

**HYPOGLYCEMIC AND ANTIOXIDATIVE EFFECTS OF
ANACARDIUM OCCIDENTALE LINN. IN DIABETIC RATS**

LETTY LING

**MASTER OF SCIENCE
UNIVERSITI PUTRA MALAYSIA**

2006

DEDICATION

Especially dedicated to.....

Almighty God,

My dearest daddy, Ling Kuok Miew and mummy, Tang Swei Ting

My brother, Han Lee and sister, Eileen

My late younger sister, Pyleen

My beloved husband, Tang Tung Kwong

Brothers and sister in Christ

Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfilment of the requirement for the degree of Master of Science

**HYPOGLYCEMIC AND ANTIOXIDATIVE EFFECTS OF
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By

LETTY LING

April 2006

Chairman : Zulkhairi Haji Amom, PhD

Faculty : Medicine and Health Sciences

Diabetes mellitus is found to be associated with oxidative damage which coexists with a reduction in the antioxidant status. The Malay folklore medicine in Malaysia believes that by consuming the decoction of the vein and leaves (vascular bundle) of *A. occidentale* L. (cashew-nut), it is able to lower blood glucose level of diabetic patients. The objective of this study was to verify the potential hypoglycemic and antioxidative effects of *Anacardium occidentale* L. leaves aqueous extract (AOE) in Type 2 diabetes rat model. Freeze-dried AOE of various doses (50 mg/kg, 250 mg/kg, 500 mg/kg and 1000 mg/kg body weight) were administered to streptozotocin induced Type 2 diabetic rats. The rats were force-fed with the extracts once daily for six weeks. Oral glucose tolerance test (OGTT) with 1.5 g/kg body weight of glucose challenge was then conducted to monitor the serum glucose level. Blood was collected through cardiac puncture to examine the levels of lipid peroxidation and the enzymatic activities in the experimental rats. The result showed an improvement in the glucose tolerance after six weeks of treatment significantly as compared to the diabetes control ($P < 0.05$). After treatment, the rats treated with AOE at all doses

have lower fasting glucose levels compared to the pre-treatment week. It was also noticed that all the doses of the leaves extract managed to delay the rise of glucose level in oral glucose tolerance curve. Thus, *A. occidentale* leaves aqueous extract had suppression effect on the increase of serum glucose levels in oral glucose load.

The AOE at all doses exhibited low lipid peroxidation product indicated by serum malondialdehyde (MDA) levels after six weeks of extract administration as compared to the pre-treatment week. The reduced MDA levels of group treated with AOE 250mg/kg, AOE 500 mg/kg and AOE 1000mg/kg are comparable to the MDA levels as obtained by the normal control groups after six weeks of extract treatment.

Serum catalase activities were found to be significantly elevated in diabetic groups treated with AOE as compared to the diabetic control groups whereas in the normal groups, the serum catalase activities in blood were much higher than the diabetic groups. Plasma superoxide dismutase (SOD) activities of AOE treated were found to be higher than the diabetes control after three weeks of administration. The SOD activities were higher than the normal groups significantly ($P < 0.05$). However, there were reductions on SOD activities at Week 6. Diabetes treated groups (AOE 50 mg/kg, AOE 250 mg/kg and AOE 500 mg/kg) showed an increased of plasma glutathione peroxidase (GPx) activities at Week 3 as compared to the pre-treatment groups.

Histological study of the pancreas showed an extensive damage of the islets of Langerhans and reduced dimensions of islet in the diabetic-induced rats. There were significant increase in the area, perimeter and diameter of pancreatic islets in both

glybenclamide and AOE treated rats. The diabetic rats treated with AOE 250 mg/kg have the highest increase in area, perimeter and diameter of the islet of Langerhans and have no significant difference compared to the normal control rats. This may suggest that AOE 250 mg/kg could improve and protect the islet Langerhans cells from oxidative degeneration resembling the normal rats.

The results of this study indicate that *A. occidentale* L. leaves might possess hypoglycemic activity. The alterations of the enzymatic antioxidant activities in the experimental animals provides evidence that the preventive effects of *A. occidentale* L. may be due to inhibition of lipid peroxidation by its antioxidant properties. Thus, *A. occidentale* L. possesses antioxidant properties which counteract the oxidative damage in diabetic subjects.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan untuk ijazah Master Sains

**KESAN HIPOGLISEMIK DAN ANTIOKSIDATIF
ANACARDIUM OCCIDENTALE LINN. PADA TIKUS DIABETIK**

Oleh

LETTY LING

April 2006

Pengerusi : Zulhairi Haji Amom, PhD

Fakulti : Perubatan dan Sains Kesihatan

Penyakit diabetes mellitus telah dikatakan berhubung kait dengan kemusnahan oksidatif yang diakibatkan oleh pengurangan status antioksidan. Masyarakat Melayu di Malaysia mempercayai perubatan traditional di mana penggunaan daun *A. occidentale* L.(gajus) berupaya menurunkan kandungan aras glukosa darah pada pesakit diabetes. Kajian ini bertujuan untuk menentukan keberkesanan ekstrak akuas daun *A. occidentale* L. (AOE) dalam menurunkan kandungan aras glukosa serum darah dan kesan antioksidannya pada model tikus teraruh diabetes jenis 2. Ekstrak daun berakuas yang telah disejukbekukan telah diberikan dos (50 mg/kg, 250 mg/kg, 500 mg/kg dan 1000 mg/kg berat badan) kepada tikus teraruh diabetes jenis 2 melalui suntikan streptozotosin. Tikus telah diberikan AOE melalui oral setiap hari sekali, selama enam minggu. Ujian toleransi glukosa secara oral (OGTT) dengan “cabaran glukosa” sebanyak 1.5 g/kg berat badan telah dijalankan untuk menentukan kesan ekstrak terhadap kandungan glukosa serum darah. Darah juga diambil melalui tusukan jantung untuk menguji aras oksidasi lipid dan aktiviti enzim antioksidan

pada tikus kajian. Keputusan daripada kajian ini telah menunjukkan dos rawatan AOE berjaya menurunkan aras glukosa darah tikus diabetik selepas enam minggu tempoh rawatan berbanding dengan kumpulan kawalan diabetes melalui ujian toleransi glukosa. Kumpulan tikus yang diberikan rawatan AOE pada semua dos menunjukkan aras glukosa serum darah yang lebih rendah sebelum diberikan “cabaran glukosa” berbanding dengan tempoh sebelum rawatan diberikan. Kajian ini mendapati bahawa AOE pada semua dos berupaya merencatkan peningkatan aras glukosa serum darah setelah diberikan “cabaran glukosa” dalam Ujian toleransi glukosa. Ekstrak akuas daun *A. occidentale* ini dicadangkan telah menunjukkan penindasan terhadap peningkatan glukosa dalam Ujian toleransi glukosa.

Tikus yang diberikan AOE pelbagai dos telah menunjukkan kandungan oksidasi lipid, iaitu aras malondialdehid (MDA) yang lebih rendah berbanding dengan sebelum rawatan diberikan selepas enam minggu tempoh rawatan. Kandungan MDA yang diperolehi daripada kumpulan rawatan AOE 250 mg/kg, AOE 500 mg/kg, dan AOE 1000 mg/kg selepas enam minggu rawatan dijalankan adalah setara seperti yang diperolehi daripada kumpulan kawalan normal.

Aktiviti katalase pada tikus diabetik yang diberikan rawatan AOE telah meningkat berbanding dengan kumpulan kawalan diabetik. Manakala, aktiviti katalase pada kumpulan normal adalah sentiasa lebih tinggi daripada kumpulan diabetik. Tikus yang diberikan rawatan AOE juga menunjukkan aktiviti superoksida dismutas (SOD) yang lebih tinggi berbanding dengan kumpulan kawalan diabetik selepas tiga minggu rawatan dijalankan dan lebih tinggi daripada kumpulan normal secara ketara ($P < 0.05$). Kumpulan diberikan dos rawatan AOE 50 mg/kg, AOE 250 mg/kg and

AOE 500 mg/kg telah menunjukkan peningkatan aktiviti glutathion peroksidases (GPx) pada minggu ke-3 berbanding dengan minggu sebelum diberikan rawatan.

Pemerhatian histologi pada pankreas yang dikaji telah menunjukkan kemusnahan pada pulau Langerhans dan dimensi pulau Langerhans telah berkurangan pada tikus diabetes yang disuntik streptozotosin (STZ). Luas permukaan, perimeter dan panjang diameter pada pulau Langerhans telah menunjukkan peningkatan yang ketara pada kumpulan tikus yang diberikan rawatan glibenklamida dan AOE. Tikus yang diberikan rawatan AOE 250 mg/kg telah menunjukkan peningkatan luas permukaan, perimeter dan panjang diameter pulau Langerhans yang tertinggi antara tikus yang diberikan dos rawatan yang lain ($P < 0.05$). Peningkatan luas permukaan, perimeter dan panjang diameter pada tikus yang diberikan rawatan AOE 250 mg/kg tiada perbezaan ketara berbanding dengan tikus kawalan. Ini menunjukkan dos AOE 250 mg/kg mungkin mampu memperbaiki dan melindungi sel pada pulau Langerhans seperti yang ditunjukkan pada tikus kumpulan kawalan.

Kesimpulannya, kajian ini telah mendapati ekstrak akuas daun *A. occidentale* L. mungkin mempunyai aktiviti hipoglisemik dan antioksidan. Kesan antioksidannya boleh dilihat melalui perubahan aktiviti antioksidan yang diperolehi pada haiwan dalam kajian ini. Perencatan oksidasi lipid pada haiwan eksperimen berkemungkinan disebabkan oleh aktiviti antioksidan yang bertindak semasa berlakunya kemusnahan oksidatif di dalam subjek diabetik.

ACKNOWLEDGEMENTS

My sincere praises and thanksgiving is accorded to Almighty God for His unfailing loves and endless grace. Almighty Lord has provided me with the strength, wisdom and guidance to finish and complete this master study.

I wish to express my deepest appreciation and gratitude to my supervisor, Dr. Zulkhairi Hj. Amom for his invaluable guidance, constant encouragement and constructive suggestions throughout the study. Thanks are also extended to the members of my supervisory committee, Professor Dr. Hamdan Hj. Mohd. Noor and Dr. Amin Ismail for their guidance, advice, and motivation throughout my study. Thanks to IRPA Grants: 54422 which supported the research and study.

Heartfelt appreciation is also due to Dr. Che Norma Mat Taib, Mr. Ramli, Mr. Nordin, Mr. Rizal, Mr. Rahman, Mdm. Siti, Mdm. Safarina, Mdm. Normah, Mdm. Juita, Miss Noridah, Miss Farhatani, Miss Hasnah, and Miss Husni for their kindly assistance, co-operation, and technical support during the period of this study.

My warmest gratitude to Pastor Elaine Goh, brothers and sisters in Christ and all my friends who helped ensure the success of this project and their endless prayers and support.

Last but not least, I wish to convey my gratitude to my family members and my dearest husband for their enormous amount of loves, understandings, sacrifices and steadfast support that have made the task of completing this master project possible.

With all of my love, thank you very much. May God bless each one of you abundantly.

Amen.

Trust in the Lord with all your heart and lean not on your own understanding;

In all your ways acknowledge him, and he will make your paths straight.

Proverbs 3: 5-6

THANK YOU

I certify that an Examination Committee has met on 12 April 2006 to conduct the final examination of Letty Ling on her Master of Science thesis entitled “Hypoglycemic and Antioxidative Effects of *Anacardium occidentale* Linn. in Diabetic Rats” in accordance with Universiti Pertanian Malaysia (Higher Degree) Act 1980 and Universiti Pertanian Malaysia (Higher Degree) Regulations 1981. The Committee recommends that the candidate be awarded the relevant degree. Members of the Examination Committee are as follows:

Abdul Manan Mat Jais, PhD

Associate Professor
Faculty of Medicine and Health Sciences
Universiti Putra Malaysia
(Chairman)

Norhaizan Mohd. Esa, PhD

Lecturer
Faculty of Medicine and Health Sciences
Universiti Putra Malaysia
(Internal Examiner)

Mohamad Taufik Hidayat, PhD

Lecturer
Faculty of Medicine and Health Sciences
Universiti Putra Malaysia
(Internal Examiner)

Jamaludin Mohamed, PhD

Professor
Faculty of Allied Health Sciences
Universiti Kebangsaan Malaysia
(External Examiner)

HASANAHT MOHD. GHAZALI, PhD

Professor/Deputy Dean
School of Graduate Studies
Universiti Putra Malaysia

Date:

This thesis submitted to the Senate of Universiti Putra Malaysia and has been accepted as fulfilment of the requirement for the degree of Master of Science. The members of the Supervisory Committee are as follows:

Zulkhairi Haji Amom, PhD

Lecturer
Faculty of Medicine and Health Sciences
Universiti Putra Malaysia
(Chairman)

Hamdan Hj. Mohd. Noor, PhD

Professor
Faculty of Medicine
Cyberjaya University College of Medical Sciences
(Member)

AMIN ISMAIL, PhD

Lecturer
Faculty of Medicine and Health Sciences
Universiti Putra Malaysia
(Member)

AINI IDERIS, PhD
Professor/Dean
School of Graduate Studies
Universiti Putra Malaysia

Date:

DECLARATION

I hereby declare that the thesis is based on my original work except for quotations and citations which have been duly acknowledged. I also declare that it has not been previously or concurrently submitted for any other degree at UPM or other institutions.

LETTY LING

Date:

TABLE OF CONTENTS

	Page
DEDICATION	ii
ABSTRACT	iii
ABSTRAK	vi
ACKNOWLEDGEMENTS	ix
APPROVAL	xi
DECLARATION	xiii
LIST OF TABLES	xvii
LIST OF FIGURES	xviii
LIST OF ABBREVIATIONS	xxii
CHAPTER	
1 INTRODUCTION	1
1.1 Diabetes Mellitus — Current Perspective	1
1.2 Objectives of the Study	6
1.3 Research Hypothesis	7
2 LITERATURE REVIEW	8
2.1 General Energy Intake, Utilization and Storage	8
2.1.1 Carbohydrates	8
2.1.2 Proteins	12
2.1.3 Lipids	13
2.2 Absorptive and Post-absorptive State	13
2.2.1 Metabolisme and Regulation during the Absorptive State	14
2.2.2 Metabolisme and Regulation during the Post-Absorptive State	17
2.3 Blood Glucose Regulation	20
2.3.1 Homeostasis	20
2.3.2 Pancreas	20
2.3.3 Insulin and Regulation of Blood Glucose Levels	23
2.3.4 Glucagon and Regulation of Blood Glucose Levels	28
2.4 Diabetes Mellitus – Clinical Aspect	30
2.4.1 Concept of Diabetes Mellitus: Definition and Description	30
2.4.2 Classification of Diabetes Mellitus	31
2.4.3 Diagnosis of Diabetes Mellitus	37
2.5 Management and Modern Therapy in Diabetes Mellitus	39
2.5.1 Insulin Therapy	39
2.5.2 Pancreas Transplants	41
2.5.3 Oral Hypoglycemic Agents (OHAs)	42
2.5.4 Education, Exercise, and Diet or Medical Nutritional Therapy (MNT)	47

2.6	Free radical, Oxidative Stress and Antioxidant Defense System	48
2.6.1	Free Radicals and Reactive Oxygen Species	48
2.6.2	Oxidative Stress	50
2.6.3	Antioxidant Defense System	52
2.7	Lipid Peroxidation	56
2.8	Enzymatic Antioxidants Activities and Lipid Peroxidation in Diabetes Mellitus	60
2.9	Plants as Treatment for Diabetes Mellitus	63
2.9.1	Plants with Hypoglycemic or Antioxidative Properties	64
2.9.2	<i>Anacardium occidentale</i> Linn.: A Plant of Many Uses	72
3	MATERIALS AND METHODS	76
3.1	Chemical Reagents	76
3.2	Experimental Design	77
3.3	Plant Materials	79
3.4	Animals Study	80
3.4.1	Mating and Breeding	81
3.4.2	Induction of Experimental Diabetic Rats	81
3.5	Experimental Treatments	83
3.6	Sampling Procedure and Data Collection	83
3.6.1	Tail Vein Blood Sampling for Blood Glucose Levels	83
3.6.2	Cardiac Puncture Blood Sampling for Antioxidative Effects	84
3.6.3	Body Weight Record	84
3.7	Estimation of Blood Glucose Levels	84
3.7.1	Anaesthesia	84
3.7.2	Preparation of Treatment and Glucose Load	85
3.7.3	Blood Collection	85
3.7.4	Oral Administration	86
3.7.5	Serum Glucose Estimation	86
3.8	Lipid Peroxidation and Enzymatic Activities	89
3.8.1	Estimation of Malondialdehyde (MDA) Level	89
3.8.2	Estimation of Catalase Specific Activity (CAT)	91
3.8.3	Estimation of Protein Concentration (Biuret Method)	92
3.8.4	Superoxide Dismutase Activity (SOD)	93
3.8.5	Glutathione Peroxidase Activity (GPx)	95
3.9	Histological Study	96
3.10	Statistical Analysis	99
4	RESULTS	100
4.1	Induction of Experimental Diabetic Rats	100
4.2	Effect of <i>A. occidentale</i> L. Leaves Extract (AOE) on Serum Glucose Levels	107
4.2.1	Effect of <i>A. occidentale</i> L. Leaves Extract (AOE) on Serum Glucose Levels in Normal	107

	Groups	
4.2.2	Effect of <i>A. occidentale</i> L. Leaves Extract (AOE) on Serum Glucose Levels in Diabetic Groups	117
4.3	Body Weight	122
4.4	Lipid Peroxidation Index, Malondialdehyde (MDA)	124
4.5	Catalase Specific Activity (CAT)	126
4.6	Superoxide Dismutase Activity (SOD)	128
4.7	Glutathione Peroxidase Activity (GPx)	130
4.8	Histological Studies of the Pancreas	132
5	DISCUSSION	139
5.1	Induction of Experimental Diabetic Rats	139
5.2	Effect of <i>A. occidentale</i> L. Leaves Extract on Serum Glucose Levels	143
5.3	Body Weight	149
5.4	Lipid Peroxidation Index, Malondialdehyde (MDA)	149
5.5	Catalase Specific Activity (CAT)	153
5.6	Superoxide Dismutase Activity (SOD)	154
5.7	Glutathione Peroxidase Activity (GPx)	156
5.8	Histological Studies of the Pancreas	157
6	GENERAL DISCUSSION	160
7	CONCLUSION	163
8	SUGGESTIONS FOR FUTURE WORK	165
	BIBLIOGRAPHY	167
	APPENDICES	182
	BIODATA OF THE AUTHOR	197

