



Research Journal of
**Medicinal
Plant**

ISSN 1819-3455



Academic
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Research Article

α -glucosidase Inhibitory Activity of *Shorea belangeran* of Kerangas Forest

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Abstract

Objective: Screening of anti-diabetic activity of some plants was conducted by having ethno botanical survey. **Methodology:** Laboratory analysis was also shown to obtain the bioactivity of *S. belangeran* (*Shorea belangeran*). Bioactivities of methanol extract from leaves and barks of *S. belangeran* are: (i) Qualitative phytochemical compound and (ii) Anti-diabetic activity. Qualitative phytochemical compound was analyzed by colour visualization method. Anti-diabetic activity was determined based on inhibitory activity to α -glucosidase enzyme. **Results:** The result displays that parts of plant of *S. belangeran* of the Kerangas forest have various phytochemical compounds. Methanol extract from the bark of *S. belangeran* has a concentration index of 50% (IC_{50}) to α -glucosidase enzyme at 0.816 ppm concentration. This appearances that the methanol extract of *S. belangeran* potentially have an anti diabetic activity, even though the IC_{50} of glucobay is in 0,167 ppm concentration. **Conclusion:** The bioactivity identification of some tree species is an important step to have a high economical value of Kerangas forest.

Key words: α -glucosidase, anti-diabetic, phytochemical, Kerangas, *Shorea belangeran*

Received: February 08, 2016

Accepted: May 06, 2016

Published: June 15, 2016

Citation: Kissinger, Ahmad Yamani and Rina Muhayah Noor Pitri, 2016. α -glucosidase inhibitory activity of *Shorea belangeran* of Kerangas forest. Res. J. Med. Plants, 10: 356-361.

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Kerangas is one of land type which is characterized by podsol soil with poor of nutrient and rich of quartz sand, low pH and often has thin peat layer above the soil surface. Growing vegetation is also limited and has a special character as the result of adaptation to limited environment¹. The area of Kerangas (heath) forest is categorized as IUCN (The International Union for The Conservation of Nature-World Conservation Union) under vulnerable status.

The general perspective assumes that recently the Kerangas forest not significantly gives a direct economical profit to people and development. A case in South Kalimantan, Kerangas forest is only used for its wood and as a source of dugouts material. Usage of Kerangas is destructive to plant, land and environment of the Kerangas. Ecological destruction of Kerangas forest and its impact on other part of socio-life should be addressed by conservation practices to the Kerangas forest.

A new paradigm in biodiversity conservation is how we can utilize the biodiversity with relatively environmental friendly method. So, the utilization does not destruct the biodiversity permanently but has a potential to have high and sustainable economical benefit value². Utilization of plant bioactivity as a medication substance is an alternative utilization relatively less destructive and more environment friendly.

Diabetes mellitus is a serious chronic condition that is a major source of ill health worldwide. It is affected around 171 milion people worldwide in 2000 and it may be increase to 366 million by 2030. More than 80% of people with diabetes live in low and middle-income countries. Statistical projection about Indonesia suggests that the No. of diabetics will rise from 8,426,000 in 2000 to 21,257,000 in the year 2030³. There were 10 million cases of diabetic in Indonesia in 2015⁴. The control of diabetic patients was achieved mainly by using oral anti-diabetic agent. Major limitations of the oral antidiabetic agent are their side effects and cost of therapy.

In developing countries, all over the world, 80% of population continues to use traditional medicine in primary medical problems. Traditional medicine provide the best health services⁵. Traditional medicines and extracts from medicinal plants have been extensively used as alternative medicine for better control and management of diabetes mellitus⁶. The major advantages of medicinal plants seem to be their efficacy, low incidence of side effects and low cost⁷. Medicinal plant are continued to be a powerful source for new drugs, contributing about 90% of newly discovered pharmaceuticals⁸. Traditional medicine provides better health

coverage for 80% of the world population, especially in the developing country⁹. The World Health Organization has recognized the importance of anti-diabetic plants in the development of economic and effective treatment for diabetes worldwide.

Shorea belangeran (also called Red Balau) is a species of plant in the Dipterocarp family. It is found in Sumatera and Kalimantan. Habitat *S. belangeran* are peat swamp forest (IUCN) and kerangas forest on poor sandy soil^{1,10}. It is a critically endangered species threatened by habitat loss (IUCN red list of threatened species). Bark of *S. belangeran* from Experimental Garden Darmaga Bogor contains oligostilbenoid¹¹. Bark extract of *S. belangeran* from peat swamp forest have antibabesial activity¹².

This research was trying to find the anti-diabetic bioactivity of some *Shorea belangeran* that grow in Kerangas forest (Fig. 1). Finding of anti-diabetic activity of Kerangas plants has been conducted based on ethno-botanical knowledge approach of people's around forest and the result of initial identification of qualitative phytochemical compound of Kerangas plant.

The result of this research is expected to give valuable information to development of bioactivity utilization of the Kerangas forest. Furthermore, it will more enrich knowledge on biodiversity of Kerangas forest for conservation interest and industrial development or people's small business in health and medication sectors based on traditional knowledge and empirical support from knowledge and technology.

MATERIALS AND METHODS

Ethno botanical survey and sample collection: The equipment and material of ethno botanical survey and sample collection consists of stationery, container sample, questionnaire, camera and GPS. Materials that were collected and tested are Leaves and bark of some tree species of Kerangas forest.

Equipment and material for chemical analysis: Equipment and material for chemical analysis are for plant extraction activity, qualitative phytochemical test and inhibitory strength test of plant sample to α -glucosidase. Chemical test was conducted in Biopharmaca Study Center Laboratory of IPB Bogor. Materials used are methanol solvent for maceration process of dry material of leaves and bark, phosphate buffer pH 7, akarbose, α -glucosidase enzyme (Sigma G3651-250UN), nitrofenil α -glucopiranosida (PNP), Na_2CO_3 0.2 M and dimethyl sulfoxide (DMSO), Na_2CO_3 , p-nitrofenol, glucobay (Bayer) dan HCL 2 N.



Fig. 1: *Shorea belangeran* in Kerangas forest

Field inventory: The location of ethno botanical study of Kerangas forest are in Banjar Regency South Kalimantan and in Palangkaraya and Sampit Central Kalimantan.

Activity stages: Selecting plant type which has a potency of natural material having anti-diabetic activity are as follows:

- **Ethno botanical survey:** Ethno botanical survey was intended to do inventory of tree species of Kerangas forest which has potency as anti-diabetes. Data collection was conducted by structured interview method¹³. Data analysis was showed descriptively using tabulation matrix
- **Identification of qualitative phytochemical compound¹⁴:** Qualitative phytochemical test was conducted using colour visualization method. The identification test of qualitative phytochemical compound can be seen in Fig. 2. The result of phytochemical analysis becomes an additional guideline in selection of plant type which has potency as anti-diabetes. Usage of literature reference on relationship between phytochemical compound content to anti diabetic capacity was conducted using advanced selection method to determine Kerangas tree species that will be tested for its potency as anti-diabetes. Data analysis was done descriptively using tabulation matrix

Test of anti-diabetic activity: From some chosen tree species, test was operated to methanol extract of leaves and bark from chosen trees. Anti-diabetic test in vitro from methanol extract of chosen plant to inhibitory strength of α -glucosidase enzyme¹⁵. Tested sample was dissolved in solvent of dimethyl sulfoxide (DMSO) with concentration of 1% (b/v). A 1.0 mg



Fig. 2: Qualitative phytochemical test

α -glucosidase was dissolved in 1 mL phosphate buffer of 100 mM (pH 7.0) then added by 200 mg SBA that has been dissolved in phosphate buffer 100 mM (pH 7.0). Before being used, 1 mL of enzyme solvent was diluted 25 times with phosphate buffer (pH 7.0). Mixed reaction consists of 500 μ L PNG 20 mM as a substrate, 980 μ L phosphate buffer solvent (pH 7) and 20 μ L of sample solvent in dimethyl sulfoxide (DMSO). Mixed reaction was incubated in 5 min and added by 500 μ L of α -glucosidase solvent then incubated in 15 min. Enzymatic reaction was terminated by adding 2000 μ L Na_2CO_3 and p-nitrophenol which is produced then read the absorbant with spectrophotometer UV in wavelength of 400 nm. Acarbose tablet (Glucobay) was dissolved in buffer and HCl_2

N (1:1) with concentration of 1% b/v as a positive control. Deposit was gathered as centrifuging and supernatant of 20 μ L was put into reaction mixture as on sample. The result of the reaction was measured by spectrophotometer UV of wavelength of 400 nm. Sample and control was positively done in two repetition (duplo). Positive control data is used as a comparison with tested sample in wavelength of 410 nm. Data analysis for inhibitory capacity of extract of chosen trees to α -glucosidase enzyme was conducted by calculating IC₅₀ value. Inhibitory level (%) is calculating using this equation:

$$\text{Percentage of inhibitory} = \frac{K - (S_1 - S_0)}{K} \times 100$$

Where:

K = Negative control absorbant

S₁ = Absorbant sample by adding enzyme

S₀ = Absorbant sample without adding enzyme

RESULTS AND DISCUSSION

Based on the information from native people around the forest of Kerangas forest, there are 27 general plants in Kerangas forest, 20 types of them are used as medication substance. Tree species used as anti-diabetes: Bark of *S. belangeran*, leaves of Rambuhatap (*Backea frutescens*) and leaves of Tabat Barito (*Ficus delteodea*).

Identification of phytochemical compound for 3 plant species which have anti-diabetes capacity based on people's information. The result of identification of phytochemical compound of some dominant trees in Kerangas forest can be seen in Table 1.

Anti-diabetes test of 2 plant species samples as comparison to tested *S. belangeran*. These species with *S. belangeran* that people believe to have role as anti-diabetes. The test shows that inhibitory capacity to α -glucosidase from methanol extract of some tree species of Kerangas can be seen in Table 2.

Table 1 displays that almost all sample contain flavonoid, phenol hydroquinone and tannin. Alkaloid is only available in 1 tree species that is bark of *S. belangeran*. Some study shows that anti-diabetes capacity is related with antioxidant capacity^{16,17}. Antioxidant capacity mostly related with the content of flavonoid, phenol hydroquinone and tannin^{18,19}. Some compounds such as terpenoid, flavonoid, phenolic, has potency as anti-diabetes²⁰. Tannin compound which is in some material has potency as anti-diabetes¹⁷. Flavonoid in extract of *Acalypha indica* is potential as anti-diabetes²¹.

Table 1: Phytochemical compound of some tree samples of Kerangas forest

Plant species	1	2	3	4	5	6	7	Part of plant
Belangiran (<i>Shorea belangeran</i>)	+	+	+	-	+	+	+	Bark
Tabat Barito (<i>Ficus delteodea</i>)	-	+	+	+	+	+	+	Leaves
Rambuhatap (<i>Backea frutescens</i>)	-	+	+	+	+	+	+	Leaves

Table 2: IC₅₀ of methanol extract of some Kerangas forest trees

Plant species	Part of plant	IC ₅₀ (ppm)
Belangiran (<i>S. belangeran</i>)	Bark	0.816
Tabat Barito (<i>F. delteodea</i>)	Leaves	29.715
Rambuhatap (<i>B. frutescens</i>)	Leaves	21.796
Positive control	Glucobay	0.167

Based on relationship between plant phytochemical compound to anti-diabetes capacity, there are 7 tree species more that potentially can be an anti-diabetes besides 3 tree species which are potentially as anti-diabetes based on people's knowledge.

Methanol extract of bark of Belangiran (*S. belangeran*) has IC₅₀ with concentration below 1 ppm. Methanol extract of leaves of Tabat Barito (*F. delteodea*), Rambuhatap (*B. frutescens*) has IC₅₀ more than 20 ppm. The IC₅₀ akarbose (glucobay) which is bigger than methanol extract sample of plant will be possible due to active compound concentration in raw extract which has not yet been fractionated.

The result indicates that 1 tree species that is usually used by people in diabetes medication (bark of *S. belangeran*) was proof at concentration of <1 ppm has inhibitory activity to α -glucosidase enzyme. Inhibitory capacity α -glucosidase enzyme is assumed to be related to synergetic effect of phytochemical compound (Table 2) which is in methanol extract of plant part²². Chemical compounds such as terpenoid, flavonoid, phenolic, tannin, has potency as anti-diabetes^{17,20,21}. Ability of those compounds as anti-diabetes is caused by the existence of conjugated double bond in the form of cyclic bound (phenol and its derivative) and straight chain bond (aliphatic).

The result of analysis of anti-diabetes from methanol extract of *S. belangeran* is better comparing to other plant extract which is usually used as traditional medication. Extract of etil acetat of pandan wangi leaves (*Pandanus amaryllifolius*) has IC₅₀ in the concentration of 94.23 ppm²³. Methanol extract of *Syzygium malaccense* detain α -glucosidase with IC₅₀ of 5.7 ppm²⁰. Cinnamon powder (*Cinnamomum cassia*) has IC₅₀ in concentration of 55.02 ppm²⁴. This finding is more enrich knowledge on plant species diversity which is potential as anti-diabetes.

Implication to conservation implementation or environmental friendly utilization is that most part of plant used for anti-diabetes are bark and leaves, so it is not permanently destruct the plant. Usage of part of plant that can

permanently harm the plant such as part of root can be addressed by *in situ* breeding action and arrangement of plant population which is relatively safe to be harvested.

CONCLUSION

Extract of bark of *S. belangeran* which is the Kerangas trees potentially as anti-diabetes has IC₅₀ below 1 ppm (0.816 ppm). Capacity of anti-diabetes from methanol extract of part of plant from Kerangas trees is assumed has synergetic effect of phytochemical compound content of each tree species.

Usage of part of plant as a source of anti-diabetes is relatively safe to maintain plant sustainability. Utilization of part of plant which is permanently merusak are part of root can be addressed with *in situ* breeding action and arrangement of plant population which is relatively safe to be harvested. Fractionation or advance test should be conducted to strengthen proof on potency of anti-diabetes from trees in Kerangas forest.

ACKNOWLEDGMENTS

Authors appreciated to Directorate General of Higher Education (DIKTI) for funding of this study. Special appreciation was extended to Biopharmaca Study Center Laboratory of IPB Bogor, Indonesia for an excellent assistant in analysis of bioactivity.

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