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The Effect of Combination Cream of Patchouli Extract and Arabica Gayo Coffee Peel Extract on Aging Skin

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ABSTRACT

Background: Indonesia is a tropical country that is exposed to sunlight throughout the year. Due to this exposure, Indonesians are more susceptible to aging. It has been demonstrated that the antioxidative chemicals in Aceh nilam and Arabica Gayo coffee peel extract can prevent the aging process of the skin. **Purpose:** To observe the effect of combination cream patchouli extract and arabica Gayo coffee peel extract on aging skin, focusing on collagen and elastin. **Methods:** *This is an exploratory study using a randomized post-test only design.* The rats were divided into two control groups and three experimental treatments, each with a different active ingredient concentration (10%, 12.5%, and 15% of patchouli and coffee peel extract). For six weeks, each group is exposed to UVB light three times a week, for a total dose of 1020 mJ/cm2. The macroscopic morphology, density, and thickness of collagen and elastin in rat skin were observed and assessed. **Result:** There were noteworthy variations observed in collagen density, collagen thickness, and elastin density, whereas no significant difference was found in elastin thickness. The macroscopic skin morphology exhibited absence of inflammation across all experimental groups. **Conclusions:** This study suggests that cream containing Aceh patchouli extract and 15% of Arabica Gayo coffee peel extract exhibits potential in enhancing the quantity and quality of collagen while preserving elastin levels and it is safe for rat skin.

Keywords: Aceh Patchouli, Arabica Gayo Coffee Peel, Collagen, Elastin, Aging skin.

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BACKGROUND

Aging is an inevitable process in a person's life. One of the causes of aging is ultraviolet (UV) light exposure. Skin indications of photoaging can be brought on by prolonged exposure to UV rays. An extrinsic aging process brought on by prolonged exposure to UV light is called photoaging. Hyperpigmentation, pigmented patches, wrinkles, telangiectasis, and precancerous lesions are indicators of photoaging. UV light is able to penetrate the epidermal layer until the dermal layer and induce skin cell DNA damage. UV light can also degrade proteins in extracellular matrix (ECM) in the dermal layer and reduce antioxidative enzymes in the skin cells. This process can impact ROS concentration in the skin cells which can further damage ECM (collagen, elastin, etc.). Therefore, collagen and elastin levels can decrease and cause the appearance of photoaging clinical manifestations.[1]

Based on a study that was conducted in Europe and Northern America, photoaging is the most common cause of premature aging in a fair-skinned population. In Europe and Northern America, photoaging prevalence in skin phototype I, II, and III populations is 80-90%. In Caucasian population, photoaging is marked with wrinkles appearance and pigmentation patches and spots. On the other hand, the Asian population, specifically Southeast Asian countries like Indonesia, Singapore, and Malaysia, has different manifestations of photoaging. This population

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tends to develop hyperpigmentation in the skin due to higher level of melanin and thicker stratum corneum compared to the Caucasian population.[2]

As a country located in the equator line, Indonesia exposed to sunlight throughout the year. Due to this, Indonesia's average UV index is relatively high.[2] Hence, it cause Indonesian at risk of having photoaging. Previous study conducted in Jakarta showed that 78 of 158 participant age 18 to 21 years old undergo premature aging.[5] This number potentially increased in later year if the antiaging product that easily accessed is not available. In order to minimize this process of photoaging, some plant extract proofed can be used preventive skin care, as these plant extract exhibit anti-photoaging properties.

Indonesia is known as an oil producing country. One of the most well-known commodities is patchouli, specifically Aceh patchouli. (*Pogostemon cablin*). Since ancient times, patchouli oil has been utilized in cuisine, cosmetics, fragrances, and pharmacy. According to the Indonesian Directorate General of Plantation in 2021, Aceh is the province with the main production of patchouli commodities in Indonesia, which accounts for 18.78% of the total production with an average production of 421.8 tons. The contents of patchouli essential oil are distributed thoroughly in all of its parts, including the stem, leaf, and root. This makes patchouli the biggest contribution to the country's plantation.[12] Through an increase in the concentration of antioxidant enzymes such glutathione peroxidase, superoxide dismutase, and catalase in the skin, previous in vivo investigations demonstrated the antioxidative activity of patchouli extract. Additionally, patchouli oil has photoprotective qualities.[7]

Indonesia is not only known as the world's largest producer of patchouli oil, but it has been designated as the world's fourth largest exporter of coffee. Therefore, coffee production in Indonesia is very high. However, the waste generated from this coffee production is of course also large. The coffee waste produced is coffee peel extract pulp, mucilage, parchment, and silverskin that can be harmful for the environment.[8] Arabica coffee is produced in the Gayo Highlands in the districts of Aceh Tengah and Bener Meriah, Aceh Province. According to the Aceh Statistics Agency in 2019, the two districts produced 64,177 tons of coffee per year and this coffee commodity serves as the main source of regional income.[9]

Coffee peel contains nutrients and compounds that can be beneficial for health. According to previous research, arabica Gayo coffee peel extract has high antioxidant levels. It is also antioxidative, antidiabetic, and anti-inflammatory. Additionally, prior studies demonstrated that the arabica Gayo coffee peel extract can preserve the thickness and density of collagen when exposed to ultraviolet B (UVB) light.[15]

Based on the previous studies mentioned, patchouli extract and coffee peel extract have antioxidative activity. Antioxidant compounds work by inhibiting the formation of ROS or forming more stable radical compounds by forming intramolecular hydrogen bonds and oxidation.[12] Antioxidants, therefore, are crucial in reducing or inhibiting photoaging symptoms including the appearance of fine lines and the development of hyperpigmentation spots by reducing oxidative stress on the skin brought on by UV exposure.[13]

Because of the abundant availability of patchouli oil and arabica Gayo coffee peel, especially in Aceh, the author is interested in examining the effect of patchouli extract and arabica Gayo coffee peel extract combination cream on skin aging. The aim of this study is assessing the effectiveness of a combination cream of Aceh patchouli extract 1% and arabica Gayo coffee peel extract in preventisng changes in collagen and elastin due to photoaging.

METHODS

Aceh patchouli was obtained from South Aceh. The part of the patchouli plant used is terna, which is all parts of the plant that are above ground level. While the arabica coffee plants grown in Gayo Lues, Aceh, produced the arabica Gayo coffee peel extract. The coffee peel extract was made with 96% ethanol solvent. Arabica Gayo coffee peel extract and Aceh patchouli extract were combined and processed into cream in oil-in-water dosage form with extract concentrations of 10%; 12.5%; and 15%. Macroscopic skin observations were made through the observation of photos of the skin of the rat's back photographed with a camera during the treatment period for 6 weeks. Using Masson-Trichrome staining, a histological analysis was conducted to measure the density and thickness of elastin and collagen. This research has been through the Ethics Committee review in Faculty of Veterinary Medicine of Syaiah Kuala University Banda Aceh, Indonesia (No.212/KEPH/2023).

The Syiah Kuala University Faculty of Veterinary Medicine's Animal Laboratory handled the treatment of the test animals. The test animals used were male Wistar rats *(Rattus norvegicus)*. The rats that were collected were 2-3 months old and weighed between 150 and 200 grams. The exclusion criteria for

the sample were rats that sick and died during the process of research. Rats were grouped into 5 groups: PP, PN, P1, P2, and P3. PP is a positive control group of rats treated with 10% vitamin C cream. PN is a negative control group treated with a base cream or cream without any active ingredients. P1 is a group of rats given a 10% concentration of patchouli and coffee peel extract as a combination cream. P2 is a group of rats that received a 12.5% concentration of patchouli cream and coffee peel extract. P3 is a group of rats given a 15% dosage of patchouli cream along with coffee peel extract. To calculate the minimal sample size in each group, The Federer formula [(n – 1) (t – 1) ≥ 15] was used according to the number of groups that was used in this research (5 groups). As the result of the calculation, each of the group consist of 6 rats. Hence, in total, there were 30 rats used in this research. Rats with their backs shaved to a size of 4 x 4 cm were given the cream. The cream was given at a dose of 0.05 mg/cm2 in the morning before irradiation and 4 hours after irradiation. UVB radiation was applied once day, three times a week. For six weeks, the total dose of radiation was 1,020 mJ/cm^2. Twenty-four hours following the second cream application, macro photos were obtained. The ethics committee gave its approval for this study's use of experimental animals at the Faculty of Veterinary Medicine, Syiah Kuala University, with reference number 212/KEPH/V/2023.

The UVB lamp used during this investigation is a Kernel KN-4003 UVB lamp. For six weeks, each group was subjected to the same UVB light three times per week. Every group was exposed to a low dose of 50 mJ/cm^2 during the first week. Every group was exposed to a low dose of 60 mJ/cm^2 during the second week. Every group received an average dose of 70 mJ/cm^2 during the third week. During the fourth week, a low dose of 80 mJ/cm^2 was administered to each group. During the fifth and sixth weeks, a mild dose of 90 mJ/cm^2 was administered to each group. For a period of six weeks, the rats received a cumulative dose of 1,020 mJ/cm^2.

The dorsal skin of the rats was photographed using a camera during the 6-week treatment period. Photographs were taken before irradiation and the first application (before treatment) and 24 hours after the second application of the cream (after treatment). The cream residue on the rat's back was removed to see if there was any edema or irritation arising on the skin of the rat's back. Observation of alterations in the rat's skin condition, including erythema and wrinkles was performed. The primary irritation index (PII) was used as the basis for evaluating criteria for changes in the skin condition of rats. The value is computed by multiplying the total number of observations by the number of rats, which is then divided by the total number of edema and erythema scores in the observation. According to the PII classification, there are four categories: mild irritation (0.5 - 1.9), moderate irritation (2.0 - 4.9), severe irritation (5.0 - 0.8), and no irritation or minor (0.0 - 0.4). Table 1 displays the PII table 1.

Table 1. Primary Irritation Index (PII)

Components	Scoring
Erythema Formation	
No Erythema	0
Very Slight Erythema (Barely visible)	1
Well-defined Erythema	2
Moderate to Severe Erythema	3
Severe Erythema (Visible Scar Formation)	4
Edema Formation	
No Edema	0
Very Slight Edema (Barely Visible)	1
Slight edema with raised margins (1 mm high)	2
Moderate edema with raised margin (approximately 1 mm)	3
Severe edema with raised margin (>1 mm)	4

The mice were euthanized after all treatments were completed. The euthanasia method used was cervical dislocation. The skin of the rat's back was excised 1x1 cm in depth reaching the muscle layer. The skin samples were transported to the Syiah Kuala University Faculty of Veterinary Medicine's Histology

Laboratory for macroscopic observation, Masson-Trichrome staining, and histological processing. Histological preparations of rat skin were photographed with a camera connected to a microscope. The results of rat skin preparation images were analyzed with the FIJI ImageJ application to measure the thickness and density of collagen and elastin.The hypothesis of this study is that application of combination of Aceh patchouli extract cream and arabica Gayo coffee peel extract able to increase the thickness and density of skin collagen and decrease the thickness and density of skin elastin. In order to verify the hypothesis, tests of comparative analysis were performed using the One-Way ANOVA and Kruskal-Wallis test. For both tests, a p-value of less than 0.05 was deemed significant for all values**.** SPSS program was used for data analysis.

RESULT

After treatment, none of the groups developed erythema or edema, suggesting there was no primary irritation. The cream formulation was categorized as having no irritation (not substantial) based on the primary irritation index (PII). Table 2 displays the PII assessment's findings.

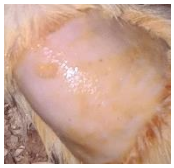
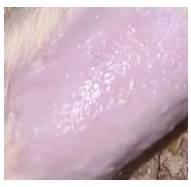
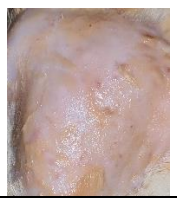
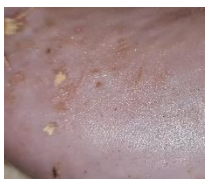
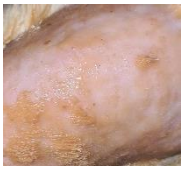
Table 2. The results of PII Scoring (n=30)

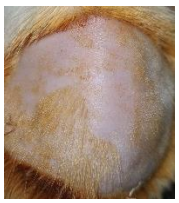
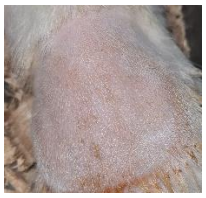
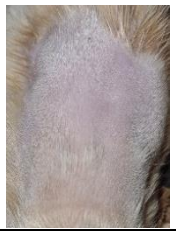
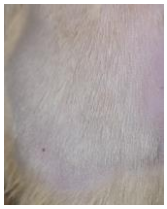
Group	PII Score		PII Score	PII Classification
	Erythema	Edema		
PP	0	0	0	No irritation (minor)
PN	0	0	0	No irritation (minor)
P1	0	0	0	No irritation (minor)
P2	0	0	0	No irritation (minor)
P3	0	0	0	No irritation (minor)

Description :
PP : Positive control group (10% vitamin C cream and UVB irradiation)
PN : Negative control group (Basic cream and UVB irradiation)
P1 : Treatment group 1 (Combination cream of Aceh patchouli extract and 10% arabica Gayo coffee peel extract and UVB irradiation)
P2 : Treatment group 2 (Combination cream of Aceh patchouli extract and 12.5% arabica Gayo coffee peel extract and UVB irradiation)
P3 : Treatment group 3 (Combination cream of Aceh patchouli extract and 15% arabica Gayo coffee peel extract and UVB irradiation)
PII : Primary Irritation Index

Macroscopic observations revealed erythema and wrinkles in every group, especially during the first week following UVB light exposure. In the second to sixth week, the PP, P1, P2, P3 groups experienced a decrease in the formation of erythema and wrinkles despite being exposed to UVB light. Macroscopic photos of mice can be viewed in Table 3.

Table 3. Macroscopic photos of rat's skin before and after treatment

Observation Time	Group				
	PP	PN	P1	P2	P3
Before Treatment					

After Treatment					

Description :
PP : Positive control group (10% vitamin C cream and UVB irradiation)
PN : Negative control group (Basic cream and UVB irradiation)
P1 : Treatment group 1 (Combination cream of Aceh patchouli extract and 10% arabica Gayo coffee peel extract and UVB irradiation)
P2 : Treatment group 2 (Combination cream of Aceh patchouli extract and 12.5% arabica Gayo coffee peel extract and UVB irradiation)
P3 : Treatment group 3 (Combination cream of Aceh patchouli extract and 15% arabica Gayo coffee peel extract and UVB irradiation)

The development of wrinkle formation can be inhibited by the application of cream with active ingredient. The negative control showed increased wrinkle formation compared to the other group. The group that received combination of arabica Gayo coffee peel extract and Aceh patchouli extract cream show less wrinkle compared to the group that receive vitamin C cream. To confirm this morphology result, further examination of the microscopic changes was conducted in these groups.

The study used the Kruskal-Wallis method to assess the collagen thickness of the subjects. The test results showed that the collagen thickness in each of the five treatment groups varied significantly. The P3 group exhibited the maximum collagen thickness, whereas the PN group displayed the lowest collagen thickness. The Kruskal-Wallis test has a p value of 0.000. This indicates that the thickness of the collagen varies significantly throughout groups. Figure 4 displays data of collagen thickness from all treatment groups.

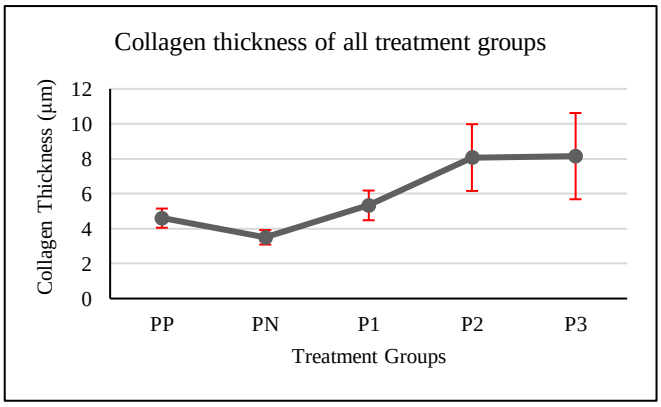


Figure 4. Collagen thickness of all treatment groups.

Description :
PP : Positive control group (10% vitamin C cream and UVB irradiation)
PN : Negative control group (Basic cream and UVB irradiation)
P1 : Treatment group 1 (Combination cream of Aceh patchouli extract and 10% arabica Gayo coffee peel extract and UVB irradiation)
P2 : Treatment group 2 (Combination cream of Aceh patchouli extract and 12.5% arabica Gayo coffee peel extract and UVB irradiation)
P3 : Treatment group 3 (Combination cream of Aceh patchouli extract and 15% arabica Gayo coffee peel extract and UVB irradiation)

The Kruskal-Wallis method was used to analyze the collagen density results. The test results showed that the collagen density of the five treatment groups varied significantly from one another. It was shown that the P3 group had the highest collagen density, whereas the PN group had the lowest. The p-

value results of the Kruskal-Wallis test was 0.000. This indicates that the collagen density varies significantly throughout groups. Table 5 displays the data of collagen density from all treatment groups.

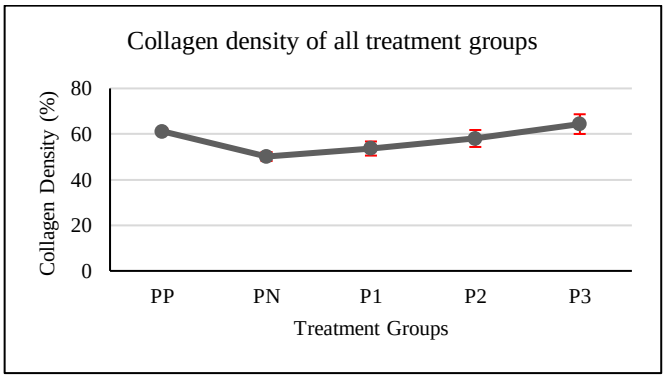


Figure 5. Collagen density of all treatment group.

Description:
PP : Positive control group (10% vitamin C cream and UVB irradiation)
PN : Negative control group (Basic cream and UVB irradiation)
P1 : Treatment group 1 (Combination cream of Aceh patchouli extract and 10% arabica Gayo coffee peel extract and UVB irradiation)
P2 : Treatment group 2 (Combination cream of Aceh patchouli extract and 12.5% arabica Gayo coffee peel extract and UVB irradiation)
P3 : Treatment group 3 (Combination cream of Aceh patchouli extract and 15% arabica Gayo coffee peel extract and UVB irradiation)

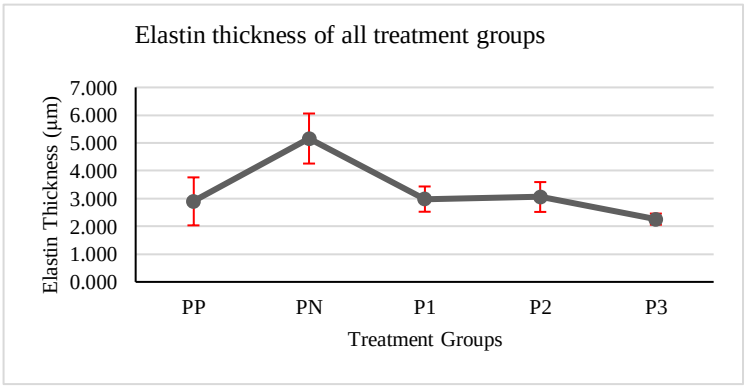


Figure 6. Elastin thickness of all treatment groups.

Description :
PP : Positive control group (10% vitamin C cream and UVB irradiation)
PN : Negative control group (Basic cream and UVB irradiation)
P1 : Treatment group 1 (Combination cream of Aceh patchouli extract and 10% arabica Gayo coffee peel extract and UVB irradiation)
P2 : Treatment group 2 (Combination cream of Aceh patchouli extract and 12.5% arabica Gayo coffee peel extract and UVB irradiation)
P3 : Treatment group 3 (Combination cream of Aceh patchouli extract and 15% arabica Gayo coffee peel extract and UVB irradiation)

The one-way ANOVA approach was utilized to analyze the elastin thickness results. The test results showed that the five treatment groups' collagen densities varied significantly from one another. With respect to elastin thickness, the PN group had the highest value, whereas the P3 group had the lowest. The One-way ANOVA test yielded a p value of 0.000. This indicates that the elastin thickness varies significantly throughout groups. Figure 6 displays the results of elastin thickness from all treatment groups.

Data on elastin density were examined using the Kruskal-Wallis technique. The elastin density of the five treatment groups did not significantly differ,

according to the test results. The PN group had the highest density of elastin, while the P3 group had the lowest density of collagen. The Kruskal-Wallis test yielded a p value of 0.842. This indicates that the elastin density does not significantly differ between the groups. Table 7 displays data of elastin density from all treatment groups.

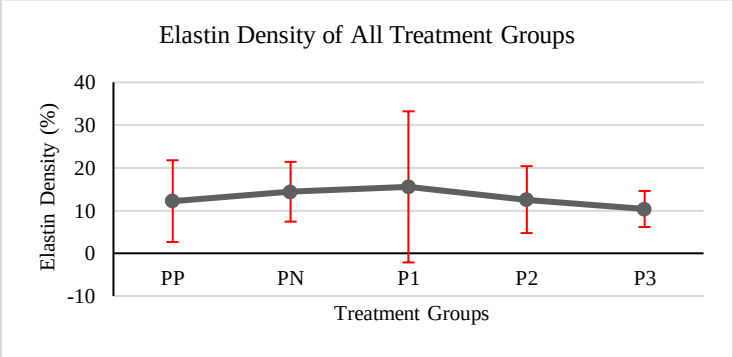
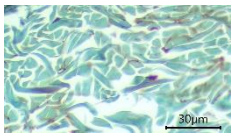
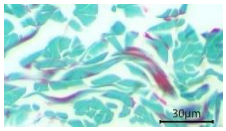
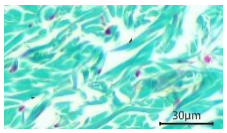
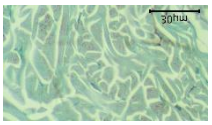
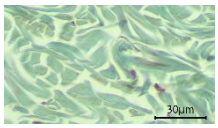


Figure 6. Elastin Density of All Treatment Groups.

Description :
PP : Positive control group (10% vitamin C cream and UVB irradiation)
PN : Negative control group (Basic cream and UVB irradiation)
P1 : Treatment group 1 (Combination cream of Aceh patchouli extract and 10% arabica Gayo coffee peel extract and UVB irradiation)
P2 : Treatment group 2 (Combination cream of Aceh patchouli extract and 12.5% arabica Gayo coffee peel extract and UVB irradiation)
P3 : Treatment group 3 (Combination cream of Aceh patchouli extract and 15% arabica Gayo coffee peel extract and UVB irradiation)

Table 4. Histological features of PP, PN, P1, P2, P3 Rat's skin. Masson-Trichrome stain. 400x magnification.

PP	PN	P1	P2	P3
				

Description :
PP : Positive control group (10% vitamin C cream and UVB irradiation)
PN : Negative control group (Basic cream and UVB irradiation)
P1 : Treatment group 1 (Cream combination of 10% arabica Gayo coffee peel extract and Aceh patchouli extract and UVB irradiation)
P2 : Treatment group 2 (Cream combination of 12.5% arabica Gayo coffee peel extract and Aceh patchouli extract and UVB irradiation)
P3 : Treatment group 3 (Cream combination of 15% arabica Gayo coffee peel extract and Aceh patchouli extract and UVB irradiation)

DISCUSSION

Rats which were given a cream containing 1% Aceh patchouli and arabica Gayo coffee peel extract did not exhibit erythema or edema on their back skin. This suggests that the cream is safe. With a pH range of 6.7 to 7.9, the cream base utilized in all groups was prepared in compliance with the Indonesian National Standard (SNI) reference no. 16-4399-1996. All ingredients used in this cream also refer to Indonesian standards. Chemical ingredients often used in cream formulations such as polyethylene glycol can trigger irritation and inflammation. These artificial substances may be more harmful to the skin. A similar study reported that the use of a scrub cream formulated with Moringa seed (Moringa oleifera) did not trigger irritation reactions because the formulation has a pH similar to the physiological pH of the skin (pH 4.5 - 6.5). A similar study also reported that the use of

arabica Gayo coffee peel extract in rats did not cause significant inflammatory signs.[14]

During the first week, wrinkles started to appear on the rat's skin. In the second week until the last day of treatment, the wrinkles on the rats of PP, P1, P2 and P3 group decreased. The wrinkles can be reduced due to the antioxidative traits that vitamin C has. It can prevent dermal collagen and elastin degradation so that wrinkles on the skin can be minimized. In addition to antioxidants properties like tannin, quinone, polyphenol, triterpenoid, and patchouli alcohol, the coffee peel extract and patchouli extract used in this study also contain antioxidant compounds that can stop dermal damage caused by reactive oxygen species, which are released when exposed to UVB light.[14]

Reactive oxygen species (ROS) can be produced at higher rates in cells in the extracellular matrix (ECM) when UVB radiation is applied repeatedly and chronically. ROS have the ability to activate cellular signaling pathways, including AP-1, MAPK, and NF-κβ, which in turn can initiate an inflammatory process. Inflammation can cause activation and infiltration of macrophage cells that are able to release MMP. MMP can degrade ECM, including collagen. Collagen degradation by MMP is proceeded in two phases. In the first phase, collagen fiber will be fragmented. In the second phase, collagen wil be phagocitized by macrophage and will be further degraded into amino acid and peptide chains. These two phases can cause collagen fiber thinning and weakening. Collagen degradarion by MMP can also decrease the level of collagen and the density of collagen.[16]

AP-1 activation can be inhibited by vitamin C that leads into the inhibition of collagen degradation chain reaction. This theory supports the findings in the study. The PP group, which was treated with 10% vitamin C has the second highest collagen thickness and density. However, previous study stated that polyphenol compounds in plants has higher antioxidative activity effectivity than vitamin C.[18]

Arabica Gayo coffee peel contains antioxidant compounds that has potential in photoprotective activity. Antioxidant compounds that were found in arabica Gayo coffee peel were tannin, falvonoid, quinone, polyphenol, and triterpenoid. These antioxidative compounds work by neutralizing ROS and reduce radical compounds into a more stable chemical compund, so the free radical chain reaction can be prevented. These compounds can also inactivate ROS catalytic enzyme, preventing the formation of ROS. It also eliminates ROS and repair the damage that has been done due to ROS.[20]

Antioxidant compounds are also found in patchouli extract. The most abundant antioxidant compound that contained in patchouli is patchouli alcohol (PA). patchouli alcohol prevents ROS formation by oxidizing hexanal compounds into hexanoic acid. According to a study by Isnaini in 2022, patchouli oil also has photoprotective activity that can maintain skin structural integrity by acting as an antioxidant. [28]

With antioxidative traits contained in coffee peel extract and patchouli, collagen fibers in dermis can be protected from collagen degradation, thus the thickness and the density of dermal collagen is maintained. This supports the findings in this study. The highest group that has highest collagen thickness and density is P3.

Elastin degradation is caused due to the increase in ROS level in the skin that is triggered by UVB light exposure. This can trigger macrophage cells, fibroblast, keratinocyte, and other cells to produce MMP enzyme as a response to oxidative stress. MMP is an enzyme that functions as degrader collagen and elastin. A study that has been done by Imokawa in 2015 stated that elastin fibers that has been exposed to UVB repetitively underwent remodelling and structural changes.[21] The findings in this study are consistant with the previous study findings. The group that has the highest elastin thickness and density is PN, which was treated with basic cream that has no active ingredient.

Vitamin C has been shown to prevent the skin's elastin from being biosynthesised in an earlier in vitro investigation. An in vitro study that was done by Phillips et al. in 1994 showed that ascorbic acid supplementation can stabilize collagen mRNA in and reduce elastin production by fibroblast.[22] The theory and findings can explain why PP group elastin thickness and density is lower than PN group.

A similar research studied how a polyphenol compound from a plant affects elastin that has been damaged due to ROS. A study by Imokawa et al. in 2015 showed thath Zingiber officinale (L.) extract can inhibit the production of elastase (MMP-12) in fibroblast. Another similar study by Thring et al. also shows that polyphenol compounds from 21 plants were anti-elastase, and the highest anti-elastase activity was in green tea extract. A research by widowati et al. in 2020 showed that mangosteen peel extract has high anti-elastase activity.[23] Those findings may be related with the reason why P3 has the lowest elastin

degradation that is marked with low elastin thickness and density in P3.

Previous animal study using patchouli oil only conclude that the application of patchouli oil can prevent aging induced by UV light.[24] Another study in China had similar result. The application of patchouli oil cream could maintain skin structural integrity in mice that was exposed to UVB light.[25] With the increasing demand of natural antiaging products, this research provide evidence of antiaging properties of arabica Gayo coffee peel extract and Aceh patchouli extract cream combination. Both of the main ingredients of this cream are readily available.

Regardless of these promising results, there were some limitations in this research. The administrated dose of UV exposure may be varied between mice as the mice actively moving in the process. In the process of cream application, the mice skin was unable to covered perfectly to prevent the skin contact after cream application with air. Currently, there is no specific staining method to observe elastin. All of these limitations have the potential to alter the result of this research.

Based on the study results indicate that cream containing Aceh patchouli extract and 15% arabica Gayo coffee peel extract can prevent collagen degradation, which is marked by the absence of collagen thickening and condensation. It also can prevent elastin degradation, which is proved by the maintenance of elastin thickness and density. Based on PII score, this combination cream does not cause any inflammatory effect on rat's skin.

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