.*, vol. 2, no. 2, pp. 1–8, 2022, <https://doi.org/10.29313/bcsp.v2i2.4366>.
- [6]. A. Kurniasari, E. Anwar, and J. Djajadisastira, "Potensi Ekstrak Biji Coklat (*Theobroma cacao* Linn) sebagai Inhibitor Tirosinase untuk Produk Pencerah Kulit," *J. Kefarmasian Indones.*, vol. 8, no. 1, pp. 34–43, 2018, <https://doi.org/10.22435/jki.v8i1.7722.34-43>.
- [7]. I. P. L. Pratyaksa, G. P. Ganda Putra, and L. Suhendra, "Karakteristik Ekstrak Kulit Buah Kakao (*Theobroma cacao* L.) sebagai Sumber Antioksidan pada Perlakuan Ukuran Partikel dan Waktu Maserasi," *J. Rekayasa Dan Manaj. Agroindustri*, vol. 8, no. 1, p. 139, 2020, <https://doi.org/10.24843/jrma.2020.v08.i01.p14>.
- [8]. M. Alfian, M. L. Maulana, and M. Mustainin, "Formulation and Physical Stability of Temulawak (*Curcuma xanthorrhiza* Roxb.) Antiaging Lotion with Natural Colorant from Strawberry Extract (*Fragaria vesca* L.)," vol. 9, no. 1, pp. 20–26, 2023, <https://doi.org/10.35311/jmpi.v9i1.289>.
- [9]. S. Pappa, A. W. Jamaluddin, and A. Ris, "Kadar Tanin Pada Kulit Buah Kakao (*Theobroma cacao* L.) Kabupaten Paliwalimandar dan Toraja Utara," *Indones. E-Journal Appl. Chem.*, vol. 7, no. 2, pp. 92–101, 2019. <https://ojs.unud.ac.id/index.php/cakra/article/view/56181>.
- [10]. I. L. Kayaputri, D. M. Sumanti, M. Djali, R. Indiarjo, and D. L. Dewi, "Kajian Fitokimia Ekstrak Kulit Biji Kakao (*Theobroma cacao* L.)," *Chim. Nat. Acta*, vol. 2, no. 1, pp. 83–90, 2014, <https://doi.org/10.24198/cna.v2.n1.9140>.
- [11]. B. Chandra et al., "Phytochemical Screening And Antioxidant Activities Of Kemangi Leaf (*Ocimum tenuiflorum* L.) Methanol Extract Using DPPH (1,1-diphenyl-2-picrylhydrazine) Method," *J. Pharm. Sci. [Volume]*, vol. 2, no. 2, pp. 1–8, 2019. <http://dx.doi.org/10.36490/journal-jps.com.v2i2.20>
- [12]. R. Mustarichie, A. N. Hasanah, G. Wilar, D. Gozali, and N. M. Saptarini, "New Hair Growth Cream Formulation with Cocoa Pod Peel (*Theobroma cacao* L.)," *Sci. World J.*, vol. 2022, 2022, <https://doi.org/10.1155/2022/2299725>.
- [13]. S. M. Utami, J. Djajadisastira, and F. C. Saputri, "Using hair growth activity, physical stability, and safety tests to study hair tonics containing ethanol extract of licorice (*Glycyrrhiza glabra* Linn.)," *Int. J. Appl. Pharm.*, vol. 9, no. iii, pp. 44–48, 2017, https://doi.org/10.22159/ijap.2017.v9s1.20_25.
- [14]. H. Mu'ani and Purwati, "Uji Stabilitas Fisik Dan Uji Aktivitas Sediaan Hair Tonic Dari Ekstrak Etanol 96% Daun Kangkung (*Ipomoea Aquatica* Forsk.) Pada Rambut Kelinci Jantan (New Zealand White)," *Indones. Nat. Res. Pharm.*, vol. 4, no. 2, pp. 23–31, 2019. <https://doi.org/10.52447/inspi.v4i2.1708>
- [15]. L. Sulastri, S. Asih, and R. Amelia, "Uji Aktivitas Penyubur Rambut Emulgel Ekstrak Etanol Buah Cabai Gendot (*Capsicum annum* Var. *Abbreviata*) Pada Mencit Putih (*Mus musculus*) Jantan" *Med. Sains*, vol. 4, no. 2, pp. 101–110, 2020. <https://doi.org/10.37874/ms.v4i2.137>.
- [16]. L. Badriyah and D. Fariyah, "Optimalisasi ekstraksi kulit bawang merah (*Allium cepa* L) menggunakan metode maserasi," *J. Sint. Penelit. Sains, Terap. dan Anal.*, vol. 3, no. 1, pp. 30–37, 2023, <https://doi.org/10.56399/jst.v3i1.32>.
- [17]. M. A. Fakhriyah and K. H. Saputra, "Potensi Daun Katuk dalam Mencegah Kerontokan Rambut," *J. Penelit. Perawat Prof.*, vol. 2, no. 2, pp. 193–200, 2020, <https://doi.org/10.37287/jppp.v2i2.107>.
- [18]. P. Kesika, B. S. Sivamaruthi, S. Thangaleela, M. Bharathi, and C. Chaiyasut, "Role and Mechanisms of Phytochemicals in Hair Growth and Health," *Pharmaceuticals*, vol. 16, no. 2, pp. 1–22, 2023, <https://doi.org/10.3390/ph16020206>.
- [19]. A. Saputra Yasir, "Uji Aktivitas Pertumbuhan Rambut Kelinci Jantan Dari Sediaan Hair Tonic Yang Mengandung Ekstrak Etanol Daun Mangkokan," *J. Farm. Malahayati*, vol. 2, no. 1, pp. 77–84, 2019. <https://doi.org/10.33024/jfm.v2i1.1551>.

- [20]. W. Muliani, F. Setiawan, and Y. P. Sukmawan, "Formulasi Dan Evaluasi Sediaan Hair Tonic Ekstrak Etanol Daun Katuk (*Sauropus Androgynus* (L.Merr.) Sebagai Pertumbuhan Rambut Pada Kelinci Jantan New Zealand White," Pros. Semin. Nas. Disem. Has. Penelit. Progr. Stud. S1 Farm. Vol., vol. 2, pp. 101–112, 2022. <https://ejurnal.universitاس-bth.ac.id/index.php/PSNDP/article/view/970/740>.



Copyright © 2023 The author(s). You are free to share (copy and redistribute the material in any medium or format) and adapt (remix, transform, and build upon the material for any purpose, even commercially) under the following terms: Attribution — You must give appropriate credit, provide a link to the license, and indicate if changes were made. You may do so in any reasonable manner, but not in any way that suggests the licensor endorses you or your use; ShareAlike — If you remix, transform, or build upon the material, you must distribute your contributions under the same license as the original (<https://creativecommons.org/licenses/by-sa/4.0/>)