UNIVERSITI TEKNOLOGI MARA

IN VITRO α-AMYLASE AND α-GLUCOSIDASE INHIBITORY ACTIVITIES OF SELECTED MALAYSIAN PLANTS AND *IN VIVO* ANTIDIABETIC PROPERTIES OF *KNEMA GLAUCA*

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ABSTRACT

Uncontrolled postprandial hyperglycaemia increases the risk of vascular diabetic complications. One of the therapeutic approaches to control postprandial hyperglycaemia is by inhibiting α-amylase and α-glucosidase enzymes activity. In this study, 50 extracts from 23 Malaysian tropical plants were assayed for α-amylase and α-glucosidase inhibitory activities. A microscale α-amylase inhibition assay was optimized and established based on method previously described by Hasenah *et al.* (2006). In comparison with the previous method, the microscale assay is simple and consists of less steps, can screen up to 22 samples at a time, decrease consumption of chemicals up to 400 % and produced reliable results. Screening of 23 Malaysian tropical plants using method described by Haseanah *et al.* (2006) found eight plant species demonstrated potent α-amylase inhibitory activity. The plants were *Burkillantus malaccensis* (stem), *Horsfieldia polyspherula* (leaves and stem), *Knema glauca* (leaves and stem), *Labisia pumila* (leaves and root) and *Phyllantus pulcher* (stem). The IC_{50} values of these extracts ranging from 1.17 to 2.78 μg/mL. Six plant species may be classified as strong α-glucosidase inhibitory activity with IC_{50} values ranging from 3.4 to 6.1 μg/mL. The plants were *Gironniera parvifolia* (stem), *H. polyspherula* (leave and stem), *K. glauca* (leaves and stem), *Leea indica* (root), *P. pulcher* (leaves and stem), *Rothmannia schoemannii* (stem). Extracts of *P. pulcher* (stem), *K. glauca* (leaves and stem) and *H. polyspherula* (leaves and stem) showed potent inhibitory activities against both enzymes. The leaves and stem extracts of *K. glauca* (*Penarahan*) belonging to family *Myristicaceae* were selected and examined further for cytotoxicity and *in vivo* antidiabetic properties. The leaves and stem extracts exhibited moderate cytotoxicity against normal cell lines of WRL-68 (human liver) with CC_{50} values of 26.55 and 89.49 μg/mL, respectively. The leaves and stem extracts showed moderate cytotoxicity towards normal cell lines of Vero (African green monkey kidney) with CC_{50} values of 32.76 and 52.05 μg/ml, respectively. Oral starch tolerance test in normal and diabetic rats showed the leaves extract at all doses (125, 250 and 500 mg/kg) significantly reduced ($P<0.05$) blood glucose level. The similar test showed the stem extract significantly reduced blood glucose level in normal and diabetic rats at dose of 500 mg/kg. Oral sucrose tolerant test showed the leaves extract at dose of 500 mg/kg significantly suppressed ($P<0.05$) blood glucose level in normal rats. Oral glucose tolerance test showed the leaves extract at all doses (125, 250 and 500 mg/kg) significantly suppressed ($P<0.05$) blood glucose level in normal rats. In diabetic rats, the extract at dose of 500 mg/kg reduced blood glucose level significantly. The stem extract demonstrated significant reduction of blood glucose level in normal and diabetic rats at a single dose 500 mg/kg. In a 2-week antidiabetic experiment showed the leaves and stem extracts did not show significant effects on fasting blood glucose level and body weight. This study revealed the potential of *K. glauca* extracts to control postprandial hyperglycemia in diabetic management.

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CHAPTER 1

INTRODUCTION

Diabetes mellitus is a well known disorder of carbohydrate metabolism that is characterized by hyperglycemia which results from defects on insulin secretion, insulin action or both (Chandra *et al.*, 2004). Statistically, more than 180 million people worldwide are suffering from diabetes with 5% deaths globally each year. Without intervention, this number will double by 2030 (World health organization, 2005). Investigations of natural antidiabetic agents are an important research area in order to provide humankind with new and safer therapeutic products. Plant based materials have been used in treatment of diabetes since ancient time. More than 1200 plants are being used traditionally as antidiabetic remedies however, only approximately 30% of the plants have been scientifically investigated (Alarcon-Aguilar *et al.*, 2002). On the other hand, many more plants are widely used in treatment of diabetes without much scientific evidence.

Blood glucose elevation in patient with *Diabetes mellitus* is correlated to postprandial hyperglycemia. Postprandial hyperglycemia happen by action of digestive enzymes which are α-amylase and α-glucosidase that act as catalysts to hydrolyze certain groups of carbohydrate into the simplest product that leads to improper regulation of glucose level in blood. One strategy in management of *Diabetes mellitus* is by retarding the digestive enzymes activity in order to suppress postprandial hyperglycemia. Development of macrovascular and microvascular diseases that are closely related to complications of postprandial hyperglycemia can