

Research Article

The Effect of Chayote (*Sechium edule*) Extract on Malondialdehyde Levels in Male Wistar Rats (*Rattus norvegicus*) Induced by Atherogenic Feed Effects of *Sechium edule* on Malondialdehyde in Atherogenic Cases

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ABSTRACT

Hypercholesterolemia and oxidative stress affect lipid peroxidation and increase malondialdehyde (MDA). *Sechium edule* contains flavonoids and phenols that have been shown to have antioxidant activity that can reduce MDA levels. To prove this, Wistar rats (*Rattus norvegicus*) fed with an experimental atherogenic diet were used as a model. The rats were divided into four groups, one group with an atherogenic diet, and the other three received an atherogenic diet and *Sechium edule* extract at doses of 20, 40, and 80 (mg/day) for 42 days. The MDA value was calculated using the Thiobarbituric acid reactive substance (TBARS) method. The results showed that *Sechium edule* extract with a topical dose of 40 mg/day effectively reduced maize MDA levels to 75.3% ($p = 0.001$).

Keywords: MDA, atherogenic feed, *Sechium edule* extract

Introduction

Atherosclerosis is a condition in which the endothelial walls of blood vessels thicken due to the presence of plaque. Fibrous plaque is formed by the accumulation of monocytes, macrophages, foam cells, connective tissue, tissue debris, and cholesterol crystals [1]. The blood vessels and atherosclerotic plaques gradually invade the endothelium and cause ischemia. This condition is strongly influenced by oxidative stress, especially the imbalance between free radicals and antioxidants [2].

The main risk factor for atherosclerosis is hypercholesterolemia. Hypercholesterolemia is characterized by an increase in low-density lipoprotein (LDL) levels to be > 190 mg/dl. Approximately 70% of cholesterol is transported as LDL [3], which is easily oxidized by free radicals and is closely involved in lipid peroxidation, particularly

malondialdehyde (MDA) [4]. MDA levels can be used as a biomarker of lipid peroxidation and as a definition of chemical oxygen stress levels [5].

Malondialdehyde, one of the end products of advanced lipid peroxidation (ALE), can react with cellular and tissue proteins to form protein complement products. The ALE protein converts LDL to a scavenger receptor on foam cell macrophages. Foam cells that accumulate in endothelial tissue turn into adipose tissue, which leads to atheroma [6].

Chayote contains bioactive compounds, such as flavonoids, phenols, vitamin C, carotenoids, and antioxidants, which are useful as antibacterial, antioxidant, and anti-inflammatory [7]. The types of flavonoids in chayote are flavones and flavanols with 3 c-glycosides and 5 O-glycosides, which can lower total cholesterol by inhibiting cholesterol in the intestine [8]. As antioxidants, phenols donate -OCH₃ and -OH groups to form stable compounds

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against free radicals [9]. Chayote fruit extract can freely inhibit and lower LDL levels as it contains high amounts of antioxidants [10]. Chayote also contains the same amount of phenolics and flavonoid levels as kiwi [11].

Male Wistar rats (*Rattus norvegicus*) are used in atherosclerosis studies as they can be observed in a short time [12]. Feeding high fiber and fat diets can increase LDL [13] and MDA [11] levels in rats.

Methods

The research was conducted at the Biomedical Laboratory, Faculty of Medicine, University of Muhammadiyah Malang. A total of 16 male Wistar rats (*Rattus norvegicus*), aged 2-3 months and with a body weight of 160-240 g, were divided into four groups of three with 1 reserve in each group.

Chayote (*Sechium edule*) extract was produced at the Chemical Engineering Laboratory, State University of Malang. In this study, *Sechium edule* var. *viren levis* was chosen and taken from Batu, Malang. Chayote extract made from dried chayote fruit, subsequently ground into powder. The powder macerated with 95% ethanol for 24 hours afterwards. The residue was macerated repeatedly until the solution became clear. The liquid extract was concentrated on a rotary evaporator at room temperature [14].

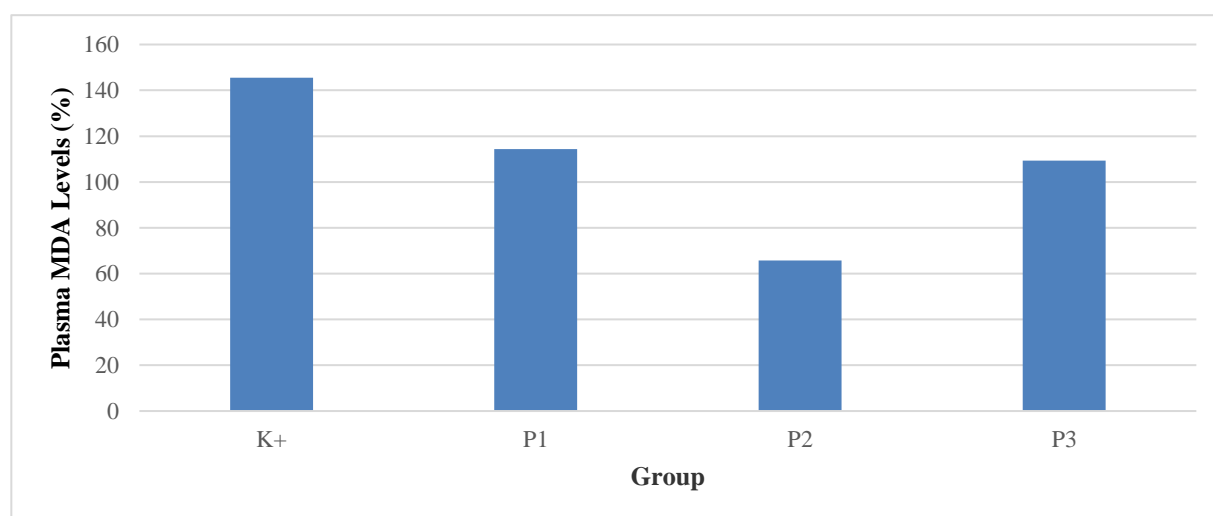
The extract was given orally at a dose of 20 mg/200 g BW/day, 40 mg/200 g BW/day, and 80 mg/200 g BW/day for 42 days. Besides, Atherogenic feed (20 g) mixed with standard feed was given once a day for 42 days. The atherogenic diet was made of 5% goat fat, 0.5% cholic acid, 2% quail eggs, and 10% fat.

Rats were adapted for seven days by being given standard feed and water, then divided into:

1. K+ group (positive control): standard feed mixed with atherogenic diet without chayote extract (*Sechium edule*) for 42 days.
2. Group P1: standard feed mixed with atherogenic diet and Chayote extract (20mg/200 gr BW/day) for 42 days.
3. Group P2: standard feed mixed with atherogenic diet and Chayote extract (40 mg/200 gr BW/day) for 42 days.
4. Group P3: standard feed mixed with atherogenic diet and Chayote extract (80 mg/200 gr BW/day) for 42 days.

After 42 days, rats were anesthetized with chloroform and dissected to draw 2 ml of blood from the left ventricle. The blood serum was used to measure the MDA levels in the Physiology Laboratory, Faculty of Health, Brawijaya University, using the TBARS method. The data were analyzed using one-way ANOVA, post hoc, and linear regression tests.

Results and Discussion



(Processed primary data, 2020)

Figure 1. Average Plasma MDA levels

Without chayote extract, rats with an atherogenic diet had the highest MDA levels of 145,493 ng/ml at the end of the treatment. According to Zhenxiang et al. [11], the normal MDA level is 74,793 ng/ml. Therefore, feeding a high fat atherogenic diet in this study increased normal MDA levels by 20 g/day in 42 days. The addition of lard can increase cholesterol content as it contains high saturated fatty acids (38-43%) and cholesterol

[15]. Another study also found that giving a diet consisting of oil, cholic acid, and cholesterol powder for 42 days could increase MDA levels in rats [11].

An atherogenic diet increases low-density lipoprotein (LDL) and free radicals [16]. The abundance of free radicals can boost ROS production due to oxidative stress and trigger lipid peroxidation, resulting in MDA [16].

Table 1. One-way ANOVA test results

Variable	Sig.
Plasma MDA Level	0.001

(Processed primary data, 2020)

The results of the one-way ANOVA test showed a significance level (p) of 0.001. P value <0.05 means that the null hypothesis (H0) is rejected and hypothesis 1 (H1) is accepted. This

indicates that there is at least one difference in the mean plasma MDA levels between the treatment and the control groups.

Table 2. Bonferroni post-hoc test results

(I) Treatment	(J) Treatment	Sig.	Explanation
K+	P1	0.339	Not significantly different
	P2	0.001	Significantly different
	P3	0.141	Not significantly different

(Processed primary data, 2020)

The post-hoc test (sig = 0.001) revealed that a dose of 40 mg/200 g BW/day of chayote extract (P2 group) could significantly reduce plasma MDA levels compared to the other two doses. This is supported by research by Agustini et al. [17] and Neeraja et al. [10], who found that administration of 40 mg/200 g BW/day of chayote fruit extract in rats reduced total cholesterol, triglycerides, and LDL levels. In addition to chayote, kiwi has also been shown to reduce MDA levels at the same dose as it contains phenols and flavonoids.

Chayote contains flavonoids in the form of flavones and flavanols, which contain 3 c-glycosides and 5 o-glycosides that can inhibit

cholesterol in the intestine. Flavonoids can also inhibit the formation of ROS, enhance antioxidant defense and regulation, and protect lipid membranes from oxidative damage, which can ultimately hinder MDA production [18]. As antioxidants, phenols donate -OCH₃ and -OH groups to form stable compounds from free radicals. In addition to reducing cholesterol esterification, phenols also inhibit the synthesis of Apo B-48 and Apo B-100 in hepatic enterocytes, thereby inhibiting LDL oxidation and lipid peroxidation [19].

The dose of 20 mg/200 g BW/day chayote extract did not affect MDA levels in the P1 group. This is consistent with the research of Agustini et

al. [17], who found that 20 mg/200g/day of chayote extract was ineffective in decreasing total cholesterol, triglyceride, and LDL levels. Meanwhile, the same amount of kiwi extract was proven can reduce MDA levels in hypercholesterolemic rats [11]. This difference may occur because the absorption coefficient of the extract is different in each species. The absorption factor is influenced by body size, age, stamina, and endurance. The absence of effects may also be due to the usage of low or less-than-ideal concentrations.

Additionally, using a larger dose (80 mg/200 g BW/day) does not ensure that it will have a more significant impact than using a lower dose (40 mg/200 g BW/day). It may be due to

keratinization, particularly the dose-responsive phenomenon, where small doses produce a stimulant effect, whereas large doses produce an inhibitory effect. Exogenous antioxidants cannot distinguish between parallel physiological oxidations. Therefore, when antioxidants are plentiful, they can also neutralize the physiological oxidants involved in this signaling pathway. Under these conditions, high doses of chayote extract produced oxidants and prevented MDA levels from rising. Based on the regression test, the administration of chayote extracts reduced MDA levels by 75.3% in mice, and 24.7% was influenced by other unexplored factors.

Table 3. Dummy Variable Linear Regression Test Results

Model	Unstandardized Coefficients		Std. Coefficients Beta	T	Sign
	B	Std. Error			
1 Constant	144.454	13.196		10.947	0.000
Dummy 1	-30.152	17.036	-0.407	-1.770	0.111
Dummy 2	-78.758	16.162	-1.164	-4.873	0.001
Dummy 3	-35.090	16.162	-0.519	-2.171	0.058

(Processed primary data, 2020)

The absorption of chayote extract is influenced by endogenous metabolism and levels of antioxidants released from the body, such as SOD (Superoxide dismutase), GSH (Glutathion), and catalase, which play a direct role in inhibiting the formation of free radicals. External factors, especially the cage environment, are the limitations of this study. The recommended room temperature for rats is 20-28 °C. However, there was no temperature gauge installed in the enclosure area of this study, so the exact temperature was unknown. Room temperature below 20 °C increased the metabolism of rats, while room temperature above 28 °C disrupted their thermoregulatory system [19]. The procedures used in this study, such as holding and lifting rats during cage repair, increased stress levels and decreased immunity in rats [21].

Conclusion

Chayote extract (*Sechium edule*) can reduce MDA levels in male Wistar rats (*Rattus norvegicus*) induced by an atherogenic diet.

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