تقييم تعدد أشكال مستقبلات النوع الثانى للانجيوتنسن وعلاقتها بشدة أمراض الشرايين التاجية

رسالة

توطئة للحصول على درجة الماجستير في

الكيمياء الحيوية الطبية و البيولوچيا الجزيئية

مقدمة من

الطبيبة/ سهام عادل بدوى

بكالوريوس الطب والجراحة

تحت إشراف

الأستاذ الدكتور/ هانى يوسف حليم

أستاذ الكيمياء الحيوية الطبية والبيولوچيا الجزيئية

كلية الطب - جامعة عين شمس

الدكتورة/ منال لويس لوقا

مدرس الكيمياء الحيوية الطبية والبيولوچيا الجزيئية

كلية الطب - جامعة عين شمس

الدكتور/ وائل محمود الكيلانى

أستاذ مساعد أمراض القلب

كلية الطب - جامعة عين شمس

كلية الطب

جامعة عين شمس

٢٠١٣

Evaluation of AngiotensinII receptor type 2 Polymorphism and its relation to the severity of Coronary Artery Diseases

Thesis

Submitted for Partial Fulfillment of Master Degree
In Medical Biochemistry and Molecular Biology

By

Seham Adel Badawy
M.B.B.Ch.
Faculty of Medicine
Ain Shams University

Under Supervision of

Prof. Hany Youssef Halim
Professor of Medical Biochemistry and Molecular Biology
Faculty of Medicine – Ain Shams University

Dr. Manal Louis Louka
Lecturer of Medical Biochemistry and Molecular Biology
Faculty of Medicine – Ain Shams University

Dr. Wael Mahmoud EL Kilany
Assistant Professor of Cardiology
Faculty of Medicine – Ain Shams University

Faculty of Medicine
Ain Shams University
2013

List of Tables

List of Figures

List of Abbreviations

ACE	Angiotensin converting enzyme
ACE2	Angiotensin converting enzyme 2
ADP	Adenosine diphosphate
AGT	Angiotensinogen
AMI	Acute myocardial infarction
Ang (1–7)	Angiotensin(1–7)
Ang I	Angiotensin I
Ang II	Angiotensin II
ARB	Angiotensin-receptor blocker
AT1R	Angiotensin II Type one Receptor
AT2R	Angiotensin II Type Two Receptor
B2Rs	Bradykinin 2 receptors
BLAST	Basic Local Alignment Search Tool
CAD	Coronary artery Disease
cGMP	Cyclic guanosine monophosphate
CHD	Coronary heart disease
CHF	Congestive heart failure
CIs	Confidence intervals
CK MB	Creatine Kinase-MB
COX	Cyclooxygenase
CRP	C- reactive protein
dATP	Deoxyadenosine triphosphate
dCTP	Deoxycytidine triphosphate
dGTP	Deoxyguanosinetriphosphate
DHB	Dihydroxy Bergamottin
DNA	Deoxyribonucleic acid

List of Abbreviations *(Cont…)*	
DNP	Double nucleotide polymorphisms
Dttp	Deoxythiamin triphosphate
ECG	Electrocardiography
EDTA	Ethylene Diamine Tetraacetic acid
EGFR	Epidermal growth factor receptor
ERK	Extracellular signal regulated kinases
GF	Growth Factor
GPIIb/IIIa	Glycoprotein llb/llla
HDL	High-density lipoprotein
ICAM	Intercellular adhesion molecule
IR	Insulin receptor
IRS	Insulin receptor substrates
JAKs	Janus kinases
KDa	Kilo Dalton
LBBB	Left bundle branch block
LDL	Low-density lipoprotein
LVH	Left ventricular hypertrophiy
MAP	Mitogen-activated protein
MCP-1	Monocyte chemoattractant protein-1
MI	Myocardial Infarction
MNPs	Multi-nucleotide polymorphisms
NAD	Nicotinamide adenine dinucleotide
NAD (P)	Nicotinamide adenine dinucleotide Phosphate
NO/cGMP	Nitric oxide/cyclic guanosine monophosphate
Ors	Odds ratios
PI3K	Phosphatidylinositol-3 kinase

List of Abbreviations *(Cont…)*	
PKC	Protein kinase C
PLA	Phospholipase A
PLD	Phospholipase D
PRR	Prorenin receptor
RAS	Renin-angiotensin system
RFLP	Restriction fragment length polymorphism
ROS	Reactive oxygen species
RVLM	Rostral ventrolateral medulla
SNPs	Single nucleotide polymorphisms
STEMI	S-T elevation myocardial infarction
TBE	Tris-borate EDTA
TG	Triglycerides
TGF	Tumor Growth Factor
TNF	Tumor necrosis factor
TNPs	Triple nucleotide polymorphisms
URL	Upper Reference Limit
UTR	Untranslated region
VLDL	Very low density lipoproteins

List of Contents

My deepest gratitude & thanks to **ALLAH** *the most merciful for guiding me through & giving me the strength to complete this work the way it is.*

I would like to express my deepest thanks & profound respect to my honored Professor **Dr. *Hany Youssef Halim,*** *Professor of medical Biochimstry and Molecular Biology. Great thanks for his guidance, support and patience that he gave me throughout the whole work. It has been an honor & privilege to work under his generous supervision.*

I find no words by which I can express my deepest thanks & appreciation to ***Dr. Manal Louis Louka,*** *Lecturer of medical Biochemistry and Molecular Biology, Faculty of Medicine, Ain Shams University, for her great support, valuable time and continuous advices which helped me to overcome many difficulties.*

I am also deeply grateful & would like to express my sincere thanks & gratitude to ***assist. Proff Dr. Wael Mahmoud EL Kilany,*** *assist. Proff of Cardiology, Faculty of Medicine–Ain Shams University for his great help, careful supervision, continuous contributions & great encouragement throughout the whole work.*

Seham Adel Badawy

Coronary heart disease (CHD) is the single largest killer of men and women in the United States. The total numbers of individuals affected by CHD or by myocardial infarction (MI) in 2003 were 13.2 million and 7.2 million, respectively. Despite recent advances in therapy for these conditions, nearly 480,000 and 170,000 patients die annually from CHD or MI, respectively ***(Thom et al., 2006)***.

Myocardial infarction in Egypt is an epidemic of enormous proportions. Contrary to the common belief, that ischemic heart disease is uncommon in developing countries, all evidence indicates that, this very serious condition is just as common in Cairo as it is in New York, Paris and in Moscow. The technology needed to combat MI is abundantly available in Egypt, yet only a minority of victims of this disease receive proper state of art treatment ***(Mehta et al., 2009)***.

Coronary artery disease (CAD) is a multifactorial disease influenced by environmental and genetic factors. Family history of premature CAD in addition to other risk factors, such as smoking, obesity, diabetes and dyslipidaemia, are all interactive factors contributing to the occurrence of the disease ***(Egred et al., 2005)***.

Although the role of these environmental factors in the development of myocardial infarction (MI) has been clearly established, the role of nonconventional risk factors remains undefined. In the last few years, great interest has been focused on genetic factors with the intention of finding common markers that could identify a subgroup of patients at higher risk of death or with a worse prognosis in which new therapeutic timings and interventions could be tested ***(Hengstenberg et al., 2002).***

The renin-angiotensin system comprises a cascade of enzymatic reactions, which results in the production of angiotensin II from the angiotensinogen substrate. The physiological effects of angiotensin II are mediated by a final common pathway, through angiotensin II binding to specific receptors located on the cell membrane ***(Timmermans et al., 1993; de Gasparo et al., 2000)***.

Aim of the work

1. Study the association between the genetic polymorphism of AT2R gene A1675G and susceptibility to acute myocardial infarction.
2. Correlation of the genotypic results to the severity of the coronary artery disease and to the different risk factors.

Myocardial Infarction

Definition :

Defined according to the "Universal Definition of Myocardial Infarction" conducted by ***Thygesen et al. (2007)*** on behalf of the Joint ESC/ACCF/AHA/WHF Task Force for the Redefinition of Myocardial Infarction as:-

i. Detection of a rise of cardiac biomarkers (preferably troponin) with at least one value above the 99th percentile of the upper reference limit (URL) together with evidence of myocardial ischemia with at least one of the following:

1. Symptoms of ischemia.
2. ECG changes indicative of new ischemia (new ST-T changes or new left bundle branch block [LBBB]).
3. Development of pathological Q waves in the ECG.
4. Imaging evidence of new loss of viable myocardium or new regional wall motion abnormality.

ii. Sudden, unexpected cardiac death, involving cardiac arrest, often with symptoms suggestive of myocardial ischemia, and accompanied by presumably new ST elevation, or new left bundle branch block (LBBB), and/or evidence of fresh

thrombus by coronary angiography and/or at autopsy, but death occurring before blood samples could be obtained, or at a time before the appearance of cardiac biomarkers in the blood.

Risk Factors:

1. Non-modifiable risk factors:

i. Older age

Older adults are more likely to die of heart disease. About 80% of heart disease deaths occur in people aged 65 or older.

ii. Gender

Men tend to have heart attacks earlier in life than women. Women's rate of heart attack increases after menopause but does not equal men's rate. Even so, heart disease is the leading cause of death for both men and women ***(Braunwald et al., 2001).***

iii. Heredity/Family history:

Increased risk is noticed if a first degree blood relative has had coronary heart disease or stroke before the age of 55 years for male relative and 65 years for female relatives ***(Braunwald et al., 2001).***