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Antioxidant potential and hypolipidemic effects of combined purple sweet potato (*Ipomoea batatas L.*) tuber extract with honey in rats given high cholesterol feed

I Wayan Putu Sutirta-Yasa,[1] I Made Jawi[2*]

ABSTRACT

Backgrounds: Flavonoids prevent cholesterol rising in blood and has antioxidant effects. The purpose of this study is to prove the hypolipidemic and antioxidant effect of purple sweet potato tuber extract with the natural honey combination, which contains flavonoids. **Methods:** this is an experimental study using randomized pre-post-test control group design. The total of 28 adult male Wistar rats, were divided into four groups. During the four weeks of treatment, Group 1 was given high-cholesterol feed. Group 2 were given high-cholesterol feed and water extract of purple sweet potato tubers 200 mg/day. Group 3 were given high-cholesterol feed and natural honey three cc/day. Group 4 were given high-cholesterol feed and water extract combination of purple sweet potato tuber with natural honey. Before the treatment, lipid profiles, malondialdehyde (MDA), and superoxide dismutase (SOD) were examined as a pre-test data. After the treatment, the blood was examined again as the pre-test data. **Results and conclusion:** the obtained data were tested using t-test and ANOVA. The results showed a significant increase in total cholesterol, LDL, and MDA in the blood ($p < 0.05$), and significantly decrease HDL in group 1 ($p < 0.05$). In group 2, 3, and four there was no significant difference in these parameters ($p > 0.05$), but significantly different with group 1 ($p < 0.05$). In conclusion, the combination of water extract of purple sweet potato tuber with honey can maintain lipid profile and MDA in the normal range, in rats with high cholesterol feed.

Keywords: Purple sweet potato, honey, lipid profile, SOD, rat
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[1]Department of Clinical Pathology, Faculty of Medicine, Udayana University
[2]Department of Pharmacology Faculty of Medicine, Udayana University, Denpasar, Bali, Indonesia

[*]Correspondending Author:
I Made Jawi, Department of Pharmacology Faculty of Medicine, Udayana University, Denpasar, Bali, Indonesia
made_jawi@yahoo.co.id

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INTRODUCTION

Natural ingredients usage from plants as herbal medicine is on the rise today. Purple sweet potato is one of the natural ingredients from plants that have been studied. Research has been conducted to prove that the purple sweet potato extract can lower blood cholesterol levels and as an antioxidant in rats and rabbits.[1,2] Purple sweet potato extract was also shown to increase the expression of SOD-2 and SOD-3 in vascular endothelial cells.[3] Research conducted in patients with hypertension also proves that purple sweet potato extract can lower blood pressure and reduce oxidative stress.[4] In the study, patients complained that the taste of the water extract of purple sweet potato makes them less adherent in taking the medication. Based on these results, we need to find an alternative to improve the taste of the purple sweet potato tuber extract and simultaneously strengthen the expected therapeutic effect.

Honey is a natural ingredient that tastes sweet and contains flavonoids which are natural antioxidants. Honey as an herbal treatment on rats for 54 weeks was found to increase and high-density lipoprotein cholesterol (HDL) significantly[5] so that the purple sweet potato extract combined with honey should be beneficial in improving the taste of purple sweet potato tuber extract and amplify antioxidants and hypolipidemic effect. This research investigated the effect of the combination of honey and sweet purple potato extract in rats given high-cholesterol diet with the measurements of MDA and SOD in blood and lipid profile.

MATERIALS AND METHODS

Animal and Experimental Design

Male Wistar rats (150 – 200g), 3-4 months old, were obtained from Animal House Facility from Department of Pharmacology Udayana University, Denpasar, Bali, Indonesia, and used in this study. All rats were maintained under standard laboratory conditions at 25 ± 2°C (temperature), 50 ± 15% relative humidity and normal photoperiod (12-hours light-dark cycle). Commercial high cholesterol pellet diet and water were provided ad libitum for those animals. The usage of these animals was approved by Institutional Animal Care and Use Committee of the Faculty of Medicine, Udayana University, Bali, Indonesia. A total of 24 Wistar rats were divided into 4 group (6 rats per group)

as follows. Group 1: The control group: consists of rats treated with high cholesterol diet for four weeks. Group 2: The treatment 1 group: consists of rats treated with high cholesterol diet and 3 ml/day liquid extract of purple sweet potato tuber for four weeks. Group 3: The treatment 2 group: consists of rats treated with high cholesterol diet and natural honey 2 ml/day for four weeks. Group 4: The treatment 3 group: consists of the rats treated with high cholesterol diet and combination of purple sweet potato extract and natural honey for four weeks.

Materials Testing

The purple sweet potato was obtained from farmers, washed with clean water and then peeled. Once peeled, the sweet potato is cut crosswise with a thickness of 2.0-2.5 cm. Sweet potato chunks are mixed with water at a ratio of 1 kg of sweet potato plus 1 liter of water and then blended and filtered through three layers of gauze. The liquid obtained from the filtration is boiled for thirty minutes. This liquid extract is ready to be used in the research.

Honey was purchased from a bee farm in java. (Madu Kusuma from Apis Mellyfera). The honey was dissolved in drinking water and prepared freshly each time it was administered

Blood Examination

Blood was taken via retroorbital plexus of all rats at baseline and after treatment for four weeks'. Blood samples were collected in plain tubes and allowed to clot at room temperature. The blood samples were centrifuged at 1500 rpm for 10 min. The supernatants (sera) were collected and stored at 20°C until further analysis. The sera of the blood samples at baseline was used for the examination of MDA, SOD, lipid profile and at the end of the study. MDA by thiobarbituric acid reactive substances (TBARS) and SOD method total antioxidant status Randox kit.

Serum triglycerides, total cholesterol, HDL-C were assessed using colorimetric kits from Azmun (Tehran, Iran). Non-HDL-C was calculated by subtracting HDL-C from total cholesterol.

Statistical Analysis The data was entered and analyzed using Statistical Package for Social Sciences (SPSS) 17.0. All data are shown as mean ± S.E.M. One-way ANOVA was applied to observe group mean differences. Post Hoc LSD test was used to observe mean differences among the groups. A p-value of <0.05 was considered as statistically significant.

RESULTS

The results of the study showed that the average weight, total cholesterol, low-density lipoprotein (LDL), HDL, triglycerides, MDA, at baseline and after treatment for four weeks, against all four groups of animals could be seen in Table No. 1-2 and Figure 1-3.

Table 1 shows the weight at the beginning of the experiment did not differ (p> 0.05) in the four experimental groups. At the end of the experiment, there was a significant increase of weight in the four experimental groups and did not differ in all four groups.

Figure 1 shows total cholesterol levels at the start of the experiment did not differ between the four groups of rats (p> 0.05). At the end of the experiment, turns out there was a significant increase in total cholesterol level (p <0.05) in the control group (given high-cholesterol feed). In group 2 (group given a water extract of purple sweet potato tuber) we found a significant increase in(p <0.05), but much lower than the control group (p <0.05). At the end of the experiment, the total cholesterol in control group was much higher than the total

Table 1 Mean weight (Pre-test and Post-test) in 4 groups of study

No	Mean Weight (kg) ± SD	
	Pretest	Post-test
(1)	1,67 ± 0,39	2,62 ± 0,42
(2)	1,65 ± 0,25	2,28 ± 0,36
(3)	1,66 ± 0,35	2,38 ± 0,38
(4)	1,65 ± 0,27	2,48 ± 0,46

Note: (1) control group, consists of rats treated with high cholesterol diet. (2) groups treated with high cholesterol diet and purple sweet potato.(3) groups treated with high cholesterol diet and natural honey.(4) groups treated with high cholesterol diet and combination of purple sweet potato and natural honey.

Table 2 Triglycerides, LDL, and HDL Mean (Pre-test and Post-test) in four experimental group

No	Triglycerides (mg/dl) ± SD		HDL (mg/dl) ± SD		LDL (mg/dl) ± SD	
	Pre-test	Post-test	Pre-test	Post- test	Pre-test	Post-test
(1)	84,07 ± 1,31	162,23 ±2,7*	64,70± 2,55	27,08± 2,7*	26,22± 1,5	70,03± 1,7*
(2)	83,71 ± 3,8	94, 91 ± 3,9*	65,68± 2,7	63,34 ± 1,7*	24,38± 2,25	23,34 ± 2,3*
(3)	84,97± 2,7	112,44 ±2,7*	64,24 ±3,4	53,33 ±4,1*	25,9 ±1,4	34,61 ±1,5*
(4)	84,82±1,5	96,25 ± 2,8*	64,87 ± 3,6	62,20 ± 2,2*	26,22 ±1,5	27,36 ± 2,4*

Note: (1) control group, consists of rats treated with high cholesterol diet. (2) groups treated with high cholesterol diet and purple sweet potato.(3) groups treated with high cholesterol diet and natural honey.(4) groups treated with high cholesterol diet and combination of purple sweet potato and natural honey.

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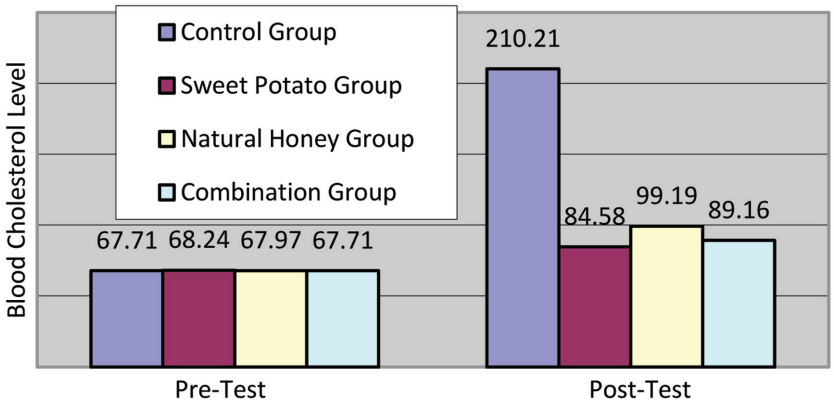

Figure 1 Comparison of Total Cholesterol pre-test and post-test on all four Group of Rats

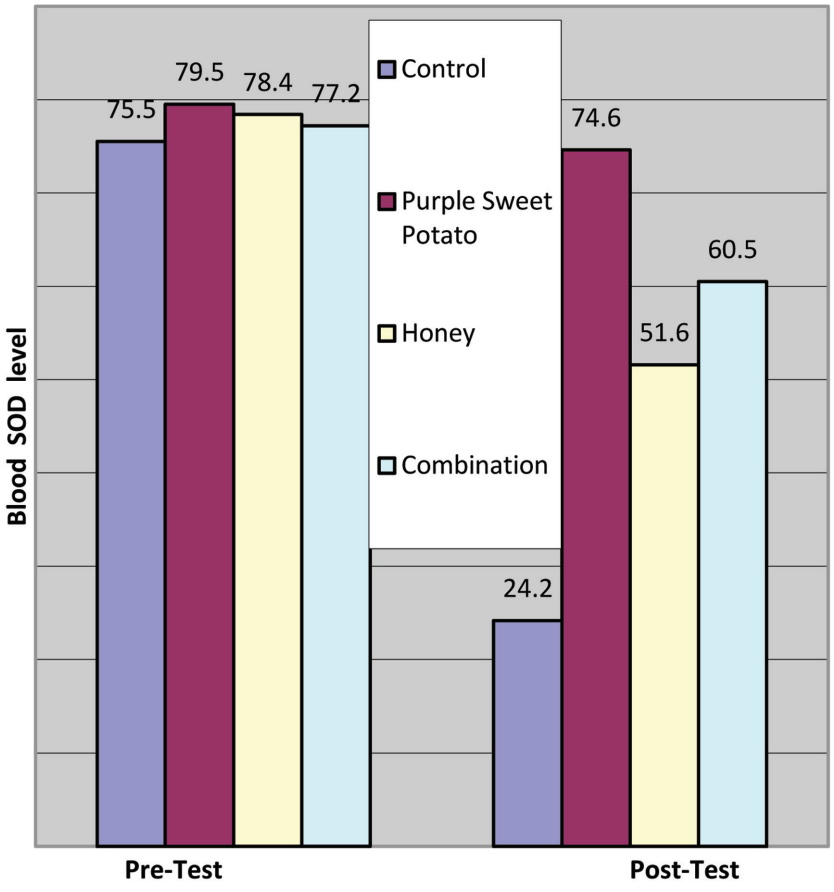

Figure 2 Comparison of Blood SOD Level Pre-test and Post-Test

cholesterol group given or honey extract or a combination of extracts with honey ($p < 0.05$).

Table 2 shows visible changes in the value of triglycerides, HDL and LDL before and after the experiment. Triglycerides increased sharply after feeding high cholesterol for one month. HDL dropped significantly, and LDL rose significantly after a high-cholesterol feed was given for one month ($P < 0.05$). In rats given high cholesterol feed and water extract of tuber sweet potato purple or honey or a combination of the extracts and honey

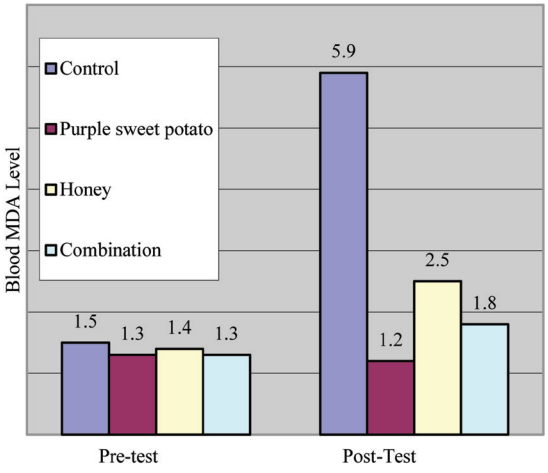

Figure 3 Comparison of Blood MDA Level Pre-test and Post-Test

for a month, it happens the opposite way, namely a decrease in triglycerides, increase in HDL and LDL significantly ($P < 0.05$), compared to the control group.

Figure 2 presented a significant decrease of SOD levels after high-cholesterol feeding without purple sweet potato extract or honey or a combination of purple sweet potato extract and honey ($P < 0.05$). Groups of rats that were given high-cholesterol feed showed a change in SOD level and the change was more prominent in the control group. ($P < 0.05$).

Figure 3 presented an increase in MDA levels were significantly higher after cholesterol feeding without purple sweet potato extract or honey or the combination of both ($P < 0.05$).Groups of rats that were given high-cholesterol feed simultaneously with the purple sweet potato extract have no change in MDA after one month of treatment ($p > 0.05$). In the group given a water extract of purple sweet potato tuber rose significantly ($P < 0.05$). The group that received purple sweet potato extract turns out to have significantly lower level of MDA ($P < 0.05$). At the beginning of the experiment, the MDA level does not differ in all four experimental groups ($p > 0.05$). Based on this study, sweet purple potato extract, natural honey and a combination of both seems to have an antioxidant effect on rats that were given high-cholesterol diet.

DISCUSSION

In this study, there was an increase in total cholesterol, LDL, and triglycerides, and decrease in HDL after feeding rats with high cholesterol diet for four weeks. There was also a significant increase in MDA level and a significant reduction in SOD level in given rats. Groups of rats that were given purple sweet potato extracts or honey or a combination of both showed a change in all the variables studied

compared to the control group. Anthocyanins are a type of flavonoid found in purple sweet potato tuber[6] (Huang, 2004) that can inhibit the absorption of cholesterol in the gastrointestinal tract and presumably inhibit cholesterol synthesis by the liver. This study is consistent with research anthocyanin extracts from the seeds of soybean (Glycine max. L) which can improve the lipid profile by lowering triglycerides and total cholesterol significantly and increased HDL.[7]

A decrease in serum cholesterol due to the administration of anthocyanin was possible by blocking the absorption of cholesterol and bile acids in the intestine. This is also consistent with the study by Fumio et al. who found that rats given nasunin, an anthocyanin from eggplant, have lower total cholesterol serum and also higher HDL level.[8] Furthermore, another study showed that the level of anthocyanin increased significantly as fast as 30 minutes to 8 hours after administration of a single dose of purple sweet potato extract. However, in a certain metabolic disease like diabetes, the absorption of anthocyanin was observed to be compromised. The study showed that the absorption anthocyanin is better in healthy rats compared to diabetic one.[9]

Rats that were given honey or a combination of it with purple sweet potato extract also have increased LDL after high cholesterol diet, but it was lower than the control group. Honey has a hypolipidemic effect through increasing adiponectin, which is a regulator of fat metabolism and glucose. Honey also prevents oxidative stress through the barriers against peroxidation lipid.[10] The exact mechanism of honey in improving the lipid profile is still unclear but presumably through blocking the lipoprotein lipase enzyme. Existing fructose in honey inhibits the lipoprotein lipase enzyme and lecithin cholesterol acyl-enzyme transferase enzyme.[10]

In this study, high-cholesterol feeding led to significant increase in MDA and decreased in SOD ($P < 0.05$). The sweet potato extract or honey or a combination of both can lower MDA and increase SOD. Flavonoids which are found in honey[11,12] or purple sweet potato extract[12] has a formula built with phenolic structure, so it can transfer the hydrogen atoms of hydroxyl free radicals, just as antioxidants. The extract of purple sweet potato contains a high amount of anthocyanin,[13] which will serve as exogenous antioxidants that can prevent the increase of blood MDA level because anthocyanins also have antioxidant effect.[14] The study is coherent with a study by Satriyasa BK et al. found that purple sweet potato extract decreased MDA and increase SOD2 gene in the kidney of diabetic rats.[15] Another study found that Balinese purple sweet potato can also reduce oxidative stress and decrease the level of interleukin-1 in hypercholesterolemic rabbits.[16] Furthermore, study by Jawi IM et al. found that besides the antioxidant effect measured by SOD and MDA, the purple sweet potato also have antihypertensive effect.[17]

CONCLUSION

Administration of purple sweet potato extract or honey or a combination of both may improve blood lipid profile in rats given high-cholesterol diet. Administration of purple sweet potato extract or honey or a combination of both may reduce levels of MDA and increase levels of blood SOD in rats given high-cholesterol diet.

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