

## Inhibitory Effect on $\alpha$ -Glucosidase by Traditional Thai Medicinal Plants

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### Abstract

$\alpha$ -glucosidase inhibitors are utilized in the treatment of type II diabetes mellitus (DM). The inhibition of intestinal  $\alpha$ -glucosidase would delay the digestion and absorption of carbohydrates and consequently suppress postprandial hyperglycemia. Thai medicinal plants are widely used in the treatment of diabetes mellitus. An attempt to verify their Inhibitory effect on  $\alpha$ -glucosidase from two different sources are reported herein. Among of them, the methanolic extract of *Amomum xanthioides* showed significant inhibitory activity against  $\alpha$ -glucosidase, the sucrase and maltase from rat intestine with IC<sub>50</sub> value 3.10 4.20 and  $\alpha$  glucosidase from baker's yeast with IC<sub>50</sub> values of 26.6 which is 25 times greater than the standard anti diabetic drug, acarbose. The preliminary observation provides the basis for further examination of Thai medicinal plants as supplement food and DM drug toward the treatment and prevention of diabetes.

**Keywords:**  $\alpha$ -Glucosidase inhibitor, Traditional Thai Medicinal Plant, non-communicable disease, hyperglycemia, Diabetes Melitus, *Amomum xanthioides*

### 1. Introduction

NCD is non-communicable disease such as heart disease, stroke, cancer, diabetes and obesity which cause 56 million deaths globally [1]. Among of them, DM type II was related to other NCDs, and is the leading cause of blindness, kidney failure and heart attack [2]. The management of DM type II usually requires the regimens including diet and medicine. Therefore,  $\alpha$  glucosidase inhibitor can retard the liberation of D-glucose absorption, reducing plasma glucose levels and suppressing postprandial hyperglycemia Consequently, drugs such as acarbose and miglitol have been approved for clinical treatment of DM [3-4].

As a part of our research for potent  $\alpha$  glucosidase inhibitors, we focused on traditional Thai medicinal plants widely used in the treatment of diabetes mellitus. The extract of *Lagerstoemia speciosa*, *Averrhoa carambola*, *Momordica charantia*, *Wedelia trilobata*, *Pseuderanthemum palatiferum*, *Moringa oleifera*, *Amomum xanthioides* *Helicteres isora*, *Aegle marmelos* and *Teminalia chebula* were tested for their inhibitory activities against  $\alpha$ -glucosidase from baker's yeast and  $\alpha$ -glucosidase (maltase and sucrase) from rat intestine [5-7].

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## 2. Materials and Methods

### 2.1 Plant Materials

The traditional medicinal plants such as *Lagerstoemia speciosa*, *Averthoa carambola*, *Momordica charantia*, *Wedelia trilobata*, *Pseuderathemum palatiferum*, *Moringa oleifera*, *Amomum xanthioides*, *Helicteres isora*, *Aegle marmelos* and *Teminalia chebula*. were collected at Phranakhon Si Ayutthaya, Thailand. All of them were identified by botanist, staff of the Faculty of Science and Technology. Plant specimens were deposited at Herbarium of Science and Technology Center, Phranakhon Si Ayutthaya Rajabhat University.

### 2.2 Extraction

The air-dried selected part of medicinal plants (500 g) were ground and extracted with 1:1 methanol:water (250 mL.) at 60°C with a soxhlet extractor for 24 hrs., and then evaporated under reduced pressure to yield methanolic extract, which was defatted by hexane. The defatted methanolic extract was tested an inhibitory activity against  $\alpha$ -Glucosidase.

### 2.3 $\alpha$ -Glucosidase assay: ( $\alpha$ glucosidase from Baker's Yeast)

Inhibitory effect against  $\alpha$ -glucosidase from baker's yeast was performed using our protocol previously reported [8-9]. Briefly, the  $\alpha$ -glucosidase (0.1 U/mL) and substrate (1 mM *p*-nitrophenyl- $\alpha$ -D-glucopyranoside) were dissolved in 0.1 M phosphate buffer, pH 6.9. A 10  $\mu$ L of test compound (1 mg/mL in DMSO) and incubated with 40  $\mu$ L of  $\alpha$ -glucosidase at 37 °C for 10 min. A 50  $\mu$ L substrate solution was then added to the reaction mixture and incubated at 37 °C for additional 20 min. The reaction was terminated by adding 100  $\mu$ L of 1 M Na<sub>2</sub> CO<sub>3</sub>, Enzymatic activity was quantified by measuring the absorbance at 405 nm (Bio-Red microplate reader model 3550 UV). The percentage inhibition was calculated by  $[(A_0-A_1)/A_0] \times 100$ , where A<sub>0</sub> is the absorbance without the sample, and A<sub>1</sub> is the absorbance with the sample. The IC<sub>50</sub> value was determined from a plot of percentage inhibition versus sample concentration. Acarbose was used as a standard control and the experiment was performed in triplicate.

### 2.4 $\alpha$ -Glucosidase assay: ( $\alpha$ - glucosidase from rat intestine)

Inhibitory activity against  $\alpha$ -glucosidase from rat intestine was determined using our developed method [8-9]. The crude enzyme solution prepared from rat intestinal acetone powder was used as a source of maltase and sucrase. Rat intestinal acetone powder (1 g) was homogenized in 30 mL of 0.9%NaCl solution. After centrifugation (12,000g  $\times$  30 min), the aliquot was subjected to assay. A 10  $\mu$ L of test compound was added with a substrate solution (10 mM maltose and 100 mM sucrose, each 20  $\mu$ L), 0.1 M phosphate buffer (pH 6.9, 30  $\mu$ L), glucose oxidase solution (80  $\mu$ L) and enzyme solution (20  $\mu$ L). The reaction mixture was incubated at 37°C (10 min for maltase and 40 min for sucrase) and its absorbance was determined at 500 nm. Percentage inhibition and IC<sub>50</sub> value was obtained using aforementioned methodology.

### 2.5 Data Analysis

The concentrate of tested compounds required to inhibited 50% of the  $\alpha$  glucosidase activity under the assay conditions was determined from dose-response curves and defined as IC<sub>50</sub> Value. Statistic significant was evaluated by one-way analysis of variene (ANOVA). A statistically significant difference was defined as  $p < 0.05$

### 3. Results and Discussion

The results of extraction were recorded as percentage yield of dried weight showed in Table 1. The extract of *Helicteres isora* revealed the highest value of percentage of weight of extract by dry weight of plant.

**Table 1** The Percentage Yield of Extract Of Medicinal Plant

Name of Plant	Family	Parts used	% wt. by wt.
<i>Lagrstoemia speciosa</i>	Lythraceae	Leaf	75.0
<i>Averrhoa carambola</i>	Oxalidaceae	Leaf	72.6
<i>Momordica charantia</i>	Cucurbitaceae	Fruit	44.5
<i>Wedelia trilobata</i>	Asteraceae	Leaf	58.2
<i>Pseuderathemum palatiferum</i>	Acathaceae	Leaf	53.0
<i>Moringa oleifera</i>	Moringaceae	Leaf	54.13
<i>Amomum xanthioides</i>	Zingiberaceae	Seed	60.5
<i>Helicteres isora</i>	Sterculiaceae	Fruit	78.80
<i>Aegle marmelos</i>	Rutaceae	Leaf	48.2
<i>Terminalia Chebula</i>	Combretaceae	Leaf	72.6

All extracts were subjected to evaluation for inhibitory effects against  $\alpha$  glucosidase from two different sources, baker's yeast (type I) and rat intestine (type 2). Comparison were made with Acarbose which is a drug proven to have anti-diabetes clinical efficacy. The inhibitory activity of medicinal extract were obtained and the IC<sub>50</sub> values were calculated (Table 2) The extract of *Lagerstoemia speciosa* inhibited  $\alpha$  glucosidase from baker's yeast with IC<sub>50</sub> values of 140 mM *Averrhoa carambola*, *Wedelia trilobata*, *Helicteres isora* and *Terminalia Chebula* revealed toward maltase and surcease whereas inhibitory activity against baker's yeast was not observed while *Momordica charantia*, *Moringa oleifera*, *Amomum xanthioides*, *Aegle marmelos* inhibited  $\alpha$ -glucosidase from both sources. Among of them *Amomum xanthioides* exhibited the highest inhibitory activity against. both sucrose and maltase with IC<sub>50</sub> value 3.10 4.20 and  $\alpha$  glucosidase from baker's yeast with IC<sub>50</sub> values of 26.6 which is 25 times greater than the standard anti diabetic drug, acarbose. In this study, acarbose and all medicinal plants extracts exhibited a less pronounced inhibition of  $\alpha$  glucosidase from baker's yeast compared to  $\alpha$  glucosidase from rat intestine which suggests distinct differences in inhibitory potency of them toward  $\alpha$  glucosidase from different species.

**Table 2** Glucosidase Inhibitory effect of Medicinal Plants

Extract	IC <sub>50</sub> (µg/mL)		
	Baker's Yeast	Maltase	Sucrase
<i>Lagerstoemia speciosa</i>	140.0±0.04	NI <sup>a</sup>	NI <sup>a</sup>
<i>Averrhoa carambola</i>	NI <sup>a</sup>	21.42±0.04	7.9±0.05
<i>Momordica charantia</i>	33.3±0.01	15.0±0.02	23±0.18
<i>Wedelia trilobata</i>	NI <sup>a</sup>	65.1±0.01	41.6±0.03
<i>Pseuderathemum palatiferum</i>	135.0±0.01	36.2±0.04	NI <sup>a</sup>
<i>Moringa oleifera</i>	52.1±0.10	74.2±0.02	258.5±0.02
<i>Amomum xanthioides</i>	26.6±0.03	3.10±0.02	4.20±0.03
<i>Helicteres isora</i>	NI <sup>a</sup>	3.71±0.21	2.15±0.48
<i>Aegle marmelos</i>	40.8±0.01	16.6±0.01	3.12±0.03
<i>Terminalia Chebula</i>	NI <sup>a</sup>	97.0±0.02	140.0±0.03
Acarbose <sup>®</sup>	502±0.04	3.60±0.02	2.10±0.14

<sup>a</sup> No inhibition, inhibitory effect less than 30% at 10 mg/mL.

#### 4. Conclusions

In conclusion, we evaluated the inhibitory activity of medicinal plant extract compared to synthetic glucosidase inhibitor acarbose. To summarize the Inhibitory effects on  $\alpha$ -glucosidase of Traditional Thai Medicinal plants, among of them, *Amomum xanthioides* was greatest inhibitory activity against glucosidase from two different sources, baker's yeast (type I) and rat intestine (type II).

The primary observation will provide the basis for further examination of Thai medicinal plant as medicinal supplement that contribute toward the treatment and prevention of diabetes.

#### 5. Acknowledgements

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