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Effect of herbal drink plants Tiwai (*Eleutherine Americana* Merr) on lipid profile of hypercholesterolemia patients

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Abstract

Tiwai (*Eleutherine americana* Merr) is a medical plant which has indicated potentially as hypercholesterolemia medicine. This research was aimed to investigate the cholesterol-lowering ability of *Tiwai* Herbal Drink (THD) in human bloods. Two weeks clinical observation was conducted with 12 volunteers. The volunteers are in age between 40 and 65 years old with a cholesterol level above 220 mg/dL, and are not currently using any cholesterol-lowering drugs. The study was divided into two periods, first week as a control (without giving THD) and the second week was treated with THD two times a day. THD were prepared in dried and brewed before drink. The serum cholesterol was measured before, during and after volunteers drank THD. The collections of blood sample are taken before and after treatment. Paired-sample t test were used to determine the comparability of value of THD and control group. The results conclude that consumption of THD for 7 days could reduce 5.33 mg/dL of total cholesterol in comparison to control but not HDL, LDL and Triacylglycerol. The ratio of HDL/LDL ranged between 0.26-0.56. The THD may reduce total cholesterol patients following a good diet with 2 times day consumption.

Introduction

Cholesterol metabolic changes can lead to hypercholesterolemia and it is major reason of cardiovascular disturbance for instance atherosclerosis and coronary heart disease (Steinberg, 2002). The increasing of per-capita income drives dietary pattern changes. Dietary pattern is related to the multiple nutritional problems and in many cases end up with unbalanced lipid profile (total blood cholesterol, LDL, HDL and triglycerides). The present study showed that pure anthocyanin resulted in a dual beneficial effect in lowering LDL-cholesterol and raising HDL-cholesterol concentrations. The use of natural antioxidants such as anthocyanin currently considered is safer because it is obtained from plant extracts. Recently, the biological activities of anthocyanin, such as antioxidant activity, protection from atherosclerosis and anti-carcinogenic activity have been investigated, and shown to have some beneficial effects in the treatment of diseases. Anthocyanins were also found to have many times more activity than common antioxidants such as ascorbate (Wang *et al*., 1997). Functional foods and nutraceuticals which lower plasma cholesterol level play an important role in reducing the risk of coronary heart disease. Development of these functional foods and nutraceuticals foods is of interest to both public and scientific community (Chen *et al*., 2011). Supplementation of Concord Grape Juice to a hypercholesterolemia diet may reduce the initiation of atherosclerosis and the enhanced sensitivity of platelets associated with the diet, care should be taken in extrapolating the findings and making recommendations for human health (Shanmuganayagam *et al.*, 2007).

Tiwai is often used by Dayak Kalimantan as cholesterol-lowering drugs, diabetes and cancer. A *Tiwai* plant is containing anthocyanin component 4.3 mg/100 g, 7.3 mg/100 g of vitamin C (Saragih, 2011) and according to (Ieyama *et al*., 2011) *tiwai* plants containing Eleutherol, Eleutherinoside A, B Eleuthoside potential as a drug. Based on the description above it is important to do a study to determine the effect of THD on lipid profile in patients.

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Material and Methods

Preparation of THD

Tiwai was obtained from local farmers. Tuber of *tiwai* herb washed with clean water to remove dirt or dust. It was cut to be 0.5 cm thick and it was wind-dried. After that, the dried herb was bags packed in the closed container.

Subject

Twelve volunteers were recruited from patient at Sungai Kapih Health Center. The study protocol was carefully explained to all subjects before they provided written informed consent as ethics. Subjects were eligible if they were between the ages of 40 and 65 years, cholesterol levels above 220 mg/dL, not currently using cholesterol-lowering drugs, willing to be a subject of study were all inclusion criteria. Exceptional criteria were subject with no history of hypercholesterolemia.

Study design

The study was conducted in September 2012. The study was divided into 2 periods, in first week was as a control (without giving THD) and second week two time consumption of THD per day, morning and afternoon. The experiment began with pretest for measuring basal serum cholesterol levels of all volunteers. Dietary consumption recalls includes carbohydrate, protein, fat, calories, cholesterol and fiber with subjects conducted 2 times for 24 hours during the first week before giving THD. Food intake was analyzed by using a computerized software program Nutri-survey and the results were compared with the table numbers nutrition Indonesia (RDA).

Collection and preparation of blood samples

Blood samples were collected at the beginning of the study pretest and posttest in the morning between 8:00 and 9:30 am. The samples were collected in tubes without anticoagulant. Blood was allowed to clot at room temperature for 30 minutes then centrifuged at 1500 x g for 10 minutes at room temperature. The serum cholesterol level (total cholesterol, triacylglycerol, LDL and HDL cholesterol) was measured by enzymatic colorimetric (Automated enzymatically analyzer Prestige 24i).

Statistical analysis

Paired-sample t tests confirmed by Wilcoxon rank-sum tests were used to determine the comparability of values for THD and control group as baseline, to compare the changes and percentage changes from baseline between treatment groups, and to compare dietary analyses indexes between groups. All statistical analyses were performed by using SPSS for Windows software version 17.

Results and Discussion

Subject characteristics

Subjects in this study is patient of health center in Sungai Kapih that has a history of hypercholesterolemia, cholesterol levels above 220 mg/dL, age of subjects between 40-49 years 6 (50%), 50-64 years 5 (41.6%) and 65 years 1 (8.4%). 10 (83.3%) woman and 2 (16.7%) man. Two (16.7%) subjects with a good nutritional status or normal Body Mass Index, overweight 6 (50%) and obesity 4 (33%).

Food intake

The average food intake can be shown in Table 1, the calorie intake before THD treatment is 2095.8 ± 275.76 kcal, while during THD treatments is 2045.6 ± 199.84 kcal. Protein intake before THD treatment was 71.9 ± 21.80 g meanwhile average protein intake during treatment THD was 72.5 ± 16.16 g. Fat intake has increased from 41.4 ± 16.36 g to 32.9 ± 10.04 g before and after treatment. The average of carbohydrate intake is increased from 356.3 ± 44.66 g to 381.5 ± 71.04 g. Meanwhile the average intake of fiber is decreased from 11.1 ± 2.45 g to 9.8 ± 2.66 g. The cholesterol intake was decrease from 195.6 ± 79.96 mg to 189.6 ± 55.56 mg before and after treatment. The consumption of a cholesterol-rich diet may increase serum cholesterol level.

Based on 2 x 24 hours food recall using Nutrisurvey shows the entire subject require carbohydrate intake, protein, calorie higher than 110% RDA. Fat intakes were sufficient in ranged 70-90%. The cholesterol intake is lower than 300 mg per day, however the fiber intake were low in ranged between 7 - 15 mg per day. All the subject was rarely consume vegetable and fruits to increase their fiber intake.

Serum lipid profile

t test resulted there is no significantly different between before THD treatment (test) and after treatment (posttest) were shown in Table 3. The average pretest total serum cholesterol was 282.08 ± 55.42 mg/dL, test 278.08 ± 65,81 mg/dL and posttest 276,75 ± 81.29 mg/dL (Table 2). Similar research conducted by Bo *et al*. (2012) also no significant differences were observed for endogenous DNA damage, peripheral arterial function and nitric oxide levels after consumption (at 1, 2,and 24 hours)

Table 1. Food intake

Food intake		Minimum	Maximum	Average
Calorie (kcal)	A	1798.8	2695.0	2095.8±275.76
	B	1791.6	2434.8	2045.6±199.84
Protein (g)	A	47.9	121.5	71.9±21.80
	B	51.3	101.8	72.5±16.16
Fat (g)	A	19.2	82.4	41.4±16.36
	B	23.9	56.0	32.9±10.04
Carbohydrate (g)	A	305.9	445.0	356.3±44.66
	B	327.0	594.8	381.5±71.04
Fiber (g)	A	7.5	15.0	11.1±2.45
	B	4.0	13.6	9.8±2.66
Cholesterol (mg)	A	95.4	284.7	189.6±55.56
	B	66.8	343.0	195.6±79.96

A (first week before THD treatment), B (during THD treatment)

Table 2. Blood lipid profile at pretest and posttest of THD for one week

	Total Cholesterol (mg/dL)	HDL Cholesterol (mg/dL)	LDL Cholesterol (mg/dL)	Triacylglycerol (mg/dL)
Pre	282.08±55.42	74.58±6.51	164.0±44.06	142.9±47.41
Test	278.08 ±65.81	70.17±9.0	166.9±50.59	137±53.57
Posttest	276.75±81.29	65.42±9.7	167.0±61.94	149.2±57.97

Tabel 3. Mean differences Lipid Profile before, during and after treatment

Lipid Cholesterol	Treatment	t	df	P-value
Cholesterol Total	before and during	0.580	11	0.574
	during and after	0.167	11	0.871
HDL	before and during	1.304	11	0.219
	during and after	1.216	11	0.249
LDL	before and during	0.532	11	0.606
	during and after	0.037	11	0.971
Triglyceride	before and during	0.565	11	0.584
	during and after	1.476	11	0.168

Table 4. Ratio Total Cholesterol, HDL and LDL profile before, during and after treatment

Treatment	Ratio Total Cholesterol /HDL	Ratio Triacylglycerol /HDL	Ratio HDL /LDL
Before	3.32 – 4.16	1.40 - 2.34	0.38 - 0.56
During	3.47 - 4.34	1.36 - 2.40	0.26 –0.52
After	3.50- 4.76	1.63 - 2.75	0.32- 0.53

blueberry.

HDL, Triglycerides, LDL showed there is no significant difference between before, during and treatment (Tabel 3). The anthocyanin compounds in THD is responsible for improve LDL and HDL. A study conducted by Qin *et al.* (2009) showed that anthocyanin supplementation in humans improves LDL- and HDL-cholesterol concentrations and enhances cellular cholesterol efflux to serum. These benefits may be due to the inhibition of cholesteryl ester transfer protein (CETP). Research done by Chang *et al.* (2006) showed extracts of Hibiscus flowers preventing LDL oxidation and macrophage death may contribute its anthocyanin components. However, it remains to be established whether dietary HAs have any effect on LDL oxidation and inhibited macrophage apoptosis *in vivo* and can thus affect the atherosclerotic process. Furthermore, the study of Basu *et al.* (2010) showed strawberry supplementation significantly decreased total and low-density lipoprotein cholesterol 5.8 ± 0.2 to 5.2 ± 0.2 mmol/L and 3.5 ± 0.2 to 3.1 ± 0.1 mmol/L, respectively. Strawberry supplementation further decreased circulating levels of vascular cell adhesion molecule-1 versus controls at 8 weeks ranged from 272.7 ± 17.4 to 223.0 ± 14.0 mg/mL).

HDL is expected increase after THD treatment; however it is not occur in this treatment. The ratio of HDL/LDL ranged between 0.26-0.56 Table 4. According to Hansel *et al.* (2007) revealed that hypercholesterolemia subjects (LDL-cholesterol concentrations ≥130 and ≤190 mg/dL; n = 194) consumed 2 low-fat portions of FM (fermented food) in the same meal daily for 6 week. Subjects were randomly assigned to 2 groups: low-fat FM enriched with 0.8 g PS ester per portion or control FM. Plasma LDL-cholesterol concentrations were reduced by 9.5% and 7.8% after 3 and 6 week, respectively, in the 1.6-g/d PS group compared with the control group, whereas plasma triacylglycerol and HDL-cholesterol concentrations were not significantly affected. Recently, other groups have also found that plant food rich in phytochemicals produced beneficial effects in improving the lipid profile. Furthermore, Hansen *et al.* (2005) found that moderate red wine consumption for 4 week was associated with a relative increase of 11–16% in HDL cholesterol in healthy human subjects. In another report, berry consumption increased serum HDL concentrations by 5.2% (Erlund *et al.*, 2008). According to Zern *et al.* (2005) showed that lyophilized grape powder modestly lowered plasma LDL cholesterol and apo-B and E in both pre- and postmenopausal women.

Some factors may occur during test and makes the sample is not significantly different. First, due to non-compliance samples to maintain a diet or food intake during treatment THD, the treatment became not optimal. In addition, the subjects had diabetes mellitus and hypercholesterolemia in a long time with a history of cholesterol 300-400 mg/dl.

The second factor, the number of sample is small (12 subjects) and the period of test is short (7 days) as well, compared to the study done by Darmono (2010) has shown that total blood cholesterol levels dropped significantly i.e. 21.13 mg/dL after treatment with Roselle tea for 6 weeks. A series of earlier studies have reported that the maximum time of anthocyanin's in plasma is 15–60 min, and excretion is complete within 6–8 h (McGhie and Walton, 2007).

Although the test results showed no statistically significant differences between lipid profiles before and after THD treatment for one week, yet from some of the subjects delivered testimony during drinking herbal tea to feel better because of cramps or a tingling feeling that once felt after THD drinking symptoms have started to decrease and some even say that the symptoms were no longer perceived. One of the subjects had checks his blood sugar levels before

and after the intervention, the results showed blood sugar levels decreased, from 250 mg to 150 mg/dL without taking any medication.

Iyeama *et al.* (2011) revealed that amount and the inhibitory activity of each naphthalene in the whole extracts, the bulb of *E. americana* inhibitory activity against a-glycosidase might be a result of compound 2 (IC_{50} = 0.5 mm, yield = 5 mg/50 g plant sample, with a characteristic structure which has never been found in other AGIs. AGIs play an important role for the treatment of diabetes. Inhibitory activity of all isolates from *E. americana* are relatively low compared to commercially available glycosidase inhibitors, such as acarbose, this research results may suggest novel alternatives for diabetes treatment management.

The results showed no significant effect after treatment THD on levels of total cholesterol, HDL, LDL, and Triglycerides. However, drinking THD twice daily in 7 days can lower total cholesterol by 5.33 mg/dL. THD contains anthocyanin whereas largest group of water-soluble pigments in the plant. In recent years, numerous studies have shown that anthocyanin's display a wide range of biological activities including antioxidant, anti-inflammatory, antimicrobial and anti-carcinogenic activities; improvement of vision; induction of apoptosis; and neuroprotective effects (Mazza, 2007).

Conclusion

Reduction of total cholesterol, LDL, HDL and Triacylglycerol were not significantly different. The large variability of subject may cause the significances of result. The result can conclude that twice a day consume of THD only reduce total cholesterol of 5.33mg/dL but not HDL, LDL and Triacylglycerol.

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References

Basu, A., Fu, X.D., Wilkinson, M., Simmons, B., Wu, M., Betts, N.M., Du, M. and Lyons, T. 2010. Strawberries decrease atherosclerotic markers in subjects with metabolic syndrome. Nutrition Research 30:462–469.

Bo, C.D., Riso, P., Campolo, J., Møller, P., Loft, S., Zacas, D.K., Brambilla, Rizolo, A. and Porrini, M. 2013. A single portion of blueberry (*Vaccinium corymbosum* L) improves protection against DNA damage but not vascular function in healthy male volunteers. Nutrition Research 33:220–227.

Chang, Y.C., Huang, K.X., Huang, A.C, Ho, Y.C. and Wang, C.J. 2006. Hibiscus anthocyanins-rich extract inhibited LDL oxidation and oxLDL-mediated macrophages apoptosis. Food and Chemical Toxicology 44:1015–1023

Chen, Y.Z., Ma, K.Y., Liang, Y., Peng, C. and Zuo, Y. 2011. Role and classification of cholesterol-lowering functional foods. Journal of Functional Foods 3:61-69

Darmono, A.M. 2010. Effect of Tea Rosella against total Cholesterol levels decrease in women post menopause. Semarang: Diponegoro University.

Erlund, I., Koli, R., Alfthan, G., Marniemi, J., Puukka, P., Mustonen, P., Mattilla, P. and Jula, A. 2008. Favorable effects of berry consumption on platelet function, blood pressure, and HDL cholesterol. American Journal of Clinical Nutrition 87:323–331.

Hansel, B., Nicolle, C., Lalanne, F., Tondu, F., Lassel, T., Donazzolo, Ferrières, J., Krempf , M., Schlienger, J.L., Verges, B., Chapman, M.J. and Bruckert, E. 2007. Effect of low-fat, fermented milk enriched with plant sterols on serum lipid profile and oxidative stress in moderate hypercholesterolemia. American Journal of Clinical Nutrition 86(3): 790-796.

Hansen, A.S., Marckmann, P., Dragsted, L.O., Nielsen, F.I., Nielsen, S.E. and Gronbaek, M. 2005. Effect of red wine and red grape extract on blood lipids, haemostatic factors, and other risk factors for cardiovascular disease. European Journal of Clinical Nutrition 59:449–455.

Ieyama, T., Maria D.P.T., Puteri, G. and Kawabata, J. 2011. a-Glucosidase inhibitors from the bulb of Eleutherine Americana. Food Chemistry 128:308–311

Mazza, G. 2007. Anthocyanins and heart health, Ann. 1st. Super. Sanita 43:369–374

McGhie, T.K. and Walton, M.C. 2007. The bioavailability and absorption of anthocyanins: towards a better understanding. Molecular Nutrition and Food Research 51:702–713.

Qin, Y., Xia, M., Ma, J., Hao, Y.T., Liu, J., Mou, H.Y.,Cao, L. and Ling, W.H. 2009. Anthocyanin supplementation improves serum LDL- and HDL-cholesterol concentrations associated with the inhibition of cholesteryl ester transfer protein in dyslipidemic subjects. American Journal of Clinical Nutrition 90: 485-492.

Saragih, B. 2011. Functional drinks herbal bags *tiwai* (*Eleutherina Americana* Merr). Journal of Research and Development area Gerbang Etam 5(1): 15-21.

Shanmuganayagam, D., Warner, T.F., Krueger, C.G., Reed, J.D. and Folts, J.D. 2007. Concord grape juice attenuates platelet aggregation, serum cholesterol and development of atheroma in hypercholesterolemic rabbits. Atherosclerosis 190: 135–142

Steinberg, D. 2002. Atherogenesis in perspective: hypercholesterolemia and inflammation as partners in crime. Natural Medicine 8:1211–1217.

Wang, H., Cao, G. and Prior, R.L. 1997. Oxygen radical absorbing capacity of anthocyanins. Journal of

Agricultural and Food Chemistry 45: 302–309.

Zern, T.L., Wood, R.J., Greene, C., West, K.L., Liu, Y., Aggarwal, D., Shachter, N.S. and Fernandez, L.M. 2005. Grape polyphenols exert a cardioprotective effect in pre- and postmenopausal women by lowering plasma lipids and reducing oxidative stress. Journal of Nutrition 135:1911–1917.