The Influence of African Leaf (Vernonia Amygdalina Delile) Extract On Blood Glucose Levels of Diabetic Rats

Veince B. Silahooy 1*, Meillisa Carlen Mainassy 1, Imanuel B D. Kapelle 2, Laury M C. Huwae 1

1 Department of Biology, Faculty of Mathematics and Natural Sciences, Pattimura University, Ambon.
2 Department of Chemistry, Faculty of Mathematics and Natural Sciences, Pattimura University, Ambon.

Abstract- Diabetes mellitus (DM) is a disease associated with lifestyle, especially food consumption. One way to reduce and treat the pathogenesis of diabetes is by providing antioxidants that are proton compounds to reduce the negative effects of ROS. African leaves contain flavonoid compounds, glycosides, saponins, tannins, and triterpenoids / steroids that function as primary antioxidants and scavenger to superoxide anions. The purpose of this research is to know the effect of African Leaves (Vernonia amygdalina Delile) on blood glucose levels of diabetic rats. The results showed that African leaves have an effect on blood glucose levels of diabetic rats. The concentration of African leaf extract which gives effect to the decrease of glucose level is 60%, at 127 mg/dl glucose level.

I. INTRODUCTION

In Indonesia, diabetes mellitus is one of the highest non-communicable diseases that cause death. Several factors associated with increased risk for DM type 2 are increased age, obesity, overeating, type of diet (increased consumption of animal fat), family history with diabetes, hypertension, hyperlipidemia and heart disease [1].

Diabetes mellitus is divided into type 1 and type 2. Type 1 diabetes is caused by cellular damage mediated by an autoimmune that damages pancreatic β cells so that cells are unable to produce insulin. This type of diabetes usually occurs starting in childhood but can also occur at any age. Type 2 diabetes mellitus is usually characterized by a decrease in insulin sensitivity (insulin resistance) or a result of a decrease in insulin production and occurs at age above 45 years and can also occur in obese people [2].

Diabetes mellitus is characterized by insufficient insulin synthesis or insulin action damage that will lead to hyperglycemia. Hyperglycemia increases ROS (Reactive Oxygen Species) consisting of free radicals and radical compounds, as well as lowering antioxidants. This condition causes oxidative stress. Free radicals are compounds that in their outermost orbits have one or more unpaired electrons, are highly volatile and highly reactive which can cause damage to cellular components such as DNA, lipids, proteins and carbohydrates. If these free radicals attack the DNA of pancreatic cells then cause DNA damage, dysfunction or decrease in the number of insulin-producing β cells in the pancreas [3]. Reference [4] explains that conditions of oxidative stress can cause insulin resistance and complications are dripped.

Some of the mechanisms that contribute to the pathogenesis of diabetes mellitus due to hyperglycemia are glycation (binding of proteins with sugar or fat), lipid peroxidation, oxidation and DNA methylation resulting in DNA adducts and glucose autoxidation. The condition of diabetes also increases inflammatory compounds such as IL-6 (Interleukin 6) and TNF-α (Tumor Necrosis Factor α). ROS can induce NF-xB (Nuclear Factor kappa β) to express the cytokine compound gene. [2]. ROS can be neutralized by the antioxidants present in the body so that biological
damage by free radicals and radical compounds that can trigger the occurrence of degenerative diseases can be avoided [5]. The mechanism of action of cellular antioxidants according to reference [6], antioxidants can interact directly with oxidants, free radicals, or oxygen single; prevent the formation of reactive oxygen species; changing the type of reactive oxygen to less toxic; prevent the ability of reactive oxygen; and repair damages that arise. Meanwhile, according to reference [5] there are four mechanisms of antioxidant action: 1) bind the reactive oxygen species (ROS) and free radical nitrogen, 2) metabolize lipid peroxide into non-radical products, 3) metal ions, and 4) reduce the oxidation potential of a molecule. Consumption of antioxidants can prevent oxidative stress and cell damage that can cause various diseases such as hypertension, diabetes mellitus, heart, cholesterol and neurodegenerative diseases.

Plants that have the potential to be developed as a treatment for diabetes are African Leaves (*Vernonia amygdalina* Delile). African leaves (family Asteraceae) grow mostly in the west African continent especially in Nigeria. The content in African leaves is saponin (vernonioside and steroidal saponin), sesquiterpenes (vernolene, vernoladol, vernolepin, vernodalin and vernomygin), flavonoids, coumarin, phenolic acids, lignans, xanthones, terpenes, peptides and luteolin. Generally, flavonoid compounds serve as primary antioxidants and scavenger to superoxide anions [7]. Flavonoids act as hydrogen donors, have the ability to stabilize and delocalize unpaired electrons in free radicals, and are capable of chelating metal ions (Fenton's termination reaction) [8]. The purpose of this study was to determine the effect of African Leaf extract (*Vernonia amygdalina* Delile) on blood glucose levels of diabetic rats.

### II. RESEARCH METHODS

A. Materials

Materials used in the research are: Leaves Africa (*Vernonia amygdalina* Delile), and Alloxan (*Sigma A7413-10G*) to make normal rats become diabetic. The equipment used in the research is weight scales for mice, glucometer (One Touch Ultra) for measurement of blood glucose level, syringes for injection of alloxan, sonde for drinking, and cage.

B. Test Material Preparation

1000 gr of dried African leaf boiled with 3 liters aquades for 10 minutes and filtered. The boiling result was concentrated with vacuum evaporator up to 500 ml. African leaf extract is packed in polyethylene plastic and stored at 4°C.

C. Rations

Rats were given standard laboratory feeds: protein (10% casein), fat (8% corn oil), mineral mix (5%), vitamin mix (1%), fiber (cellulose 1%), and carbohydrate (tapioca starch) up to 100%. Water and feed were given ad libitum during the study period (AOAC 1990).

D. Alloxan induction for diabetic rats

After passing the adaptation period, 30 rats were made into diabetes by induction using alloxan monohydrate. Induction was performed by intraperitone injection; alloxan dose used was 2.5 ml.

E. Testing the influence of African Leaf

Blood glucose measurement by glucometer using electrochemical method, that is based on potential measurement (electric power) caused by reaction from glucose with glucose reagent material on strip electrode. The test strip contained chemicals: glucose oxidase 29.1% w/w, 32.0% w/w hexacyanoferate (III) and 38.9% w/w non-reactive ingredients.

The principle works: the blood sample is absorbed into the end of the test strip based on the capillary reaction. When the blood fills the reaction chamber on the test strip, the potassium fericyanide is decomposed and the sample glucose is oxidized by the glucose oxidase enzyme, causing a decrease in the oxidation number (potassium hexanoanoferate (III) to potassium hexanoanoferate (II)). The application of constant voltage quantities of the meter oxidizes potassium hexanoanoferate (II) back in potassium hexanoanoferate (III), and gives electrons. The electron is generated to produce a current proportional to the glucose level in the sample. After 60 seconds, the glucose concentration in the sample is displayed on the monitor screen. How to measure blood glucose in experimental mice: the tail of the test rat was warmed with warm water, then pierced with a needle and dripping blood worn on the glucometer strip. Blood glucose levels are expressed in mg/dL.

### III. RESULT AND DISCUSSION

1000 grams of African leaves are smoothed and strained, then added 3 liters of aquades to obtain 500 ml African leaf extract (Fig. 1). The extract was then diluted into several different concentrations (12%, 24%, 36%, 48% and 60%).
A. The Effect of African Leaf

Preparation of mice to diabetes was done by induction using alloxan monohydrate, induction was done by intraperitone injection. African leaf extract was then fed with sonde and measured glucose levels during processing time.

The results showed elevated glucose levels before treatment and post-SZT addition treatment (Fig 1). This is because the effect of adding SZT has not been working within 60 minutes. Based on the results can be made histogram: the influence of African leaves on glucose levels (Fig 2). Effect of African leaf extract concentration during 60 minute process time showed 60% extract concentration gave the best result. When compared with other concentrations, at a concentration of 60% can maintain glucose levels at normal levels, ie 122 mg/dl before treatment to 127 mg/dl after treatment (Table 1).

![Image of African Leaf and extract](image_url)

**Figure 1. African leaf material, extracting process, evaporation and extract**

<table>
<thead>
<tr>
<th>Sample code</th>
<th>Extract concentration (%)</th>
<th>SZT Volume (ml)</th>
<th>Glucose levels before treatment (mg/dl)</th>
<th>Glucose levels after treatment within 1 hour (mg/dl)</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>12</td>
<td>2.5</td>
<td>119</td>
<td>168</td>
</tr>
<tr>
<td>B</td>
<td>24</td>
<td>2.5</td>
<td>117</td>
<td>154</td>
</tr>
<tr>
<td>C</td>
<td>36</td>
<td>2.5</td>
<td>120</td>
<td>149</td>
</tr>
<tr>
<td>D</td>
<td>48</td>
<td>2.5</td>
<td>121</td>
<td>139</td>
</tr>
<tr>
<td>E</td>
<td>60</td>
<td>2.5</td>
<td>122</td>
<td>127</td>
</tr>
<tr>
<td>Control</td>
<td>0</td>
<td>0</td>
<td>121</td>
<td></td>
</tr>
</tbody>
</table>
IV. CONCLUSION

African leaves have an effect on blood glucose levels of diabetic rats, and the concentration of African leaf extract which gives effect to the decrease of glucose level is 60% extract with glucose content 127 mg/dl.

ACKNOWLEDGEMENT

For the Rector of Pattimura University, Research and Service Institute of Pattimura University, Ministry of Research, Technology and Higher Education who has financed the research In accordance with the Rector's Decree No: 605/UN13/SK/2017.

REFERENCES


Figure 2. Effect of African leaf extract on glucose level