

RESEARCH ARTICLE

Purple Sweet Potato Yogurt Affects Lipid Metabolism and Reduces Systemic Inflammation and Oxidative Stress in High Fat Diet Mice

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Abstract

BACKGROUND: Purple sweet potato yogurt (PSPY) is a functional food which is rich in anthocyanin and probiotics. However, the currently available data on its potentially protective effect on anthropometry, lipid metabolism, oxidative stress, and pro-inflammatory markers is very minimal, especially in mice. This study was performed to investigate those effects on balb/c mice models (*Mus musculus*) given a high-fat diet (HFD).

METHODS: Balb/c mice were treated with or without standard diet, HFD, ethanol extract, yogurt, and PSPY according to the group. The changes of anthropometry were analyzed using Lee Index. After three months, the interscapular brown adipose tissue (iBAT) was morphologically observed with hematoxylin and eosin (H&E) staining. The blood serum was used for evaluation using cholesterol oxidase-peroxidase aminoantypirin

(CHOD-PAP) for lipid profile, enzyme-linked immunoassay (ELISA) for tumor necrosis factor (TNF)- α and interleukin (IL)-6, and thiobarbituric acid reactive substance (TBARS) procedure for malondialdehyde (MDA).

RESULTS: Lee Index revealed a decrease in time ($p < 0.0001$). The PSPY group showed a decrease in iBAT weight ($p < 0.05$), lipid profiles including LDL ($p < 0.05$) and total cholesterol ($p > 0.05$), TNF- α and IL-6 ($p > 0.05$), and MDA ($p > 0.05$). Adipocytes' density showed a significant increase ($p = 0.001$).

CONCLUSION: This research finding indicates that PSPY affects lipid metabolism and has a potential protective effect of reducing systemic inflammation and oxidative stress.

KEYWORDS: anthocyanin, high-fat diet, lee index, lipid metabolism, oxidative stress, purple sweet potato yogurt, systemic inflammation

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Introduction

Lifestyles have an impact on changes in people's dietary behavior. People nowadays have difficulty controlling the amount and type of food consumed. The changes can increase the high-fat diet (HFD) consumption while lacking

daily activities or a sedentary lifestyle. It makes the fat consumed will be built-up in the body and cause an abnormal fat accumulation in the adipose tissue.(1) This abnormal fat accumulation increases the body mass index (BMI) and waist circumference of a person by whom to be categorized as overweight or obese.(2,3) High consumption of HFD is also known to increase the risk of developing metabolic

syndrome.(4) The presence of systemic inflammation characterizes the state of the metabolic syndrome itself in the body, increasing tumor necrosis factor (TNF)- α , interleukin (IL)-6, and other pro-inflammatory profiles. The elevation of serum soluble tumor necrosis factor receptor (sTNFR)-2 and IL-1 β in adult males with central obesity. (5) The increased levels of proinflammation occur due to an imbalance in oxidant and antioxidant levels that cause oxidative stress. As a result, many molecular markers are used to assess inflammatory and oxidative stress level.(6) This situation causes metabolic syndrome to be a high-risk factor for other serious diseases such as type 2 diabetes mellitus, cardiovascular disease and stroke, non-alcoholic fatty liver disease (NAFLD), non-alcoholic steatohepatitis (NASH), and cancer.(7)

There was an increase in the prevalence of overweight from 11.5% to 13.6% and obese from 14.8% to 21.8% compared to 2013.(8) Furthermore, it is also reported that dyslipidemia prevalence is between 9-25%. In addition, high levels of total cholesterol (TC) in a person are estimated to be the cause of 4.5% of total deaths and 2% of total Disability Adjusted Life Years (DALY).(9) Thus, many therapeutic approaches to overcoming dyslipidemia are rapidly explored.

One commonly used therapy to reduce lipid levels in the blood is by taking β -Hydroxy β -methylglutaryl-CoA (HMG-CoA) reductase drugs, statins. This type of drug is widely used because of its good effect on reducing lipids in the blood and its affordable price. However, this pharmacological therapy can have various side effects, such as myopathy, diabetes, and hemorrhagic stroke.(10) Therefore, prevention and treatment are being carried out nowadays with beneficial dietary therapies such as increasing the consumption of fruits and vegetables that contain antioxidant and anti-inflammatory components.(11) Previous studies showed an anti-obesity effect of fruit with high antioxidant content by reducing oxidative stress and assisting in inhibition of intestinal cholesterol absorption. (12,13)

One of the natural ingredients known to have antioxidant and anti-inflammatory properties is purple sweet potato (*Ipomoea batatas* (L.) Lam.). Anthocyanin contains in purple sweet potato is a bioactive component, a flavonoid derivative with antioxidant properties such as anticancer, antidiabetic, and anti-inflammatory.(14) In addition, the polyphenolic content in purple sweet potato increases the growth of lactic acid bacteria such as *Lactobacillus spp.* and *Bifidobacteria spp.*, which are usually contained in yogurt.(15)

Yogurt, as one of the most common dairy products, utilizes a combination of a starter culture of probiotic bacteria and prebiotic growth substrate. The combination of probiotics and prebiotics in yogurt aims to improve the quality of yogurt, which is called synbiotic yogurt.(16) Many vegetable and fruit products can be used as a source of prebiotics to be combined with lactic acid bacteria to form a synbiotic yogurt.(17) Yogurt consumption showed beneficial effects in reducing lipid levels in the blood, increasing protection in the intestine, and improving body health.(18)

This research used purple sweet potato-based yogurt (PSPY) made from purple sweet potato puree mixed with low-fat milk and bacterial starter (*Lactobacillus bulgaricus* ATCC 11842, *Lactobacillus acidophilus* ATCC 4356, and *Bifidobacterium longum*), which reported to be safe to consume based on *in vivo* study.(19) Previous studies have shown that purple sweet potato can reduce carbohydrate content in yogurt and improve its sensory properties.(20) Nonetheless, the data in present-day about the potential protective effect of the product on anthropometry, lipid metabolism, oxidative stress, and pro-inflammatory markers is still inadequate.

The effects of anthocyanin and probiotics of the PSPY product still needs to be elucidated. Thus, this research aimed to identify the differences in the effect of ethanol extract of purple sweet potato as prebiotics, yogurt as probiotics, and the combination of both into PSPY as synbiotics on lipid metabolism. Also, the exploration was conducted to clarify the protective effects against systemic inflammation and oxidative stress in hyperlipidemic induced mice fed with HFD.

Methods

Preparation of Bacterial Starter

The probiotics used in this study are from Advanced Biomedical Laboratory, Division of Microbiology and Parasitology Laboratory, Faculty of Medicine, Universitas Padjadjaran, Indonesia. The instruments were sterilized and put in an autoclave at 121°C with a pressure of 1 atm (15lbs) for 15-20 minutes. Then, rejuvenated the bacteria culture (*Lactobacillus bulgaricus* ATCC 11842, *Lactobacillus acidophilus* ATCC 4356, and *Bifidobacterium longum*). Each bacteria culture was inoculated and incubated in a 5% CO₂ incubator at 37°C for 24 hours in low-fat pasteurized milk (PT Ultrajaya, Jakarta, Indonesia) to make the bacterial starter. Following that, all bacterial starter cultures were

combined with milk (10% v/v) and incubated at 37°C for 24 hours. Finally, the fermented product became the mother culture of PSPY.(21)

Preparation of PSPY

Purple sweet potatoes were obtained from a plantation in Tanjungsari, Sumedang, West Java, Indonesia. Five grams of purple sweet potatoes were steamed until soft and then separated from the skin. Three grams of steamed purple sweet potato were peeled and cut into small pieces before it was mashed into a puree using a blender. Warm water was added with a ratio of 1:1 of the weight of sweet potatoes. Subsequently, the puree was filtered using cotton fabric to harvest the purplish liquid from the puree. Around 60% of low-fat pasteurized milk was added to 30% purple sweet potato puree. Later, the starter cultures of three bacteria were added to as much as 10%. The mixture was incubated in a CO₂ incubator at 37°C for 24 hours.(21)

Experimental Animals

This research used male mice (*Mus musculus* balb/c) aged 4-6 weeks with a weight of 25-35 grams. The mice were obtained from the animal development unit in PT. Biofarma, Cisarua, Cimahi, West Java, Indonesia, and acclimated in plastic cages (20 x 30 x 14 cm) with husks around 3 cm high as the base. The treatment was conducted at the Animal Laboratory of the Department of Biomedical Sciences, Faculty of Medicine, Universitas Padjadjaran. and the research procedure was approved by the Research Ethics Committee of Faculty of Medicine, Universitas Padjadjaran (No.1081/UN.6KEP/EC/2019).

Animals Observation

This research was conducted for thirteen weeks. The first week was for adaptation, followed by the induction of hyperlipidemia with HFD in the next four weeks. The treatment was applied during the last eight weeks. The mice were divided into five groups of treatment. Group I was fed a standard diet (SD) as a negative control, Group II was fed with 50 grams of HFD, Group III was fed with 50 grams HFD and 0.3 mL ethanol extract of purple sweet potatoes, Group IV was fed with 50 grams of HFD and 0.3 mL yogurt, and Group V was fed with 50 grams HFD and 0.3 mL PSPY.

The dosage of PSPY was calculated based on the daily recommended dose of dairy product consumption for humans, which is 2-3 cups (400-700 mL).(22) The conversion of human dosage to mice was calculated by multiplying the human dose by 12.3, which is the coefficient number of human-equivalent doses.(23) Each treatment was

given by oral gavage tube into the mice's stomachs in two batches for each group. Half of the mice were treated in the morning around 7-10 am and the other half in the late afternoon around 3-5 pm.

Analysis of Lee index

The changes in body weight and naso-anal length were calculated using the Lee Index formula as an anthropometric status of the mice. The decrease in the Lee Index was used to indicate the treatment effects in this research. Every week for thirteen weeks, the mice's body weight and naso-anal length were measured to calculate the anthropometry of the Lee index. The Lee index was calculated using the following formula used in the previous study.(24)

Histology Analysis of Brown Adipose Tissue (BAT)

At the end of the treatment period, the mice were sacrificed with cervical dislocation method. The interscapular subcutaneous brown adipose tissue was taken and weighed. The tissue was fixed with 4% formaldehyde, embedded in paraffin, then cut into 4 µm using a microtome. The tissue was placed in the glass object and stained using hematoxylin & eosin (HE). The tissue was seen using a binocular microscope and Optilab 3.0 (Miconos, Sleman, Indonesia) with a 40X magnification. The morphological changes were analyzed by seeing the density level of the adipocyte cells in every image. The cell density was measured by counting the adipocyte cells in one field. The larger the adipocyte cells indicate, the smaller the density level and vice versa. The density level range was from +1 to +4.(25)

Analysis of Blood Lipid Profile

The lipid profiling was conducted using two methods. The first method was used to follow up on lipid profiles during the intervention. The mice's blood was taken from their tail, and the lipid profile was evaluated using the strip method from the familyDr.TM kit (CHOL Monitoring System, CE0197, Taiwan). The mice had fasted for 8 hours before the measurement. The second lipid profile evaluation method was done at the end of the intervention. The mice were sacrificed, and blood serum was checked using the CHOD-PAP enzymatic method before being analyzed with a spectrophotometer at a wavelength of 505 nm. Sample measurements were always included with standard lipid measurements.(26)

Analysis of TNF-α and IL-6

At the end of the intervention (after the 13th week of the research period), about 1.0 mL of blood was inserted into

the microtube. The blood was then centrifuged, and the serum was taken. The pro-inflammatory markers (TNF- α and IL-6) were examined by ELISA kit from ThermoFisher Scientific (No Cat. BMS607-3 (TNF- α) and (BMS603/2 (IL-6), Waltham, Massachusetts, United States). Thus, samples were read using a 450 nm microwell spectrophotometer. The examination was carried out in duplicate.

Analysis of Malondialdehyde (MDA)

Blood samples were taken at the end of the intervention. As much as 1.0 mL of blood was inserted into the microtube and then centrifuged at 3000 rpm at 4°C for 10 minutes to take the serum. MDA tests were performed using the thiobarbituric acid reactive substance (TBARS) method. The TBARS examination was carried out in cold temperatures. The mixture was homogenized and then incubated at 96°C for 1 hour. The mixture was cooled in the 4°C refrigerator and then centrifuged at 3500 rpm at 4°C for 10 minutes twice. Then the supernatant was taken and diluted using aquadest up to 2000 μ L and then examined using a spectrophotometer on the 532nm wavelength. The examination was carried out in duplicate.

Statistical Analysis

This was an experimental analytic study. The interscapular BAT (iBAT) weight data were analyzed by one-way ANOVA, and the score of iBAT density was analyzed by the Kruskal-Wallis test.(26) The data were processed using Microsoft Excel 2016 (Microsoft Corporation, Albuquerque, NM, United States) and SPSS version 25 (IBM Corporation, Armonk, NY, USA). The weight and Lee Index were analyzed with a general linear graph of the means and repeated measures MANOVA.

Results

PSPY Effects on the Anthropometry Parameters

The mean of body weight and Lee Index in time were described in Table 1. The data showed an increased value in all groups during the adaptation period and HFD induction. Further, the HFD induction period was shown in the mean differences before the initial treatment minus the initial HFD induction (5th week – 1st week). A decrease occurred in the mean differences between the end and the initial treatment (13th week – 5th week). The most significant decrease occurred in the mean of the Group V (Wilk's Λ =0.013, $F(13,14)$ =81.308, p <0.0001).

PSPY Effects on Lipid Metabolism

There were three analytical methods used to evaluate the effect of PSPY on lipid metabolism. The methods included iBAT analysis, enzymatic lipid profile, and total cholesterol strip analysis. iBAT analysis measured the weight and the histology appearance (Figure 1).

The average weight in the Group V showed a slight difference compared to Group I and Group II (p =0.023). The average of Group I was 0.374 grams, Group II was 0.438, and Group V was 0.42 grams (Figure 2). Histological analysis was done by categorizing the density appearance of adipocyte cells in each field. The highest density has a score of +4, where adipocyte cells appear to be denser and more numerous with smaller sizes. Density with a score of +3 has adipocyte cells that look denser with a larger size. Density with a score of +2 has a more tenuous density with larger adipocyte cell sizes. Density with a +1 score has fewer cells but larger adipocyte cells.

Table 1. Mean differences of body weight and Lee Index during the period of induction and treatment.

Variable	Group	Mean \pm SD				Mean Differences	
		Initial Adaptation (Week-0)	Initial Induction (Week-1)	Initial Treatment (Week-5)	End of Treatment (Week-13)	Period of HFD Induction	Period of Treatment
Body weight (gram)	Group I	30.06 \pm 3.59	36.67 \pm 4.04	40.81 \pm 4.09	42.66 \pm 4.65	4.14	1.85
	Group II	28.29 \pm 3.31	32.75 \pm 4.69	39.35 \pm 4.41	37.43 \pm 6.74	6.6	-1.92
	Group V	36.91 \pm 3.88	36.49 \pm 4.54	43.58 \pm 4.92	38.39 \pm 6.37	7.09	-5.19
Lee index	Group I	320 \pm 16	338 \pm 7	349 \pm 5	348 \pm 5	11	-1
	Group II	323 \pm 16	341 \pm 13	347 \pm 13	340 \pm 20	6	-7
	Group V	351 \pm 13	345 \pm 7	358 \pm 11	342 \pm 12	13	-16

Data were presented as the mean \pm SD of each group at a certain period. Data were analyzed using repeated measures one-way ANOVA (p <0.0001).

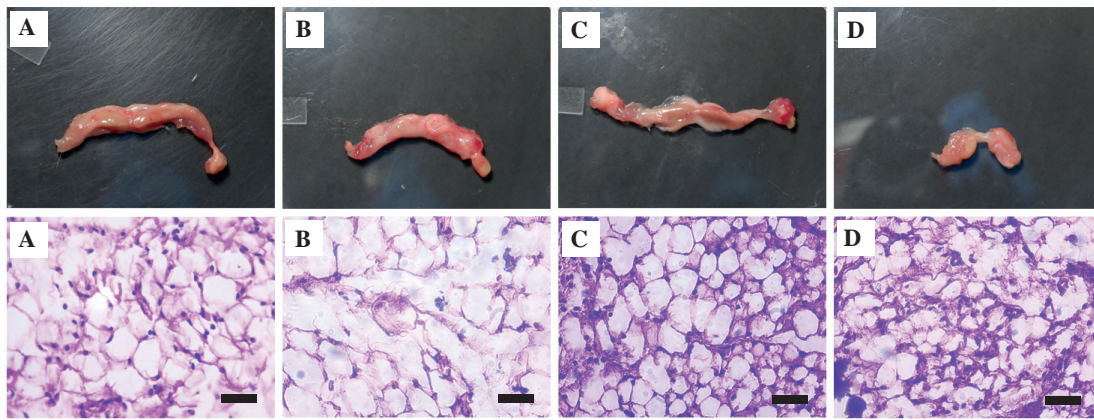


Figure 1. iBAT section and histology appearance in HE staining. A: Group fed with standard diet (Group I) showing an adipocytes density level +2; B: Group fed with HFD (Group II) showing an adipocytes density level +1; C: Group fed with HFD + PSPY (Group V) with an adipocytes density level +3 and; D: Group fed with HFD + PSPY (Group V) with an adipocytes density level+4. Black bar: 50µm.

The analysis was performed using the Kruskal-Wallis non-parametric test. Total figures studied were from 126 pictures from different fields. One object-glass contained 3-5 slices and three fields of photographs taken in each slice. Adipocyte cell morphology density showed a significant difference with $p=0.001$. Based on the density level mean results, Group V had the highest mean rank of density value (121.24), followed by Group I (100.42) and Group II (77.73).

The Mann-Whitney post hoc test was performed to determine the mean difference between groups. The results of the Mann-Whitney test also revealed the most significant difference in Group V compared with the other groups ($p<0.05$). The lipid profiles from each group were examined in this study. The results of serum lipid profile analysis were shown in Table 2 and Figure 3. Using the One-Way ANOVA method, statistical test results showed significant differences

($p=0.028$) for LDL. There was a mean difference compared with Group II. In Group III, there was an average difference of 6.749, while for Group V was -12.567, and Group IV was -28.050.

Meanwhile, statistical tests of total serum cholesterol showed no significant differences ($p=0.199$). However, there was an average difference compared to Group II. Those in the treated groups (Group III, IV, and V) tended to experience a decrease except for Group IV. In Group III, there was a mean difference of 10.230; in Group V amounted to 15.160, and in Group IV amounted to -3.553. In this study, total cholesterol was also examined throughout the research period using cholesterol strips. The total cholesterol strip test was measured twice, before and after treatment. Pre and post mean in Group I were 106.5 and 113.25, Group II were 117 and 125.8, Group III were 114.2 and 110.4, Group IV was 111 and 110.17, and Group V of 116,4 and 109 (Table 3).

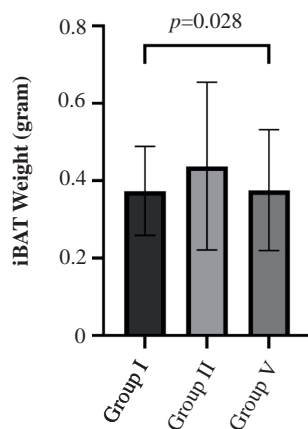


Figure 2. The average iBAT weight. Each bar represented the mean±SD. Data were analyzed using one-way ANOVA, significant if $p<0.005$.

PSPY Effects on Pro-inflammatory Markers

The ELISA test results showed the changes in cytokine levels in the mice's blood. Changes that occur in the form of an indication of decreased systemic inflammatory processes that occur due to HFD behavior. Comparison of the amount of reduction was shown in Figure 4A and 4B. TNF- α level in Group I was 1.15 pg/mL, Group II was 1.09 pg/mL, and Group V was 0.84 pg/mL. IL-6 levels in Group I was 84.05 pg/mL, Group II was 119.22 pg/mL, and Group V was 83.13 pg/mL.

PSPY Effects on Oxidative Stress

MDA is a secondary derivative component of free radicals derived from the fat oxidation process. In this study, the

Table 2. Statistical analysis of serum lipid profile.

Group	LDL (mg/dL)			Total Cholesterol (mg/dL)		
	Mean	Mean Difference ^a	<i>p</i> -value*	Mean	Mean Difference ^a	<i>p</i> -value*
Group 1	46.373	24.244		57.575	41.905	
Group 2	70.616			99.48		
Group 3	63.866	6.749	0.028	89.249	10.23	0.199
Group 4	98.667	-28.051		103.034	-3.554	
Group 5	83.183	-12.567		84.319	15.16	

Data were presented as the mean and mean difference of each group with the *p*-value of each test. ^aMean difference compared to group fed with HFD (Group II). *Data were analyzed by one-way ANOVA, significant if *p*<0.05.

PSPY administration showed a decrease in MDA amount in the blood after being tested using the TBARS method. These results can be seen in Figure 4C. The MDA level in Group I was 0.0298 $\mu\text{mol/L}$, Group II was 0.0318 $\mu\text{mol/L}$, and Group V was 0.0315 $\mu\text{mol/L}$. This showed that PSPY was able to reduce the lipid peroxidation product.

Discussion

Consumption of HFD in a certain period can increase a lipogenesis *de novo*, affecting the anthropometry changes of body mass index from normal to overweight.(27) Our data had shown an increase in the body weight and anthropometry in all groups during HFD induction. Thus, this research hypothesizes that the consumption of PSPY prevented the production of fatty acids with its bioactive content.

The test results showed the anthocyanin level was 143.06 mg/L. It means that in this research, PSPY contained 42.9 μg anthocyanin at 0.3 mL dose. The period of 8 weeks of treatment with a 0.3 mL dose of PSPY was revealed to reduce the weight and anthropometry of the mice that were calculated with the Lee Index (Table 1). It also showed

a decrease in weight and an increase in density level in the morphological appearance of iBAT. Meanwhile, statistically, the lipid profile showed no significant difference in total cholesterol (*p*=0.199), but on the other side, there were significant differences in LDL (*p*=0.028). The high carbohydrate content of purple sweet potato helps the growth of lactic acid bacteria from yogurt fermentation. (28) This synbiotic system can help regulate the hepatic lipogenic enzymes and increase the short-chain fatty acids (SCFA) production. The production of esterase, lipase, and co-enzyme A has also been investigated to reduce cholesterol.(29) Anthocyanin in PSPY are antioxidants that activate AMPK and inactivate the HMG-CoA reductase enzyme. The inactivation of the enzyme will inhibit the formation of cholesterol.(30) The previous study showed a beneficial effect of anthocyanin supplementation in decreasing LDL-C concentration in dyslipidemic patients. It was reported to be partially mediated via cholesteryl ester transfer protein (CETP), which is a plasma protein that mediates the exchange of cholesteryl ester from HDL with triglycerides.(31)

As previously mentioned, consumption of HFD in a certain period can increase lipogenesis *de novo* in adipose

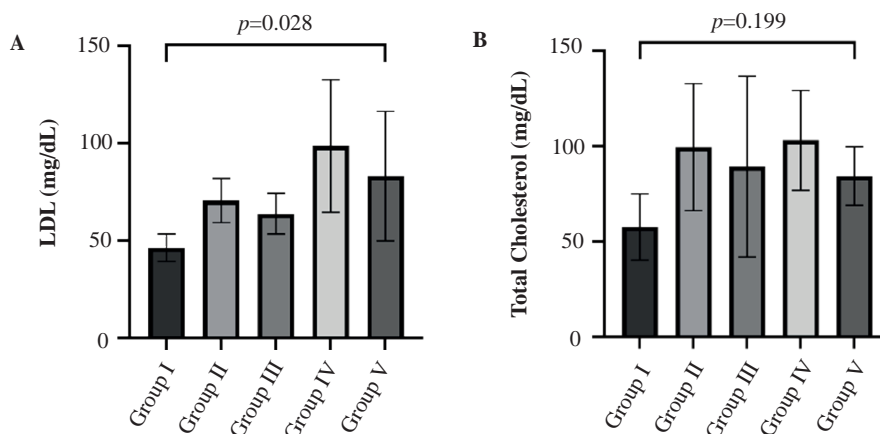


Figure 3. Result of serum lipid profile analysis. A: LDL; B: Total cholesterol. Results are shown as the means \pm SD from each group.

Table 3. Pre, post, and mean difference analysis of total cholesterol strips (mg/dL).

Group	Mean Total Cholesterol Strip		Mean Difference
	Pre	Post	
Group I	106.5	113.25	-6.75
Group II	117	125.8	-8.8
Group III	114.2	110.4	3.8
Group IV	111	110.17	0.83
Group V	1164	109	7.4

Data were presented as the mean and mean differences of each group.

tissue.(32) Many studies on animals about the specific type of adipose tissue and its role in body weight regulation have been conducted. In rodents, the interscapular brown adipose tissue is the largest depot in the body. One study stated that brown adipose tissue helps maintain metabolic homeostasis. (33) Brown adipose tissue has abundant Uncoupling Protein 1 (UCP-1) mitochondria compared to the white type. UCP-1 in mitochondria in brown adipose tissue requires up to 100% oxygen consumption, in contrast to the other cell, which only requires up to 20%.(33)

Anthocyanin down-regulates the expression of genes in adipogenesis. On the other hand, it can increase the expression of UCP-1 through cAMP activation. UCP-1 is fundamental to increasing lipolysis to release FFA from lipid droplets as β -oxidation fuel and the TCA cycle process in cells to obtain oxygen.(33) This research showed an increase in adipocyte density in the histology appearance of the PSPY group. It means that browner adipose tissue presented with numerous lipid droplets inside the cell (as shown in Figure 1). The droplets inside the brown adipose tissue aid in obtaining oxygen to maintain the TCA cycle process.(33)

PSPY used in this research was made from purple sweet potato puree mixed with low-fat milk and bacterial starter (*Lactobacillus bulgaricus* ATCC 11842, *Lactobacillus acidophilus* ATCC 4356, and *Bifidobacterium*

longum). The results of giving PSPY to HFD mice showed a change in the form of a decrease in levels of pro-inflammatory cytokines (TNF- α and IL-6) with $p>0.05$. Although it is not statistically significant, this finding is still in line with other researches. The study was done on mice with inflammation of neurons in the brain. The treatment was done by administering purple sweet potato color (PSPC). The research showed a significant decrease in pro-inflammatory cytokine levels, TNF- α , IL-6, and IL-1 β , in mouse brain tissue. It concluded that purple sweet potato could reduce brain inflammation due to HFD and improve memory function.(34) Another study used the obese mice model, given the treatment of the anthocyanin extract of black soya beans. It also showed a consistent result in a significant decrease in pro-inflammatory cytokines levels, TNF- α , and IL-6 mesenteric mice fat tissue.(35)

This research concludes that anthocyanin can show its ability as an anti-inflammatory.(35) Inflammatory events due to HFD are caused by two processes, specifically by hyperplasia and hypertrophy of adipocyte tissue and dysbiosis of intestinal microbiota. HFD can cause intestinal microbiota dysbiosis by changing the number or ratio of several important species that play a role in maintaining the balance in the intestinal lumen. In the HFD condition, the ratio of Bifidobacteria bacteria decreases with an increase in the number of Enterobacteriaceae and Firmicutes.

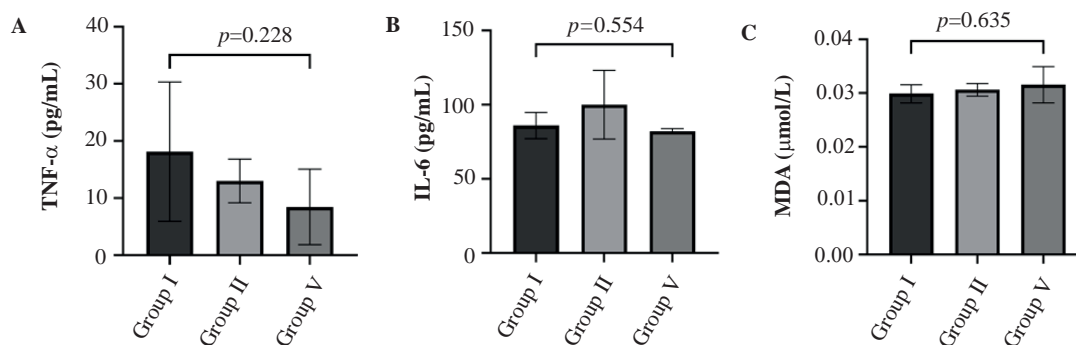


Figure 4. Cytokines and lipid peroxide levels. A: TNF- α levels; B: IL-6 levels; C: MDA levels.

Increased Enterobacteriaceae and Firmicutes cause the LPS (endotoxin) in the lumen to increase, which leads to local inflammatory reactions. The inflammatory reaction will trigger tight junction damage and endotoxin leakage into the blood and finally cause a systemic inflammatory effect. (36,37) Previous research reported that improved microbiota composition in the intestine can reduce inflammation.(38) Based on these findings and theories, PSPY as a synbiotic product can reduce the systemic inflammatory processes due to HFD better than purple sweet potato ethanol extract, which only contains the antioxidant substance.

The fat oxidation process was observed in this research by examining MDA levels. The results of giving PSPY showed a change in the form of a decrease in MDA values with $p > 0.05$, which indicated statistically not significant. Nevertheless, the results of this study are in line with several other studies. This research was conducted in a diabetic rat model with anthocyanin treatment of Kawi Mountain purple sweet potato and showed significant MDA degradation results and concluded that purple sweet potato could improve spatial memory function through MDA levels.(39) Another study used old mice treated with purple anthocyanin extracts showed a significant decrease in MDA values. It concluded that purple sweet potatoes could inhibit the aging process.(40) Based on these findings, PSPY has the potential to reduce levels of MDA in the blood due to HFD. Further study is needed to elucidate more detail on the molecular mechanism in how the PSPY could affect the lipid profile at the molecular level.

Conclusion

It is revealed that PSPY might have a potential effect on anti-obesity. PSPY consumption in high-fat diet mice significantly decreased the body weight measurement and Lee Index analysis during the treatment period. PSPY also affects iBAT, shown by a decrease in weight and an increase of density level in the morphological appearance. These purple sweet potato-based treatments affect the lipid profile on total cholesterol and LDL concentrations. Hence, PSPY can potentially improve lipid profile by reducing pro-inflammatory cytokines and MDA levels.

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Authors Contribution

AFK planned and designed the study and analyzed the data. CN, RS, and JA were equally participated in the collection of the samples and tests. AFK, CN, RS, JA, PHAR, ER, and WAS were involved in drafting and revising the manuscript. AFK, WAS and NA performed data processing and critical review of the article. All authors read and approved the final manuscript.

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