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The Effectiveness of Kombucha Coffee (*Coffea canephora*) Extract Antioxidant Moisturizer on UV-B Induced Skin Epidermal Thickness

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ABSTRACT

Background: The exposure to UV-B (280-320 nm) in Indonesia's tropical sunlight leads to the accumulation of free radicals, which in turn causes the formation of dry skin and thickening of the epidermis, a sign of skin photoaging. Topical intervention with moisturizers containing antioxidants is one approach to prevent further damage. The addition of kombucha coffee extract from fermented robusta coffee (*Coffea canephora*) with SCOBY (symbiotic cultures of bacteria and yeasts) can be a source of antioxidants. **Purpose:** To evaluate the effect of *Coffea canephora* extract as an antioxidant moisturizer in reducing epidermal thickness on mice's back skin exposed to UV-B irradiation. **Methods:** This research is a true experimental design with a post-test only control design. The sample used in this research is thirty male white mice (*Rattus norvegicus*), which were divided into six groups and were UV-B irradiated for two weeks with total dosage of 980 mJ/cm^2.At the end of the treatment, skin samples were excised and stained histologically with Mollory Azan (MA) to evaluate the thickness of the epidermis. Data obtained were analyzed statically with SPSS. **Result:** The Kruskal-Wallis test demonstrated significant results (0.0001, $p<0.05$) across all groups, indicating the efficacy of the kombucha coffee extract antioxidant moisturizer in reducing epidermal thickness. **Conclusion:** The application of *Coffea canephora* extract antioxidant moisturizer provided photoprotection against UV-B induced hyperplasia skin epidermis with the concentration of 5% and 10% extract.

Keywords: Antioxidants, epidermal thickness, Kombucha coffee, photoaging.

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BACKGROUND

Chronic exposure of skin to Ultraviolet B (UV-B) that can penetrate skin's outermost layer leads to dysfunction of the stratum corneum in maintaining skin water content.[1] Dry skin due to photoaging (skin aging caused by ultraviolet radiation) can be prevented with moisturizer. Not only skin hydration, UV-B-induced skin damage also stimulates epidermal thickening and collagen degradation due to accumulation of free radicals, which can be seen in the formation of skin wrinkles.[2] The addition of antioxidants to moisturizer can reduce free radical levels and prevent further damage from photoaging. One of the Indonesian natural sources of antioxidants is robusta coffee, which contain various bioactive compounds such as caffeine, chlorogenic acid, and flavonoids that act as free radical scavengers.[3] Fermentation can be used to enhance the production of bioactive compounds in coffee, thereby increasing antioxidant capacity. The production of kombucha, traditionally derived from fermented tea, can be innovatively adapted using alternative substances such as c offee.[4]

Fermentation utilizes biofilm-shaped symbiotic cultures of lactic and acetic acid bacteria with yeast

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called SCOBY. Under aerobic fermentation, microbes convert sugar in sweetened coffee into a primary energy source in producing antioxidant substances. Acetic acid and flavonoids, which are products of acetic acid bacteria, play a role in reducing free radicals and inflammation.[5] Moreover, the lactic acid that lactic acid bacteria produce nourishes the skin by inducing the production of ceramide and keratinocytes. Therefore, those substances can improve the protective function of the stratum corneum and prevent ageing.[6]

Based on the description above, research is aimed at evaluating the potential of moisturizer containing antioxidants in kombucha coffee (*Coffea canephora*) extract to protect and prevent epidermal thickening due to UV-B-induced free radicals.

METHODS

This study employs a true experimental design featuring a post-test only control design, utilizing simple random sampling as the sampling technique. This study involved six groups, each comprising five mice: a neutral group (N) that received no treatment or irradiation; a negative control group (C-), which underwent UV-B irradiation without any treatment; a positive control group (C+), which was subjected to UV-B irradiation and treated with a base moisturizer; and three treatment groups that were UV-B irradiated and treated with an antioxidant moisturizer containing kombucha coffee extract at concentrations of 5% (T1), 10% (T2), and 15% (T3). The analysis of data collected from epidermal thickness measurement histologically was conducted using the Kruskal-Wallis test and pairwise comparison tests.

The criteria for inclusion in this study consisted of male white mice aged 2-3 months, weighing between 150-200 grams, and exhibiting both anatomical and physiological health. The exclusion criteria for this study included mice that did not meet the weight requirements, those that exhibited health issues during the adjustment period and experiment, as well as any that were physically deformed. The study took place between March and August 2024 at the Integrated Hyperbaric-Biomolecular Laboratory within the Faculty of Medicine at Hang Tuah University. The sample utilized in this study consisted of thirty mice; however, there were six dropouts, with one mouse from each group. The sample that fully adhered to this study consisted of 24 mice.

The research began by making kombucha coffee extract, starting with preparing a liquid coffee substrate by brewing 55 grams of ground coffee, 100 grams of sugar, and 1 liter of hot water. Once the liquid had cooled, we added 200 ml of kombucha starter and the SCOBY to a glass container, then covered it with a cloth containing small pores. The fermentation process took 8 days at room temperature and was kept away from direct sunlight. Extraction was performed with rotary evaporator at 80°C, and the thick extract was processed into a moisturizer with a formulation as shown in Table 1.

Table 1. Formulation of moisturizer in different Groups

Components	Formulation (%b/w)			
	C+	T_1	T_2	T_3
Kombucha coffee extract	0	5	10	15
Oil Phase				
Cetyl alcohol	7	7	7	7
Cera alba	5	5	5	5
Span 80	3	3	3	3
Water Phase				
Glycerin	5	5	5	5
Methylparaben	1.8	1.8	1.8	1.8
Propylene glycol	3	3	3	3
Propylparaben	0.02	0.02	0.02	0.02
Tween 80	3	3	3	3
Aquadest	Ad 50ml	Ad 50ml	Ad 50ml	Ad 50ml

C+ =control positive group; T_1 = treatment group 1 with 5% kombucha extract; T_2 = treatment group 2 with 10% extract; T_3 = treatment group 3 with 15% extract

A hotplate stirrer was used to separately heat the oil and water phases to 70°C, creating a moisturizer. We mixed the two phases until we obtained a homogeneous cream preparation, ensuring no lumps formed. Thick kombucha coffee extract was added while continuing to stir until homogeneous and cooled.

The experiment started with keeping the mice in a controlled environment with a 12-h light/dark cycle. Following a week-long acclimatization phase, mice were intramuscularly anesthetized with a 0.5 mL mixture of ketamine and xylazine to cleanly shave back hairs 4x4 cm in size before dividing into groups (n = 5 mice per group). The dorsal skin of C-, C+, and T groups underwent UV-B radiation (UV-B Lamp Exoterra UVB150 Tube 60cm 18W) with a dosage given each day of 70mJ/cm^2.[7] Application of the equation[8] as follow was used in order to determine the necessary irradiation time to achieve targeted UV-B dose. In this research, 17 minutes of exposure resulted in a dose of 70mJ/cm^2.

$$\text{Irradiation time(s)} = \frac{\text{irradiation dose (mJ/cm}^2\text{)}}{\text{irradiation intensity (mW/cm}^2\text{)}} \quad 8$$

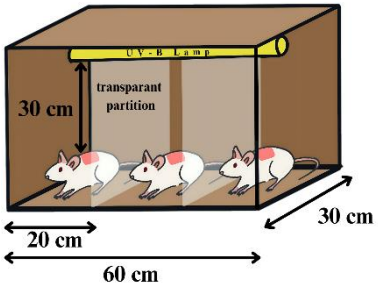

Figure 1. UV-B irradiation box design with a size of 60cm long and 30cm wide which is divided into 3 rooms with transparent partitions (20 cm wide in each room) and equipped with UV-B lamps 30cm above the mice.

Irradiation was conducted using a UV-B lamp (Exoterra UVB150 Tube 60cm 18W) within an irradiation box, as illustrated in Figure 1. The irradiation intensity was quantified at 0.07 mW/cm² utilizing the irradiance meter. The positive control group (C+) received a base moisturizer, whereas the treatment groups were administered kombucha coffee extract moisturizer at varying concentrations: T1 with 5% extract, T2 with 10% extract, and T3 with 15% extract. The application occurred bi-daily, administered 20 minutes prior to and 4 hours following irradiation. The negative control group (C-) was subjected to irradiation without any application being administered. The treatment occurs daily for a duration of two weeks, as illustrated in Figure 2. To verify that the rats satisfy the inclusion criteria throughout the treatment, their body weight will be measured on days 8 and 14.

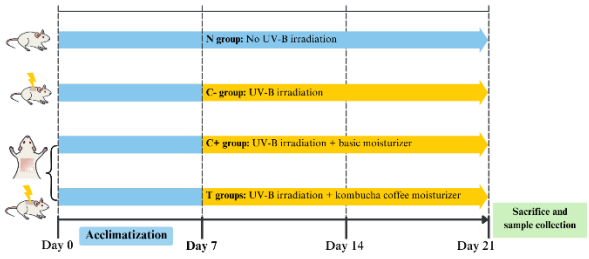

Figure 2. Illustrative depiction of experimental process.

The histology preparation was obtained from a biopsy of approximately 3 mm mouse back skin tissue, which was performed under anesthesia with xylazine and ketamine to prevent pain and discomfort. The excised skin tissues were immersed in 10% Neutralized Buffered Formaldehyde (NBF) solution and brought to the Histology Laboratory of Faculty of Medicine, Airlangga University, Surabaya. Following the formalin fixation and paraffin embedding of skin samples, we created 5-millimeter sections and stained them with Mollory Azan (MA). The epidermal thickness was observed and photographed using an Olympus IX71 digital camera (Olympus, Japan) at 200× magnification and measured digitally with ImageJ. Data obtained were analyzed statically with SPSS.

The Ethics Committee at Hang Tuah University, with ethics certificate number I/074/UHT.KEPK.03/VIII/2024, has reviewed this research.

RESULT

Table 2. Average epidermis thickness and statistical analysis results

Group	Epidermal thickness (μm)	P-value
C-	155.37	0.0001
C+	106.42	
N	49.98	
T_1	55.78	
T_2	54.52	
T_3	78.48	

C- = control negative group; N= neutral group; C+ = control positive group; T_1 = treatment group 1 with 5% kombucha extract; T_2 = treatment group 2 with 10% extract; T_3 = treatment group 3 with 15% extract

Table 2. shows the results of Kruskal-Wallis test in each groups have $p < 0.05$, which means there is significant different on epidermal thickness.

Table 3. Post Hoc Pairwise Comparison test results

Group	Comparison	P-value
C-	N	0.0001*
	C+	0.0001*
	T1	0.0001*
	T2	0.0001*
	T3	0.018*
N	C+	0.0001
	T1	1**
	T2	1**
	T3	0.014
Treatment groups		
T1	T2	1**
	T3	0.092
T2	T3	0.005

C- = control negative group; N= neutral group; C+ = control positive group; T_1 = treatment group 1 with 5% kombucha extract; T_2 = treatment group 2 with 10% extract; T_3 = treatment group 3 with 15% extract

*There is a difference of data values in both groups
**Data values in both groups have no difference

The results of the post hoc test indicated a variation in data values when the significance level (p-value) fell below 0.05. The results of this test demonstrated that each group exhibits distinct values when compared to the control negative group. The neutral group and the two treatment groups, 1 and 2, exhibit no differences in their values, nor do the values within these two treatment groups differ from each other.

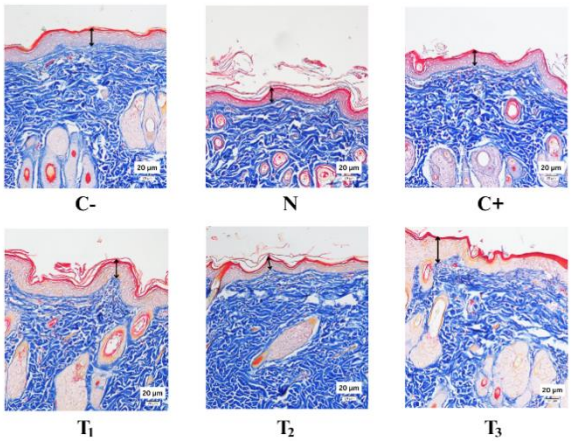

Figure 3. Histological examination of MA-stained dorsal mice skin samples. The black arrow indicated epidermal thickness from the stratum corneum to stratum basale layer (microscope magnification 200x with a scale of 200 μm). C- = control negative group; N= neutral group; C+ = control positive group; T_1 = treatment group 1 with 5% kombucha extract; T_2 = treatment group 2 with 10% extract; T_3 = treatment group 3 with 15% extract.

DISCUSSION

Adequate levels of UV radiation in sunlight provide health benefits, including the maintenance of the skin's circadian rhythm, which plays a role in the skin's barrier function (wound healing, antimicrobial action, immune defense, and hair follicle growth).[9] Skin structural integrity and physiological function can be adversely affected by prolonged exposure to UV light, and leading to photoaging. Photoaging is the aging of the skin as a result of photodamage caused by sun exposure.[10] It damages collagen and elastin fibers, leading to keratinocyte proliferation and epidermal hyperplasia.[11] This action is the body's mechanism to prevent further damage and malignancy, but it leads to thickening of the epidermis, which is one of the signs of photoaging. In addition, another effect of disrupting the epidermis is to decrease the skin's hydration levels, resulting in dry skin.[12] The use of moisturizers can reduce the impact, coupled with antioxidant content that can counteract free radicals caused by UV. This combination will slowly improve the structure of the skin.

The results of significant data analysis show that there is an effect in the use of moisturizers containing antioxidants in kombucha coffee extract on the back skin of rats exposed to UV-B rays. This research also concentrated on post hoc analysis results for the control, negative, neutral, and treatment groups. There was a difference between the control negative (C-) group, which received only irradiation without treatment, and the neutral group. These findings indicated exposure to UV-B light with a total dose of 980 mJ/cm^2 for 2 weeks successfully increased the thickness of the epidermis. Moreover, the negative control group is also significantly different from other groups.

These findings align with research of Hassan et al.(2015), which used a 15W 312 nm VILBER-LOURMAT-FRANCE lamp with 20 minutes of irradiation per day for 16 days in 4 weeks, showing an increase in epidermal thickness 12.979 times thicker than normal epidermis.[13] Epidermal thickness, which is a biomarker of photoaging, also occurred in the study of J. Zhang et al. (2014). With UV-B irradiation for 8 weeks, it was found that UV-B irradiation caused an increase of 4.23 times in epidermal thickness compared to the non-irradiated control group.[14,15]

Application of moisturizers containing antioxidants does not work directly in stopping the hyperplasia and thickening of the epidermis. Antioxidants work by neutralizing reactive oxygen species to suppress the pathways that stimulate inflammation and collagen degradation. In addition, antioxidants such as caffeine and chlorogenic acid stimulate the synthesis of collagen, which plays a role in the thickness of the dermis and indirectly signals to the body that the condition of the skin inside is improving.[16] The skin will gradually stop the process of epidermal thickening, allowing the epidermis to return to its normal thickness.

The comparison results between the neutral group and treatment 1 and treatment 2 groups show no difference in value data, as shown in Table 2. It indicates that the treatment given, which is the application of moisturizer containing 5% (T_1) and 10% (T_2) of kombucha coffee extract, a had similar epidermal condition with the neutral group. In order to consider which treatment group had better results, the comparison between treatment groups 1 and 2 was done. The result indicates that there is a similarity in the values of those two groups.

In conclusion, our study demonstrated that antioxidant moisturizer containing kombucha coffee (*Coffea canephora*) extract effectively prevents UV-B-

induced epidermal thickness, with the lower doses (5% and 10%) showing a more preferable anti-photoaging effect. In contrast, the 15% cream did not result in significant changes in epidermal thickness. These promising results were only seen in animal studies. Therefore, further clinical studies involving human participants are necessary to evaluate the safety and effectiveness of this treatment on human skin.

REFERENCES

1. Amalia RS, Rahmayunita G, Yusharyahya N, Sunardi D, Hoemardani ASD. Asupan Cairan dan Kelembapan Kulit. Medika Jurnal Kedokteran Indonesia 2021; 7(2): 91–8.
2. Kang MK, Kim DY, Oh H, Kim SI, Oh SY, Na W, et al. Dietary Collagen Hydrolysates Ameliorate Furrowed and Parched Skin Caused by Photoaging in Hairless Mice. Int J Mol Sci. 2021; 22(11): 6137.
3. Hasan AEZ, Utami NF, M. Extraction of Robusta Coffee Beans (Coffea Canephora) From Wonosobo by Ultrasonic Waves and Anticancer Tests. FITOFARMAKA: Jurnal Ilmiah Farmasi 2022; 12(1): 89-99.
4. Bishop P, Pitts ER, Budner D, Thompson-Witrick KA. Kombucha: Biochemical and microbiological impacts on the chemical and flavor profile. Food Chemistry Advances 2022; 1: 100025.
5. Neffe-Skocińska K, Karbowiak M, Kruk M, Kołożyn-Krajewska D, Zielińska D. Polyphenol and antioxidant properties of food obtained by the activity of acetic acid bacteria (AAB) – A systematic review. J Funct Foods 2023; 107: 105691.
6. Taufiqurrahman M, Agus ASR, Butar-Butar MET, Pijaryani I. Pemanfaatan Probiotik Asam Laktat dalam Susu Kuda Sumbawa sebagai Bahan Baku Tabir Surya. Jurnal Pharmascience 2024; 11(1): 125–34.
7. Widiyanto B, Yuniarifa C, Purnamasari R. Efek Perlindungan Ekstrak Bunga Telang (Clitoria Ternatea L.) Terhadap Densitas Kolagen Dari Paparan Sinar Uvb. Jusindo. 2024; 6(1).
8. Geng R, Kang SG, Huang K, Tong T. Dietary Isoeugenol Supplementation Attenuates Chronic UVB-Induced Skin Photoaging and Modulates Gut Microbiota in Mice. Nutrients 2024 ; 16(4): 481.
9. Lubov J, Cvammen W, Kemp M. The Impact of the Circadian Clock on Skin Physiology and Cancer Development. Int J Mol Sci 2021; 22(11): 6112.
10. Mayangsari E, Mustika A, Nurdiana N, Samad N. Comparison of UVA vs UVB Photoaging Rat Models in Short-term Exposure. Med Arch 2024; 78(2): 88.
11. Liu TH, Lin WJ, Cheng MC, Cheng YC, Lee CC, Lin JS, et al. Exploration of the Anti-Photoaging Mechanisms of Lactiplantibacillus plantarum TWK10 in a UVB-Induced Mouse Model. Appl Sci 2024; 14(20):9497.
12. Son DJ, Jung JC, Choi YM, Ryu HY, Lee S, Davis BA. Wheat Extract Oil (WEO) Attenuates UVB-Induced Photoaging via Collagen Synthesis in Human Keratinocytes and Hairless Mice. Nutrients. 2020; 12(2):300.
13. Hassan SMA, Hussein AJ, Saeed AK. Role of Green Tea in Reducing Epidermal Thickness upon Ultraviolet Light-B Injury in BALB/c Mice. Adv Biol 2015; 2015: 1–6.
14. Zhang C, Lu Y, Ai Y, Xu X, Zhu S, Zhang B, et al. Glabridin Liposome Ameliorating UVB-Induced Erythema and Lethery Skin by Suppressing Inflammatory Cytokine Production. J Microbiol Biotechnol 2021; 31(4): 630–6.
15. Zhang J an, Yin Z, Ma L wen, Yin Z qiang, Hu Y yan, Xu Y, et al. The Protective Effect of Baicalin against UVB Irradiation Induced Photoaging: An In Vitro and In Vivo Study. PLoS One 2014; 9(6): e99703.
16. Xue N, Liu Y, Jin J, Ji M, Chen X. Chlorogenic Acid Prevents UVA-Induced Skin Photoaging through Regulating Collagen Metabolism and Apoptosis in Human Dermal Fibroblasts. Int J Mol Sci 2022; 23(13): 6941.