



## THE HYPOGLICEMIC EFFECT OF AVOCADO SEED (*PERSEA AMERICANA* MILL) AND HISTOPATHOLOGIC PROFILE

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### ABSTRACT

Avocado (*Persea Americana* Mill) are plants that can be used in the treatment of diabetic mellitus empirically. In everyday life, avocado seed is often not utilized. The aim of this study is to investigate the antidiabetic activity of 96% ethanolic extract of avocado seed (*Persea Americana* Mill) on wistar rats induced by alloxan. Twenty five rats were divided into 8 treatment groups. Negative control group was treated by distilled water, the positive control group was treated by glibenclamide at dose of 0.45 mg / bw, group I, II, and III were treated by 96% ethanol extract of avocado seeds at doses of 300; 600 and 1200 mg / bw respectively. Before they were treated, all rats were treated by alloxan 150 mg / bw intraperitoneal. Day 3 after induction, the rats were treated by extract for 7 days orally. Measurement of glucose was done at day 0, 3 and 10. Blood glucose levels on day 7 were analyzed by Kruskal-Wallis test followed by Mann-whitney with 95% confidence interval. The results show the 96% ethanol extract of avocado seeds at doses of 300; 600 and 1200 mg / bw can reduce blood glucose levels on rats induced by alloxan.

**KEY WORDS:** Avocado (*Persea Americana* Mill), Extract, Diabetic Mellitus, Alloxan,



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## INTRODUCTION

In 2011 was estimated the number of people with diabetes mellitus (DM) is 366 million in the world and in 2030 was estimated to become 552 million<sup>1</sup>. In 2010 Indonesian population were about 237.6 million and it is the country with the 4<sup>th</sup> largest population in the world<sup>2</sup>. Approximately 7.6 million Indonesian people were suffering from diabetes. This leads Indonesia have the 7<sup>th</sup> of the country with largest DM population in the world<sup>1</sup>. Diabetes Mellitus is a disease in which pancreas lack of insulin production or ineffective use of insulin<sup>3</sup>. The lack of insulin cause decrease inglucoseup take into cells and an increase in glucose from the liver to circulation<sup>4</sup>. Today the traditional medicine is often used as self-medication. Before the modern medicine was invented, the use of traditional medicine in Indonesia has been going on since thousands years ago. Traditional medicine is widely used to treat chronic diseases such as hypertension and diabetes mellitus<sup>5</sup>. Empirically, Avocado (*Persea Americana* Mill) is widely used to treat diabetes. Avocado contains saponins, flavonoids, tannins, phenol, alkaloids, steroid and minerals such as phosphorus, iron, potassium, magnesium, and zinc<sup>6</sup>. The ethanolic extract of avocado seed contain polyphenols, tannins, flavonoids, triterpenoids, quinones, monoterpenoid, and sesquiterpenoid<sup>7</sup>. Flavonoid is found in many plants. Flavonoid plays an important role in the prevention of diabetes and it's complication<sup>8</sup>. The plants containing flavonoids have shown a beneficial effect on diabetes mellitus. The mechanism of this are: 1). Flavonoid reduce the absorption of

glucose and 2). It can increase glucose tolerance<sup>9</sup>.

## MATERIALS AND METHODS

A total of 25 rats were divided into 5 groups. Each group consisted of 5 rats. All rats were fasted for 8 hours, but they were still drinking. On day 0, all rats were measured glucose levels. Then each group get the treatment. Normal control group was treated by distilled water; The Positive control group was treated by 0.63 mg / kgbw glibenclamide; Group III, IV and V were treated by ethanolic extract of avocado seed at doses of 300 mg / kg rat; 600 mg / kg and 1200 mg / kg bw respectively on days 4 during 7 days. On day 3 after they were induced by alloxan monohydrate intraperitoneally, their blood glucose was measured again. The use of alloxan refers to research conducted by Sharma *et al.*, 2013<sup>10</sup>. Rats with glucose levels more than 200 mg / dL were given an appropriate treatment groups for 7 days. On the day 10, the blood glucose levels of all rats were measured. All blood sampling were done on tail lateral vein. Blood glucose levels on day 10 were analyzed with Kruskal Wallis test followed by Mann Whitney test with 95% confidence level. Two rats of each group were killed and their pancreases were taken for histopathological examination

## RESULTS

The results of measurements of blood glucose levels in the five treatment groups are shown in this table.

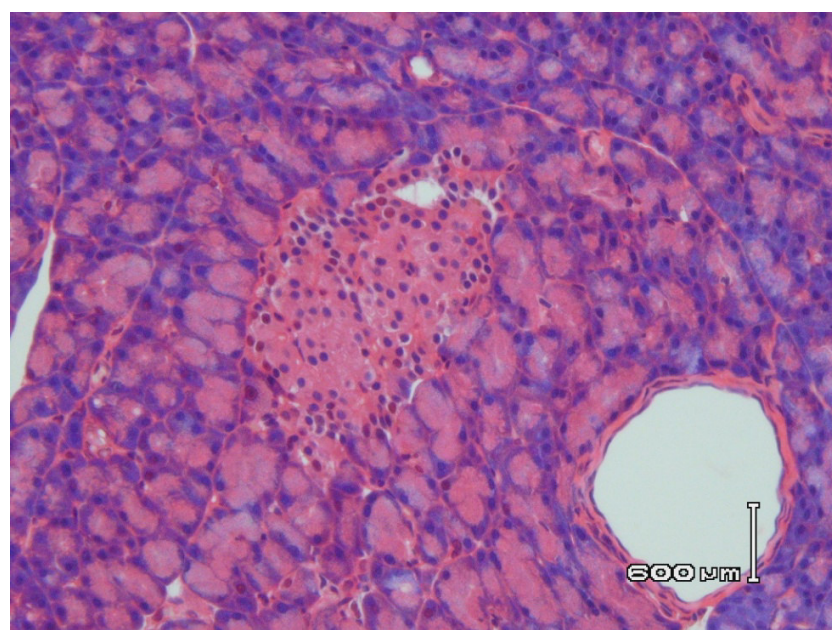
**Table 1**  
**The mean of blood glucose level of rats**

Groups	Glucose on day 0 (mg/dL)	Glucose on day 3 (mg/dL)	Glucose on day 10 (mg/dL)
Negative control (aquadest)	118±16,85	217,6±39,22	246,8±64,25
Positive control (Glibenklamid 0.63 mg/kg orally)	109,6±13,63	252,8±25,54	88,6±11,17**
The 96% ethanolic extract of seed avocado dose 3000 mg/200 g bw	120,4±9,86	249,2±14,93	145,4±6,84**
The 96% ethanolic extract of seed avocado dose 600 mg/200 g bw	153,8±26,1	235±23,94	143,2±64,93*
The 96% ethanolic extract of seed avocado dose 120 mg/200 g bw	84,2±15,64	212,6±19,56	134,8±27,2**

\*\* $P < 0.01$  and \* $P < 0.05$  statically significant from negative group by Mann Whitney test

Table shows that on the negative control group, the blood glucose levels increased from day 3 to 10 post alloxan induction. This is because aquadest have no effect of lowering the blood glucose levels. The positive control (glibenclamide) shows the decreasing blood glucose level significantly. Glibenclamide can decrease blood glucose levels by increasing insulin release from pancreatic beta cells through inhibition / blocking ATP-dependent K + channel<sup>11</sup>. Group III, IV and V showed a decrease in blood glucose levels. This

suggests that glibenclamide and the 70% ethanolic extract of avocado seed have antidiabetic effects. The p value of Kruskal-Wallis test of blood glucose level on day 10 is 0.003 ( $p < 0.05$ ). This means there is a difference in blood glucose levels of the five treatment groups. The result of Mann Whitney test show that the 96% ethanolic extract of seed avocado dose 3000; 600 and 120 mg/200 g bw can reduced blood glucose level significantly. The result on histological examination can be seen in figure 1.



**Figure 1**

***Histological examination of rat pancreas on day 10 of The 96% ethanolic extract of avocado seed at dose of 3000mg/200 gbw. It can be seen that there are vacuolization in the insula islet cells and aciner cell and inflammatory.***

## DISCUSSION

The results of this study show that the 96% ethanolic extract of avocado seed is capable of lowering blood glucose levels in male Wistar rats induced by alloxan significantly ( $P < 0.05$ ). This study is in line with the previous studies. Several previous studies related to this study include: The ethanolic extract of avocado leaves (*Persea Americana* Mill) (Lauraceae) has a hypoglycemic effect in rats induced by streptozotocin<sup>12</sup>. The aqueous extract of avocado seed (*Persea Americana* Mill) at doses of 300 mg and 600 mg / kg bw was able to reduce blood glucose levels in normal mice and rats induced by alloxan. The decline of blood glucose levels in rats induced by alloxan are 73.26-78.24%, meanwhile in normal mice are 34.68-38.9%<sup>13</sup>. The aqueous extract of avocado seed at dose of 20; 30 and 40 g / L was able to lower blood glucose levels in albino rats induced by alloxan and has a protective effect on the kidneys, pancreas and hepar<sup>14</sup>. The chloroform, methanol and N-hexane of avocado seed can lower blood glucose levels in mild hyperglycemia diabetic rats induced by alloxan<sup>15</sup>. The aqueous extract of avocado seed has a hypoglycemic effect in rats induced by alloxan<sup>16</sup>, and in rabbits induced by alloxan<sup>17</sup>. The ethanolic extract of fruit of *Persea Americana* Mill at dose of 300mg/kgbw/day orally for 30 days can reduce blood glucose, glycosylated hemoglobin, blood urea and serum creatinine hyperglycemic rats induced by streptozotocin<sup>18</sup>. Some presumed the mechanism of hypoglycemic effects of a medicinal plant, among others: The plant is able to inhibit the activity of insulinase or the plant increases pancreatic beta cells by

activating cellular regeneration<sup>19,20</sup>, some plants contain compounds insulin like substance<sup>20</sup>, and some plants have antioxidant effects<sup>22,23</sup>. This study does not determine the mechanism of the lowering blood glucose. Based on histological examination show the 96% ethanolic extract of avocado seed is able to lower blood glucose levels but it cannot repair pancreatic beta cells damaged by alloxan. Some suspected the mechanism of hypoglycemic effect of these extracts, among others: the content of flavonoids and saponins in avocado seed. Flavonoids have antioxidant effects that protect pancreas from oxidative stress by alloxan<sup>24</sup>. Saponin suspected inhibit glycolysis and glycogenesis liver<sup>25</sup>. Tiwari & Rao supposed that flavonoids, tannins, and saponins have a hypoglycemic effect by inhibiting the action of sodium-glucose transporter 1 (S-GLUT1)<sup>26</sup>.

## CONCLUSION

The oral treatment of 96% ethanolic extract of avocado seed (*Persea Americana* Mill) at doses of 300; 600 and 120 mg/kgbw respectively for 7 days can lower blood glucose level on wistar strain rat induced by alloxan. The 96% ethanolic extract of the avocado seed at a dose of 300 mg / kgbw cannot repair the insulin Langerhans cell vacuolization on wistar strain rats induced by alloxan.

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