



UNIVERSITI PUTRA MALAYSIA

**BENEFICIAL PROPERTIES OF *Garcinia atroviridis* Griff.
FRUIT AND LEAF AQUEOUS EXTRACT IN EXPERIMENTAL
ATHEROSCLEROSIS IN RABBITS**

NURSAKINAH BINTI ISEMAAIL

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RABBITS**



By

NURSAKINAH BINTI ISEMAAIL

**Thesis Submitted to the School of Graduate Studies, Universiti Putra Malaysia, in
Fulfilment of the Requirements for the Degree of Master of Science**

June 2012

DEDICATION

I would love to dedicate this accomplishment to my lovely parents, Isemail bin Basri and Fatimah binti Ibrahim; to my precious siblings, Nizamuddin, Nurfadhilah, Najmi and Nurnaimah; to my much beloved husband, Muhammad Nor Fazali bin Fazil; and last but certainly not least to my dear lifelong best friend, Hasnah binti Bahari.



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

BENEFICIAL PROPERTIES OF *Garcinia atroviridis* Griff. FRUIT AND LEAF AQUEOUS EXTRACT IN EXPERIMENTAL ATHEROSCLEROSIS IN RABBITS

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Chairman: Assoc. Prof. Zulkhairi Amom, PhD

Faculty: Medicine and Health Sciences

Numerous plants were proven beneficial in preventing atherosclerosis. The plant *Garcinia atroviridis*, locally known as asam gelugur had been traditionally utilized for various medicinal purpose. The present study aimed to evaluate the potential benefit of *G.atroviridis* fruit and leaf aqueous extracts supplementation by assessing the lipid profiles, antioxidant and liver enzymes, and atherosclerosis development in atherosclerosis-induced rabbits. The fruit and leaf samples were collected fresh from parts of Kedah, Malaysia; cut into pieces, dried in an oven at 30°C for 2 days and powdered. Then, 100 g of fruits and leaves powder were soaked separately in 1000 ml distilled water, incubated in water bath at three temperatures and time settings (40°C for 12 hours; 60°C for 6 hours; 100°C for 15 min) to optimize their antioxidant ability, filtered and spray dried. The antioxidant activity was evaluated using the 1,1-diphenyl-2-picrylhydrazyl (DPPH) scavenging and Ferric Reducing Antioxidant Power assay. Total phenolic content was measured by the Folin Ciocalteu method. The leaf extract (89.2%)

had higher DPPH scavenging activity than vitamin C (88.8%) ($p \geq 0.05$) and almost comparable to synthetic antioxidant butylated hydroxytoluene (94.6%). The fruit and leaf extracts prepared at 60°C for 6 hours, and 100°C for 15 min, respectively offered the highest antioxidant activity and phenolic contents, and therefore were supplemented to the rabbits. The extracts were force-fed once daily for 12 weeks at the dose of 50 mg/kg (F50), 100 mg/kg (F100) and 200 mg/kg (F200) for fruits; and 5 mg/kg (L5), 55 mg/kg (L55) and 105 mg/kg (L105) for leaves. Three control groups were excluded from the extract supplementation. All rabbits were fed a 0.5% cholesterol enriched diet, except the normal control group. The simvastatin control group was prescribed 10 mg/kg simvastatin while the positive control group was without any supplementation. Blood samples were collected via marginal ear vein at the beginning and every 4 weeks. As the experiment end, the leaf (L105) and fruit (F50, F100 and F200) supplementations significantly ($p < 0.05$) retained the enzyme superoxide dismutase at its initial level. All extracts supplementations significantly reduced ($p < 0.05$) lipid profiles (total cholesterol, triglycerides and low density lipoprotein) and liver enzymes (alkaline phosphatase, gamma-glutamyl transpeptidase and aspartate aminotransferase). Both extracts had effectively prevented liver damage and atherosclerotic plaque formation with at least 25% lower atherogenic index than the positive control group. Hence, this study signifies the potential benefits of *G. atroviridis* fruit and leaf extracts as antioxidants, hypocholesterolemic, hepatoprotective, and anti-atherosclerotic agents.

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

FAEDAH KANDUNGAN EKSTRAK AIR BUAH DAN DAUN *Garcinia atroviridis* Griff DALAM EKSPERIMEN ATEROSKLEROSIS DALAM ARNAB

Oleh

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Jun 2012

Pengerusi: Prof. Madya Zulhairi Amom, PhD

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Pelbagai tumbuhan telah terbukti berkesan mengurangkan risiko penyakit aterosklerosis. Tumbuhan *Garcinia atroviridis*, dikenali sebagai asam gelugur telah digunakan secara tradisi untuk pelbagai tujuan kesihatan. Kajian ini bertujuan menilai potensi faedah pengambilan ekstrak buah dan daun *G. atroviridis* sebagai makanan tambahan, dengan mengkaji profil lemak, enzim antioksidan dan hati, serta pertumbuhan aterosklerosis dalam arnab yang dirangsang ke arah atherosklerosis. Sampel buah dan daun segar dikumpul dari beberapa kawasan di Kedah, Malaysia; dipotong, dikeringkan di dalam ketuhar pada suhu 30°C selama 2 hari dan dihancurkan. Kemudian, 100 g serbuk buah dan daun direndam secara berasingan dalam 1000 ml air suling, dieram dalam pengukus pada tiga tetapan suhu dan masa (40°C selama 12jam; 60°C selama 6jam; 100°C selama 15 minit) bagi mengoptimalkan antioksidannya, ditapis dan dikeringkan secara semburan. Aktiviti antioksidan diukur menggunakan ujian perambatan 1,1-diphenyl-2-picrylhydrazyl (DPPH) dan asai kemampuan menurunkan ferum (FRAP). Jumlah kandungan fenolik diukur

melalui kaedah Folin Ciocalteu. Aktiviti antioksidan ekstrak daun (89.2%) adalah lebih tinggi daripada vitamin C (88.8%) ($p \geq 0.05$) dan hampir sama dengan antioksidan tiruan *butylated hydroxytoluene* (94.6%). Ekstrak buah yang disediakan pada suhu 60°C dalam tempoh penderaman 6 jam, serta ekstrak daun yang disediakan pada 100°C dalam 15 minit mempunyai aktiviti antioksidan dan kandungan fenolik paling tinggi, maka ia diberikan sebagai makanan tambahan kepada arnab. Ekstrak tersebut disuapkan secara paksa sekali sehari selama 12 minggu pada dos 50 mg/kg (F50), 100 mg/kg (F100) dan 200 mg/kg (F200) bagi buah; 5 mg/kg (L5), 55 mg/kg (L55) dan 105 mg/kg (L105) bagi daun. Tiga kumpulan kawalan telah dikecualikan dari pemberian ekstrak. Semua arnab diberi makanan yang diperkaya 0.5% kolesterol, kecuali kumpulan kawalan normal. Kumpulan kawalan simvastatin (SC) telah diberi 10 mg/kg simvastatin manakala kumpulan kawalan positif tidak diberi apa-apa rawatan. Sampel darah dikumpul melalui salur vena telinga pada permulaan dan setiap 4 minggu. Setelah eksperimen berakhir, didapati pemberian daun (L105) dan buah (F50, F100 and F200) telah mengekalkan enzim *superoxide dismutase* secara signifikan ($p < 0.05$) di tahap asal. Semua pemberian ekstrak telah menurunkan profil lemak (jumlah kolesterol, trigliserida dan lipoprotein ketumpatan rendah) serta enzim hati (*alkaline phosphatase*, *gamma-glutamyl transpeptidase* dan *aspartate aminotransferase*) secara signifikan ($p < 0.05$). Kedua-dua ekstrak itu juga telah berkesan menghalang kerosakan hati dan pembentukan plak aterosklerosis, dengan index aterosklerosis sekurang-kurangnya 25% lebih rendah dari kumpulan kawalan positif. Maka, kajian ini telah menunjukkan potensi faedah buah dan daun *G.atroviridis* sebagai antioksidan, agen pengawal kolesterol, pelindung organ hati dan anti-aterosklerosis.

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APPROVAL

I certify that an Examination Committee has met on 18th of June 2012 to conduct the final examination of **Nursakinah binti Isemaail** on her Master of Science thesis entitled "**Beneficial Properties of *Garcinia atroviridis* Griff Fruit and Leaf Aqueous Extract in Experimental Atherosclerosis in Rabbits**" in accordance with Universiti Pertanian Malaysia (Higher Degree) Act 1980 and Universiti Pertanian Malaysia (Higher Degree) Regulations 1981. The Committee recommends that the student be awarded the Master of Science.

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DECLARATION

I declare that the thesis is my original work except for quotations and citations which have been duly acknowledged. I also declare that it has not been previously, and is not concurrently, submitted for any other degree at Universiti Putra Malaysia or at any other institution.



NURSAKINAH BINTI ISEMAAIL

Date: 18 June 2012

TABLE OF CONTENTS

	Page
DEDICATION	ii
ABSTRACT	iii
ABSTRAK	v
ACKNOWLEDGEMENT	vii
APPROVAL	viii
DECLARATION	x
LIST OF TABLES	xv
LIST OF APPENDICES	xvi
LIST OF FIGURES	xvii
LIST OF ABBREVIATIONS	xx
CHAPTER	
1 INTRODUCTION	1
2 LITERATURE REVIEW	
Introduction	6
Garcinia Atroviridis	7
Atherosclerosis	9
Lipids, cholesterol and lipoproteins	14
Cholesterol Uptake	15
Lipids Exogenous Pathway	17
Lipids Endogenous Pathway	19
Low Density Lipoprotein	19
High Density Lipoprotein	21
Hypercholesterolemia	22
Free radicals and oxidation	23
Low Density Lipoprotein Oxidation	24
Lipid Peroxidation	26
Antioxidants	30
Endogenous Antioxidant	31
Exogenous Antioxidant	33
Phenolic Compounds	34
Flavonoids	36
Experimental Research on Atherosclerosis	38
Conclusion	40

3	GENERAL MATERIALS AND METHODS	
	Equipments	41
	Plant Materials and Extraction	41
	Experimental Animal Procedures	43
4	NUTRITIONAL COMPOSITION AND <i>IN VITRO</i> ANTIOXIDANT ACTIVITY OF <i>Garcinia atroviridis</i> FRUITS AND LEAVES	
	Introduction	45
	Materials and Methods	47
	Chemicals and Reagents	47
	Proximate and Mineral Contents	47
	Plant Materials and Extraction	47
	DPPH radical scavenging	48
	Ferric Reducing Antioxidant Power	49
	Total phenolic content	49
	Statistical Analysis	50
	Results	
	Proximate and Mineral Contents	51
	DPPH radical scavenging	53
	Ferric Reducing Antioxidant Power	55
	Total phenolic content	57
	Discussion and Conclusion	59
5	<i>IN VIVO</i> ANTIOXIDANT STATUS OF RABBITS FED A CHOLESTEROL-ENRICHED DIET SUPPLEMENTED WITH <i>Garcinia atroviridis</i> FRUIT AND LEAF AQUEOUS EXTRACT	
	Introduction	64
	Materials and Methods	67
	Commercial Kits and Equipments	67
	Plant Materials and Extraction	67
	Experimental Animal Procedures	67
	Superoxide Dismutase (SOD)	68
	Glutathione Peroxidase (GPx)	68
	Total Antioxidant Activity (TAS)	68
	Statistical Analysis	69
	Results	
	Superoxide Dismutase (SOD)	70
	Glutathione Peroxidase (GPx)	73
	Total Antioxidant Activity (TAS)	76
	Discussion and conclusion	79

6	LIPID PROFILES OF RABBITS FED A CHOLESTEROL-ENRICHED DIET SUPPLEMENTED WITH <i>Garcinia atroviridis</i> FRUIT AND LEAF AQUEOUS EXTRACT	
	Introduction	84
	Materials and Methods	86
	Commercial Kits and Equipments	86
	Plant Materials and Extraction	86
	Experimental Animal Procedures	86
	Total Cholesterol	87
	Low Density Lipoprotein	88
	High Density Lipoprotein	89
	Tryglycerides	90
	Atherogenic Index (AI)	91
	Statistical Analysis	91
	Results	
	Total Cholesterol	92
	Low Density Lipoprotein	95
	High Density Lipoprotein	98
	Tryglycerides	101
	Atherogenic Index (AI)	104
	Discussion and conclusion	107
7	LIVER FUNCTION STATUS OF RABBITS FED A CHOLESTEROL-ENRICHED DIET SUPPLEMENTED WITH <i>Garcinia atroviridis</i> FRUIT AND LEAF AQUEOUS EXTRACTS	
	Introduction	111
	Materials and Methods	
	Commercial Kits and Equipments	113
	Plant Materials and Extraction	113
	Experimental Animal Procedures	113
	Alkaline phosphatase (ALP)	114
	Alanine aminotransferase (ALT)	114
	Aspartate aminotransferase (AST)	115
	Gamma-glutamyl transpeptidase (GGT)	115
	Statistical Analysis	116
	Results	
	Alkaline phosphatase (ALP)	117
	Alanine aminotransferase (ALT)	120
	Aspartate aminotransferase (AST)	123
	Gamma-glutamyl transpeptidase (GGT)	126
	Discussion and conclusion	129

8	EFFECTS OF SUPPLEMENTING <i>Garcinia atroviridis</i> FRUIT AND LEAF AQUEOUS EXTRACTS ON ATHEROSCLEROSIS PROGRESSION AND LIVER HISTOLOGY	
	Introduction	133
	Materials and Methods	
	Commercial Kits and Equipments	135
	Plant Materials and Extraction	135
	Experimental Animal Procedures	135
	Atherosclerotic Plaques	136
	Histology of Aorta and Liver	137
	Results	
	Atherosclerotic Plaques	138
	Histology of Aorta	141
	Histology of Liver	143
	Discussion and Conclusion	145
9	GENERAL DISCUSSION	
	General discussion	148
10	CONCLUSION	
	Conclusion	152
	Recommendations for Future Research	154
	REFERENCES	156
	APPENDICES	177
	BIODATA OF STUDENT	180