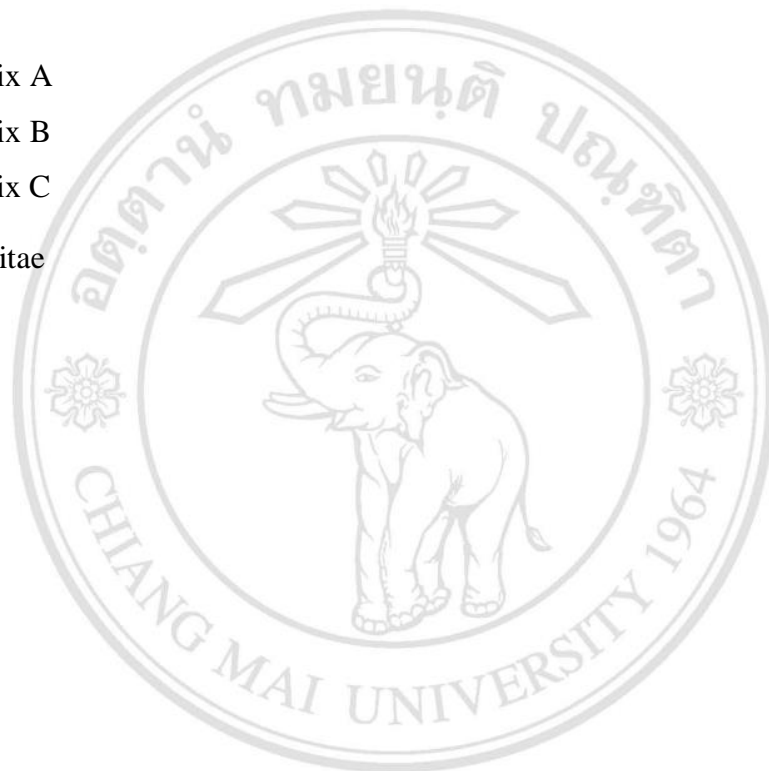


CONTENTS

	Page
Acknowledgement	d
Abstract in Thai	e
Abstract in English	g
Lists of Tables	l
Lists of Figures	m
List of Abbreviations	o
List of Symbols	r
Chapter 1 Introduction	1
1.1 Principle, theory and rational/hypothesis	1
1.2 Literature review	2
1.2.1 Ischemia/reperfusion injury	2
1.2.2 Mechanism of I/R injury	3
1.2.3 Mitochondrial function during I/R injury	4
1.2.4 Treatment of I/R injury	6
1.2.5 Autonomic nervous system	6
1.2.6 Vagus nerve anatomy	8
1.2.7 Alteration of autonomic control in ischemic heart disease	10
1.2.8 Vagus nerve stimulation (VNS) system	11
1.2.9 Effects of VNS on myocardial infarct size after I/R injury	12
1.2.10 Effects of VNS on anti-arrhythmic effects	14
1.2.11 Effects of VNS on LV function and hemodynamic parameters	16
1.2.12 Clinical application of VNS on cardiovascular disease	19
1.3 Purposes of the study	21

1.4 Hypothesis of the study	21
Chapter 2 Materials and methods	22
2.1 Animal preparation	22
2.2 Ischemic/reperfusion (I/R) protocol	22
2.3 Vagus nerve stimulation (VNS) protocol	23
2.4 Experimental protocols	23
2.5 Evaluation of cardiac functions	24
2.6 Evaluation of rhythm disturbances	25
2.7 Evaluation of left ventricular (LV) functions parameters	25
2.8 Infarct size determination	26
2.9 Isolated cardiac mitochondria	26
2.10 Transmission electron microscopy for cardiac mitochondrial morphology	27
2.11 Western blot analysis	27
2.12 HPLC-based assay of malondialdehyde (MDA) concentration	28
2.13 Cardiac inflammatory and anti-inflammatory cytokine assay	29
2.14 TUNEL assay	29
2.15 Statistical Analysis	30
Chapter 3 Results	31
3.1 The effect of VNS on the electrocardiographic parameters during the ischemic/reperfusion period	31
3.2 The effect of VNS on the occurrence of cardiac arrhythmia during the ischemic/reperfusion period	34
3.3 The effect of VNS on LV function during the ischemic/reperfusion period	36
3.4 The effect of VNS on myocardial infarct size after the ischemic/reperfusion period	38
3.5 The effect of VNS on cardiomyocyte apoptosis	40
3.6 The effect of VNS on oxidative stress activity (MDA)	43
3.7 The effect of VNS on pro-inflammatory and anti-inflammatory	44

3.8 The effect of VNS on mitochondrial function	45
3.9 The effect of VNS on mitochondrial dynamics	47
3.10 The effect of VNS on mitochondrial biogenesis	49
Chapter 4 Discussion and conclusion	50
References	61
Appendix	74
Appendix A	75
Appendix B	79
Appendix C	84
Curriculum Vitae	86



ลิขสิทธิ์มหาวิทยาลัยเชียงใหม่
 Copyright© by Chiang Mai University
 All rights reserved

LIST OF TABLES

	Page
Table 1-1 Fiber type and function of vagus nerve	9
Table 2-1 Criteria of arrhythmia score	25

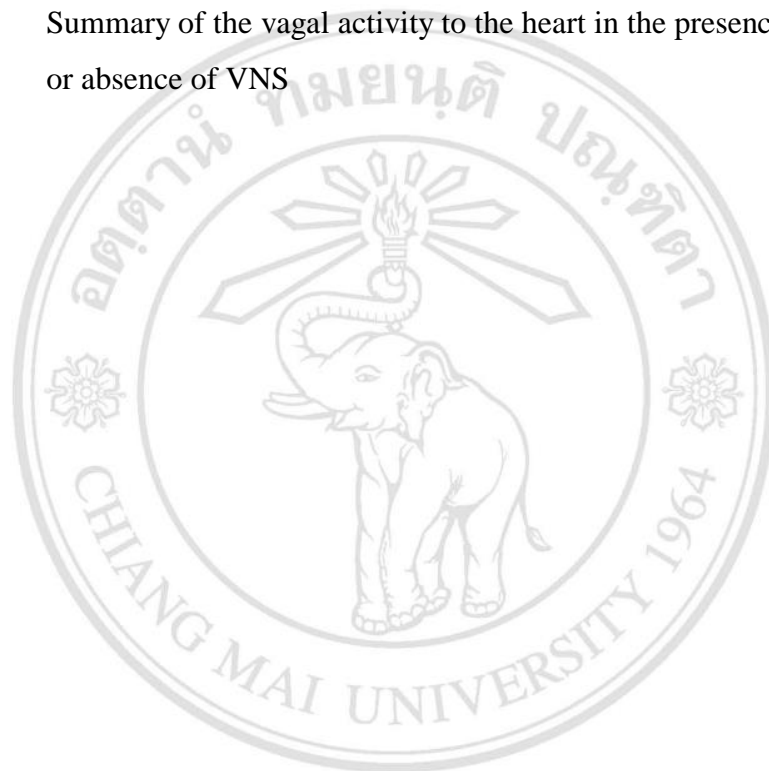


ลิขสิทธิ์มหาวิทยาลัยเชียงใหม่
Copyright© by Chiang Mai University
All rights reserved

LIST OF FIGURES

		Page
Figure 1-1	Summarize the mechanism of ischemia/reperfusion injury	4
Figure 1-2	Schematic representation of the autonomic nervous system	7
Figure 1-3	Illustration of the vagus nerve anatomy and vagal nuclei connections	10
Figure 1-4	Illustration of the VNS system	11
Figure 1-5	Illustration of myocardial damage in I/R injury	12
Figure 2-1	Schematic design of experimental protocols	24
Figure 3-1.1	Effect of VNS on the electrocardiographic parameters during the ischemic and the reperfusion periods	32
Figure 3-1.2	Effect of VNS on the electrocardiographic parameters during the ischemic and the reperfusion periods	33
Figure 3-2	Effects of VNS on the occurrence of ventricular arrhythmias during the ischemic/reperfusion period	35
Figure 3-3	The effect of VNS on LV function during I/R injury	37
Figure 3-4	The effect of VNS on myocardial infarct size after the ischemic/reperfusion period	39
Figure 3-5.1a	Effect of VNS on the TUNEL-positive cells	40
Figure 3-5.1b	Representative of the TUNEL assay	41
Figure 3-5.2	Effect of VNS on cardiomyocyte apoptosis	42
Figure 3-6	Effect of VNS on oxidative stress biomarkers in myocardium	43
Figure 3-7	Effect of VNS on pro-inflammatory and anti-inflammatory markers	44
Figure 3-8	Effect of VNS on cardiac mitochondria function after ischemia and reperfusion periods	46
Figure 3-9	Effect of VNS on mitochondrial dynamics	48

Figure 3-10	Effect of VNS on mitochondrial biogenesis	49
Figure 4-1	Summary of the molecular mechanism of I/R injury	56
Figure 4-2	Summary of the cardioprotection of VNS against I/R injury	57
Figure 4-3	Summary of the molecular mechanism underlying the cardioprotection of LC-VNS, LtVNX and RtVNX against I/R injury	58
Figure 4-4	Summary of the vagal activity to the heart in the presence or absence of VNS	59



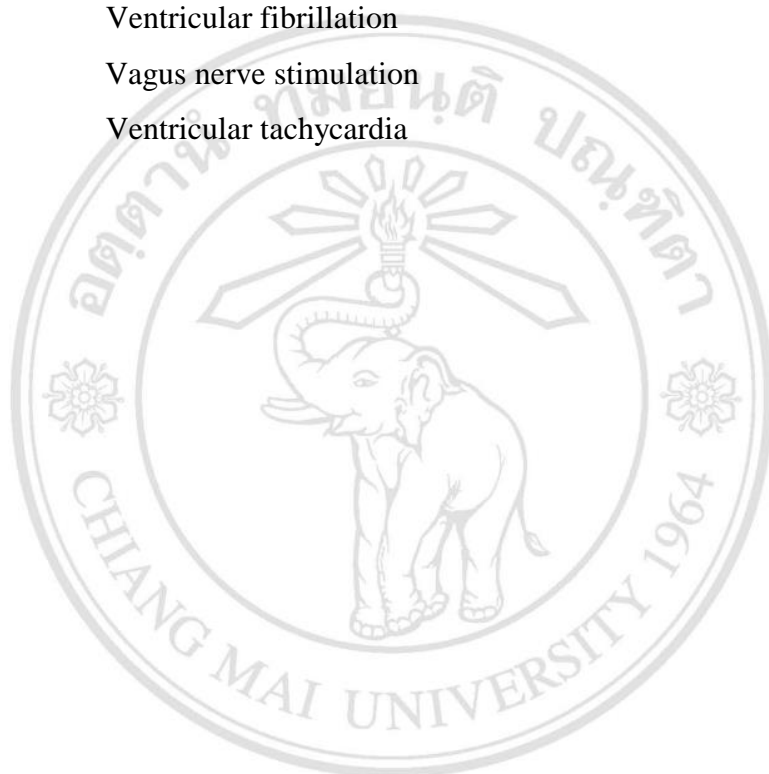
ลิขสิทธิ์มหาวิทยาลัยเชียงใหม่
 Copyright© by Chiang Mai University
 All rights reserved

LIST OF ABBREVIATIONS

AAR	Area at risk
ANOVA	Analysis of variance
ADP	Adenosine diphosphate
ATP	Adenosine triphosphate
BP	Blood pressure
CABG	Coronary artery bypass grafting
CAD	Coronary artery diseases
CAP	Cholinergic anti-inflammatory
CPT1	Carnitine palmitoyltransferase 1
CVD	Cardiovascular disease
Cx43	Connexin 43
DCFH-DA	Dichlorohydrofluorescein diacetate
Drp1	Dynamin related protein 1
ECG	Electrocardiogram
EDP	End diastolic pressure
EDV	End diastolic volume
EF	Ejection fraction
ESP	End systolic pressure
ESV	End-systolic Volume
HR	Heart rate
H ₂ O ₂	Hydrogen peroxide
I	Ischemic
IHD	Ischemic heart disease
IL-10	Interleukin-10
IMAC	Mitochondrial inner membrane anion channel
i.p.	Intraperitoneal

I/R injury	Ischemia/reperfusion injury
JC-1	5,5',6,6'-tetrachoro-1,1',3,3'-tetraethyl-imidacarbocyanine iodide
LAD	Left anterior descending coronary artery
LC-VNS	Left cervical vagus nerve stimulation
LtVNX	Left vagus nerve transection combined with left cervical vagus nerve stimulation
LV	Left ventricle
$\Delta\Psi_m$	Mitochondrial membrane potential change
MDA	Malondialdehyde
MFN2	Mitofusin-2
MMP	Mitochondrial membrane potential
mPTP	Mitochondrial permeability transition pore
NSS	Normal saline solution
OPA1	Optic atrophy type 1
PCI	Percutaneous coronary intervention
pH	Potential of hydrogen
PGC1 α	Peroxisome proliferator-activated receptor-gamma coactivator 1alpha
P-V loop	Pressure-volume loop
PVC	Premature ventricular contraction
R	Remote area
ROS	Reactive oxygen species
RtVNX	Right vagus nerve transection combined with left cervical vagus nerve stimulation
SCD	sudden cardiac death
SE	Standard error
SOD	Superoxide dismutase
SV	Stroke volume
SW	Stroke work
TNF- α	Tumor necrosis factor- α

TBST	Tris-Buffered Saline and Tween
TEM	Transmission electron microscopy
Tpe	T-wave peak to end
TTC	2,3,5-Triphenyltetrazolium chloride
TUNEL	Terminal Deoxynucleotidyl transferase-mediated dUTP nick end labeling
VF	Ventricular fibrillation
VNS	Vagus nerve stimulation
VT	Ventricular tachycardia



ลิขสิทธิ์มหาวิทยาลัยเชียงใหม่
 Copyright© by Chiang Mai University
 All rights reserved

LIST OF SYMBOLS

%	Percentage
γ	Gamma
α	Alpha
β	Beta
κ	Kappa
μ	Micro
μl	Microliter
$\Delta\Psi$	Mitochondrial membrane potential change
$^{\circ}\text{C}$	Degree Celsius
g	Gram
kg	Kilogram
mg/g	Milligram per gram
mg/dl	Milligrams per deciliter
min	Minute
mm	Millimeter
mmHg	The millimeter of mercury
mmHg/s	The millimeter of mercury per second
mmol/l	Millimoles per liter
mM	Millimolar
ng/ml	Nanograms per milliliter
nm	Nanometer