

ORIGINAL ARTICLE

Effect of Tender Coconut Water on Blood Lipid Levels in High Fat Diet Fed Male Rats*Siti Thomas Z^{1*}, Danis Pertiwi², Bagus S.A³, Nuri S³, Brillian Jelita E.M³, Alfiza N.S³**¹Department of Public Health Sciences, ²Department of Clinical Pathology, ³Student of Faculty of Medicine, Sultan Agung Islamic University (UNISSULA) Semarang***Abstract:**

Background: Dyslipidemia is a lipid metabolic disorder characterized by an increased levels of Triglycerides (TG), Total Cholesterol (TC), Low-Density Lipoprotein (LDL) cholesterol, and decreased High-Density Lipoprotein (HDL) cholesterol levels.

Aim and Objectives: To investigate the effect of the administration of tender coconut water on blood lipid levels in male wistar rats. **Material and Methods:** Twenty four male wistar rats were randomly divided into four group of six animals each. Group K1 served as control, Group K2 was fed quail egg yolk (1g/rat/day). Group K3 was given coconut water (8 ml/rat/day) orally. Group K4 was given simvastatin (0.18 mg /rat /day) orally for 21 days. **Results:** that increased levels of TC, LDL and TG with decreased level of HDL were shown in group K2 compared with group K1. Decreased levels of TC, LDL and TG levels were observed significantly in both groups K3 and K4 than K2. Administration of 8ml coconut (200g/kg b w for 21 days) could lower TC, LDL and TG levels with an increased HDL level ($p < 0.05$). **Conclusion:** The administration of tender coconut water has shown lower total cholesterol, LDL cholesterol and triglycerides levels and increase HDL serum levels in male wistar rats fed a high fat diet.

Keywords: Tender Coconut Water, Total Cholesterol, LDL, HDL, Triglycerides

Introduction:

Dyslipidemia is a lipid metabolic disorder characterized by increased levels of Triglycerides (TG), Total Cholesterol (TC), Low-Density

Lipoprotein (LDL) cholesterol, and decreased High-Density Lipoprotein (HDL) cholesterol levels [1].

Blood lipids including TC, LDL, HDL and TG are needed to maintain the healthy status of cells, however high levels of lipid profile increase the risk of heart disease. Ideally, TC should be < 200 mg / dL, LDL < 130 mg/dL, HDL > 40 mg/dL and TG < 150 mg/dL. Patients with hyperlipidemia have a higher risk to develop Coronary Heart Disease (CHD) [2]. The main risk factors for cardiovascular disease includes dyslipidemia, high levels of TC, LDL with low level of HDL which may lead to atherosclerosis. CHD is a term commonly used to describe the formation or accumulation of plaque in coronary arteries causing heart attacks [3]. Major health problem in developed and developing countries includes heart and blood vessels related diseases. Annually, these lipid abnormalities increases high mortality in the world. In Indonesia, the number of heart and blood vessel diseases continues to increase [4]. Till date, for the treatment and prevention of hyperlipidemia, synthetic drugs have been used. But, they are costly, non economic and non affordable for patients. So, alternative complementary curative efforts or preventive measures are necessary to minimize lipid abnormalities in patients that CHD could be prevented [5].

Some studies have suggested that foods containing natural antioxidants can be used as alternative strategy to reduce the prevalence of degenerative disease [6]. Tender Coconut Water (TCW) is a nutritious healthy beverage that is easily available, as natural isotonic beverage containing nearly similar to plasma [7]. Indonesia is a country with the world's leading palm oil producer [8]. The macro and micro nutrients contained in coconut water reduce lipid profile, protecting the heart and liver [9]. TCW has a therapeutic effect as it contains various nutrients such as minerals, vitamins, antioxidants, amino acids, enzymes and growth hormones [10]. Coconut water is called "fluid of life", referred to as TCW. It is called tender because it is soft like jelly [7]. Previous research studies have shown that the administration of tender coconut water as much as 4 ml/ 100 g body weight in rats for 3 weeks can reduce levels of LDL, TGA and increase HDL levels in mice fed a high-fructose diet [11].

Material and Methods:

This study was designed as experimental research with Post-Test Control Group Design [14].

Ethical Clearance:

This study received ethical clearance from the Bioethics Committee of Medical Research / Medical Faculty of Sultan Agung Islamic University, Semarang (263 / XI / 2015 / Bioethics Commission).

Tender Coconut Water:

TCW used was coconut *varieties viridis* (green coconut) aged 5-7 months old and obtained from several different sites. The appearance of coconut flesh (endosperm) was soft, jellylike thin. The administered dose was chosen as 8ml/200g of body weight/day for 21 days [11]. Examination of

the composition of tender coconut water was carried in LPPT Gajah Mada University Yogyakarta with TLC and HPLC methods shown in Table 1.

Table 1: Composition of Tender Coconut Water (TCW) used for Study

Constituents	Levels
Asam Amino (µg/mL)	
- L- Aspartic	115.60
- L-Glutamic	316.60
- L- Glutamin	<0.05
- L-Threonine	45.80
- L-Glycine	34.2
- L-Arginine	43
- L-Alanine	218.20
- L-Tyrosine	49
- L-Thryptophan + L-Methionine	9.8 11.2
- L-Valine	253
- L-Phenylalanine	<0.05
- L-Isoleucine	<0.04
- L-Leucine	<0.14
- L-Lycine	99.00
- L-Histidine + Serine	
Vitamin C (mg/L)	32.5
Polyphenol (mg/mL)	7.53
Selenium (µg/Kg)	<0.01
Mineral (mg/Kg)	
- Cu (tembaga)	0.17
- Fe (besi)	2.19
- Mg (Magnesium)	225.35
- Mn (Mangan)	4.09
- Zn (Seng)	1.91
- Na (Natrium)	601.47
- K (Kalium)	1668.37
- P (Phospor)	64.11

Source : LPPT Gajah Mada University Yogyakarta, 2016

Experimental Animals:

The high-fat diet was the quail egg yolk. The quail egg yolk was administered orally with feeding tube at the dose of 1g/day.

Male wistar rats were chosen as aged 2 months, weighing between 180-220g. After one day adaptation, 24 male rats randomly were divided into four groups of six rats each and fed the following diet:

Statistical analysis:

Statistical analysis was done by one-way ANOVA with Tukey's test. The results were presented as the mean value ± SD for the control and treated groups. Difference among the means for the groups were assayed using Duncan's Multiple Range Test to determine which mean values were significantly different at p < 0.05 [15].

Group 1 (K1) :	fed standard diet + distilled water <i>ad libitum</i> (Control Group)
Group 2 (K2) :	fed standard diet + distilled water <i>ad libitum</i> + 1g /rats/quail egg yolk (Negative Control Group)
Group 3 (K3) :	fed standard diet + distilled water <i>ad libitum</i> +1g /rats/day quail egg yolk + 8 ml/rats/day of tender coconut water (Treated Group)
Group 4 (K4) :	fed standard diet+ distilled water <i>ad libitum</i> +1g / rats/ day quail egg yolk + 0.18 mg/rats/day of simvastatin (Positive Control Group)

The duration of experimental period was continued for 21 days. On 22nd day, rats were previously fasted for 12 hours and blood were collected by retro-orbital puncture and serum were analyzed for TC, LDL, HDL and TG at PAU Laboratory Gadjah Mada University, Yogyakarta.

Result:

The effect of tender coconut water on TC, LDL cholesterol, HDL and TG levels shown in Table 2. Table 2 shows that increased levels of TC, LDL and TG with decreased level of HDL were shown in group K2 compared with control group K1.

Table 2: Effects of TCW on mean TC, LDL, HDL and TG levels in groups

Group	TC level (mg/dL)	LDL level (mg/dL)	HDL level (mg/dL)	TG level (mg/dL)
K1	85.18±1.66	24.28±1.18	75.61±2.81	40.44±4.78
K2	181.95±4.19*	74.34±2.07*	31.71±5.71*	108.5±2.93*
K3	136.79±2.37*	42.72±0.65*	45.85±2.54*	63.17±1.56*
K4	107.88±2.74*	34.70±1.01*	60.98±3.64*	54.67±2.28*

*Significantly different from control (p<0.05)

Decreased levels of TC, LDL and TG levels were observed significantly in both groups of TCW fed K3 and positive control K4 than negative group K2. Administration of 8ml coconut (200g/kgbw for 21 days) could lower TC, LDL and TG levels with an increased HDL level ($p < 0.05$).

Discussion:

Coconut water used in this study was TCW from coconut cultivar group, *vareitas viridis* (*green coconut*), 5-7 months, total sugar contents increases from 5 months to reach the maximum at 7 months [16]. Results have shown that the administration of TCW at a dose of 8ml/200g body weight/day for 21 days significantly decreased the levels of TC, LDL and TG and increased HDL levels in rat fed high fat diet compared to control. The findings of this present study are in conformity with other studies by Sandhya *et al.* (2006) [12], Johnkennedy *et al.* (2013) [13], Agbafor *et al.* [17] and Mohammed [18]. Epidemiological studies have show that high levels of HDL have protective value against cardiovascular diseases such as Ischemic stroke and Myocardial Infection [17]. Coconut water contains several biologically active components including free amino acid L-arginine, ascorbic acid, minerals such as calcium, magnesium and potassium [19]. Similarly, the presence of L-arginine in coconut water could have a cardio protective effect through its production of nitric oxide, which favours vasorelaxation. A similar hypolipidemic effect of coconut water and lovostatin (a lipid-lowering drug) was detected in rats fed a fat/cholesterol-enriched diet [20].

The ability of TCW in lowering total cholesterol levels was due to the compounds polyphenols, vitamin C, and L-arginine [11,21]. The high content of L-arginine in tender coconut water can be used to reduce the free radical generation, increase antioxidant activity and inhibit the process of lipid peroxidation [16].

Constituents minerals as calcium, magnesium and potassium decrease serum cholesterol and triglycerides levels. Magnesium intake prevents cholesterol deposition in the aortas of mice. The high potassium diet, by protecting endothel cells can greatly decrease the cholesterol ester deposition during hypercholesterolemia [9].

Polyphenols in lowering total cholesterol levels play role in two ways by reducing fat absorption in the digestive system and increase fat excretion into feces, so less amount of cholesterol absorption and in the circulation. TCW contains both ascorbic acid and vitamins of B group those are nicotinic acid, pantothenic acid, biotin, riboflavin, folic acid and thiamine. Lower blood levels of vitamin B6 and folate can increase the risk for atherosclerosis and other vascular diseases. The concentration of ascorbic acid ranges from 2.2 to 3.7 mg per ml, which gradually diminishes as the kernel surrounding the water begins to harden. Vitamins play a vital role in many metabolic pathways, healthy cellular activity which therefore can guide against infections. It is also essential for the structure of bones, cartilage, muscle and blood vessels, and maintenance of capillaries and gums and the absorption of iron. Vitamin containing fruits helps to reduce risk of cardiovascular disease, stroke, and cancer [20].

Vitamin C is responsible for keeping the activity of cholesterol 7 alpha-hydroxylating system containing cytochrome P-450. Decrease in TC and reduced accumulation of TC in the liver, blood plasma, and the arteries caused by the increased activity of cholesterol 7 alpha-hydroxylating system containing cytochrome P-450 thus stimulating change cholesterol into bile acids increased [22]. The effect on the stimulation of bile acid sequestrants like cholestyramine or pectin strengthen the effects of vitamin C to

reduce the solubility of fat and block the absorption of cholesterol. Bile acids are typically 95% reabsorbed also inhibited. Bile acid synthesis will also be encouraged which will compete with cholesterol synthesis in the liver [22].

Vitamin C in tender coconut water proved to have protective activity on HDL cholesterol, playing a role in the process of reverse cholesterol transport. Lecithin transforms cholesterol HDL into mature cholesterol ester. Cholesterol ester in HDL is transferred back to the liver and is then converted into bile acids. Apolipoprotein AI in HDL may alter HDL is oxidized to lecithin. Vitamin C can increase the ability of Apolipoprotein AI, so that the process of increasing reverse cholesterol transport [23].

HDL can prevent the oxidation of LDL, and become a free radical scavenger through the function of an enzyme called HDL-associated paraoxonase. Vitamin C can prevent the loss of activity paraoksonase, so that cell damage due to reduced oxidized LDL [23]. Vitamin C can increase the enzyme in endothelial Nitric Oxide Synthase (NOS) resulting in the formation of nitric oxide in the blood vessels will be increased, where the function of nitric oxide is a vasodilator on the blood vessels and inhibit the aggregation palatelet of the blood vessels. An increase of the amount of nitric oxide in the endothelium will be

instrumental in increasing the proliferation of LPL enzyme in serum, which later this enzyme will hydrolyze triglycerides into glycerol and free fatty acids [24]. Vitamin C also can reduce the amount of cholesterol and TG in rats and can increase intracellular tetrahydrobiopterin (BH₄) and subsequent enhancement of NOS activity [9]. Compound L-Arginine does not contribute directly to the reduction in total cholesterol levels, but the defects of Nitric Oxide (NO) signaling that minimized the benefits of these compounds so Xanthine Oxide (XO) of supraoxide and lipoprotein oxidation will not be formed so that the dysfunction endothelial can be reduced [23]. Coconut water is the richest natural source of cytokinins. Cytokinins can retard the effect of aging in plant cell as well as human cell, produce beneficial effects, and reduce the risk of cardiovascular diseases [13, 25]. They inhibit platelet clots that may lead to heart attacks and strokes. Cytokinins has potential for treatment of degenerative brain diseases like demential and Alzheimer's disease. Coconut water has lipid lowering effect [25].

Conclusion:

The oral administration of TCW could lower TC, LDL and TG levels and increase HDL serum levels in male rats fed high fat diet.

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