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Antioxidant Maja Fruit (Aegle marmelos (L) Carrea) Lowering Blood Sugar Mus Musculus

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Abstract: *Maja fruit crops (Aegle Marmelos (L) Correa) is very popular with the culture of the Majapahit kingdom, the kingdom is the biggest in Indonesia in the 13th century. Maja fruit tastes bitter with light green skin. Because of the bitter taste of this fruit then nothing like it, but provide tremendous benefit in lowering blood sugar. This study found that the method of testing the antioxidant activity of maja fruit extract samples was very strong with values reaching the IC50 range of 50-100 µg/mL. Trials of mice were also carried out and proven to reduce sugar levels. Also interesting from the results of this study produce maja fruit in the form of a drink that no longer tastes bitter but changes the taste to be rather sweet, warmer when drinking it as there are natural alcohol levels that are quite safe for the body.*
Keywords: *antioxidant; aegle marmelos (L) carrea; blood sugar; mus musculus*

I. Introduction

Maja fruit (Aegle marmelos (L) Correa) is one type of plant that is considered unique among the people of Indonesia. The word Majapahit itself comes from two words, namely: Maja and Bitter which means Maja fruit that tastes bitter. Besides that, it turns out that in other parts of this Maja plant it holds a myriad of extraordinary benefits for health that are as traditional treatment materials such as fever, constipation, dysentery, diarrhea, hepatitis, tuberculosis, inflammation of the nasal mucous membranes, brain disorders, liver disorders, itching (scabies, ulcers, boils, eczema), anti-inflammatory, hearing loss, urinary disorders, and abortive (Kapoor 1990, Singh & Malik 2000 in Tasyrifah, 2010). The fruit is also useful as an anti-inflammatory and larvicidal (Angajala. et al, 2014).

In history, Maja fruit is closely related to the kingdom of Majapahit, with its characteristic white flesh and light green skin (Fatmawati, 2015). Even in India, the tree of the fruit is recognized as a sacral nutraceutical medicinal tree (Verma et al, 2014). The fruit of this Maja tree has extraordinary benefits such as an effective ingredient in cleaning ferrous metals and disinfectants. Maja rind extract effectively inhibits corrosion rate on API 5L carbon steel (Rasitiani., Et al, 2018). This fruit is also effective in removing rust and dirt on rusty metal tools (Jamili et al., 2011), besides Maja, there is pine sap extract which serves to reduce iron corrosion from seawater (Hidayat and Sumarji, 2016), due to ozonation and adsorption methods can work effectively in reducing levels of Fe and Mn (Hidayanti, 2015). In the world of Maja fruit plantations, it is useful to increase cocoa yields because it can reduce the intensity of attack borer on cocoa fruit (Sjam, 2006), as fuel in the manufacture of ethanol by distillation process produces 90% content (Batutah, 2017).

In (Chavda et al., 2012) stated, Maja fruit contains tannins by 9% contained in the flesh of the fruit and 20% in the rind. In addition to the bitter taste of Maja fruit also has a sweet, fragrant, and sharp taste in the throat.

Maja flesh contains substances such as balm oil, marmelosin (C13H12O), and 2-furocoumarins-psoralen. In the fruit, roots, and leaves of Maja are antibiotics. Therefore, the effectiveness of Maja fruit needs to be explored in depth so that it is not only used as a name. However, in the Maja fruit contains many antioxidant properties of one of them.

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Antioxidants are needed by the body to overcome and prevent oxidative stress. Various natural ingredients native to Indonesia contains antioxidants with different active ingredients. The use of natural materials native to Indonesia as an antioxidant needed to improve the quality of public health at a relatively affordable cost (Development Agency (RKD), 2007), one of which is contained in the Maja fruit.

Maja fruit extract can produce antioxidants, so Maja fruit extract can be used as a blood sugar-lowering (Diabetes Mellitus). Diabetes mellitus is a disease that is generally always characterized by increased glucose levels in the blood as a result of a deficiency of insulin secretion or a decrease in the biological effects of insulin or both, resulting in abnormalities in the body to metabolize carbohydrates, lipids, and proteins.

In the state of diabetes mellitus (DM) of the body relative insulin deficiency blood glucose levels so that arrangements be disrupted. Diabetes is caused by the disruption of the body's ability to use glucose into the cells. High levels of glucose in the blood can damage the small blood vessels in the kidneys, heart, eyes, and nervous system. Untreated diabetes can cause heart disease, stroke, kidney disease, blindness, and nerve damage in the legs.

Based on the above research fruits Aegle Marmelos Maja (L) Correa) as the lowering of blood sugar has not been studied, therefore, this study aims to determine how potential Maja fruit in a drop in blood sugar (diabetes mellitus).

II. Research Methods

2.1 Antioxidant Activity Test

A stock solution is made by measuring the extra samples of fruit Maja (*Aegle Marmelos (L) Correa*) by 30 mL of methanol is diluted with 70%, homogenized and then the volume up to 100 mL. Furthermore, dilution is carried out again by making 5 series of solution concentrations (50, 100, 150, 200, 250 ppm).

To determine the antioxidant activity of each concentration of the sample solution taken 1 mL using a micropipette put into a test tube, then add 4 mL of 50 μM DPPH solution. The mixture is homogeneous and left for 30 minutes in a dark place, absorption is measured with a UV-Vis spectrophotometer at a wavelength of 517 nm.

The antioxidant activity of the sample is determined by the amount of DPPH radical uptake resistance by calculating the percentage (%) of DPPH absorption inhibition using the formula (Molyneux, 2004):

$$\% \text{ Inhibisi} = \frac{(\text{Abs blanko} - \text{Abs sample}) \times 100\%}{\text{Abs blanko}}$$

Information:

Abs blanko: absorbance of DPPH radical

Abs sample: absorbance of the sample in a DPPH radical

IC50 values of each sample concentration were calculated using the linear regression equation formula (Andayani et al., 2008). The concentration of the sample as the x-axis and % inhibition as the y-axis. From the equation: $Y = a + bX$

To determine the value of IC50 can be calculated by formula:

$$IC50 = \frac{(50-a)}{b}$$

Information:
Y: % inhibition
a : Intercept (Y-line intersection)
b : Slope
X: Concentration

In this study using the DPPH method (1,1-diphenyl-2-picrylhydrazil), where the DPPH method is a method that can be used to determine antioxidant activity in the sample to be tested by looking at its ability to counteract DPPH free radicals. The advantage of this method is that the method is simple, easy, fast, sensitive, and requires small samples. Easy to apply because the DPPH radical compound used is relatively stable compared to other methods (Rahmawati *et al*, 2016).

Preparation of DPPH solution (1,1-diphenyl-2- *picrylhydrazil*). DPPH solution was weighed as much as 1.97 mg DPPH and dissolved with methanol in a flask to 100 mL to obtain a solution with a concentration of 50 µM (Molyneux, 2004).

2.2 Trials to Mice *(Mus musculus)*

The animal test used in this study was in mice (*Mus musculus*) able-bodied males, aged 3 months and weighing between 20-30 grams. 16 mice were used and divided into 4 treatment groups (Pitriya, 2017).

Mice (*Mus musculus*) is conditioned to have above-normal blood sugar levels by providing a dose of 0.1 mL glucose solution orally. The treatment of each group has blood drawn through the end of male mice (*Mus musculus*) to get out of his blood. Then blood dripped on the glucometer strips. Within 10 seconds the blood glucose levels in mice (*Mus musculus*) will be measured automatically and the results can be read on the monitor glucometers Figure 1.

Figure 1. *Glukometer*

To reduce sugar levels in mice (*Mus musculus*) which have been conditioned sugar levels above the norm, given treatment. P0: feed, P1: feed + glucose 0.1 mL + maja fruit extract (*Aegle Marmelos (L) Correa*) 0.1 mL, P2: feed + glucose 0.1 mL + fruit extract Maja (Aegle Marmelos (L) Correa) 0.2 mL, P3: feed + glucose 0.1 mL + fruit extract Maja (*Aegle Marmelos (L) Correa*) 0.3 mL.

Next, All the test animals were incubated for 3 hours. Blood glucose levels were measured again after 3 hours of incubation. The second repetition performed 3 hours after the first treatment is completed, the third iteration is done after 3 hours of treatment the second and fourth repetitions performed 3 hours after the third treatment.

III. Result and Discussion

3.1 Antioxidant Activity Test Results

Antioxidant Activity Test on fruit samples Maja (*Aegle Marmelos* (L) *Correa*) is carried out with 5 concentrations of 50, 100, 150, 200, and 250 ppm, and made four repetitions. The first measurement of absorbance of DPPH forms in advance to calculate% inhibition and form DPPH absorbance values obtained at a wavelength of 517 nm is equal to 0.681.

Maja fruit and absorbance values (*Aegle Marmelos* (L) Correa) obtained are used to calculate% inhibition using the formula above. Then do regression between% inhibition by the concentration of the sample, the results obtained by the standard curve.

Table 1. Maja fruit antioxidant activity test results *(Aegle marmelos (L) Correa)*

Sample Concentration (ppm)	Average of Abs.	% inhibition	IC_{50} (inhibition Concentration of 50%)	Antioxidant Activity
50	0,510	24,898	0,107 μg/mL	Very strong (IC50 ≤ 50 μg/mL)
100	0,301	55,742		
150	0,219	67,800		
200	0,149	77,861		
250	0,110	83,724		

A value obtained was 20.074 (intercept) and b value of 279.54 (slope) and the R2 value 0.8991 which is based on the literature, the R2 value close to 1 indicates the data obtained is excellent (Hastono & Sabri, 2011). Furthermore, IC 50 values are calculated using the formula above and obtained a value of 0.107 mg / mL, According to Molyneux (2004), that the smaller the IC50 value indicates the higher antioxidant activity.

The test results Maja fruit antioxidant activity (*Aegle Marmelos (L) Correa*) is very strong, which is said to be very strong because of IC50 ≤ 50 mg / mL. As stated in the study Phongpainchit et al., (2007) that the antioxidant activity is very strong if IC50 ≤ 50 mg / mL, a strong if IC50 values of 50- 100 mg / mL, while if the IC50 value of 100-150 mg /mL, low if the IC50 value of 151-200 pg / mL, whereas if the IC50 value of ≥ 200 mg / mL, the antioxidant activity are extremely low.

3.2 The results of trials to Mice *(Mus musculus)*

In Figure 2 can be seen after the mice (*Mus musculus*) received induction treatment with the oral diabetes given glucose solution at a dose of 0.1 mL managed to increase blood glucose levels, be above normal blood sugar levels (70-110 mg / dL), ie P0 131 mL, P1 149 mL, P2 143 mL, and P3 141 mL.

After mice (*Mus musculus*) receive treatment to reduce the sugar content, the results can be seen in Figure 2, namely P0 to 119 mL to 102 mL P1, P2 and P3 to 104 mL to 95 mL. From the results of this trial, the levels of mice sugar (Mus musculus) have decreased.

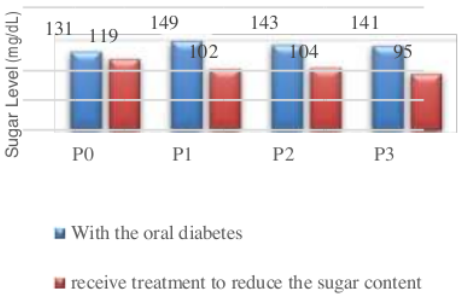

Figure 2. *Shows Sugar Levels from the early administration of Glucose and Extra Maja*

IV. Conclusion

1. The results of antioxidant activity tests on Maja fruit extracts (*Aegle marmelos (L) Correa*) of 0.107 μg / mL were stated to be very strong. Where it is stated very strongly because of IC50 ≤ 50 mg / mL.
2. While the results of trials on mice (*Mus musculus*), Maja fruit extract (*Aegle marmelos (L) Correa*) can reduce sugar levels.

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