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Influence of Hawthorn (*Crataegus Oxyacantha*) Leaves Extract Administration on Myocardial Infarction Induced by Isoproterenol in Rats.

Thesis

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بِسْمِ اللَّهِ الرَّحْمَنِ الرَّحِيمِ



صدق الله العظيم

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DEDICATION.....

This work is dedicated for the soul of my father and grandmother, may God bless their souls.

I'd like to express my love to all my family; my mother, my uncle Fth-Allah, my sisters "Samira", "Shimaa", & "Fatma" and my brother Mostafa for their encouragement, support, patience and endless love when I was physically and psychologically away. I owe for them a lot.

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Abstract

Hawthorn (*Crataegus Oxyacantha*) is one of the most important traditional medical plants which have been used in treating various cardiovascular diseases (CVDs) such as myocardial infarction (MI). MI is one of the most frequent CVDs causes of death all over the world. This study had been designed to investigate the influence of ethanolic hawthorn leaves extract (EHLE) administration at 200 mg/ kg body weight/day on isoproterenol (ISO)-induced MI by measuring different biological and biochemical parameters in rats. In the present study, 50 adult male white albino rats Sprague-Dawley strain weighing 185 ± 15 g were used and divided into five groups comprising of ten rats in each group. **Group I:** served as healthy untreated rats. **Group II:** healthy rats treated with EHLE at tested dose by gastric tube three times weekly. **Group III: (MI group):** rats were injected with ISO subcutaneously at dose of 85mg/kg body weight/day on two consecutive days with 24 hours intervals. **Group IV:** MI rats treated with EHLE. **Group V:** rats treated with EHLE pre and post MI induction. The results of EHLE bioactive components analysis revealed that each one gram of the tested extract contains 70.048 ± 0.5254 mg as gallic acid equivalent (GAE) for total phenols, and 154.417 ± 0.7406 mg as quercetin equivalent (QE) for total flavonoids, as well as high free radical scavenging activity, where 0.05 ml of the tested EHLE scavenged 5 ml of diphenyl-2-picryl hydrazyl (DPPH) radical by $89.55 \pm 0.5683\%$, also flavonoids and phenolic compounds were analyzed by HPLC method. The biological trail showed significant ($P \leq 0.05$) reduction in rats' body weight change, feed intake and feed efficiency ratio (FER) and increment in both feed conversion ratio (FCR) and relative heart weight in MI group when compared with healthy untreated rat group. ISO induced significant ($P \leq 0.05$) increment in serum cardiac enzyme activities of AST, LDH, CK, and CK- MB as well as serum specific MI biomarkers of cTnI and Gal-3 levels and oxidative stress markers of NO and MDA, in association with a significant reduction ($P \leq 0.05$) in cardiac enzymatic and non-enzymatic anti-oxidant system. While, EHLE administration either post or pre and post ISO injection showed a significant improvement in all biological and biochemical parameters, confirming its cardio-protective effects. These results were confirmed by the histopathological examination of the heart tissues in different rat groups.

List of Abbreviations

<i>Abbreviation</i>	<i>Full term/ words</i>
AAP	4-Aminophenazone
AC	Aortic Constriction
ACE	Angiotensin-Converting Enzyme
ACS	Acute Coronary Syndrome
ADP	Adenosine Di-phosphate
AEC	Alcoholic Extract of <i>Crataegus oxyacantha</i>
ALNA (n-3)	Alfa Lenolenic Acid (omega 3 fatty acid)
AMI	Acute Myocardial Infarction
AST	Aspartate aminotransferase
ATP	Adenosine Tri-phosphate
b. wt.	Body weight
BHA	Butylated HydroxyAnisole
BHT	Butylated Hydroxyl Toluene
BMD	Bone Mineral Density
BMI	Body Mass Index
BSA	Bovine Serum Albumin
Ca⁺²	Calcium
CAD	Coronary Artery Disease
CAT	Catalase
CHD	Coronary Heart Disease
CHF	Congestive Heart Failure
CK	Creatine Kinase
CK-MB	Creatine Kinase-MB
CNS	Central Nervous System
COC	<i>Crataegus oxyacantha</i> extract
COE	<i>Crataegus orientalis</i> ethanolic extract
Conc.	Concentration
COX-2	Cyclooxygenase-2
CT	Computerized Tomography
cTnC	Cardiac Troponin C

cTnI	Cardiac Troponin I
cTns	Cardiac Troponins
cTnT	Cardiac Troponin T
CVDs	Cardiovascular Diseases
d. water	Distilled water
DASH	Dietary Approaches to Stop Hypertension
DHBS	3,5-Dichloro -2-Hydroxy Benzene Sulfonic acid
DM	Diabetes Mellitus
DNA	De-oxy ribonucleic Acid
DPPH	1,1-Di-Phenyl-2-Picryl Hydrazyl radical
DTNB	5,5` Di-thiobis -2- Nitrobenzoic acid
ECG	Electrocardiography
Echo	Echocardiography
EHLE	Ethanolic Hawthorn Leaves Extract
ELISA	Enzyme Linked Immune-Sorbent Assay
eNOS	endothelial Nitric Oxide Synthase
ERK ½	Extracellular signal- Regulated Kinase ½
FCR	Feed Conversion Ratio
FER	Feed Efficiency Ratio
FMLP	N-Formylmethionyl-leucyl-phenylalanine
4PLC	Four Parameter Logistic Curve
G6P	Glucose 6 Phosphate
GAE	Gallic Acid Equivalent
Gal-3	Galectin-3
GOT	Glutamate Oxaloacetate Transferase
GPx	Glutathione Peroxidase
GR	Glutathione Reductase
GSH	Reduced Glutathione
GSSG	Oxidized Glutathione
GST	Glutathione -S- Transferase
H&E	Hematoxylin and Eosin
H₂O₂	Hydrogen Peroxide
HCl	Hydrochloric acid
HDL-C	High Density Lipoprotein Cholesterol

HF	Heart Failure
HIF-1	Hypoxia-Inducible Factor-1
HK	Hexokinase
HMG-CoA reductase	Hydroxyl-3-Methyl-Glutaryl-Coenzyme A reductase
HPLC	High Performance Liquid Chromatography
HR	Heart Rate
HRP	Horse Radish Peroxidase
HW	Heart Weight
I.P.	Intra-peritoneal
I/R injury	Ischemia/Reperfusion injury
IHD	Ischemic Heart Disease
IL-6	Interleukin-6
IL-8	Interleukin-8
iNOS	inducible Nitric Oxide Synthase
ISO	Isoproterenol
JNK ½	c- Jun N- terminal Kinase ½
LA (n-6)	Linoleic Acid (omega 6 fatty acid)
LD₅₀	Median Lethal Dose
LDH	Lactate Dehydrogenase
LDL-C	Low Density Lipoprotein Cholesterol
L-NAME	Nitro-L-Arginine-Methyl Ester
LP (a)	Lipoprotein (a)
LPL	Lipoprotein Lipase
LPO	Lipid Peroxidation
LV	Left Ventricular
MAP	Mean Arterial Pressure
MAPK	Mitogen- Activated Protein Kinase
MDA	Malondialdehyde
MI	Myocardial Infarction
MK-886	Montelukast anti-leukotriene
MO	Microvascular Obstruction
MRI	Magnetic Resonance Imaging
NADH	Nicotinamide-Adenine-Dinucleotide reduced

NADPH	Nicotinamide-Adenine-Dinucleotide Phosphate reduced
NaHEK	Sodium-Hydrogen Exchanger Kinase
NBT	Nitroblue Tetrazolium
NEDA	N- (1- naphthyl) - Ethylenediamine
NF-kB	Nuclear Factor-kB
nNOS	neuronal Nitric Oxide Synthase
NO	Nitric Oxide
Nrf2	Nuclear factor-erythroid-2–related factor2
O²⁻	Superoxide
ODs	Optical Densities
OPC	Oligomeric Procyanidins/ proanthocyanidins
p38 MAPK α/β	p38 Mitogen- Activated Protein Kinase alpha/beta, also called Mitogen- Activated Protein Kinase 14
p90RSK	p90 Ribosomal S90 Kinase
PBS	Phosphate Buffered Saline
PDE	Phosphodiesterase
PFA-100	Platelet aggregation as assessed by platelet Function Assay or Analyzer
PKB	Protein Kinase B
PLA2	Phospholipase A2
PMS	Phenazine Metho-Sulphate
PPARγ	Peroxisome Proliferator- Activator Receptor gamma
QEs	Quercetin Equivalents
RHW	Relative Heart Weight
RNS	Reactive Nitrogen Species
ROS	Reactive Oxygen Species
S.C.	Subcutaneous
S.D	Standard Deviation
SNS	sympathetic Nervous system
SOD	Superoxide Dismutase
SPSS	Statistical Package for Social Science
STEMI	ST- segment Elevation Myocardial infarction
TAG	Triacylglycerol
TBA	Thiobarbituric Acid

TC	Total Cholesterol
TCA	Tri-chloroacetic Acid
TCR	Tincture of <i>Crataegus</i>
TMB	3,3',5,5' - Tetra-Methyl-Benzidine
TNF-α	Tumor Necrosis Factor alpha
VT	Ventricular Tachycardia
WBCs	White Blood Cells
WHO	World Health Organization
XO	Xanthine Oxidase

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