ROLE OF SILDENAFIL AS A FACTOR MODULATING SKELETAL MUSCLE CONTRACTILITY IN CASES OF INDUCED DIABETIC RATS

Thesis

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ABSTRACT

Aim of work: This study was designed to evaluate the effect of sildenafil citrate on skeletal muscle contractility in type 2 diabetic rats.

Materials and methods: Diabetes was induced by feeding rats with a high fat diet (HFD) for 2 weeks followed by an intraperitoneal injection of streptozotocin (35 mg/kg body weight). The considered rats were divided into four groups containing ten animals for each, group1: normal control, group 2: diabetic group, group 3: diabetic treated with 5mg/kg/day sildenafil for 4 weeks and group 4: diabetic treated with 10mg/kg/day sildenafil for 4 weeks. By the end of the experimental period rats were sacrificed, blood samples were taken and serum was isolated for estimation of glucose, insulin, TNF-α, IL-1β, IL-6 and lipid profile parameters. In addition, diaphragm contractility was determined in the studied groups.

Results:

In the diabetic group, levels of glucose, TNF-α, IL-1β, IL-6, glycosylated hemoglobin, HOMA-IR, total cholesterol, triglycerides, LDL-cholesterol and vLDL-cholesterol were significantly increased, while contractility index, serum HDL-cholesterol and insulin levels were decreased. Both doses of sildenafil significantly reversed these alterations. Moreover, supplementation with either dose significantly ameliorated the diminished diaphragm contractility.

Conclusion: The current study showed that sildenafil citrate has pronounced useful effects on skeletal muscle contractility via ameliorating the deteriorated metabolic parameters in type 2 diabetic rats.

Key words:

sildenafil, insulin resistance, muscle contractility index, oxidative stress.

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Contents

List of Abbreviations

ADA	American Diabetes Association
AMPK	AMP activated protein kinase
ANOVA	Analysis of variance
ATGL	Adipose triglyceride lipase
BH_4	Tetrahydrobiopterin
CaM	Calmoduline
cGMP	Cyclic guanosine monophosphate
CMV	Cytomegalovirus
CNG	Cyclic nucleotide-gated channels
CRP	C-reactive protein
DAG	Diacylglycerol
DCCT	Diabetes Control and Complications Trial
DGC	Dystrophin glycoprotein complex
DM	Diabetic mellitus
DTNB	Dithiobis-2-nitrobenzoic acid
ED	Erectile dysfunction
EDRF	Endothelium-derived relaxing factor
EDTA	Ethylenediamine tetraacetic acid
EMEA	European Agency for the Evaluation of Medicinal Products
eNOS	Endothelial nitric oxide synthase
ERK	Extracellular signal regulated kinase
FAD	Flavin adenine dinucleotide
FDA	Food and Drug Administration
FFA	Free fatty acids
FMN	Flavin mononucleotide
GAD	Glutamic acid decarboxylase
GK	Glucokinase
GLUT	Glucose transporter
GPO	Glycerol-3-phosphate oxidase
GP_x	Glutathione peroxidase
GSH	Glutathione
GTP	Guanosine triphosphate

HDL	High density lipoprotein
HFD	High fat diet
HK	Hexokinase
HOMA-IR	Homeostasis model of insulin resistance
HSL	Hormone-sensitive lipase
ICE	Interleukin-1 converting enzyme
IDDM	Insulin dependent diabetes mellitus
IDF	International Diabetes Federation
IL	Interleukin
iNOS	Inducible nitric oxide synthase
IR	Insulin receptor
IRS	Insulin receptor substrates
LDL	Low-density lipoprotein
LPL	Lipoprotein lipase
LPS	Lipopolysaccharide
LSD	Least significant difference
MAPK	Mitogen-activated protein kinase
MDA	Malondialdehyde
MHC	Major histocompatibility complex
MRDM	Malnutrition related Diabetes Mellitus
MS	Metabolic Syndrome
NADPH	Nicotinamide adenine dinucleotide phosphate
NANC	Nonadrenergic noncholinergic
NIDDM	Non-insulin dependent diabetes mellitus
nNOS	Neuronal nitric oxide synthase
NO	Nitric oxide
NOS	Nitric oxide synthase
OD	Optical Denisty
PC	Phosphatidylcholine
PPAR	Peroxisome proliferator activated receptor
PE	Phosphatidylethanoloamine
PGC	PPARγ coactivator
PI3K	Phosphatidylinositol 3-kinase
PKB	Protein kinase B
PKC	Protein kinase C
PLC	Phospholipase C

PS	**Phosphatidylserine**
POLG	**Polymerase DNA gamma**
PTP	**Tyrosine phosphatase**
ROIs	**Reactive oxygen intermediates**
ROS	**Reactive Oxygen Species**
sGC	**Soluble guanylate cyclase**
SH2	**Src-homology- 2**
STZ	**Streptozotocin**
T2DM	**Type 2 diabetes mellitus**
SOCS	**Suppressors of cytokine signaling proteins**
SOD	**Superoxide dismutase**
SR	**Soluble receptor**
SSBP	**Single strand binding protein**
TBARS	**Thiobarbituric acid reacting substance**
TCA	**Trichloroacetic acid**
TNF-α	**Tumor necrosis factor-alpha**
VLDL	**Very low-density lipoprotein**
VSMC	**Vascular smooth muscle cells**
WHO	**World Health Organization**

List of tables

List of figures

List of tables

List of figures

INTRODUCTION

The incidence and prevalence of diabetes mellitus (DM), and its attendant complications are rising to epidemic levels **(Zimmet *et al*., 2001; Deshpande *et al*., 2008)**. Diabetes mellitus is a chronic disease of disturbed carbohydrate, fat, and protein metabolism and can result in significant progressive and permanent physical disability **(Gregg *et al*., 2002; Turcotte and Fisher, 2008)**. In 2002, DM was reported as the sixth leading cause of death, and the risk of mortality for individuals with DM is twice that of individuals of the same age without DM **(National Diabetes Fact Sheet, 2005)**. Several comorbid conditions are related to and accompany DM, including coronary heart disease **(Gregg *et al*., 2002; Von Korff *et al*., 2005)**, obesity **(Gregg *et al*., 2002)**, arthritis , stroke **(Gregg *et al*., 2000)**, depression **(Von Korff *et al*., 2005)**, and visual impairments **(Gregg *et al*., 2002)** and have been identified as contributors to DM-related disability **(Park *et al*., 2006 and 2007)**.

The full extent of the combined effects of obesity and DM and their respective complications on alterations in body composition and muscular function has not been thoroughly explored. The major tissue affected by disturbances in glucose metabolism is skeletal muscle, and

deficits in metabolic signaling in this tissue contribute to systemic insulin resistance **(Wei *et al*., 2008; Turcotte and Fisher, 2008)**.

Approximately 60% to 70% of people with DM have mild to severe forms of nervous system disease **(National Diabetes Fact Sheet, 2005)**. It has been estimated that nearly 50% of all people with DM will develop somatic sensory, motor, or autonomic peripheral neuropathy (PN) or a combination of these types of neuropathy 25 years or later after diagnosis. Individuals with DM and PN develop rapid muscle weakness and motor dysfunction **(Andersen *et al*., 1997)**. There are only a few studies examining skeletal muscle strength in diabetes, thus the current study was designed to evaluate the effect of sildenafil citrate, a phosphodiesterase 5 inhibitor, on skeletal muscle contractility via direct and indirect stimulation in high fat diet/streptozotocin diabetic rats.