The effects of exercise on plasma adiponectin concentration and adiponectin receptor expression in normal and type 2 diabetic rats

Thesis

Submitted in partial fulfillment of the requirements

for the Degree of Master of Science in

Physiology

By

OMNIA MOHAMED ABDEL MAKSOUD KHALAFALLA

Under supervision of

Prof. Dr.
Magda Mahmoud Al Hamzawi
Professor of Physiology
Cairo University

Dr.
Sahar Mohamed Kamal
Ass. Professor of Physiology
Cairo University

Dr.
Laila Ahmed Rashed
Ass. Professor of Biochemistry
Cairo University

Faculty of medicine
Cairo University
200٨

ACKNOWLEDGEMENT

All praise and thanks are due to ALLAH© The Most Beneficent© and The Most Merciful.

I would like to express my sincere thanks from the heart and deep appreciation to ***Prof© Dr© Magda Mahmoud Al Hamzawy©*** *Professor of Physiology, Faculty of Medicine, Cairo University, for her great guidance and generous helping throughout the course of study, supervision of the work, as well as unlimited encouragement, valuable advices and stimulating criticism.*

I am grateful to ***Dr© Sahar Mohamed Kamal****, Assistant Professor of Physiology, Faculty of Medicine, Cairo University, and* ***Dr© Laila Ahmed Rashed****, Assistant Professor of Biochemistry, Faculty of Medicine, Cairo University, for their sincere help, kind advices and guidance through the work as well as sharing in supervision.*

I would also like to express my deep thanks and appreciation to ***Prof© Dr© Ibrahim Mohammady Ibrahim****, the Head of Physiology department, Faculty of Medicine, Cairo University, for his great support and kind guidance. Deep appreciation is given to the staff members of Physiology department, Faculty of Medicine, Cairo University, for their valuable support.*

Last but not least, special deep appreciation from the heart is given to my dear family for their generous support and sincere encouragement.

ABSTRACT

Adiponectin is an adipocytokine which is strongly related to insulin sensitivity and the development of type 2 diabetes mellitus. Adiponectin receptor 1 (ADIPOR1) is abundantly expressed in the skeletal muscles and is strongly related to insulin sensitivity.

Both exercise and adiponectin are found to exert similar insulin sensitizing effects. Thus, this work aimed to study the effect of 4 weeks swimming exercise training on adiponectin concentration, as well as ADIPOR1 gene expression in the soleus muscle, in normal and type 2 diabetic insulin resistant rats. Type 2 diabetes was induced using a combination of high fat diet and low dose streptozotocin. Study groups included: normal control rats, normal exercise-trained rats, diabetic control rats, and diabetic exercise-trained rats.

Results of the present study revealed significant decrease in the levels of both adiponectin and ADIPOR1 in type 2 diabetic rats, which were significantly improved with exercise training, as shown in the comparison of the diabetic control and the diabetic exercise-trained rats. On the other hand, exercise training didn’t significantly affect adiponectin and ADIPOR1 in normal rats, as revealed from comparing the results of the normal control and the normal exercise trained rats.

We concluded that the exercise-induced improvement in insulin sensitivity in type 2 diabetic rats may be mediated, at least in part, through adiponectin and/or ADIPOR1 upregulation.

Key words: adiponectin, type 2 diabetes mellitus, exercise

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ADA: American Diabetes Association

ADIPOR1: Adiponectin Receptor 1

ADIPOR2: Adiponectin Receptor 2

AMPK: 5'-AMP-activated Protein Kinase

APPL1: Adaptor protein containing Pleckstrin homology domain, Phosphotyrosine-binding domain and Leucine zipper motif

DM: Diabetes Mellitus

FFA: Free Fatty Acid

GLUT4: Glucose Transporter-4

HMW: High Molecular Weight

HOMA: Homeostasis Model Assessment

IL: Interleukin

IRS: Insulin Receptor Substrate

PPAR: Peroxisome Proliferator-Activated Receptor

T2DM: Type 2 Diabetes Mellitus

TNF-α: Tumour Necrosis Factor-alpha

Adiponectin is an adipocytokine secreted mostly from the adipose tissue (**Maeda** *et al*., **1996**). It exerts its multiple effects through binding with its receptors; adiponectin receptor 1 (ADIPOR1) and adiponectin receptor 2 (ADIPOR2) (**Yamauchi** *et al*., **2003**). Adiponectin has recently attracted much attention because of its multiple actions as an anti-inflammatory (**Kadowaki** and **Yamauchi, 2005**), and anti-atherogenic agent (**Ouchi** *et al*., **2001**).

Furthermore, several studies have established that diabetes susceptibility is linked to abnormalities in adiponectin secretion from adipocytes (**Hotta** *et al*., **2001;** and **Spranger** *et al*., **2003**). This has triggered further interest to assess the possible role of adiponectin in the onset of diabetes. The results of these studies revealed that a significant relationship, which links the levels of circulating adiponectin with insulin sensitivity, may exist (**Koenig** *et al*., **2006;** and **Snijder** *et al*., **2006**).

Moreover, it appears that adiponectin can possibly be altered by various factors (**Arita** *et al*., **1999; Kondo** *et al*., **2002;** and **Pischon** *et al*., **2005**). Among these factors, exercise training has drawn the attention towards the exact role of regular exercise on the production of adiponectin (**Tang** *et al*., **2005**), and/or the expression of its receptors (**Bluher** *et al*., **2006**), in human and animal subjects.

In an attempt to clarify this role, several studies reported positive effects of exercise on adiponectin (**Bluher** *et al*., **2006;** and **Tang** *et al*., **2005**), while others concluded the reverse (**Boudou** *et al*., **2003;** and **Yokoyama** *et al*., **2004**).

In view of this contradiction as regards the possible association of adiponectin with the beneficial effects of exercise training, the experiments of the present study were designed to further explore the impact of exercise in modulating the release of adiponectin.

Based on the knowledge that exercise training improves beyond doubt insulin sensitivity in diabetic subjects (**Sigal** *et al*., **2004**), a second objective of our present work was to study if the improvement of insulin sensitivity associated with exercise is mediated through adiponectin or not.

Since ADIPOR1 was established to be abundantly expressed in the skeletal muscles (**Kadowaki** and **Yamauchi, 2005**), which is one of the major sites to be affected by insulin resistance and diabetes, we further extended our present work by estimating the gene expression of this receptor in response to regular exercise in both normal and type 2 diabetic insulin resistant rats.

DIABETES MELLITUS

The term diabetes mellitus refers to a group of diseases that, through various mechanisms, cause hyperglycemia (**Fowler, 2007**). The American Diabetes Association (ADA) defines diabetes as "a group of metabolic diseases characterized by hyperglycemia, resulting from defects in insulin secretion, insulin action, or both" (**American Diabetes Association, 2006**).

TYPES OF DIABETES MELLITUS

The ADA classifies diabetes mellitus, according to its etiology, into four broad categories including: type 1 diabetes, type 2 diabetes, gestational diabetes mellitus, and other specific forms of diabetes (**American Diabetes Association, 2006**).

Type 1 Diabetes

In type 1 diabetes there is absolute insulin deficiency caused by autoimmune destruction of β-cells. It may be classified into type 1A if autoimmune markers are found, and type 1B when autoimmune markers cannot be detected (**The Expert Committee on the Diagnosis and Classification of Diabetes Mellitus, 1997**). Type 1 diabetes is thought to be due to a combination of environmental factors and genetic susceptibility. Many triggers, such as cow's milk and food additives, are proposed for developing type 1 diabetes in genetically susceptible individuals (**Helgason** and **Jonasson, 1981;** and **Oute** *et al*., **1999**).

Type 2 Diabetes

Type 2 diabetes (T2DM) is the most common metabolic disorder worldwide, comprising 90% or more of the cases of diabetes. It is generally characterized by insulin resistance and relative insulin deficiency with increased hepatic glucose production (**Kahn, 1994**). The progressive nature of T2DM is mostly due to the steady reduction of the insulin secretory capacity (**Butler** *et al*., **2003**).

Gestational diabetes mellitus

Gestational diabetes mellitus refers to any degree of glucose intolerance with onset or first recognition during pregnancy (**American Diabetes Association, 2006**). Gestation associated with insulin resistance, is mostly due to increased maternal adipose tissue, and anti-insulin effects of placental hormones (**Buchanan** and **Xiang, 2005**). Women who experience gestational diabetes mellitus may develop T2DM within 10 years after delivery (**Kim** *et al*., **2002**).

Other specific forms of diabetes

The ADA describes more than 56 other specific types of diabetes. The autosomal-dominant defects in β-cell function, known as maturity-onset diabetes of the young, may manifest themselves in infancy or childhood and cause impaired insulin secretion (**American Diabetes Association, 2006**). Moreover, diseases of the exocrine pancreas, such as pancreatitis and cyctic fibrosis, may lead to insulin deficiency. Eventually, the severity of diabetes is proportionate to the degree of pancreatic injury (**Fowler, 2007**).

AETIOLOGY OF TYPE 2 DIABETES MELLITUS

Type 2 diabetes is a heterogenous disorder with a multifactorial etiology, where considerable controversy surrounds its exact primary cause. Most forms of T2DM are associated with complex inheritance patterns, affecting both insulin secretion and action (**Ek** *et al*.**, 2001**).

Multiple environmental factors have been associated with a higher incidence of T2DM. Indeed, high fat and/or high sucrose diets are known to induce insulin resistance in rodents, although the underlying mechanisms are not fully understood (**Gonsolin** *et al*.**, 2007;** and **Ryu** and **Cha, 2003**). High dietary intake of the monounsaturated fatty acid oleic acid, which is abundant in olive oil, was found to improve insulin sensitivity, whereas the intake of saturated fatty acids has proved to promote the opposite (**Marshall** *et al*.**, 1997;** and **Soriguer** *et al*.**, 2004**).

Moreover, low levels of habitual physical activity, poor aerobic fitness, and obesity have been associated with an increased risk of T2DM (**Katzmarzyk** *et al*., **2007;** and **Rana** *et al*., **2007**). Furthermore, insulin secretion and to some extent insulin action may be declined with age (**Fernandez-Castaner** *et al*.**, 1996**).

Human studies have depicted the strong genetic influence on insulin secretion and insulin resistance to be even highly dependent on these environmental factors (**Poulsen** *et al*., **2005**). This strong genetic influence has been suggested by greater concordance rate for T2DM observed in monozygotic compared with dizygotic twin pairs (**Medici** *et al*., **1999**).

PATHOGENESIS OF TYPE 2 DIABETES MELLITUS

Impaired insulin secretion (**Cunningham** *et al*.**, 1996**), insulin resistance (**Haring, 1991**), and increased hepatic glucose production (**DeFronzo, 1992**), are the three major players involved in the development of T2DM.

In T2DM, there is impaired ability for carbohydrate uptake and oxidation in response to insulin, with a shift to lipid fuel utilization to compensate for the diminished capacity for glucose uptake (**Shepherd** and **Kahn, 1999**). Those patients have elevated serum triglyceride and fatty acid levels, and increased intramyocellular triglyceride, the so called "metabolic inflexibility," *i.e.*, the inability to modulate the level of lipid oxidation to meet the metabolic demands (**Kelley** and **Mandarino, 2000**).

Insulin resistance

Normally, circulating insulin binds to its tyrosine kinase receptors present on the surface of adipocytes and muscle cells (**Rosen, 1986;** and **Shulman, 1999**). This binding triggers autophosphorylation of the receptors, resulting in activation of several phosphorylation-dephosphorylation cascades, mostly involving insulin receptor substrates (IRS) which mediate most of the metabolic actions of insulin (**Saltiel** and **Kahn, 2001**).

Insulin resistance implies that higher-than-normal concentrations of insulin are required to maintain normoglycemia, with diminished ability of insulin to stimulate glucose disposal and inhibit hepatic glucose production (**Kahn, 1994**).