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The Atherogenic Index of Plasma Treated with Red Guava (*Psidium guajava* L.)

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Abstract. Red guava contains fiber especially high soluble fiber (pectin). Soluble fiber in the body is hypocholesterolemia. This research aimed to know the effect of administering red guava on AIP (Atherogenic Index of Plasma) in hypocholesterolemia rats. This research was a pure experiment using the design of randomized pretest-posttest control group design to Sprague Dawley rats made into hypocholesterolemia. The rats were divided into four groups: group 1 given standard food, group 2, 3, and 4 given high cholesterol food. Group 3 and 4 were added with red guava powder as many as 0.72 g and pectin equal to the one in red guava powder. Lipid profile was checked by using Enzymatic Colorimetric Test CHOP-PAP and GPO-PAP including the rate of total cholesterol, LDL, HDL, and triglycerides. AIP was counted based on $\log(TG/HDL)$. The data analysis used t-test and ANOVA with a 0.05 significance level. The administration of red guava in hypocholesterolemia rats was significantly proven to be able to lower lipid profile by lowering total cholesterol (32%), LDL (43%), triglycerides (18%), and by increasing HDL (18%). These results were similar to the decrease by administering pectin as many as 32%, 47%, and 20% respectively. The results of counting AIP showed that the administration of red guava and pectin could lower AIP score significantly as many as 0.16 and 0.17 respectively which increased in the groups getting high cholesterol food. The administration of red guava in hypocholesterolemia rats was proven to be able to lower AIP.

Keywords: Red guava, soluble fiber, lipid profile, AIP

1. Introduction

Guava (*Psidium guajava* L.) is a kind of tropical plants from America, especially in Brazil and Antilles. Today, guava spreads until Southeast Asia, including Indonesia with many varieties and local names such as Pasar Minggu guava, Bangkok guava, Palembang guava, sukun guava, apple guava, sari guava, red guava, getas red guava. Guava is widely cultivated in Central Java, especially in Kendal Regency [1].

Generally, guava is consumed as a fruit, although it actually has more benefits. Many people use its leave as a traditional medicine for many illnesses [2], as well as the fruit which is believed to be



able to overcome many health problems, such as diarrhea medicine, antiinflammation, antidiabetic, cardioactive and antioxidant source. Red guava is also thought to help cure dengue fever [3].

Guava contains high dietary fiber, especially soluble fiber (pectin) besides vitamin C which is twice more compared to sweet oranges. The soluble fiber content in guava is the highest of other tropical fruits and higher than fiber content in cerealia family [4]. Dietary fiber, especially soluble fiber (pectin,) in the body is hypocholesterolemia which can fight against coronary heart disease (CHD) by decreasing cholesterol [5]. Maryanto and Muis said that guava (white flesh part) can reduce the rate of total cholesterol, LDL-cholesterol and triglycerides significantly [6].

Coronary heart disease develops as a result of an interaction among many risk factors, particularly the abnormality of lipid profile in the blood (dyslipidemia) consisting of high total cholesterol, LDL (hypercholesterolemic), triglycerides, and low HDL. The common action to know the risk of CHD is by checking blood lipid profile consisting of total cholesterol, LDL, HDL, and triglycerides [7].

Until now, LDL cholesterol is the most common risk factor used as an indicator of CHD. Epidemiological and clinical studies consistently show that the concentration of LDL cholesterol in plasma relates to an increase in CHD risk; however, LDL rate is not the only predictor, triglycerides rate is also needed to be observed [8]. An epidemiological study for the group age of more than 45 years old in Semarang showed that triglycerides rate similar to or more than 150 mg/dL had the risk 2.8 times bigger to get CHD than the ones having triglycerides less than 150 mg/dL. CHD risk because of triglycerides will increase in the people consuming high simple carbohydrates [9]. Another predictor to know the risk of CHD is by counting AIP (atherogenic index of plasma). AIP is a number showing log value (TG/HDL). This index shows that the increase in triglycerides will increase the risk of CHD. A reference is made by Froblich and Dobiasova as follows, $AIP < 0.11 = \text{low risk}$; $0.11 - 0.21 = \text{intermediate risk}$; and $> 0.21 = \text{increasing risk}$ [10].

This research aimed to know the effect of administering red guava on AIP (atherogenic index of plasma) in hypercholesterolemic rats. The advantage of this research is to make scientific information about the potentials of red guava in lowering the risk factor of getting CHD by counting AIP.

2. Methods

This research was a real experiment using randomized pre-test post-test control group design. The subjects were 28 male Sprague Dawley rats with the age of two months old obtained from LPPT UGM having weight about 150-160 grams. The rats were divided into four groups: group 1 given the standard food, group 2, 3, and 4 given high cholesterol food. Group 3 and 4 were added with red guava powder as many as 0.72 grams and pectin equal to the pectin content in getas red guava. To measure the minimal sample of each group the Federer formula was used and got 7 for each group [11].

The rats were adapted, given the food of AIN 93 standard for 3 days, continued by normal and high food of AIN 93 standard. High cholesterol food was given by adding 1% cholesterol crystal and sodium colat for 14 days (the rats became hypocholesterolemic). Then, they were randomized and grouped in an individual cage and continued for 28 days. The rats were fed as many as 13 g/day averagely for all groups [12]. This treatment was done by giving the water suspension of red guava powder using a tube. This treatment referred to the need of dietary fiber for a male adult according to FDA recommendation [13] and was converted for 200 grams rats according to Laurence and Bocharch as many as 0.72 grams [14].

Red guava powder was made by crushing dried flesh into powder, then it was analyzed for its fiber content by using *Enzymatic-Gravimetric* (AOAC *Official Methods*) method [15]. The analysis was carried out in Food and Nutrition Laboratory in Unika Soegijapranata Semarang. The parameter examination was done by taking the blood sample of the rats through sinus retro-orbitalis using hematocrit pipette as many as 1 mL for all groups. The examination of lipid profile (total cholesterol, LDL, HDL, and triglycerides) was done twice, after and before doing the treatment of guava and pectin using *Enzymatic Colorimetric Test CHOD-PAP* and *GPO-PAP* method [16]. AIP (*atherogenic index of plasma*) was counted based on $\log(TG/HDL)$ [10]. The maintenance of the rats and

parameter examination were undertaken in the Study Center of Food and Nutrition UGM. The data of all groups were analyzed by using one-way ANOVA continued by Post Hoc Test in a 0.05 significance level.

3. Results and Discussion

3.1. Results

The examination of lipid profile aimed to know the risk of CHD by counting AIP. The results of examining lipid profile before and after the treatment are shown in Table 1-4

3.1.1. Lipid profiles

Lipid profiles examination was performed to find out the effect of red guava flour on total cholesterol, LDL, HDL, and triglyceride contents (Tables 1 to 4).

Table 1. Total cholesterol contents by treatment group

| Group | Mean (mg/dL) | | | T-test (<i>p-value</i>) |
|----------------------------|--------------|-------|--------------|------------------------------|
| | Initial | Final | Δ (%) | |
| Normal feed (N) | 103.9 | 113.0 | 10.0 (10%) | < 0.001 |
| High cholesterol feed (HC) | 173.3 | 187.7 | 14.4 (8%) | < 0.001 |
| Red guava treatment (G) | 179.7 | 122.0 | -57.8 (32%) | < 0.001 |
| Pectin treatment (P) | 176.4 | 120.0 | -56.4 (32%) | < 0.001 |

Table 2. LDL contents by treatment group

| Group | Mean (mg/dL) | | | T-test (<i>p-value</i>) |
|----------------------------|--------------|-------|--------------|------------------------------|
| | Initial | Final | Δ (%) | |
| Normal feed (N) | 24.3 | 31.5 | 7.2 (30%) | < 0.001 |
| High cholesterol feed (HC) | 57.5 | 62.3 | 4.8 (8) | < 0.001 |
| Red guava treatment (G) | 60.0 | 34.5 | -25.5 (43%) | < 0.001 |
| Pectin treatment (P) | 56.3 | 29.8 | -26.5 (47%) | < 0.001 |

Table 3. HDL contents by treatment group

| Group | Mean (mg/dL) | | | T-test (<i>p-value</i>) |
|----------------------------|--------------|-------|--------------|------------------------------|
| | Initial | Final | Δ (%) | |
| Normal feed (N) | 72.1 | 67.3 | -4.8 (7%) | < 0.001 |
| High cholesterol feed (HC) | 57.7 | 50.6 | -7.1 (12%) | < 0.001 |
| Red guava treatment (G) | 59.2 | 69.7 | 10.6 (18%) | < 0.001 |
| Pectin treatment (P) | 62.3 | 74.6 | 12.3 (20%) | < 0.001 |

Table 4. Triglyceride contents by treatment group

| Group | Mean (mg/dL) | | | T-test (<i>p-value</i>) |
|----------------------------|--------------|-------|--------------|------------------------------|
| | Initial | Final | Δ (%) | |
| Normal feed (N) | 72.4 | 82.0 | 13 (12%) | < 0.001 |
| High cholesterol feed (HC) | 110.4 | 127.5 | 17.1 (15%) | < 0.001 |
| Red guava treatment (G) | 108.3 | 88.6 | -19.7 (18%) | < 0.001 |
| Pectin treatment (P) | 104.9 | 85.3 | -19.6 (19%) | < 0.001 |

The results showed an increase in total cholesterol, LDL and triglycerides in the group given normal food (N) and high cholesterol (HC); while the rate of HDL cholesterol decreased. The groups with the treatment of guava (G) and pectin (P) got the decrease in total cholesterol, LDL and triglycerides; while the rate of HDL cholesterol increased.

3.1.2. AIP (atherogenic index of plasma)

AIP was calculated according to the formula, $\log(TG/HDL-C)$ on the lipid profile before and after the treatment to know the status of atherogenic plasma based on lipoprotein in blood [10].

Table 5. AIP Score by treatment group

| Group | Mean (mg/dL) | | | T-test (<i>p</i> -value) |
|----------------------------|--------------------|---------------------|--------------|------------------------------|
| | Initial * | Final * | Δ (%) | |
| Normal feed (N) | 0.130 ^a | 0.131 ^a | 0.001 (7%) | < 0.001 |
| High cholesterol feed (HC) | 0.282 ^b | 0.401 ^b | 0.12 (43%) | < 0.001 |
| Red guava treatment (G) | 0.263 ^b | 0.104 ^{ac} | -0.16 (61%) | < 0.001 |
| Pectin treatment (P) | 0.227 ^c | 0.058 ^c | -0.17 (70%) | < 0.001 |

* Different superscript notations show that there are significant differences in treatment effects

Table 5 shows the decrease in AIP significantly in group G (61%) and P (70%) from the category of the increasing risk (AIP > 0.21) to the low risk (AIP < 0.11), but it increased in group N and HC. Therefore, it can be said that red guava and pectin were proven to be able to lower AIP in hypocholesterolemic rats. The different test of AIP in the treatment groups showed no difference between the treatment groups (G and P). Therefore, red guava and pectin had a similar potential to lower AIP (Table 6).

Table 6. The different change test of AIP between treatment groups

| Group | Δ average \pm SD* |
|----------------------------|--------------------------------|
| Normal feed (N) | 0.001 \pm 0.004 ^a |
| High cholesterol feed (HC) | 0.12 \pm 0.022 ^a |
| Red guava treatment (G) | -0.16 \pm 0.017 ^b |
| Pectin treatment (P) | -0.17 \pm 0.015 ^b |

* Different superscript notations show that there were significant differences in treatment effects

3.2. Discussion

The analysis about the possibility to get CHD was based on the existence of lipoprotein which was atherogenic called as *Index Atherogenic of Plasma* (AIP). It was proven that red guava (G) and pectin (P) had decreased AIP significantly, *p* value < 0.001 (Table 5). The results of the different test in table 6 show no difference between the treatment groups (G and P). These results interpreted that red guava had the same ability as pectin in lowering AIP in hypocholesterolemic rats from the category of the increasing risk (AIP > 0.21) to the low risk (AIP < 0.11). AIP describes the balance between lipoprotein which is atherogenic (triglycerides) and which is protective (HDL). This research proved that red guava and pectin could lower atherogenic lipoprotein which was triglycerides significantly with *p* value < 0.001. It also proved that red guava and pectin increase lipoprotein HDL significantly with *p* value < 0.001 (Table 3 and 4). AIP relates to the size of pro-atherogenic and anti-atherogenic cells related to the *Fractional Esterification Rate* (FER). Frohlich and Dobiášová in their research said that FER had a strong connection to the changes of HDL, triglycerides. High concentration of triglycerides related to the existence of VLDL particles and small dense LDL. On the other side, the increase in HDL related to the protective characteristics such as inhabiting cholesterol esterification causing the decreasing FER. The decrease in HDL increased FER. FER value is an independent indicator in angiographic. Small dense LDL is more easily oxidated than LDL, which then it forms foam cells. This process is the beginning of atherosclerosis [17]. Another research of Dobiasova reported that the increase in triglycerides plasma concentration related to the increase in CHD followed by the increase in small dense LDL population. The related research of Supriyono discussing the risk factor of CHD in dr. Kariadi hospital and Tlogorejo hospital mentioned that the people with triglycerides rate similar to or more than 150

mg/dL had the risk 2.8 times bigger to get CHD than the ones with triglycerides rate less than 150 mg/dL [18].

The decrease in AIP is basically the decrease in lipoprotein rate which is pro-atherogenic (triglycerides) and LDL and the increase in anti-atherogenic (HDL). It can be concluded that the decrease in AIP is a mechanism of repairing lipid profile as a cause of administering red guava and pectin.

4. Conclusion

The administration of red guava in hypocholesterolemic rats was proven to lower AIP (atherogenic index of plasma).

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