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Inhobitory Potential of *Poligala paniculata* L. Against α-glucosidase

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Abstract : There are many Indonesian plants used as herbal medicine. For example, herbal medicine to treat the diabetes mellitus, such as *Poligala paniculata* L. In this point, we tested the ethanolic extracts of *Poligala paniculata* L. with respect to a glucosidase inhibition activity. *Polygala paniculata* L.used for herbal medicine as antihypertension and anticholesterol, and potential for hyperglycemia.The sample has extracted with maseration method using ethanol solvent. Antidiabetic assay with inhibitory of enzyme α-glucosidase. Our results show the extract of *P. paniculata*have a potential as antidiabetic with IC_{50}0.518 µg/mL. It is higher than the glucobay 0,017 µg/mL. These results demonstrate the ability of *P. paniculata* extract is still lower than the positive control, but this results contribute to understand and the mechanism of action of these plants on glucose metabolism.
Key word : Antidiabetic, enzyme α-glucosidase, *Poligala paniculata* L.

Introduction

Diabetes is the world's oldest diseases, diabetes is associated with the metabolism of glucose in the blood. Medically, the notion of diabetes mellitus aspect extends to a series of symptoms that arise in a person caused by an increase in blood sugar levels (hyperglycemia) due to lack of insulin[1]. These disorders can occur due to damage pancreatic beta cells and is unable to supply insulin as needed or some other thing that is not yet known[2]. Insulin is a hormone that is released from pancreatic beta cells and responds to various stimuli, especially glucose[3]. Insulin breaks down sugar into a monomer-monomer so easily fit into the muscle into muscle sugar. Impaired insulin production or function may affect sugar metabolism resulting in increased concentrations of glucose in the bloodstream.

The prevalence of diabetes mellitus according to the WHO (World Health Organization), Indonesia ranks the 4th largest in the world[1], and based on research results Wild *et al.* (2004) reported that in 2000, Indonesia is a fourthranks after India, China and the United States with 8.4 million the number of people, and is predicted to increase until 21.3 million in 2030. It underlies to provide special handling or even seek alternative treatment for diabetes mellitus.

A-glucosidase enzyme is a key enzyme in the digestion of carbohydrates in the small intestine. A-glucosidase inhibition may inhibit the digestion of carbohydrates by reducing post-prandial glucose. The reaction of α-glucosidase (an enzyme) with carbohydrates (substrate) will be broken down into disaccharides and oligodisakarida, this process occurs at α-glukopiranosida hydrolysis, thus producing α-D-glukopiranosida of non reducing sugars[4].

The development of diabetes mellitus treatment has been done, one of them is the use of natural materials and the development of traditional medicine that is more minimal side effects as antidiabetic drugs through the mechanism of inhibition of the enzyme α-glucosidase. One of the local plants (Indonesian plant) that can be developed as an antidiabetic drug candidate is *Polygala paniculata* L.

Polygala paniculata L. is one of the rare and endemic plants in South Sulawesi precisely in Enrekang. Enrekang community use *Polygala paniculata* L.) as an herb, that is also believefpotential of this plant is very large for the treatment of hyperglycemia (diabetes mellitus).

Based on the empirical use of the potential of herbal extracts as antidiabetic poligala grass, so we need more research on how the activity antidibetes poligala grass herb extract (*Polygala paniculata* L.) through inhibition of the enzyme α-glucosidase.

Material and Methods

Plant : *Poligala paniculata* L. has determinated in divisi of Botanical, Laboratory of Pharmacognosy-Phytochemistry, Faculty of Pharmacy, Universitas Muslim Indonesia.

Chemical : α-glucosidase, acarbose, sodium carbonate, phosphate buffer, PNPG.

α-glukosidase inhibitory assay

The α-glukosidase inhibitor assay has been used refers to Saijyo *et al.* (2008)[5] with some modified. In a 96- well plate reader, a reaction mixture containing 30 µL of extract solution with varying consentrations (0,05; 0,1; 0,5; 1 dan 5 µg/mL) and 36 µL of phosphate buffer, 17 µl of PNPG as a substrate and pre-incubated for 5 min at 37°C, and then 17 µL of α-glukosidase was added. After further incubation at 37°C for 15 min, the reaction was stopped by adding 100 µl of sodium carbonate (200 mM). All the enzyme, inhibitor and substrate solutions were made using the same buffer. Acarbose was used as a positive control and water as negative control. The yellow colour produced (due to pnitrophenol formation) was quantitated by colorimetric analysis and reading the absorbance at 405 nm.

The % inhibition has been obtained using the formula:
% Inhibition = 1-B/A

Explanation :

B = Control – Blank Control
A = Sample – Sample Control

IC_{50} value is defined as the concentration of extract inhibiting 50% of alpha-glucosidase activity under the stated assay conditions. In case of significant inhibition, IC_{50} values were determined by linear regression by fitting to a sigmoidal dose-response equation with variable slope. All values are represented as Mean ± Standard Deviation. The IC_{50} was calculated using the equation :

IC_{50}= y – a / b

Result and Discussion

Diabetes mellitus is a group of metabolic diseases due to insulin both in insulin secretion, insulin action or both (American Diabetes Association, 2009)[6].

Alpha-glucosidase is an enzyme that speeds up the metabolism polysaccharides or oligosaccharides into monosaccharides and raise blood glucose levels. This enzyme catalyzes the hydrolysis of terminal non-reducing glucose residues that bind α-1,4 on a variety of substrates and produced α-D-glucose. α-glucosidase hydrolyze α-glycosidic bond in oligosaccharides and α-D-glycosides[7]. This was the key to the final process in the breakdown of carbohydrates inhibition of enzyme activity can lower postprandial blood glucose levels[8].

Compound existence at one particular plant extract ingredient that has ability to inhibitory α-glucosidase enzyme, give a chance to develop a natural medicine in the effort therapy of diabetic[9].

The potency of extracts to be traditional medicine depends on their chemical compounds (secondary metabolite). Phenolic and flavonoid occur widespread in some plants. Some studies have been reported the biological activities of those compounds. Thus, the determination of phenolic and flavonoid total is need to conduct[12,13].

Test of inhibition of α-glucosidase enzyme involved four test substances are samples, control samples, blank and blank control. Test material samples were made with increasing concentrations of the concentrations of 0.05; 0.1; 0.5; 1 and 5 mg / mL. The samples were then measured its absorbance resulting absorbance variation different and produce regression equation as the equation used to determine the IC_{50} of the sample.

Table 1. Absorbance of *P. paniculata*

Concentration	Absorbance	
	control sampel	**Sampel**
Kontrol	1.030	1.065
0.05	1.286	1.344
0.1	1.292	1.355
0.5	1.272	1.343
1	1.267	1.350
5	1.253	1.337

Table 2. Absorbance of Glukobay

Concentration	Absorbance	
	Control sampel	**Sampel**
Control	0.651	0.705
0.1	0.427	0.480
0.5	0.209	0.263
1	0.113	0.166
5	0.026	0.079
10	0.011	0. 065

The activity of a compound can be seen from the change in percent inhibition increased along with the increase in concentration. This happens because of the ability of extracts in competing with enzyme to the substrate so that the action of the enzyme can be inhibited. The ability of inhibition of α-glucosidase enzyme activity is expressed in IC_{50}.

The test results obtained extract of *P. paniculata* IC_{50} value of 0.518 mg / mL. This value is higher than glucobay that is 0,017 pg / mL. These results demonstrate the ability of *P. paniculata* extract is still lower than the positive control.

Table 3. IC_{50} Value*P. paniculata*

Concentration (µg/mL)	% Inhibition (%)	Equation	IC_{50} (µg/mL)
0.05	39.65	y=a+bx y= 40.959+17.445x R= 0.9	0.518
0.1	44.44		
0.5	50.70		
1	57.83		
5	58.33		

Table 4. IC_{50}Value of Control Glucobay

Concentration (µg/mL)	% Inhibition (%)	Equation	IC_{50} (µg/mL)
0.1	34.409	y=a+bx y=34.73+13.70x R= 0.9	0.017
0.5	67.896		
1	82.642		
5	96.006		
10	98.310		

The absorbance lower which is an indication that the lower glucose production and inhibitory effect on the enzyme α-glucosidase higher.

Alpha-glucosidase is an enzyme that speeds up the metabolism polysaccharides or oligosaccharides into monosaccharides and raise blood glucose levels. This enzyme catalyzes the hydrolysis of terminal non-reducing glucose residues that bind α-1,4 on a variety of substrates and produced α-D-glucose. α-glucosidase hydrolyze α-glycosidic bond in oligosaccharides and α-D-glycosides[7]. This was the key to the final process in the breakdown of carbohydrates, inhibition of enzyme activity can lower postprandial blood glucose levels[8,10,11].

Conclusions

Poligala paniculata L. have a potential as antidiabetic with inhibitor of α-glucosidaseIC_{50}0.518 µg/mL

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