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The Antidiabetic Activity of *Cocor Bebek* Leaves' (*Kalanchoe pinnata* Lam.Pers.) Ethanolic Extract from Various Areas

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ABSTRACT

The anti-diabetic activity of *Cocor Bebek* leaves' (*Kalanchoe pinnata* Lam.Pers.) ethanolic extract from Bogor city, *Kabupaten* Bogor, and Tangerang Selatan (south of Tangerang) city has been studied. The study was conducted *in vitro* using α-glucosidase inhibitor method. The results of the study showed that the IC_{50} of the extract from Bogor city, *Kabupaten* Bogor, and Tangerang Selatan city is 40.94 ppm, 33.58 ppm and 16.12 ppm, respectively. Meanwhile, the IC_{50} of quercetin which has antidiabetic activity is 10.22 ppm. The results showed that *Cocor Bebek* leaves' (*Kalanchoe pinnata* Lam.Pers.) ethanolic extract had anti-diabetic activity with IC_{50} less than 100 ppm. However, the activity is lesser than quercetin.

Keywords: *Kalanchoe pinnata Lam.Pers., antidiabetic activity, a glucosidase inhibitor*

BACKGROUND

Cocor Bebek (*Kalanchoe pinnata* Lam.Pers.), belongs to *sukulen* plants which has thick and watery leaves [1, 2]. The plant lives in tropical area. In Vietnam it is called *Thuoc bong*, *cay song don*, *truong sinh*, *diep sinh can*, *da bat tur*, or *tau pua sung*; while in the Philippines it is called *Katakataka*, *siempre viva*, *abisrana*, *aritana*, or *karitana* [3, 4]. In Indonesia the plant is more popular as an ornamental plant than medicinal one [2]. *K. pinnata* contains alkaloid, triterpene, glycoside, flavonoid, steroid and lipid. In Guatemala and India, the plant is used to treat diabetes [5].

Diabetes mellitus is a chronic metabolic disorder involving the metabolism of carbohydrates, fats and proteins, in which the disorder is characterized by the increase in blood sugar (hyperglycemia). There are two types of diabetes mellitus, namely, insulin-dependent diabetes mellitus/IDDM (type 1) and non-insulin dependent diabetes mellitus/NIDDM (type 2). Treatment for the type 2 is by regulating blood sugar levels by means of using hypoglycemic, α-glucosidase inhibitor and a diet that limits the use of sugar [6, 7].

Glucosidase is an enzyme that is needed in the process of carbohydrate metabolism. It is located on the banks of the surface of intestinal cells and is a key enzyme in the metabolism. Moreover, it is needed in the process of glycoprotein and glycolipid formation [8]. The enzyme breaks down carbohydrates into glucose in the human intestine [9]. Compounds that can inhibit the activity indicate that the compound has a possibility as anti-diabetes by lowering blood sugar levels [10].

Because of the use of *K. pinnata* as anti-diabetic treatment, an evidence obtained from in vitro testing using α-glucosidase inhibitor methods is needed. The method uses spectrophotometry by reacting substrate p-nitrophenyl α-D-glucopiranoside, the enzyme α-glucosidase and the ethanol leaves extract [8]. Considering that the pharmacology activity of natural products are influenced by environmental factor where they live, *Cocor Bebek* leaves were taken from different areas in order to compare its activity.

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MATERIALS AND METHODS

Ethanol extract of K.pinnata Preparation

Fresh *K. pinnata* leaves were obtained from Bogor city (BALITRO), *Kabupaten* Bogor (Pabuaran), and Tangerang Selatan city (PUPIPTEK area–Serpong). The species determination was done in Research Center for Biology—LIPI, Cibinong. The leaves were dried by using oven at 50° C, then blended and

macerated by using ethanol 70% for 3 x 24 hours. After that, They were filtered and evaporated by using a rotary evaporator, then dried by using water bath at 50° C until they were viscous. 1%, 0,5 % and 0,25 % (b/v) solution of *K. pinnata* extract were, then, prepared.

α-Glucosidase Inhibitory Activity of K. pinnata's Extract

α-glucosidase activity was determined by using Lee & Lee's (2001) method, that is, Spectrophotometry method at 400 nm wave length. α-glucosidase, bovine serum albumin (Wako Pure Chemical Industries, Ltd.), p-nitrophenyl α-D-glucopiranoside (Wako Pure Chemical Industries Ltd.), Bufer fosfat pH 7, DMSO, Na_2CO_3, and Quercetin were used as the standard. α-glucosidase enzyme was diluted 1/10 times before assay. 200 mg bovine serum albumin and 1.0 mg α-glucosidase were diluted in buffer phosphat pH (7.0). The mixing solution contains 250µl solution of 20 mM p- nitrophenyl α-D-glucopiranoside, 495 µl buffer phosphat (pH 7.0), and 5 µl sample. Then, it was pre-incubated at 37° C for a while. Reaction was started by adding 250 µl α-glucosidase, then incubated it at 37° C for a while. The reaction was stopped by adding 1000µl Na_2CO_3 solution. The amount of p-nitrofenol obtained was measured at 400 nm wave length. The percentage of the inhibitory activity was counted by using this formula:

$$\text{Inhibitory activity (\%)} = \frac{(C-S)}{C} \times 100$$

where C = absorbance of enzyme activity without inhibitor (absorbance of DMSO), and S = absorbance of enzyme activity with sample examined.

RESULT AND DISCUSSION

IC_{50} (ppm) were counted from the regression equation (concentration vs inhibition percentage). 1% concentration of sample in reaction mixture is 25 ppm. The graphs are shown in Figure 1 until 4.

Tabel 1. IC50 ethanolic extract of *K. pinnata* (Lam.Pers.)

Sample	IC50
Quercetin	10.22
CB EtOH Bogor City	40.94
CB EtOH *Kabupaten* Bogor	33.58
CB EtOH Tangerang Selatan City	16.12

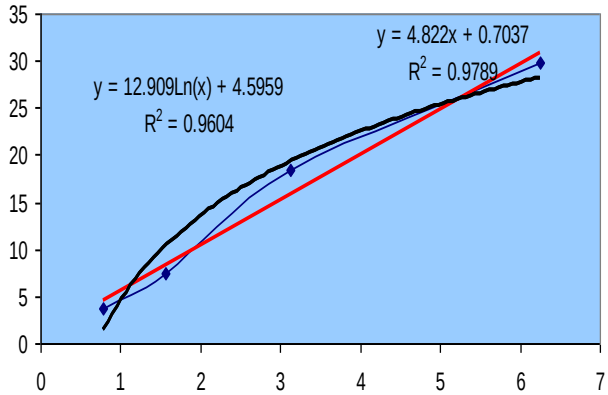

Figure 1. Regression equation of quercetin as standard

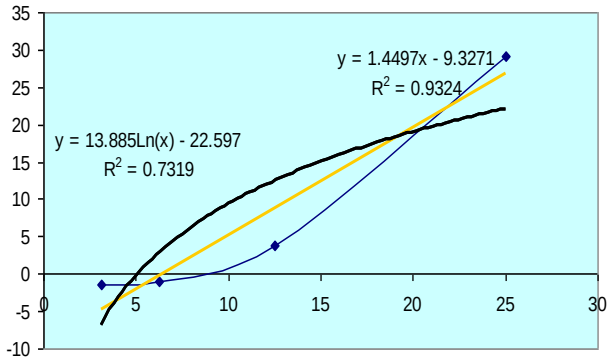

Figure 2. Regression equation of *Cocor Bebek*'s ethanolic extract from Bogor City

Ethanol was used as solvent in order to obtain less polar compound until the polar compound reached maximum. The ethanol can extract alkaloid, sterol, saponin, flavonoid Anthraquinone and glicoside compound from plants. Quercetin, a flavonoid compound which has antioxidant property, was used as the standard because of its anti-diabetic activity as shown by the research carried by Vessal et al. (2003) in which they used streptozocin induced diabetic rat [11].

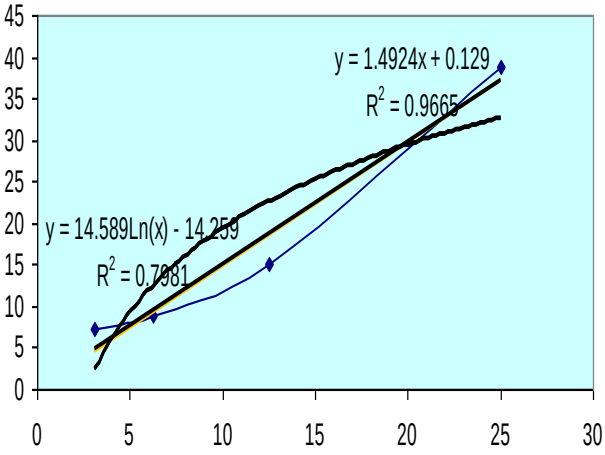

Figure 3. Regression equation of *Cocor Bebek*'s ethanolic extract from *Kabupaten* Bogor

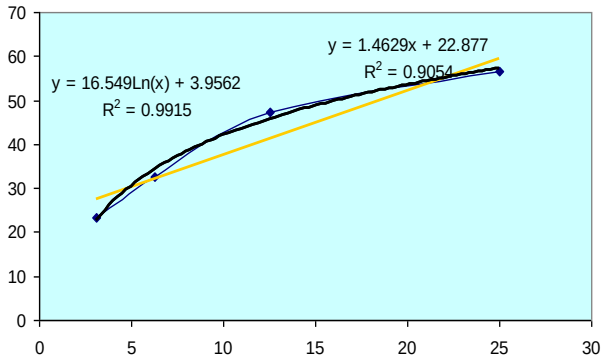

Figure 4. Regression equation of *Cocor Bebek*'s ethanolic extract from Tangerang Selatan city

The results of this study showed that IC_{50} of the extract from Bogor city, *Kabupaten* Bogor, and Tangerang Selatan city is 40.94 ppm, 33.58 ppm, and 16.12 ppm, respectively. Meanwhile, quersetin, which has antidiabetic activity, has IC_{50} 10.22 ppm. This study shows that the anti-diabetic activity of *K. pinnata* ethanolic extract varies. Its habitat and the sunlight can influence flavonoid compound in the plant such as quercetin [12]. The sunlight can improve flavonoid constituent in the plant as a result of adaptation because of the near wave length effect of the sunlight [13, 14]. Temperature and water supply at certain season can also influence flavonoid constituent in leaves [15, 16].

CONCLUSION

The result of this study shows that *K. pinnata*'s ethanolic extract has anti-diabetic properties and the activity is influenced by the habitat of the plant. The drier the climate, the stronger the activity will be.

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