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Purple Sweet Potato (*Ipomea batatas L.*) Cream Extract Increase Superoxide Dismutase (SOD) - Malondialdehyde (MDA) Ratio and Higher Collagen Expression in Rats (*Rattus novergicus*) Exposed to Ultraviolet B

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ABSTRACT

Introduction: Aging can occur in all parts of the body. Superoxide dismutase (SOD) enzyme is an enzyme that interacts with collagen, especially in types I and IV. UVB rays can trigger the presence of free radicals and lipid peroxidation, which form malondialdehyde (MDA) compounds. This study aimed to determine the effect of cream application of purple sweet potato (Ipomoea batatas L.) extract on increasing the ratio of SOD to MDA levels and increasing collagen in rats exposed to UVB.

Methods: This was experimental, using a post-test only with a control group design. The irradiation was carried out for 4 weeks with a total radiation dose of 840 mJ/cm2. Purple sweet potato cream concentration was 4%, 8%, and 16%. Rat blood was taken to check the ratio of SOD/MDA and collagen levels using the ELISA kit and Sirius red staining. This study used descriptive analysis and ANOVA using SPSS.25.sav.

Results: The highest ratio of SOD/MDA levels was found in the treatment group with an 8% purple sweet potato extract concentration of 3.346 ± 0.113. In the collagen expression, the highest percentage was found at 8% levels of 83,290 ± 1,885%. There was a significant mean difference between the treatment and control groups, with the treatment group having a higher result than the control group ($p<0.001$).

Conclusion: The administration of purple sweet potato cream extract could inhibit aging by increasing the ratio of SOD/MDA levels and collagen expression.

Keywords: *Ipomoea batatas L.*, SOD, MDA, collagen, aging process.

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INTRODUCTION

Aging is a natural process that can be found in all living creatures, and it is unavoidable. Aging can occur in all parts of the body, including the skin. Skin is the largest organ in the body, weighing 15% of the total body weight. Skin is one of the vital organs in the body that has a protective function against external physical, chemical, and biological exposure. Skin has a structure that has various functions, such as regulating temperature (thermoregulation) and regulating fluid loss in a person's body.[1] Skin aging can be classified into two factors, namely intrinsic and extrinsic. Ultraviolet light is one of the extrinsic factors that influences the aging process more quickly and can damage human skin tissue. The aging process due to exposure to light is often called photoaging.[2,3]

Aging is a process that occurs when tissues start to experience a slow decline in function. It is a condition that occurs due to several factors or is multifactorial. However, in general, skin aging is influenced by intrinsic factors, which are influenced by the chronological phenomena of the individual, such as increasing age. Generally, it progresses slowly, resulting in changes in the tissue structure of the skin. Meanwhile, extrinsic factors occur due to external environmental influences such as exposure to ultraviolet light or photoaging.[4,5]

Intrinsic skin aging is a condition that occurs synergistically and simultaneously. This process involves several stages, including the decreased ability of skin cell proliferation activity, decreased function of skin extracellular matrix synthesis, and the activity of collagen degrading enzymes in the skin layer. Increased dermis. Skin cells with vital roles, such as fibroblasts, keratinocytes, and melanocytes, decrease cell number as we age. A reduction in the number of these cells will cause changes in the tissue structure of the skin, where fibroblast cells experience a decrease in cell number, which will induce a decrease in collagen biosynthesis in the dermis layer of the skin.[3,4,6] Meanwhile, extrinsic factors that induce aging are influenced by temperature, individual diet and lifestyle, smoking habits, environmental pollutants, and infrared and ultraviolet radiation (UVR). Extrinsic skin aging is characterized by elastosis in the upper

dermis layer, damage to the fibril tissue, and moderate inflammatory infiltrates.[7]

These two factors influence the aging process and can cause a decrease in structural integration and morphological changes in the skin so that the skin becomes increasingly susceptible to developing skin diseases, from benign conditions to malignancies. Looking at the clinical signs of aging that are visible on the skin, previous studies show that there are skin changes that can be felt when aging, including irregular skin pigmentation, telangiectasis, wrinkles, dry skin, atrophy, and a decrease in the thickness of the dermal and epidermal layers.[7]

Ultraviolet radiation, whether emitted naturally from the sun or artificial sources, is a substance that can cause changes in the skin. Ultraviolet rays are present in the environment in abundant amounts and can cause skin aging and lead to malignancies. Based on the electromagnetic spectrum, ultraviolet light has varying wavelengths and is classified into three categories: UVA, UVB, and UVC.[8] UVC rays are rays with the shortest wavelength (100-290 nm). The ozone layer completely absorbs these rays, so they do not affect the skin. UVB rays with a wavelength of 290-320 nm are ultraviolet rays that can affect the skin's or epidermis's superficial layers and cause sunburn. Meanwhile, UVA rays, which have the largest wavelength among other ultraviolet rays, namely 320-400 nm, are known to have minimal effects on the skin, but because of the large wavelength, their penetration ability penetrates the skin longer when compared to other ultraviolet rays (around 20% at a wavelength of 365 nm).[9]

Previous studies found that UVB rays are one of the causes of skin malignancies, where UVB radiation has a carcinogenic effect three to four times higher than UVA.[9] UVB rays can cause skin inflammation through the induction of cytokines and several vasoactive and neuroactive mediators in the skin, which trigger an inflammatory response and cause sunburn.[8] If there is chronic light exposure, ultraviolet light can also form reactive oxygen species (ROS), which can cause a faster-aging process.[2] UV light can induce mutations through the role of ROS such as superoxide anions, hydrogen peroxide, and hydroxyl radicals where oxidation of nucleotide bases can cause mispairing and cause mutagenesis.[8]

Cells have complex mechanisms related to the detoxification of ROS to prevent free radical changes into macromolecules and DNA. Catalase is one of the antioxidant enzymes known to detoxify hydrogen peroxide, where superoxide dismutase (SOD) can deactivate superoxide anions.[8] The SOD enzyme is an antioxidant enzyme that has a role in capturing superoxide radicals. Previous studies have shown that SOD interacts with collagen, especially types I and IV, which can protect and inhibit collagen-mediated wounds.[10]

UVB rays that induce ROS can later trigger the presence of hydroxyl and hydroperoxyl radicals. Changes in lipid peroxidation occur through a series of stages, such as a cyclization reaction, which will later form the compound malondialdehyde (MDA). This compound is a highly mutagenic product of lipid peroxidation and is known for its role as a biological marker of lipid peroxidation of omega-3 and omega-6 fatty acids. An increase in MDA in body cells can result in functional and biochemical changes in the body and cellular degradation, which can also affect cell death.[2]

Aging is also indicated by a decrease in collagen expression in the body. Collagen degradation is one of the central changes visible in the skin, where this condition can be exacerbated by the activation of matrix metalloproteinase (MMPs). Previous studies showed that the expression of collagen and SOD is related to the aging process in living creatures, where procollagen I mRNA expression increased in young extracellular SOD transgenic mice.[10] Collagen is the largest part of the dermis layer, which occupies 70% of the skin mass. Collagen has a vital role in the skin; if its amount decreases in the body or is damaged, it can cause changes in the morphology and structure of the skin, resulting in clinical signs such as wrinkles, sagging skin, and decreased skin elasticity. Aging, which then causes ROS, also affects collagen production, where ROS can inhibit collagen synthesis.[3]

Until now, research on the SOD to MDA levels ratio is still very limited. The ratio of MDA to SOD levels is one of the indicators to be considered in oxidative stress conditions.[11] Oxidative stress is a condition indicated by an imbalance between free radicals and the antioxidant regulatory system in an individual's body. Free radicals can trigger lipid peroxidation, producing an unstable final product, malondialdehyde (MDA). Lipid peroxidation in the body can disrupt cell membrane permeability, increasing the risk of cell death.[12]

In oxidative stress conditions, MDA in the blood will influence the reduction in SOD levels, where SOD itself neutralizes tissue damage.[11,12] To reduce the negative effects of aging, currently, many modalities are being developed using natural raw materials that use plants, one of which is purple sweet potato (*Ipomoea batatas L.*). Purple sweet potatoes contain flavonoid compounds, namely anthocyanins, which are components rich in antioxidants that can prevent premature skin aging.[13] Compared with other sweet potato types, purple sweet potato has the highest anthocyanin content (110.51 mg/100g). Apart from that, the availability of purple sweet potatoes in Indonesia reaches 2.3 million tons per year, so they are abundant. They can be used for various needs, such as producing extracts. Previous studies extracting sweet potatoes showed average anthocyanin levels of 521.84-729.74 mg/100g. Meanwhile, the total phenol content of ethanol extract of purple sweet potato skin ranged between 4785.71-5134.92 ppm GAE (Gallic Acid Equivalent).[14]

Anthocyanin is an antioxidant compound that can inhibit ROS formation by capturing free radicals formed due to exposure to ultraviolet light. This compound cuts the chain oxidation reaction of free radicals such as superoxide, hydrogen peroxide, and hydroxyl radicals.[13] Another study shows that the anthocyanin content in sweet potatoes can suppress lipid peroxidation, which is associated with cellular and tissue level oxidative damage due to free radicals, which was evaluated using malondialdehyde (MDA) levels in the blood.[15] Other studies also show that purple sweet potatoes can increase the amount of dermis collagen in types I and III when given in topical preparations.[13,16]

Ipomoea batatas L. has been said to be able to suppress MDA and increase collagen synthesis associated with aging due to UVB light exposure, but previous studies discussing similar topics are still very limited. On the other hand, topical use of extracts is said to have several advantages, including practical use and good penetration and stability.[17] Therefore, this study aimed to determine the effect of application of purple sweet potato (*Ipomoea batatas L.)* extract cream on increasing the ratio of SOD to MDA levels and increasing collagen expression in mice exposed to UVB. Currently, several studies related to herbal products are continuing to be carried out due to findings showing that they contain anthocyanins, which have high antioxidant activity, so that they can prevent the aging process of the skin. Based on this, this study aimed to determine the effect of topical administration of purple sweet potato extract in reducing the aging process in mice exposed to UVB light.

METHODS

Study Design

This experimental study uses a post-test only with a control group design, which is analytical descriptive. Research and data collection were only carried out at the end of the treatment so that no comparisons were made between before and after the treatment was carried out.

Data Collection

The research was conducted in February 2022 – September 2022 at several locations. This research used experimental animals in the form of Wistar strain rats (*Rattus norvegicus*) obtained from the Integrated Biomedical Laboratory Unit, Faculty of Medicine, Udayana University and which met the inclusion criteria which included male Wistar strain white rats (*Rattus norvegicus*), healthy and without physical defects, aged 2 -3 months, and the rat's body weight is 100-150 grams. Meanwhile, exclusion criteria include dead mice before and during treatment and sick mice during treatment. Before treatment, 54 experimental animals were given standard care. Then, random sampling was carried out and put into five groups, namely the negative control group (K-), negative control group (K1), UVB control group (K2), UVB control with base cream (placebo) (K3), treatment group with purple sweet potato extract with a concentration of 4% (P1), treatment group with purple sweet potato extract with a concentration of 8% (P2), treatment group with purple sweet potato extract with a concentration of 16% (P3). The negative control group did not receive any intervention. Treatment groups 1, 2, and 3 were exposed to UVB light and smeared with purple sweet potato extract cream (4%, 8%, and 16%). The mice's blood was examined using an ELISA kit and Sirius red staining to check the SOD/MDA ratio and collagen levels.

Data Analysis

Data were analyzed using SPSS for Windows version 25.0 software. Test data normality with the Shapiro-Wilk Test. One-way ANOVA is used to investigate if the data distribution is normal. Followed by post hoc analysis to determine significant comparisons between the two groups. If the data is not normally distributed (p-value<0.05), then Kruskal-Wallis analysis is carried out. The p-value is interpreted as significant if p-value<0.05.

RESULTS

Histological description of collagen tissue in the skin of experimental animals

Based on the results of reading the picro-sirius red histochemical staining preparations, there was a picture of denser and thicker collagen tissue in the treatment group compared to the negative control group, as indicated by the difference in the amount of non-collagen tissue (yellow) and collagen (red). In addition, the collagen tissue in the treatment group was dark red compared to the control group, which was brighter red, indicating a denser collagen structure. The collagen network in the positive control group (K1+ and K2+) seemed less structured and less dense than the other test groups. The picture of picro-sirius red histochemical staining in each test group is shown in Figure 1.

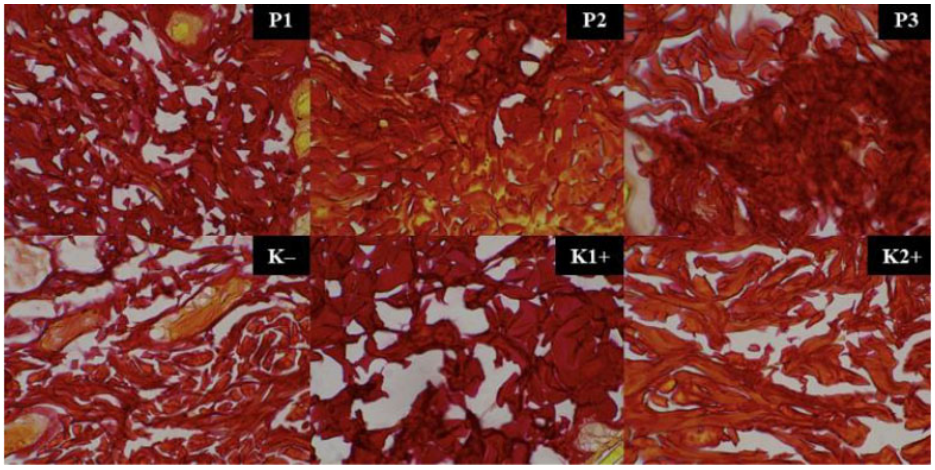


Figure 1. Histological image of collagen tissue (red arrow) in experimental animals using Sirius red staining.

Table 1. Phytochemical Results of Purple Sweet Potato Cream Extract

Anthocyanin (mg/100g)	IC50(ppm)
7.1669	194.26

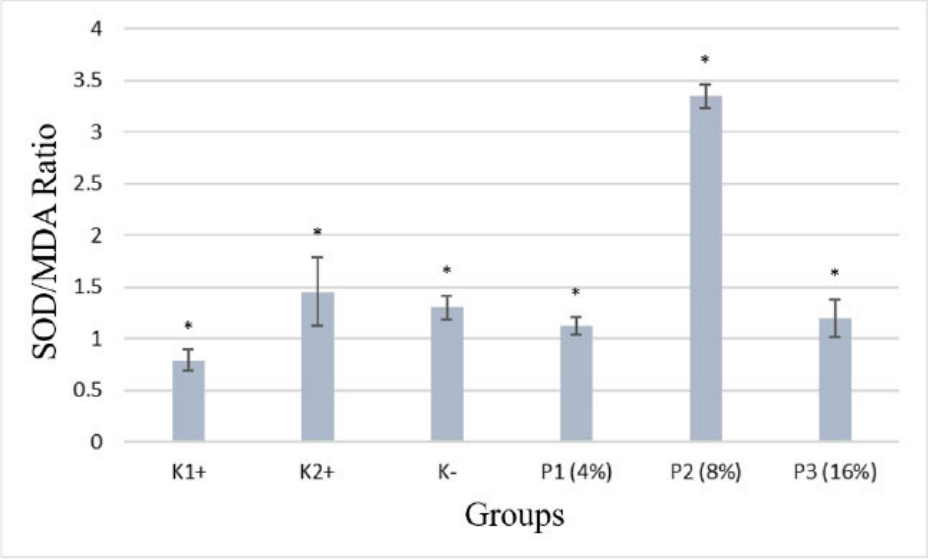
Table 2. *Shapiro-Wilk* Test of SOD/MDA ratio

No.	Group	p-value
1.	Negative control (K–)	0.210
2.	Positive control 1 (K1+)	0.451
3.	Positive control 2 (K2+)	0.122
4.	Intervention 1 (P1)	0.369
5.	Intervention 2 (P2)	0.798
6.	Intervention 3 (P3)	0.273

Table 3. ***Mean Differences in SOD/MDA Levels***

Group	N	Mean ± SD	p-value
K1+	9	0.820 ± 0.093	<0.001*
K2+	9	1.408 ± 0.281	
K-	9	1.236 ± 0.107	
P1 (4%)	9	1.155 ± 0.088	
P2 (8%)	9	3.391 ± 0.116	
P3 (16%)	9	1.187 ± 0.149	

*Analysis was carried out using One-Way ANOVA. Results were considered significant if p-value < 0.05.



*Significant differences between test groups

Figure 2. Differences in the Mean Ratio of SOD/MDA Levels in Each Group of Experimental Animals.

Table 4. ***Post hoc* test of LSD SOD/MDA ratio in Animals**

	Mean Differences	95% CI		p-value
		Minimum	Maximum	
K1+ vs K2+	0.587	-0.733	-0.441	<0.001*
K1+ vs K-	0.415	-0.562	-0.269	<0.001*
K1+ vs P1	0.335	-0.481	-0.188	<0.001*
K1+ vs P2	2.571	-2.717	-2.424	<0.001*
K1+ vs P3	0.366	-0.512	-0.220	<0.001*
K2+ vs K-	0.171	0.025	0.318	0.022*
K2+ vs P1	0.252	0.106	0.398	0.001*
K2+ vs P2	1.983	-2.129	-1.836	<0.001*
K2+ vs P3	0.221	0.074	0.367	0.004*
K- vs P1	0.081	-0.065	0.227	0.272
K- vs P2	2.155	-2.301	-2.008	<0.001*
K- vs P3	0.049	-0.096	0.195	0.500
P1 vs P2	2.236	-2.382	-2.089	<0.001*
P1 vs P3	0.031	-0.177	0.114	0.668
P2 vs P3	2.204	2.058	2.350	<0.001*

Phytochemical Results of Purple Sweet Potato Cream Extract

In this study, an intervention was carried out using purple sweet potato extract after mice were given treatment for 4 weeks. Providing purple sweet potato extract with concentrations of 4%, 8% and 16%. Apart from checking the SOD and MDA parameters as markers to determine the effectiveness of purple sweet potato cream, the anthocyanin levels in purple sweet potatoes were reviewed in this study. The results of the phytochemical analysis showed that the anthocyanin content was 7.1669 mg/100g with IC50=194.26 (Table 1).

The measurement SOD/MDA ratio

Normality Test

Bivariate analysis used One Way ANOVA comparative analysis to determine differences in mean SOD/MDA levels between test groups. Before carrying out the One-way ANOVA test, parametric assumption tests are first carried out, namely the normality and data homogeneity tests. However, if the results of the normality test and homogeneity test show that the data is not normally distributed or homogeneous, then an alternative test, namely the Mann-Whitney test, can be used.

The normality test is used to determine whether the data used is normally distributed or not normally distributed. The normality test in this study used Shapiro-Wilk (number of samples per group <50). The data normality test results showed that the SOD/MDA ratio distribution in the test group is normal (p-value>0.05), as described in Table 2.

Homogeneity Test

The homogeneity test is the second stage that must be carried out in bivariate analysis. This stage is carried out to determine whether the data is homogeneous or inhomogeneous. The data homogeneity test in this study used Levene's test. The results of the homogeneity of variance test for the SOD/MDA ratio data show that the data is homogeneous (p-value=0.208), so the One-Way ANOVA test will be used and continued with Post Hoc LSD analysis.

Test for Differences in Mean SOD/MDA Levels

Based on the results of analysis using One-Way ANOVA, it was found that there was a statistically significant difference in the mean ratio of SOD/MDA levels between groups of experimental animals (p-value<0.05) (Table 1). An illustration of the differences in the ratio of SOD/MDA levels is shown in Figure 2. Next, to find

Table 5. Shapiro-Wilk test of Collagen Expression

No.	Group	p-value
1.	Negative control (K–)	0.070
2.	Positive control 1 (K1+)	0.448
3.	Positive control 2 (K2+)	0.455
4.	Intervention 1 (P1)	0.786
5.	Intervention 2 (P2)	0.158
6.	Intervention 3 (P3)	0.413

Table 6. Differences in Mean Percentage of Collagen Expression in Each Group of Experimental Animals

Group	N	Mean ± SD	p-value
K1+	9	74,846 ± 2,040	<0,001*
K2+	9	73,530 ± 1,633	
K-	9	63,501 ± 3,293	
P1 (4%)	9	68,046 ± 1,553	
P2 (8%)	9	83,834 ± 1,714	
P3 (16%)	9	78,441 ± 1,304	

*Analysis was carried out using One-Way ANOVA. Results were considered significant if p-value < 0.05.

out which groups had significant mean differences, a Post Hoc test was carried out using the LSD test, and the results were obtained as attached in Table 3.

Based on the results of the Post Hoc test, it was found that there were significant mean differences between several groups of experimental animals (p-value<0.05), as shown in Table 4.

The Measurement of Percentages of Collagen Expression

Normality Test

Bivariate analysis in this study used one-way ANOVA comparative analysis to determine the difference in the mean percentage of collagen expression between test groups. Before carrying out the One-way ANOVA test, parametric assumption tests are first carried out, namely the normality and data homogeneity tests. However, if the results of the normality test and homogeneity test show that the data is not normally distributed or homogeneous, then an alternative test, namely the Mann-Whitney test, can be used.

The normality test is used to determine whether the data used is normally distributed or not normally distributed. The normality test in this study used Shapiro-Wilk (number of samples per group <50). The results of the data normality test showed that the percentage distribution of collagen expression in the test group is normal (p-value > 0.05), as described in Table 5.

Homogeneity Test

The homogeneity test is the second stage that must be carried out in bivariate analysis. This stage is carried out to determine whether the data is homogeneous or inhomogeneous. The data homogeneity test in this study used Levene's test. The homogeneity of variance test results for the percentage of collagen expression data show that the data is homogeneous (p-value = 0.076), so the One-Way ANOVA test will be used and continued with Post Hoc LSD analysis.

Test for Differences in Mean Collagen Expression

Based on the results of analysis using One-Way ANOVA, it was found that there was a statistically significant difference in the mean percentage of collagen expression between groups of experimental animals (p-value<0.05) (Table 3). An illustration of the percentage of collagen expression is shown in Figure 3. Next, to find out which groups had significant mean differences, a Post Hoc test was carried out using the LSD test, and the results were obtained as attached in Table 6.

Based on the results of the Post Hoc test, it was found that there were significant mean differences between several groups of experimental animals (p-value<0.05), as shown in Table 7.

DISCUSSION

Exposure to ultraviolet (UV) light is one of the factors of extrinsic skin aging that can result in photoaging. In this regard, UVB rays can cause skin inflammation through the induction of cytokines and several vasoactive and neuroactive mediators in the skin, which trigger an inflammatory response and cause sunburn. Cells have complex mechanisms related to the detoxification of ROS to prevent free radical changes into macromolecules and DNA. Catalase is one of the antioxidant enzymes known to detoxify hydrogen peroxide, where superoxide dismutase (SOD) can deactivate superoxide anions. UVB rays that induce ROS can later trigger the presence of hydroxyl and hydroperoxyl radicals. Changes in lipid peroxidation occur through a series of stages, such as a cyclization reaction, which will later form the compound malondialdehyde (MDA). Apart from that, the expression of collagen has a vital role in the skin, where if the amount is reduced in the body or is damaged, it can cause changes in the morphology and structure of the skin.[8]

Anthocyanin is an antioxidant in various natural ingredients, including purple sweet potato (*Ipomoea batatas L.*). In this study, the anthocyanin content in purple sweet potatoes was 7.1669 mg/100g with IC50=194.26 ppm. However, several studies have different anthocyanin levels in purple sweet potatoes. A study examining purple sweet potatoes obtained in Saree, Aceh Besar (light purple sweet potatoes and dark purple sweet potatoes). Purple sweet potatoes with a deep color contain 61.85 mg/100 g of anthocyanin, 17 times higher than the anthocyanin content of light purple sweet potatoes of 3.51 mg/100 g. Deep purple sweet potato has antioxidant activity of 59.25%, greater than light purple sweet potato at 56.64%.[18] Another study found that the anthocyanin level in purple sweet potatoes taken from Sidikalang, North Sumatra, was 62.138 mg/100g.[19]

Inhibitory concentration (IC50) is a parameter commonly used to measure the

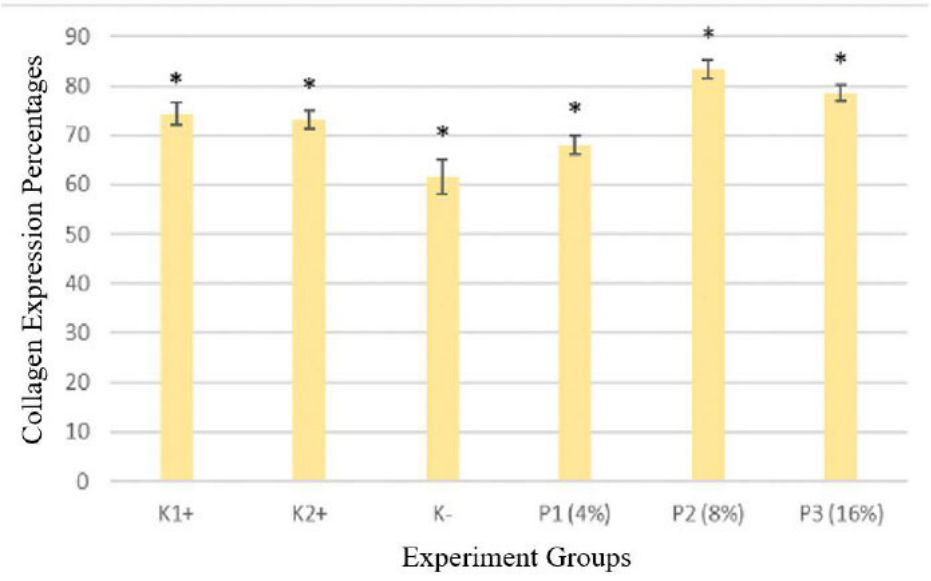


Figure 3. Differences in the Percentage of Collagen Expression in Each Group of Experimental Animals.

Table 7. LSD Post Hoc Test Percentage of Collagen Expression in Experimental Animals

	Mean Differences	95% CI		p-value
		Minimum	Maximum	
K1+ vs K2+	1.315	-0.608	3.239	0.176
K1+ vs K-	11.344	9.420	13.268	<0.001*
K1+ vs P1	6.800	4.875	8.724	<0.001*
K1+ vs P2	8.988	-10.912	-7.064	<0.001*
K1+ vs P3	3.595	-5.519	-1.671	<0.001*
K2+ vs K-	10.028	8.104	11.952	<0.001*
K2+ vs P1	5.484	3.560	7.408	<0.001*
K2+ vs P2	10.304	-12.228	-8.380	<0.001*
K2+ vs P3	4.911	-6.835	-2.987	<0.001*
K- vs P1	4.544	-6.468	-2.620	<0.001*
K- vs P2	20.333	-22.257	-18.409	<0.001*
K- vs P3	14.940	-16.864	-13.015	<0.001*
P1 vs P2	15.788	-17.712	-13.864	<0.001*
P1 vs P3	10.395	-12.319	-8.471	<0.001*
P2 vs P3	5.393	3.469	7.317	<0.001*

antioxidant activity of a material. IC50 is calculated by measuring the antioxidant concentration required to reduce the initial 2,2-diphenyl-1-picrylhydrazyl (DPPH) concentration to 50%. This causes a low IC50 to mean a material has high antioxidant activity and vice versa. The antioxidant activity is very strong if the IC50 value is less than 50 ppm. If it is in the range of 50-100 ppm, then the antioxidant activity is classified as strong; if it is in the range of 100-150 ppm, then the antioxidant activity is moderate; if the IC50 is in the range of 150-200 ppm, then the antioxidant activity is classified as weak, and if the IC50 value is more than 200 ppm, then the antioxidant is said to be very weak. In this study, the IC50 results for purple sweet potato (*Ipomoea batatas L.*) were 194.26 ppm.

The results of another study in Bali, which obtained purple sweet potatoes from the Tabanan Regency area, found that the anthocyanin level in the water extract was 146 mg/ml and in the 70% ethanol extract, it was 119 mg/ml.[15] Another study that evaluated the content of purple sweet potato using the high-performance liquid chromatography (HPLC) technique with linear gradient elution showed that it contains anthocyanins and anthocyanidins, which have antioxidant effects.[20] It is known that anthocyanins can reduce free radicals through single electron transfer reactions and through splitting hydrogen atoms from phenolic groups.[21] From the results of purple sweet potato anthocyanin levels in various studies, different results were obtained because there are several influencing factors, such as the origin of the purple sweet potato obtained and the age of the purple sweet potato. However, only a few studies contain this information.

Based on the results of the analysis in this study, it was found that there was a statistically significant difference in the mean ratio of SOD/MDA levels between groups of experimental animals. Based on the results of measurements and analyses that have been carried out, it was found that the highest increase in the ratio of SOD/MDA levels was found in the group of experimental animals given purple sweet potato extract cream at a concentration of 8%. The ratio of superoxide dismutase (SOD) to malondialdehyde (MDA) levels is defined as the ratio between superoxide dismutase (SOD) levels and malondialdehyde (MDA) levels.[22] Until now, studies that specifically examine the effect of cream application of purple sweet potato extract (*Ipomoea batatas L.*) in increasing the ratio of SOD to MDA levels in mice exposed to UVB are still very limited. The results of this study are supported by Kim et al.'s study evaluating changes in the intracellular and extracellular activity of three antioxidant enzymes, namely superoxide dismutase (SOD), guaiacol-type peroxidase (POD), and glutathione peroxidase (GPX) in suspension cultures of sweet potato (*Ipomoea batatas L.*) during cell growth. The study found that the extracellular activity of the three enzymes (SOD, POD, GPX) in the suspension culture medium of sweet potato (*Ipomoea batatas L.*) was much higher than the intracellular activity. In other words, there is an increase in the activity of the superoxide dismutase (SOD) enzyme.[23] Lu et al.'s study also found that the administration of anthocyanins significantly increased levels of superoxide dismutase (SOD) and catalase. At the same time, MDA and monoamine oxidase

(MAO) activity decreased significantly.[24]

Anthocyanins have high antioxidant activity, and one source of anthocyanins is purple sweet potato.[15] In an in vivo study by Zhi et al., MDA levels in the control group exposed to UVB light increased significantly by 31.02% compared with the normal control group not exposed to UVB. Interestingly, after receiving the purple sweet potato extract modality, the increase in MDA in the skin caused by UVB rays decreased significantly by 53.70% and 63.85-68.29% from initial levels (p-value<0.05).[25] Various studies have been conducted to test the effectiveness of purple sweet potatoes as a source of antioxidants that reduce MDA levels in various other pathological conditions, apart from skin aging/photoaging problems.[15,16]

Malondialdehyde (MDA) is useful as a marker of oxidative stress, while superoxide dismutase (SOD) is an antioxidant enzyme. Malondialdehyde (MDA) is the end product of lipid peroxidation and is often used to define oxidative stress. Superoxide dismutase (SOD) is an enzyme that converts superoxide anion radicals into hydrogen peroxide and molecular oxygen and plays an important role in controlling cellular ROS levels.[26] In other words, a higher ratio of SOD to MDA levels will have a better and more beneficial impact because it shows the body's protection against oxidative stress. A previous study by Kamal et al. examined blood superoxide dismutase (SOD) and plasma malondialdehyde (MDA) as indicators of lipid peroxidation in 97 workers exposed to asbestos and 42 healthy male controls. The study found that the MDA, SOD, and MDA/SOD ratios in workers exposed to asbestos were significantly higher than controls.[27]

Until now, no recent studies have shown the ratio of SOD to MDA levels. Previous studies conducted research separately, and no studies specifically calculated the ratio or ratio of SOD to MDA levels. This is also in line with Lu et al.'s study, which reported that the administration of anthocyanins was able to increase SOD levels and reduce MDA levels significantly.[24] The results of this study are also in line with the study of Zhi et al. which reported that after being given purple sweet potato extract, there was a significant decrease in MDA levels in the skin caused by UVB rays, namely 53.70% and 63.85-68.29% of the levels. Baseline (p-value<0.05).[25] Other research also shows the effectiveness of purple sweet potatoes as a source of antioxidants that reduce MDA levels in various conditions, including skin aging/photoaging problems.[15,16] Previous studies have been limited and only raised topics related to the relationship between the MDA and SOD ratio and other diseases. Previous studies showed an increase in the MDA/SOD ratio found in tissues in leprosy infection patients with PB and MB types when compared with the control group.[28]

Based on the results of the analysis in this study, it was found that there was a statistically significant difference in the average percentage of collagen expression between groups of experimental animals (p-value<0.001). Based on the results of the measurements and analysis that have been carried out, it was found that the highest increase in the collagen expression ratio was found in the group of experimental animals given purple sweet potato extract cream at a concentration of 8%. The results of previous research also showed differences in mean collagen expression between groups (p-value<0.05). Furthermore, in this study, type I and III collagen expression between the treatment and control groups showed significant differences (p-value<0.05). This condition can occur because MMP-1 activity is inhibited by purple sweet potato extract, so degradation of collagen expression is also inhibited. Increased MMP-1 activity can be influenced by UV-B radiation, which can affect the loss of collagen fibers in the skin. Continuous UV-B radiation can cause ongoing collagen degradation so that collagen formed disorganizedly will accumulate in aging skin due to photoaging.[13]

UVB radiation can have various effects, one of which is the formation of ROS. The accumulation of ROS in the body will trigger the activation of pro-inflammatory cytokines. It can further increase MMP-1 in fibroblast cells and cause a decrease in collagen in the dermis layer. ROS induction also influences the degradation of TGF-β, which also plays a role in reducing collagen production. The content of purple sweet potato, namely anthocyanin, has a role in inhibiting the formation of ROS caused by exposure to UV rays on the skin. The mechanism that is currently known is that anthocyanins play a role in cutting oxidation reactions from free radicals or can capture free radicals (free radical scavengers), such as hydrogen peroxide, superoxide, singlet oxygen, and peroxide. Furthermore, anthocyanins, which can inhibit the progression of ROS, can also influence collagen expression in the skin.[13,29]

The anthocyanins contained in purple sweet potato extract can influence the increase in collagen expression in the skin through inhibition of MMP-1. In this study, purple sweet potato extract was administered topically and showed significant differences in collagen expression in the experimental animal group. Like previous research, topical administration of purple sweet potato extract showed a significant difference between collagen expression in the experimental animal groups. This study further divided collagen types, both type I and type III collagen, which generally gave significant results between the experimental animal groups. This study also used different dose concentrations, where the highest dose of purple sweet potato extracts showed a high increase in collagen expression. This is in line with the principle that the amount of active substance absorbed by the skin per unit area of the skin surface is directly proportional to the concentration gradient of the active substance. Furthermore, previous research stated that topical administration of purple sweet potato extract could show a greater increase in the amount of collagen due to better MMP-1 inhibition in research using the purple sweet potato modality.[13]

This study can provide information regarding the effect of topical purple sweet potato extract on the SOD/MDA ratio and collagen expression in skin exposed to UV-B light using one of the true experimental methods, namely post-test-only control group design, which shows a significant effect. Through the differences between the experimental group and the control group. As for this study, the research limitations were the limitations of previous studies that discussed the SOD/

MDA ratio in more detail and identified other purple sweet potato components that could influence photoaging. However, this can be one of the components that can support the writing of this scientific work, where this research can also be one of the initial foundations for developing research on similar topics in the future.

CONCLUSION

This study showed that topical application of purple sweet potato (*Ipomoea batatas L.*) extract cream with concentrations of 4%, 8%, and 16% can inhibit the skin aging process in mice (*Rattus novergicus*) exposed to UVB by increasing SOD/MDA expression ratio and collagen expression increased significantly in the skin tissue of experimental animals compared to the control group. Further research needs to be carried out regarding the isolation of anthocyanin, flavonoid and other components from purple sweet potatoes, which can then be tested for their respective effectiveness against photoaging.

DISCLOSURE

Author Contribution

All authors have contributed to these research processes, including arranging the study design, data collection, data analysis and interpretation of the data, drafting of the article, critical revision of the article for important intellectual content, final approval of the article, and collection and assembly of data.

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Conflict of Interest

There is no conflict of interest for this manuscript.

Ethical Consideration

This research was approved by the Health Research Ethics Committee of Udayana University No: 1503/UN14.2.2.VII.14/LT/2022.

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REFERENCES

1. Yousef H, Alhajj M, Sharma S. Anatomy, Skin (Integument), Epidermis. In Treasure Island (FL); 2023.
2. Pratama GMCT, Hartawan IGNBRM, Indriani IGAT, Yusrika MU, Suryantari SAA, Sudarsa PSS. The potency of Spirulina platensis Extracts as Sunscreen on Ultraviolet B Exposure. J Med Heal. 2020;2(6):205–17. DOI: http://dx.doi.org/10.28932/jmh.v2i6.2484.
3. Zahruddin A. Penuaan Kulit: Patofisiologi dan Manifestasi Klinis (Skin Aging: Pathophysiology and Clinical Manifestation). Berk Ilmu Kesehat Kulit dan Kelamin. 2018;30(3):208–15.
4. Ahmad Z, Damayanti. Penuaan Kulit : Patofisiologi dan Manifestasi Klinis. Berk Ilmu Kesehat Kulit dan Kelamin – Period Dermatology Venereol [Internet]. 2018;30(03):208–15.
5. Zhang S, Duan E. Fighting against Skin Aging: The Way from Bench to Bedside. Cell Transplant. 2018;27(5):729–38. DOI: https://doi.org/10.1177/0963689717725755.
6. Hwang KA, Yi BR, Choi KC. Molecular mechanisms and in vivo mouse models of skin aging associated with dermal matrix alterations. Lab Anim Res. 2011 Mar;27(1):1–8. DOI: https://doi.org/10.5625/lar.2011.27.1.1.
7. Tsatsou F, Trakatelli M, Patsatsi A, Kalokasidis K, Sotiriadis D. Extrinsic aging: UV-mediated skin carcinogenesis. Dermatoendocrinol. 2012 Jul;4(3):285–97. DOI: https://doi.org/10.4161/derm.22519.
8. D'Orazio J, Jarrett S, Amaro-Ortiz A, Scott T. UV radiation and the skin. Int J Mol Sci. 2013;14(6):12222–48. DOI: https://doi.org/10.3390/ijms140612222.
9. Pandel R, Poljšak B, Godic A, Dahmane R. Skin Photoaging and the Role of Antioxidants in Its Prevention. ISRN Dermatol. 2013;2013:1–11. DOI: https://doi.org/10.1155/2013/930164.
10. Lee MJ, Agrahari G, Kim HY, An EJ, Chun KH, Kang H, et al. Extracellular Superoxide Dismutase Prevents Skin Aging by Promoting Collagen Production by Activating AMPK and Nrf2/HO-1 Cascades. J Invest Dermatol. 2021;141(10):2344-2353.e7. DOI: https://doi.org/10.1016/j.jid.2021.02.757.
11. Herdiani N, Wirjatmadi B, Adriani M. Administration of red rosella's petals extract (*Hibiscus sabdariffa*) increase the level of superoxide dismutase (SOD) Wistar rats which induced by repeatedly heated cooking oil. In 2017. DOI: https://doi.org/10.30742/JIKW.V4I2.9.
12. Putra H, Annis F, Purnomo Y. Efek Ekstrak Etanol Daun Gedi Merah (Abelmoschus Manihot (L.) Medik) Terhadap Kadar Sod Dan Mda Pada Aorta Tikus Model Diabetes Melitus Tipe 2. J Kesehat Islam Islam Heal J. 2020;9(1):1. DOI: http://dx.doi.org/10.33474/jki.v9i1.8861.
13. Nilla PY, Hussaana A, Chodidjah. Pengaruh Krim Ekstrak Ubi Ungu (Ipomoea Batatas Var Ayumurasaki) terhadap Matriks Metalloproteinase-1 dan Jumlah Kolagen Dermis Tipe I dan Tipe III (Penelitian Eksperimental Pada Mencit BALB/c yang Dipapar Sinar UV-B). J Tunas Med Kedokt Kesehat. 2021;7(1):1–19.
14. R. DL, L. LNP, V. PNLP, G. WIMA. Uji Aktivitas Antioksidan Ekstrak Etanol Kulit Ubi Jalar Ungu (Ipomoea Batatas (L.) Lam) dengan Metode Ferrous Ion Chelating (FIC). J Farm Udayana. 2014;3(1).
15. Jawi IM, Yasa IWPS, Mahendra AN. Antihypertensive and Antioxidant Potential of Purple Sweet Potato Tuber Dry Extract in Hypertensive Rats. Bali Med J. 2016;5(2):65. DOI: https://doi.org/10.15562/bmj.v5i2.217.
16. Dwi Primayanti IDAI, Aman IGM, Bagiada IMA. Ipomoea batatas Syrup Decrease Malondialdehyde and Increase Nitrous Oxide Plasma Levels Among Moderate Smoker Workers at Denpasar. Bali Med J. 2012;1(3):125–30. DOI: https://doi.org/10.15562/bmj.v1i3.28.
17. Ansel. Pengantar bentuk sediaan farmasi. Glob Shad Africa Neoliberal World Order. 2005;44(2):8–10.
18. Husna N El, Novita M, Rohaya S. Anthocyanins Content and Antioxidant Activity of Fresh Purple Fleshed Sweet Potato and Selected Products. Agritech. 2013;33(3):296–302. DOI: https://doi.org/10.22146/AGRITECH.9551.
19. Prasetyo HA, Winardi R. Antioksidan pada Pembuatan Tepung dan Cake Ubi Jalar Ungu (Ipomoea batatas L.). J Agrica Ekstensia. 2020;14(1):25–32.
20. Lachman J, Hamouz K, Šulc M, Orsák M, Pivec V, Hejtmánková A, et al. Cultivar differences of total anthocyanins and anthocyanidins in red and purple-fleshed potatoes and their relation to antioxidant activity. Food Chem. 2009;114(3):836–43. DOI: https://doi.org/10.1016/J.FOODCHEM.2008.10.029.
21. Mattioli R, Francioso A, Mosca L, Silva P. Anthocyanins: A Comprehensive Review of Their Chemical Properties and Health Effects on Cardiovascular and Neurodegenerative Diseases. Molecules. 2020 Aug;25(17). DOI: https://doi.org/10.3390/molecules25173809.
22. Ngestiningsih D, Rahayu RA, Batubara L. Effect of Superoxide Dismutase (SOD) Supplementation on Plasma Levels of Malondialdehyde (MDA), Total Cholesterol and LDL Cholesterol in the Elderly. J Biomed Transl Res Vol 5, No 2 December 2019. DOI: https://doi.org/10.14710/jbtr.v5i2.4679.
23. Kim YH, Kim Y, Cho E, Kwak S, Kwon S, Bae J, et al. Alterations in intracellular and extracellular activities of antioxidant enzymes during suspension culture of sweet potato. Phytochemistry. 2004 Sep;65(17):2471–6. DOI: https://doi.org/10.1016/j.phytochem.2004.08.001.
24. Lu X, Zhou Y, Wu T, Hao L. Ameliorative effect of black rice anthocyanin on senescent mice induced by D-galactose. Food Funct. 2014;5(11):2892–7. DOI: https://doi.org/10.1039/c4fo00391h.
25. Zhi Q, Lei L, Li F, Zhao J, Yin R, Ming J. The anthocyanin extracts from purple-fleshed sweet potato exhibited anti-photoaging effects on ultraviolent B-irradiated BALB/c-nu mouse skin. J Funct Foods. 2020;64:103640. DOI: https://doi.org/10.1016/j.jff.2019.103640.

26. Camkurt MA, Fındıklı E, İzci F, Kurutaş EB, Tuman TC. Evaluation of malondialdehyde, superoxide dismutase and catalase activity and their diagnostic value in drug naïve, first episode, non-smoker major depression patients and healthy controls. Psychiatry Res. 2016;238:81–5. DOI: https://doi.org/10.1016/j.psychres.2016.01.075.
27. Kamal AA, el Khafif M, Koraah S, Massoud A, Caillard JF. Blood superoxide dismutase and plasma malondialdehyde among workers exposed to asbestos. Am J Ind Med. 1992;21(3):353–61. DOI: https://doi.org/10.1002/ajim.4700210308.
28. Abdel-Hafez HZ, Mohamed EEM, Abd-Elghany AA. Tissue and blood superoxide dismutase activity and malondialdehyde level in leprosy. J Eur Acad Dermatology Venereol. 2010;24(6):704–8. DOI: https://doi.org/10.1111/j.1468-3083.2009.03496.x.
29. Załuski D, Smolarz HD. Plant inhibitors of metalloproteinases and the possibility of their application in the prevention of photoaging. Ann Umcs, Pharm. 2009;22:89–96. DOI: https://doi.org/10.2478/V10080-008-0184-9.

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