COPYRIGHT© INTERNATIONAL ISLAMIC UNIVERSITY MALAYSIA

ANTIDIABETIC EFFECTS OF ETHANOLIC EXTRACT OF *P. MACROCARPA (SCHEFF) BOERL.* FRUITS IN STREPTOZOTOCIN-INDUCED RATS

BY

MD. ABUL KALAM AZAD

A thesis submitted in fulfilment of the requirement for the degree of Master of Pharmaceutical Sciences (Pharmacology)

Kulliyyah of Pharmacy
International Islamic University Malaysia

MARCH 2015

ABSTRACT

Traditionally *P. macrocarpa* fruits have been used as an herbal anti diabetic remedy for a long time in South-East Asia. The purpose of this study was to evaluate the cytotoxicity, acute oral toxicity and antidiabetic effects of *Phaleria macrocarpa* (EEPM) fruits in streptozotocin-induced Sprague-Dawley rat model. The plant fruits were extracted using 70% ethanol followed by cold maceration. Brine shrimp lethality bioassay, using sea water was prepared by dissolving 38 g sea salt in 1 liter distilled water for two days, this is allowed to hatch the shrimp to nauplii and potassium dichromate was used as a positive control. In an acute oral toxicity study, twelve male adult Sprague-Dawley rats (10 weeks) weighing 180-200 g were divided into Group-I (Control- 10% normal saline) and Group-II (extract), n=6. The fruit extract (5000 mg/kg/b.w) was given orally to each rat and observation carefully at 4 and 6 hr intervals for any physical changes. In antidiabetic study, a total of thirty-six healthy adult male rats were divided into six groups (n=6). Diabetes was induced under light ether anesthesia by a single dose (65 mg/kg/b.w) of intraperitoneally injected streptozotocin. Their glycemic status (Oral glucose Tolerance Test) was re-evaluated intermittently at 0, 30, 60, 90 and 120 min, respectively. Blood sugar level (mg/dl) and body weight of each rat in the respective groups were repeatedly measured on day 0, 7, 14, 21, 28 and 35 of the experiment. The findings of the present toxicity study suggest that the ethanol extract of EEPM fruits is non-toxic. It was found that the EEPM at 50, 100 and 200 mg/kg and glibenclamide (0.5 mg/kg) reduced the blood glucose level (hyperglycemia due to glucose load 2 g/kg p.o.) significantly after 2 hr of oral administration, when compared to the diabetic control group. The repeated oral administration of EEPM daily up to 35 days exhibited significant ($p< 0.01$) blood glucose activity in STZ-induced diabetic rats compared with diabetic control. At the end of 35 days of treatment, the blood glucose level of normal control, drug control and diabetic control was 132.16±5.79, 134.33±7.18 ($p< 0.01$) and 514.83±7.96 respectively. In the treatment groups, the dose of EEPM 200 mg/kg (392.66±3.2 to 174.33±4.3 mg/dl, $p< 0.01$) was shown to be more effective than EEPM 100 mg/kg (392.5±3.9 to 240.5±9.2, $p< 0.05$) and EEPM 50 mg/kg 395.66±4.4 to 284.66±4.8 ($p< 0.05$). However, all selected doses showed antidiabetic activity gradually in STZ-induced diabetic rats. In histopathological examination results showed the pancreas of diabetic control were degranulated and dilated islet cells, whereas the drug control group showed granulated, nonappearance of dilation and hyperplasticity of islets. In treatment groups (EEPM at 100 and 200 mg/kg) also showed granulated pancreatic islets and prominent hyper plasticity islets. Light micrographs of rat kidney tissue in treatment groups showed various regions of the kidneys of treated animals with absence of matrix expansion and glomerular basement membrane thickening suggesting normal histoarchitecture of pancreas and renal. Biochemical aspects of the treated animal group were almost similar to the drug control group except the EEPM 50mg/kg group. In conclusion, EEPM may also serve as a good alternative in the present armamentarium of antidiabetic drugs.

Keywords: *P. macrocarpa, Toxicity, Antidiabetic, Histopathology, Kidney, Pancreas, Streptozotocin.*

خلاصة البحث

تستخدم ثمار *P. macrocarpa* (Scheff.) Boerl. (الميزوكارب والقشرة) كعلاج تقليدي ضد الداء السكري منذ مدة طويلة في بلدان منطقة آسيان. الهدف من هذه الدراسة هو لتقييم السمية الخلوية والسمية الحادة بعد الإعطاء الفموي وكذلك التأثير المضاد للسكري باستخدام نموذج جرذان سبارغ-دولي المحرضة بالستربتوزوتوسين. تم استخلاص الفواكه باستخدام الإيتانول 70% ومن ثم النقع البارد. تمت المعايرة الحيوية عن طريق تموت الجمبري البحري، تم تحضير ماء البحر بتذويب 38 غ من الملح البحري في ليتر من الماء المقطر لمدة يومين، هذا يسمح بفقس يرقات الجمبري وتم استخدام ديكرومات البوتاسيوم كمعينة ضابطة موجبة. تمت دراسة السمية الحادة بعد الإعطاء الفموي، 12 من ذكور الجرذان (10 أسابيع) بوزن (180-200 غ) مقسمة إلى مجموعة-1 (ضابطة- 10% محلول ملحي عادي) ومجموعة-2 (خلاصة)، العدد=6. تم إعطاء خلاصة الفواكه (5 غ/كغ/مرتي أسبوعياً) فموياً إلى كل جرذ ومراقبتها بانتظام كل 4 و 6 ساعات لملاحظة أي تغيرات فيزيائية. في دراسة التأثير المضاد للسكري،استخدم 36 جرذاً ذكراً بالغاً (سلالة سبارغ دولي) (8-12 أسبوعاً) بوزن 180-200 غ. تم استخدام 36 جرذاً (6 طبيعية و 30 مصابة بالسكري) في هذه التجربة. تم تقسيمها إلى 6 مجموعات من 6 جرذان في كل مجموعة. تم تحريض الداء السكري تحت التخدير الخفيف بالإيتر عن طريق حقنة وحيدة في البريتوان من الستربتوزوتوسين 65 ملغ/كغ. تم تقييم حالتها السكرية (OGTT) بشكل متقطع بعد 0 ساعة، و30، 60، 90 و120 دقيقة على الترتيب. تم قياس مستوى سكر الدم (ملغ/دل) ووزن الجسم (غ) لكل جرذ في المجموعات المذكورة أعلاه بشكل متكرر في اليوم 0 قبل التحريض، وفي الأيام 3، 7، 14، 21، 28 و 35 من التجربة وتسجيلها بشكل مناسب. أظهر الاختبار الفيتوكيميائي وجود القلويدات، الكربوهيدرات، الغليكوزيدات، السابونينات، التربين، الستيروئيدات، الفنولات والفلافونوئيدات. في الـOGTT أدى الـPME بجرعة 50، 100 و200 ملغ/كغ والغليبنكلاميد (0.5 ملغ/كغ) إلى خفض مستوى سكر الدم (ارتفاع سكر الدم نتيجة الحمل الغلوكوزي 2 غ/كغ .p.o) على نحو هام بعد ساعتين من الإعطاء الفموي، مقارنة بالمجموعة الضابطة السكرية. عند تكرار الإعطاء الفموي للـPME يومياً لمدة 35 يوماً أظهرت فعالية مضادة للسكر هامة في الجرذان السكرية المحرضة بالستربتوزوتوسين مقارنة بالمجموعة الضابطة السكرية. في نهاية الـ35 يوماً من المعالجة، جرعة 200 ملغ/كغ (PME) كانت أكثر فعالية من 100 و50 ملغ/كغ ومستوى سكر الدم كان قد انخفض من 392.66±3.20 إلى 174.33±4.32 ملغ/دل ($p< 0.01$). من ناحية أخرى، في اليوم 35، مستوى سكر الدم للمجموعة الضابطة الطبيعية، الضابطة الدوائية والضابطة السكرية كان 132.16±5.79 و 134.33±7.18 ($p< 0.01$)، 514.83±7.96. وكان بنكرياس المجموعة الضابطة السكرية خال من الحبيبات ومتسع خلايا الجزر بينما المجموعة الضابطة الدوائية أظهرت جزر متحببة وخالية من التمدد ومرونة عالية هامة. في مجموعات المعالجة (PME100 and 200 mg/kg) أظهرت أيضاً جزراً بنكرياسية متحببة وجزراً سائدة شديدة المرونة. الصور المكرونية الضوئية لنسج الكلى عند الجرذان في مجموعات المعالجة أظهرت عدم وجود التمدد وتثخن الغشاء القاعدي الكبيبي، مما يدل على تنسج طبيعي للكلى. القيم الكيميائية الحيوية في مجموعات المعالجة كانت تقريباً طبيعية مقارنة بالمجموعة الضابطة الدوائية باستثناء المجموعة المعالجة بـ50 ملغ/كغ. كخلاصة، من الممكن أن تستخدم الخلاصة المدروسة كبديل جيد للأدوية المتوفرة ضد الداء السكري.

الكلمات المفتاحية: *P. macrocarpa*، سمية، مضاد للسكري، الإمراضية النسيجية، كلية، بنكرياس، ستربتوزوتوسين.

APPROVAL PAGE

I certify that I have supervised and read this study and that in my opinion; it conforms to acceptable standards of scholarly presentation and is fully adequate, in scope and quality, as a thesis for the degree of Master of Pharmaceutical Sciences (Pharmacology).

……..
Wan Mohd Azizi bin Wan Sulaiman
Supervisor

…….
Muhammad Taher
Co-supervisor

I certify that I have read this study and that in my opinion, it conforms to acceptable standards of scholarly presentation and is fully adequate, in scope and quality, as a thesis for the degree of Master of Pharmaceutical Sciences (Pharmacology).

……..
Zainul Amiruddin Zakaria
External Examiner

…….
Suzanah Abdul Rahman
Internal Examiner

This thesis was submitted to the Department of Basic Medical Sciences and is accepted as a fulfillment of the requirement for the degree of Master of Pharmaceutical Sciences (Pharmacology).

……
Noriah Mohd Noor
Head, Department of Basic Medical Sciences

This thesis was submitted to the Kulliyyah of Pharmacy and is accepted as a fulfillment of the requirement for the degree of Master of Pharmaceutical Sciences (Pharmacology).

……..
Siti Hadijah Binti Shamsudin
Dean, Kulliyyah of Pharmacy

DECLARATION

I hereby declare that this thesis is the result of my own investigation, except where otherwise stated. I also declare that it has not been previously or concurrently submitted as a whole for any other degrees at IIUM or other institutions.

MD. Abul Kalam Azad

Signature……. Date….....

INTERNATIONAL ISLAMIC UNIVERSITY MALAYSIA

DECLARATION OF COPYRIGHT AND AFFIRMATION OF FAIR USE OF UNPUBLISHED RESEARCH

Copyright ©2015 by International Islamic University Malaysia. All rights reserved.

ANTIDIABETIC EFFECTS OF ETHANOLIC EXTRACT OF *P. MACROCARPA (SCHEFF) BOERL*. FRUITS IN STREPTOZOTOCIN-INDUCED RATS

No part of this unpublished research may be reproduced, stored in a retrieval system, or transmitted, in any form or by any means, electronic, mechanical, photocopying, recording or otherwise without prior written permission of the copyright holder except as provided below.

1. Any material contained in or derived from, this unpublished research may be used by others in their writing with due acknowledgement.

2. IIUM or its library will have the right to make and transmit copies (print or electronic) for institutional and academic purposes.

3. The IIUM library will have the right to make, store in a retrieval system and supply copies of this unpublished research if requested by other universities and research libraries.

Affirmed by Md. Abul Kalam Azad

……..……..…… ……..

Signature Date

ACKNOWLEDGEMENTS

All praises to Allah (SWT) the most merciful and the most gracious in giving me the capability and gallantry to complete this study without any labyrinth. This work would have never been possible without the assistance, understanding and guardianship of many people to whom I owe my sincere thanks and accumulation. May Allah the Almighty bless and reward them all abundantly.

I take this opportunity to express my profound gratitude and deep regards to my guide, Assistant Professor Dr. Wan Mohd Azizi Bin Wan Sulaiman for his exemplary guidance, monitoring and constant encouragement throughout the course of this valuable research project. The blessing, help and guidance given by him time to time shall carry me a long way in the journey of life on which I am about to embark.

I also take this opportunity to express a deep sense of gratitude to my co-supervisor, Associate Professor Dr. Muhammad Taher, for his cordial support, valuable information and guidance, which helped me in completing this task through various stages.

My sincere thanks and appreciation goes to my Deans of Kulliyyah of Pharmacy, Assistant Professor Dr. Siti Hadijah bt. Shamsudin. I am also grateful to my former Deans of Kulliyyah of Pharmacy, Associate Professor Dr. Mohammad bin Awang for his kind assistance and advice.

I would like to express my cordial grateful and thanks to Associate Professor Dr. Sheikh Farid Uddin Akter for his kind assistance and encouragement during my struggle period.

I also would like to express my special gratitude and thanks to Assistant Professor Dr. Norazian Mohd Hassan for his kind help for taxonomic identification of my research plant.

I wish to thank to all helpful laboratory staff in Kulliyyah of Pharmacy and Kulliyyah of Medicine, especially to Sr. Sri Viorwarti Noerdin and Mohd Hanif bin Mohd Kasmuri for their cooperation in laboratory technical assistance.

Sincere thanks go to my special teammate, my beloved friend Tg Muhamad Faris for his warm support during my research journey. Not to forget all my friends in postgraduate studies and all those who contributed to this research, I feel very much grateful for their cooperation during the period of my assignment.

Special thanks to my beloved parents, brothers and sisters for their constant encouragement without which this assignment would not be possible.

Lastly, big thanks to IIUM to give me an opportunity to enroll, academic scholarship for financial support and providing me the research facilities to complete my study here.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF ABBREVIATIONS

ADA	American Diabetes Association
AGI	α-glucosidase inhibitor
AHPA	American Herbal Products Association
ALT	Alanine Aminotransferase
ALP	Alkaline Phosphatase
ALX	Alloxan
ANOVA	Analysis of Variance
AST	Aspartate Aminotransferase
b.w.	Body weight
BB	Bio Breeding
BSLB	Brine Shirmp Letality Bioassay
CO2	Carbondioxide
cm	centimeter
DM	Diabetes Mellitus
DMEM	Dulbecco's Modified Eagle Medium
DMSO	Dimethyl Sulfoxide
DNS	5-dinitro salicylic acid
dL	Deciliter
Eds./ed.	Editions/edition
e.g	(exempli gratia); for example
et al	(et alia); and others
EtOH	Ethanol
EEPM	Ethanol Extract of Phaleria macrocarpa
FBS	Foetal Bovine Serum
Figure	Figure
FFA	Free fatty acid
g	Gram
GDM	Gestational Diabetes Mallitus
GLUT4	Glucose transporter type 4
H2O	Water
HCl	Hydrogen chloride
HDL	High Density Lipoprotine
H&E	haematoxylin-eosin
HSD	Honestly Significant Didderence
IDDM	Insulin Dependent Diabetes Mellitus
IFG	Impaired Fasting Glucose
IGT	Impaired Glucose Tolerance Test
IACUC	Instituitional Animal Care and Committeee
IIUM	International Islamic University Malaysia
LDL	Low Density Lipoprotine
LC50	Lethal Concentration 50
LD50	Median Lethal Dose 50
mL	millitre
mM	millimolar

Mmol/L	Millimoles per litre
MTT	3-(4,5-dimethylthiazol-2-y)2,5-diphenyltetrazolium bromide
mg	Milligram
N	Normality
n	Sample size
NaCl	Natrium Chloride/Sodium Chloride
NaOH	Sodium Hydroxide
NHMS III	National Health Morbidity Survey III
NIDDM	Non-Insulin-Dependent Diabetes Mellitus
No.	Number/Numbers
N.O	Not Observe
NOD	Non-obese diabetic mouse
ODM	Oral Diabetes Medications
OGTT	Oral Glucose Tolerance Test
OECD	Organization for Economic Co-operation and Development
PAS	Periodic Acid Schiff
PM	Phaleria macrocarpa
PBS	Phosphate Buffer Saline
PH	ATP
RPM	Revolutions per minute
SEM	Standard Error of Means
STZ	Streptozotocin
SD	Sprague-Dawley
TZD	Thiazolidinedione
T.BIL	Total Bilirubin
T.PROT	Total Proteins
USA	United States of America
WHO	World Health Organization
µL	Microliter
µm	Micrometer

LIST OF SYMBOLS

A	Alpha
B	Beta
®	Registered trademark
$	Dollar
P	The probability of obtaining the result
*	Statistical significance denotation
°C	Degree Celcius
%	Percent
<	Less than
>	More than
≤	Less than or equal to
≥	Greater than or equal to
=	Equality
×	Multiplication
+	Positive, plus
–	Negative, minus
±	Approximation

LIST OF EQUATIONS

CHAPTER ONE

INTRODUCTION

1.1 BACKGROUND OF THE STUDY

Diabetes mellitus is one of the most serious global health problems (Zimmet et al., 2001) and it is one of the commonest endocrine and metabolic disorders often characterized by "chronic hyperglycemia". Currently, it has been considered as the fifth leading cause of death worldwide. It has become a pandemic and threatening issue globally for public health in view of its associated fatal vascular complications and lack of effective long-term treatment. In addition, it accounts for the highest contribution to the direct health care cost, and socioeconomic burden of every society, community and nations at large (Reusch, 2003).

Physiologically, it is a disturbance of carbohydrate, fat and protein metabolism accompanied by an insufficiency of insulin secretion and/or action from beta cells of islets of Langerhans. In complicated diabetes mellitus cases, it can affect the eyes (i.e, diabetic retinopathy), kidneys, nerves (i.e, diabetic neuropathy), arteries and skin (Fujisawa et al., 2004).

In 1995, among the adult population (above 20 years), the diabetes rate was 4.0 % and this percentage will reach at 5.4 % in 2025 (WHO, 1999). Globally, the incidence was estimated 215 million in 2010 and it will be raised to 366 million by 2030 (Wild et.al., 2004).

Currently, the Malaysian Diabetes Association has estimated that 1.6 million Malaysian adults are suffering from diabetes among the total estimated population of 28.3 million. This enumeration is anticipated to extend 2.48 million by the year of 2030 (Letchuman et al., 2010). The highest incidence rate was attributed to several

factors, including population growth, aging, rapid urbanization, consumption of high energy rich diet, obesity, physical inactivity and genetic predisposition among others.

Worldwide, nearly 80% of diabetic patients are almost dependent on traditional or herbal medicines mainly due to cost effectiveness, traditional uses and little or no side effects. The World Health Organization (WHO) has enlisted more than 21,000 medicinal plants with their antidiabetic and antioxidant activity, and the major compounds are in the form of carbazoles and flavonoids (Bhaskar et al., 2008). The intake of these compounds as food supplements and others play important roles in aspect of health protection and prevention of degenerative illnesses. Yet, just a few of these herbaceous have obtained appropriate scientific screening.

1.2 JUSTIFICATION OF THE STUDY

Based on the literature review, there have been no toxicity and long term antidiabetic studies on *Phyleria macrocarpa (Scheff.) Boerl.* (PM) *in vivo* method. Therefore, the crude extract of PM fruits shall focus on determining safety issues and for treating, preventing and controlling diabetes mellitus effectively.

1.3 SCOPES OF THE STUDY

The current study aimed to determine the toxicity and antidiabetic activities of ethanol crude extract of EEPM fruits (meso- & peri-carp) by *in vivo* method using male Sprague Dawley rat models.

1.4 RESEARCH HYPOTHESIS

The EEPM fruit extract has the potential to reduce the elevated blood glucose level and improve regeneration of pancreatic islet cells and renal parenchyma in streptozotocin-induced diabetic rats

1.5 RESEARCH OBJECTIVES

a) a) To carry out qualitative preliminary phytochemical screening of EEPM fruit.

b) To conduct toxicity study of EEPM fruits.

c) To evaluate the antidiabetic effects of EEPM fruits on blood plasma glucose.

CHAPTER TWO

LITERATURE REVIEW

2.1 OVERVIEW

The trend of diabetes mellitus in Malaysia among adults aged over 30 years old had elevated from 7.6% to 11.9%. The data showed a positive relation with the increase obesity (Zanariah et al., 2010).

Diabetes mellitus is an incurable metabolic disorder, which transpired when the level of blood glucose is higher than normal, resulting from pancreatic dysfunction and/or lack of insulin production by itself ß-cells that leads to distinguished complications (WHO, 1999).

According to American Diabetes Association (ADA, 20012), There are four main types of Diabetes mellitus. Type 1 DM was known as insulin-dependent diabetes (IDDM), resulting from absolute insulin deficiency due to ß-cell destruction either immune-mediated or idiopathic. It mostly (more than 90%) occurs as a result of autoimmune islet cell destruction (Carver and Abrahamson, 2009).

Type 2 Diabetes mellitus is also called non-insulin dependent diabetes (NIDDM) which is a comparative insulin deficiency owing to the insulin secretion defect and insulin resistance (Wright, 2003).

Insulin resistance (IR) is a physiological condition in when cells fail to respond to the normal physiological actions of the hormone insulin is called insulin . The body produces insulin, but the cells in the body become resistant to insulin and are unable to use it as effectively, leading to hyperglycemia (Chiu et al., 2007). Beta cells in the pancreas subsequently increase their production of insulin, further contributing to hyperinsulinemia. One of insulin's functions is to regulate delivery of glucose into

cells to provide them with energy. Insulin resistant cells cannot take in glucose, amino acids and fatty acids. Thus, glucose, fatty acids and amino acids 'leak' out of the cells. A decrease in the insulin / glucagon ratio inhibits glycolysis, which in turn decreases energy production. The resulting increase in blood glucose may raise levels outside the normal range and cause adverse health effects, depending on dietary conditions (Sesti, 2006). Certain cell types such as fat and muscle cells require insulin to absorb glucose. When these cells fail to respond adequately to circulating insulin, blood glucose levels rise (Shulman, 2000).

Diabetes Care (2003) reported that about 90% to 95% of all diagnosed cases of diabetes are an adult-onset type diabetes mellitus. In this case, it begins as insulin resistance, adult-onset diabetes is a disorder in which the cells do not use insulin properly and as the need for insulin rises; the beta cells slowly loses its ability to produce insulin. It is associated with older age, obesity, family history of diabetes, history of gestational diabetes, impaired glucose metabolism, physical inactivity, and race/ethnicity. Type three diabetes mellitus is a little that has been proposed a certain specific condition and/or syndromes like Alzheimer’s disease which results from resistance to insulin in the brain. It is, the less common types of diabetes mellitus than others. The fourth type of diabetes mellitus is regarded as gestational diabetes (GDM) and it is usually diagnosed during pregnancy (ADA, 2012).

Type 2 Diabetes Mellitus is diagnosed on the basis of fasting plasma glucose (FPG) and two hour plasma glucose (2hPG) concentration by using the oral glucose tolerance test (OGTT). The ADA expert committee defined the category of pre-diabetic state is an impaired fasting glucose (IFG) and/ or impaired glucose tolerance (IGT). The diabetes level for FPG and 2hPG are 7.0 and 11.1 mmol/L, respectively (Drouin et al., 2009)

In normal condition, the fasting plasma glucose (FPG) concentration is from 100 to 125 mg/dl (=5.6 and <7.0 mmol/ml). It will be considered as a diabetes condition when the impaired fasting glucose (IFG) and impaired glucose tolerance (IGT) value is more than 126 mg/dl and 200 mg/dL respectively (ADA,2004).

2.2 IMPORTANCE OF MEDICINAL PLANTS AND TRADITIONAL MEDICINE IN THE TREATMENT OF TYPE II DIABETES MELLITUS

Up to now, there are many kinds of chemical or biochemical antihyperglycemic agents that have been developed for patients to control or/and lower blood glucose to a normal level (Croom, 1983). Even though, there is a lot of controversy concerning about the safety and efficacy of herbal remedies, however, many people are using traditional and complementary medicine to treat diabetes and other diseases, (Fugh-Berman, 2000).

It must be noted that a big portion of the population has been using traditional herbal medicinal plants which have antidiabetic properties and a variety of compounds have been isolated (alkaloids, glycosides, terpenes, flavonoids, etc.). However, to develop a traditional herbal remedy as a useful modern medicine, still we need to more precise in investigating and standardizing the crude or isolated compound. To date, a biguanide is a derivative of an active natural product which isolated from the plant *Galega officinalis L*. (Witters, 2001).

Secondary metabolites are organic compounds that are not directly involved in the normal growth, development, or reproduction of an organism. Secondary metabolites often play an important role in plant defense against herbivory and other interspecies defenses. Humans use secondary metabolites as medicines, flavorings, and recreational drugs.

Generally, most of the active compounds isolated from plants as a secondary metabolites like alkaloids, flavonoids, triterpenoids, polysaccharides, glycopeptides, aminobutyric acid derivatives, steroids, iridoids, phenolics, peptides, alkyldisulfides and inorganic ions.

There have been a rich historical record from an ancient physicians and aborigine people that traditional and complementary medicine provide important clues for identifying and developing synergistic activity of selected crude extract but it has largely neglected. Therefore, substantial effort is needed to better screening the antidiabetic mechanism of action of most plant extracts.

2.3 THE PHALERIA MACROCARPA (SCHEFF.) BOERL.

P. macrocarpa (Scheff.) Boerl. known as Mahkota Dewa grows all over Malaysia. It grows in tropical areas throughout the year and reaches a height of around 1-6 m. It is a complete tree (stem, leaves, flower and fruit) and the fruit shape is eclipsed with a diameter of around 3 cm. The color of the fruit is green before ripening and red when fully ripe. The plant can be found planted in home gardens as an ornamental plant in gardens or as a shade plant. This chronic shrub grows upright with 1 to 2.5 m tall, round and trunk, It is rough, brown, woody and gummy, branching sigmoidal. The single leaf located opposite, short-stemmed, lancelets or oblong shape, tapered tip and base, flat edge, pinnate venation, smooth, dark green, 7-10 cm long, 2-5 cm wide. Flowers come out throughout the year, lying scattered on the stems or leaves armpits, tube shape, small, white, and fragrant. Fruits are round, 3-5 cm in diameter, smooth, grooved, when young, green and red color after ripening. The pulp is white, fibrous and watery. Seeds round, hard, brown, rooted riding and yellow-brown (Backer et al., 1965; Hendra et al., 2011).