### Study of Presence of Pancreatic Islet Cells Antibodies in A Sample of Egyptian Women with Gestational Diabetes and its Relation to the Development of Type 1 Diabetes Mellitus

#### Thesis

Submitted for Partial Fulfillment of Doctoral Degree in **Internal Medicine** 

#### By

#### **Wesam Ahmed Mohamed**

Master degree of internal medicine

### Supervised By

#### Prof. Dr. Hanan Mohmed Ali Amer

Professor of Internal Medicine and Endocrinology Faculty of Medicine-Ain Shams University

### Dr. Rania Sayed Abd El-Baky

Assistant professor of Internal Medicine and Endocrinology Faculty of Medicine-Ain Shams University

### Dr. Merhan Samy Nasr

Assistant professor of Internal Medicine and Endocrinology Faculty of Medicine-Ain Shams University

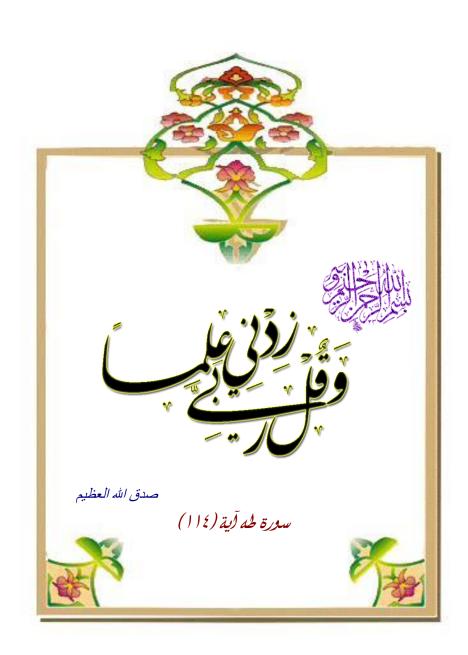
### Dr. Laila Mahmoud Ali Hendawy

Lecturer of Internal Medicine and Endocrinology Faculty of Medicine-Ain Shams University

### Dr. Mohamed Osama Taha

Lecturer of Gynacology and Obestetric Faculty of Medicine-Ain Shams University

> Faculty of Medicine Ain Shams University 2015





# Acknowledgement

No words can express my deepest appreciation and profound respect to Professor **Prof. Dr. Hanan Mohmed Ali Amer,** Professor of Internal Medicine and Endocrinology, Ain Shams University, for her continuous guidance, support and constructive criticism through the work. SHe has generously devoted much of her time and her effort for planning and supervision of this study.

Also, my profound gratitude to **Dr. Rania Sayed Abd El-Baky**, Assistant professor of Internal Medicine and Endocrinolog, Ain Shams University, for her kind supervision and support. It was great honor to work under her supervision.

Also, my profound gratitude to **Dr. Merhan Samy Nasr** Assistant professor of Internal Medicine and Endocrinology, Ain Shams University, for her great care and support.

I would like also to thank **Dr. Laila Mahmoud Hendawy** Lecturer of Internal Medicine and Endocrinology,
Ain Shams University Hospitals, for her support and help
during this work.

I would like also to thank **Dr. Mohamed Osama Taha** Lecturer of Gynacology and Obestetric Ain Shams University Hospitals, for his support and help during this work.

I would like also to thank **Dr. Magdy Abbas Abd Elaziz**, Ass. Chairman of Biochemistry, Ain Shams University Hospitals, for his support and help during this work.

Last but not least, I dedicate this work to my family, whom without their sincere emotional support, pushing me forward this work would not have ever been completed.

Wesam Ahmed Mohmed

### **Contents**

List of Abbreviations	i
List of Tables	iii
List of Figures	
Introduction and Aim of the Work	1
Review of Literature	3
(1) Diabetes mellitus	
(2) Gestational diabetes mellitus (GDM)	15
(3) Type I diabetes mellitus	50
(4) Pancreatic Islet cell autoantibodies and gestation	nal
diabetes	70
Subjects and Methods	92
Results	103
Discussion	133
Summary and Conclusion	143
Recommendations	147
References	148
Arabic Summary	

### **List of Abbreviations**

**2hPP** 2- hour postprandial plasma glucose

**ADA** American Diabetic Association

**BG** Blood glucose

**BMI** Body mass index

**CAMP** Cyclic adenosine monophosphate

CD Cluster of differentiation

DAAS Diabetes autoantibodies

**DAFNE** Dose Adusiment For Normal Eating

**DAISY** The Diabetes Autoimmunity Study in The

young

**DBP** Diastolic blood pressure

**DPT-1** The Diabetes prevention Trail**DRA** diabetes releated autoantibodies

**FBG** Fasting blood glucose

**GADA** Glutamic acid decarboxylase antibodies

GDGS Guidline Development Groups
GDM Gestational diabetes mellitus

**GLP** Glucagon like peptide

**GPCRS** G protein coupled receptors

**ACOG** American college of obstericians and

gynecologist

**HCS** Human chorionic somato-mammo-tropin

**hDAF** Human decay accelerating factor

HDL High-density lipoproteinHMD Hyaline membrane diseaseHNF Hepatocyte nuclear factor

### List of Abbreviations (Cont.)

**HOMA-IR** Homeostatic model assessment of insulin

resistance

**IA2** Insulinoma- associated autoantibodies

IAAS Insulin autoantibodiesICA Islet cell autoantibodies

**IDDM** Insulin dependent diabetes mellitus

IPF Insulinoma promotor factorIPS CELLS Induced pluripotent stem cells

IU International unit

**LADA** Latent autoimmune diabetes of adults

MAbs Monoclonal antibodiesMNT Medical nutrition therapy

MODY Maturity onset diabetes of the young

**NICE** National institute for Health and clinical

Excellence

NPH Neutral protamine Hagerdon
OGTT Oral glucose tolerance test
PCOS Polycystic ovary syndrome

**PERV** Porcine endogenous retro-viruses

PP Pancreatic polypeptidesSBP Systolic blood pressure

**SMBG** self-monitoring of blood glucose

**TEDDY** The Environmental Determinants in Diabetes

of the young

**TH-CELL** T-helper cell

VIP Vasoactive intestinal peptideWHO World Health Organization

**ZN T8** Zinc transporter 8

### List of tables

Table	Title	Page
	Tables of the Review & Subjects	
1	High and low Risk for Gestational diabetes	22
2	Diagnostic tests of Gestational diabetes mellitus	24
3	Diagnosis of GDM with 100gm or 75gm glucose load	25
4	Maternal and perinatal complications of GDM	27
5	Blood glucose targets for women with GDM	39
6	Diagnosis of GDM with a 75-g glucose load	95
	Tables of the results	
1	Descriptive data regarding all study population	111
2	Comparison between Group I (ICA positive	112
	group) and Group II (ICA negative group) as regard different parameters using t- test	
3	percentage of islet cell antibodies among studied population and number of diabetic patients within 6 month after delivery	113
4	Comparison between Group Ia (25 diabetic patients ) and GroupIb (41non dibetic subjects) (follow up 6 monthes postpartum) using t- test	114
5	Number of diabetic patients within 1 year after delivery:	115
6	comparison between diabetic patients (9 patients)&non diabetic subjects (32 subjects)1 year after delivery	116
7	Comparison between Group Ic (34 diabetic patients) and Group Id (32 non diabetic subjects ) (follow up 12 month postpartum) using t- test	117

# **List of Figures**

figures of the Review & Subjects  1 Overview of the most significant symptoms of Diabetes mellitus  2 Algorithm for prenatal GDM risk assessment  3 Structure of Islet of Langerhans  72  4 Types of cells of Islet of Langerhans  75  Figures of the Results  1 percentage of patients with positive anti islet cell antibodies among the studied population using Pie Chart  2 percentage of ICA positive diabetics and non diabetic subjects 6 monthe post partum using Pie Chart  3 percentage of ICA positive diabetics and non diabetic subjects 1 year post partum using Pie Chart  4 Comparison between group I & group II regarding age  5 Comparison between group I & group II regarding BMI  6 Comparison between group I & group II regarding FBG  7 Comparison between group I & group II regarding 2H post prandial plasma glucose  8 Comparison between group I & group II 125	Fig.	Title	Page	
symptoms of Diabetes mellitus  2 Algorithm for prenatal GDM risk assessment  3 Structure of Islet of Langerhans  72  4 Types of cells of Islet of Langerhans  75  Figures of the Results  1 percentage of patients with positive antiselet cell antibodies among the studied population using Pie Chart  2 percentage of ICA positive diabetics and non diabetic subjects 6 monthe post partum using Pie Chart  3 percentage of ICA positive diabetics and non diabetic subjects 1 year post partum using Pie Chart  4 Comparison between group I & group II regarding age  5 Comparison between group I & group II regarding BMI  6 Comparison between group I & group II regarding FBG  7 Comparison between group I & group II 123 regarding FBG  8 Comparison between group I & group II 124				
Algorithm for prenatal GDM risk assessment  Structure of Islet of Langerhans  Types of cells of Islet of Langerhans  Figures of the Results  I percentage of patients with positive anti islet cell antibodies among the studied population using Pie Chart  percentage of ICA positive diabetics and non diabetic subjects 6 monthe post partum using Pie Chart  percentage of ICA positive diabetics and non diabetic subjects 1 year post partum using Pie Chart  Comparison between group I & group II regarding age  Comparison between group I & group II 122 regarding BMI  Comparison between group I & group II 123 regarding FBG  Comparison between group I & group II 124 regarding 2H post prandial plasma glucose  Comparison between group I & group II 125	1	Overview of the most significant	13	
assessment  3 Structure of Islet of Langerhans 72 4 Types of cells of Islet of Langerhans 75  Figures of the Results  1 percentage of patients with positive anti islet cell antibodies among the studied population using Pie Chart  2 percentage of ICA positive diabetics and non diabetic subjects 6 monthe post partum using Pie Chart  3 percentage of ICA positive diabetics and non diabetic subjects 1 year post partum using Pie Chart  4 Comparison between group I & group II regarding age  5 Comparison between group I & group II 121 regarding BMI  6 Comparison between group I & group II 123 regarding FBG  7 Comparison between group I & group II 124 regarding 2H post prandial plasma glucose  8 Comparison between group I & group II 125		symptoms of Diabetes mellitus		
3 Structure of Islet of Langerhans 72 4 Types of cells of Islet of Langerhans 75  Figures of the Results  1 percentage of patients with positive anti islet cell antibodies among the studied population using Pie Chart  2 percentage of ICA positive diabetics and non diabetic subjects 6 monthe post partum using Pie Chart  3 percentage of ICA positive diabetics and non diabetic subjects 1 year post partum using Pie Chart  4 Comparison between group I & group II regarding age  5 Comparison between group I & group II regarding BMI  6 Comparison between group I & group II regarding FBG  7 Comparison between group I & group II regarding FBG  8 Comparison between group I & group II 124  Tegarding 2H post prandial plasma glucose  8 Comparison between group I & group II 125	2	Algorithm for prenatal GDM risk	49	
Figures of the Results  1 percentage of patients with positive anti islet cell antibodies among the studied population using Pie Chart  2 percentage of ICA positive diabetics and non diabetic subjects 6 monthe post partum using Pie Chart  3 percentage of ICA positive diabetics and non diabetic subjects 1 year post partum using Pie Chart  4 Comparison between group I & group II regarding age  5 Comparison between group I & group II regarding BMI  6 Comparison between group I & group II regarding FBG  7 Comparison between group I & group II regarding FBG  7 Comparison between group I & group II regarding 2H post prandial plasma glucose  8 Comparison between group I & group II 125		assessment		
Figures of the Results  1 percentage of patients with positive anti islet cell antibodies among the studied population using Pie Chart  2 percentage of ICA positive diabetics and non diabetic subjects 6 monthe post partum using Pie Chart  3 percentage of ICA positive diabetics and non diabetic subjects 1 year post partum using Pie Chart  4 Comparison between group I & group II regarding age  5 Comparison between group I & group II regarding BMI  6 Comparison between group I & group II 123 regarding FBG  7 Comparison between group I & group II 124 regarding 2H post prandial plasma glucose  8 Comparison between group I & group II 125		Structure of Islet of Langerhans		
1 percentage of patients with positive anti islet cell antibodies among the studied population using Pie Chart 2 percentage of ICA positive diabetics and non diabetic subjects 6 monthe post partum using Pie Chart 3 percentage of ICA positive diabetics and non diabetic subjects 1 year post partum using Pie Chart 4 Comparison between group I & group II regarding age 5 Comparison between group I & group II regarding BMI 6 Comparison between group I & group II regarding FBG 7 Comparison between group I & group II regarding 2H post prandial plasma glucose 8 Comparison between group I & group II 125	4	Types of cells of Islet of Langerhans	75	
islet cell antibodies among the studied population using Pie Chart  2 percentage of ICA positive diabetics and non diabetic subjects 6 monthe post partum using Pie Chart  3 percentage of ICA positive diabetics and non diabetic subjects 1 year post partum using Pie Chart  4 Comparison between group I & group II regarding age  5 Comparison between group I & group II regarding BMI  6 Comparison between group I & group II regarding FBG  7 Comparison between group I & group II regarding 2H post prandial plasma glucose  8 Comparison between group I & group II 125		Figures of the Results		
population using Pie Chart  2 percentage of ICA positive diabetics and non diabetic subjects 6 monthe post partum using Pie Chart  3 percentage of ICA positive diabetics and non diabetic subjects 1 year post partum using Pie Chart  4 Comparison between group I & group II regarding age  5 Comparison between group I & group II regarding BMI  6 Comparison between group I & group II regarding FBG  7 Comparison between group I & group II regarding YBG  7 Comparison between group I & group II regarding YBG  8 Comparison between group I & group II 124  124  125	1	percentage of patients with positive anti	118	
percentage of ICA positive diabetics and non diabetic subjects 6 monthe post partum using Pie Chart  percentage of ICA positive diabetics and non diabetic subjects 1 year post partum using Pie Chart  Comparison between group I & group II regarding age  Comparison between group I & group II 122 regarding BMI  Comparison between group I & group II 123 regarding FBG  Comparison between group I & group II 124 regarding 2H post prandial plasma glucose  Comparison between group I & group II 125				
non diabetic subjects 6 monthe post partum using Pie Chart  3 percentage of ICA positive diabetics and non diabetic subjects 1 year post partum using Pie Chart  4 Comparison between group I &group II regarding age  5 Comparison between group I &group II 122 regarding BMI  6 Comparison between group I &group II 123 regarding FBG  7 Comparison between group I &group II 124 regarding 2H post prandial plasma glucose  8 Comparison between group I &group II 125		population using Pie Chart		
partum using Pie Chart  3 percentage of ICA positive diabetics and non diabetic subjects 1 year post partum using Pie Chart  4 Comparison between group I &group II regarding age  5 Comparison between group I &group II 122 regarding BMI  6 Comparison between group I &group II 123 regarding FBG  7 Comparison between group I &group II 124 regarding 2H post prandial plasma glucose  8 Comparison between group I &group II 125	2	1	119	
percentage of ICA positive diabetics and non diabetic subjects 1 year post partum using Pie Chart  Comparison between group I &group II 121 regarding age  Comparison between group I &group II 122 regarding BMI  Comparison between group I &group II 123 regarding FBG  Comparison between group I &group II 124 regarding 2H post prandial plasma glucose  Comparison between group I &group II 125				
non diabetic subjects 1 year post partum using Pie Chart  4 Comparison between group I &group II 121 regarding age  5 Comparison between group I &group II 122 regarding BMI  6 Comparison between group I &group II 123 regarding FBG  7 Comparison between group I &group II 124 regarding 2H post prandial plasma glucose  8 Comparison between group I &group II 125				
using Pie Chart  4 Comparison between group I &group II 121 regarding age  5 Comparison between group I &group II 122 regarding BMI  6 Comparison between group I &group II 123 regarding FBG  7 Comparison between group I &group II 124 regarding 2H post prandial plasma glucose  8 Comparison between group I &group II 125	3	_	120	
4 Comparison between group I &group II 121 regarding age 5 Comparison between group I &group II 122 regarding BMI 6 Comparison between group I &group II 123 regarding FBG 7 Comparison between group I &group II 124 regarding 2H post prandial plasma glucose 8 Comparison between group I &group II 125				
regarding age  5 Comparison between group I &group II 122 regarding BMI  6 Comparison between group I &group II 123 regarding FBG  7 Comparison between group I &group II 124 regarding 2H post prandial plasma glucose  8 Comparison between group I &group II 125			101	
5 Comparison between group I &group II 122 regarding BMI 6 Comparison between group I &group II 123 regarding FBG 7 Comparison between group I &group II 124 regarding 2H post prandial plasma glucose 8 Comparison between group I &group II 125	4		121	
regarding BMI  6 Comparison between group I &group II 123 regarding FBG  7 Comparison between group I &group II 124 regarding 2H post prandial plasma glucose  8 Comparison between group I &group II 125	~	6 6	100	
6 Comparison between group I &group II 123 regarding FBG 7 Comparison between group I &group II 124 regarding 2H post prandial plasma glucose 8 Comparison between group I &group II 125	5		122	
regarding FBG  Comparison between group I &group II 124 regarding 2H post prandial plasma glucose  Comparison between group I &group II 125			102	
7 Comparison between group I &group II 124 regarding 2H post prandial plasma glucose 8 Comparison between group I &group II 125	6		123	
regarding 2H post prandial plasma glucose  8 Comparison between group I & group II 125	7	3 3	104	
glucose  8 Comparison between group I & group II 125	/		124	
8 Comparison between group I & group II 125				
	Q		125	
	0	regarding fasting insulin level	123	

# List of Figures (Cont.)

Fig.	Title	Page
9	Comparison between group I & group II	126
	regarding HOMA-IR	
10	Comparison between group Ia &group	127
	Ib Regarding FBG	
11	Comparison between group Ia &group	128
	Ib Regarding 2h postprandial plasma	
	glucose	
12	Comparison between group Ia &group	129
	Ib Regarding fasting insulin level	
13	Comparison between group Ic &group	130
	Id Regarding FBG	
14	Comparison between group Ic &group	131
	Id Regarding 2h postprandial plasma	
	glucose	
15	Comparison between group Ic &group	132
	Id Regarding fasting insulin level	

### Introduction

Gestational diabetes mellitus (GDM), defined as any degree of glucose intolerance with onset or first recognition during pregnancy, affects 3-10% of pregnancies (National Diabetes Information Clearinghouse, 2011). All pregnant women need to be screened for gestational diabetes. The timing of the screening depends on risk factor assessment. Pregnant women with not known history of diabetes are screened at 24-28 weeks gestation. Women at high risk for GDM are screened at the first prenatal visit. A 75-g 2-hour OGTT is the test of choice in both groups (American Diabetes Association, 2011).

Established risk factors for GDM are advanced maternal age, obesity, prior history of GDM or delivery of a large-forgestation-age infant (above 4000 grams or 4500 grams), presence of glucosuria, diagnosis of PCOS (polycystic ovary syndrome) and strong family history of diabetes in a first degree relative (**Perkins et al., 2007**).

Women with unmanaged gestational diabetes are at increased risk of latent autoimmune diabetes after pregnancy. Worldwide-reported incidence rates vary from 6 to 62% (*O'Sullivan* and Hiscock, 2010). The risk is highest in women who needed insulin treatment, had antibodies associated with diabetes (such as antibodies against glutamate decarboxylase, islet cell antibodies), women with more than two previous pregnancies and women who were obese (Löbner et al., 2007). Women requiring insulin to manage gestational diabetes have a 50% risk of developing diabetes within the next five years (Järvelä et al., 2010).

The appearance of diabetes-related autoantibodies has been shown to be able to predict the development of type 1

### Introduction and Aim of the Work

diabetes before even hyperglycemia arises, the main ones being islet cell autoantibodies (ICAs), insulin autoantibodies (IAA), glutamic acid decarboxylase autoantibodies (GAD). By definition, the diagnosis of type 1 diabetes can be made first at the appearance of clinical symptoms and or signs, but the emergence of autoantibodies may itself be termed "latent autoimmune diabetes". The time interval from emergence of autoantibodies to frank type 1 diabetes can be a few months up to years (**Knip et al., 2009**).

**Previous** studies have shown that pancreatic during GDM for autoantibodies are predictive development of type 1 diabetes (Füchtenbusch et al., 2009). And especially when present together with ICAs, confer a higher predictive value for the development of disease. The prevalence of ICAs in GDM varied in previous studies up to 38% (Rubenstein et al.,2011).

### **Aim of the Work:**

To study the presence of anti-Islet cells antibodies in a sample of Egyptian females with Gestational diabetes mellitus and to follow these females to estimate the risk of later development of type 1 diabetes within 6 month & one year after delivery.

### (1)Diabetes Mellitus

Diabetes mellitus is a group of metabolic diseases characterized by hyperglycemia resulting from defects in insulin secretion, insulin action, or both. The chronic hyperglycemia of diabetes is associated with long-term damage, dysfunction, and failure of different organs, especially the eyes, kidneys, nerves, heart, and blood vessels (World Health Organization Retrieved ,2014)

Several pathogenic processes are involved in the development of diabetes. These range from autoimmune destruction of the  $\beta$ -cells of the pancreas with consequent insulin deficiency to abnormalities that result in resistance to The basis of the insulin action. abnormalities in carbohydrate, fat, and protein metabolism in diabetes is deficient action of insulin on target tissues. Deficient insulin action results from inadequate insulin secretion and/or diminished tissue responses to insulin at one or more points in the complex pathways of hormone action. Impairment of insulin secretion and defects in insulin action frequently coexist in the same patient, and it is often unclear which abnormality, if either alone, is the primary cause of the hyperglycemia (Shoback et al.,2011).

### **Epidemiology of diabetes mellitus:**

Globally, as of 2010, an estimated 227 to 285 million people had diabetes, with type 2 making up about 90% of the cases (**Vos et al.,2012**). This is equal to 3.3% of the population with equal rates in both women and men. In 2011 it resulted in 1.4 million deaths worldwide making it the 8th leading cause of death. This is an increase from 1 million deaths in 2000 (**Diabetes Fact sheet ,2013**).

Its rate has increased, and by 2030, this number is estimated to almost double. Diabetes mellitus occurs throughout the world, but is more common (especially type 2) in more developed countries. The greatest increase in rates is, however, expected to occur in Asia and Africa, where most people with diabetes will probably be found by 2030. The increase in rates in developing countries follows the trend of urbanization and lifestyle changes, perhaps most importantly a "Western-style" diet. This has suggested an environmental (i.e., dietary) effect, but there is little understanding of the mechanism at present, though there is much speculation, some of it most compellingly presented (Wild et al.,2004).

### **Classification of Diabetes Mellitus:**

# Diabetes can be classified into the following general categories:

Diabetes mellitus is classified into four broad categories: type 1, type 2, gestational diabetes, and "other specific types". The "other specific types" are a collection of a few dozen individual causes (Shoback et al.,2011).

### • Type 1diabetes mellitus:

This form, previously called "insulin- dependent diabetes" or "juvenile-onset diabetes," accounts for 5-10% of diabetes and is due to cellular mediated autoimmune destruction of the pancreatic b-cells. (**Dabelea et al.,2014**).

Autoimmune markers include islet cell autoantibodies, autoantibodies to insulin, autoantibodies to GAD (GAD65), autoantibodies to the tyrosine phosphatases IA-2 and IA-2b, and autoantibodies to zinc transporter 8 (ZnT8). Type 1

diabetes is defined by the presence of one or more of these autoimmune markers. (Ziegler el al.,2013).

The disease has strong HLA associations, with linkage to the DQA and DQB genes. These HLA-DR/DQ alleles can be either predisposing or protective. The rate of beta-cell destruction is quite variable, being rapid in some individuals (mainly infants and children) and slow in others (mainly adults). (Sorensen et al.,2013)

Children adolescents and may present with ketoacidosis as the first manifestation of the disease. Others have modest fasting hyperglycemia that can rapidly change to severe hyperglycemia and/or ketoacidosis with infection or other stress. Adults may retain sufficient beta-cell function to prevent ketoacidosis for many years; such individuals eventually become dependent on insulin for survival and are at risk for ketoacidosis. At this latter stage of the disease, there is little or no insulin secretion, as manifested by low or undetectable levels of plasma C-peptide. Immune-mediated diabetes. (Sosenko et al., 2013).

### • Diabetes mellitus type 2:

Type 2 diabetes mellitus is characterized by insulin resistance, which may be combined with relatively reduced insulin secretion. The defective responsiveness of body tissues to insulin is believed to involve the insulin receptor. However, the specific defects are not known. Diabetes mellitus cases due to a known defect are classified separately. Type 2 diabetes is the most common type (Shoback et al.,2011).

In the early stage of type 2, the predominant abnormality is reduced insulin sensitivity. At this stage, hyperglycemia can be reversed by a variety of measures