Allelic variants of Insulin receptor substrate -1 Gene and its Relation to Polycystic Ovary Syndrome

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By

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Abstract

Polycystic ovary syndrome is one of the most common diseases affection women in child bearing period, its consequences range from subtle cosmetic problems as acne and hirsutism to the most detrimental as metabolic syndrome and higher susceptibility to develop diabetes mellitus, this study aimed at elucidating the correlation between gly972arg and ala512pro polymorphisms of the IRS-1 gene and their relation to PCOS. The gly972arg was found to confer positive relative risk to PCOS, while the ala512pro was not encountered in any of the subjects of this study, confirming its rarity in women of Caucasian descent.

Keywords

Polycystic ovary syndrome, Insulin receptor substrate gene-1, Insulin resistance, PCR-RFLP

List of contents

List of abbreviations

ACTH	Adrenocorticotropic hormone
AES	androgen excess society
Ala	Alanine
AR	androgen receptor
Arg	Arginine
ARMS	Amplification refractory mutation system
Asn	Asparagine
Asp	Aspartate
Beta HCG	Beta human chorionic gonadotropin
BMI	Body mass index
Bp	Base pair
CAD	Coronary artery disease
CAPN-10	Cysteine protease calpain-10
CYP	A suffix for naming any of the Cytochrome P family members
DHEA	Dehydroepiandrosterone
DNA	Deoxyribonucleic acid
EDTA	Ethylenediaminetetraacetic Acid
ELK1	Ets *(E-twenty six)* Like gene1
FGIR	Fasting glucose insulin ratio
FOC	Failure of conception
FSH	Follicle stimulating hormone
FSIVGTT	Frequently sampled intravenous glucose tolerance test
G:I ratio	Glucose to insulin ratio
Gab	Grb 2-associated binder
Gly	Glycine
GnRH	Gonadotropin releasing hormone
GLUT	Glucose transporter
HDL-C	High density lipoprotein-cholesterol
HEK	Human embryonal kidney
HOMA	Homeostasis model assessment
IGF	Insulin like growth factor
IL	Interleukin
INS	Insulin gene
INSR	Insulin receptor gene
IRS	Insulin receptor substrate gene
ISI	Insulin sensitivity index
ITT	Insulin tolerance test
LDL-C	Low density lipoprotein-cholesterol

LH	Luteinizing hormone
mRNA	messenger ribonucleic acid
Myc	myelocytomatosis viral oncogene
NIH	National institute of health
OGTT	Oral glucose tolerance test
PAI	Plasminogen activator inhibitor
PCOM	polycystic ovarian morphology
PCOS	polycystic ovary syndrome
PCR	Polymerase chain reaction
PH	Pleckstrin homology
PI3K	Phosphatidylinositol kinase-3
PPAR	peroxisome proliferator-activated receptor**s**
Pro	Proline
PTB	Phosphotyrosine-binding
QUICKI	Quantitative sensitivity check index
RAS	Rat sarcoma gene
Real time PCR	Real time polymerase chain reaction
RFLP	Restriction fragment length polymorphism
Ser	Serine
SHBG	Sex hormone binding globulin
Shc	Src Homologous and Collagen-Like Protein Shc Gene
SNP	Single nucleotide polymorphism
SSCP	Single stranded conformation polymorphism
StAR	Steroidogenic acute regulatory protein
SYBR	Synergy Brands
T2DM	Type 2 diabetes mellitus
Thr	Threonine
TNF	Tumor necrosis factor
TNFRS 1b	Tumor necrosis factor receptor superfamily 1b
TSH	Thyroid stimulating hormone
UBF1	Upstream binding transcription factor-1
UTR	Untranslated region
VNTR	Variable number of tandem repeats
WBISI	Whole body insulin sensitivity index
WGA	Genome wide association study
Y	Year

List of tables

List of figures

Introduction

Polycystic ovarian syndrome (PCOS) is one of the most common reproductive system disorders with a prevalence estimated between 5 and 10 percent of females in child bearing period (Asunción et al., 2000). An international consensus group proposed that the syndrome can be diagnosed by the determination that at least two of the following criteria are present: oligo-ovulation or anovulation (usually manifested as oligomenorrhea or amenorrhea), elevated levels of circulating androgens (hyperandrogenemia) or clinical manifestations of androgen excess (hyperandrogenism), and polycystic ovaries as defined by ultrasonography, after the exclusion of other medical conditions that cause irregular menstrual cycles and androgen excess(Rotterdam consensus, 2003).

The aetio-pathogenesis of this syndrome is not well known. Several pathogenetic hypotheses have been proposed to explain the full array of symptoms and signs.

A genetic abnormality causing PCOS is supported by the observation that different members of the same family are often affected (Fratantonio et al., 2005), family studies in first-degree relatives of women diagnosed with PCOS revealed clustering of the disease (Diamanti-Kandarakis et al., 2008) and about half of the sisters of PCOS women have elevated serum testosterone concentrations (Azziz et al., 2004) with a higher incidence of insulin resistance noted in mothers and sisters of women with PCOS (Yildiz et al., 2003).

The main genes that may play a possible role are those involved in steroidogenesis, gonadotropin release regulation and action, insulin secretion and action, and adipose tissue metabolism (Fratantonio et al., 2005).

Women with the polycystic ovary syndrome almost always have some aberration in gonadotropin secretion as compared with women who have normal menstrual cycles (Waldstreicher et al., 1988). However, since gonadotropin concentrations vary over the menstrual cycle and are released in a pulsatile fashion into the circulation, thus, in routine clinical practice, abnormal gonadotropin levels need not be documented to diagnose the polycystic ovary syndrome (Ehrmann, 2005).

Insulin plays both direct and indirect roles in the pathogenesis of hyperandrogenemia in the polycystic ovary syndrome. Insulin acts synergistically with luteinizing hormone to enhance the androgen production of theca cells. Insulin also inhibits hepatic synthesis of sex hormone–binding globulin, the key circulating protein that binds to testosterone, and thus it increases the proportion of testosterone that circulates in the biologically available free state. Because women with the polycystic ovary syndrome typically have hyperinsulinemia, the concentration of free testosterone is often elevated when the total testosterone concentration is at the upper range of normal or only modestly elevated (Ehrmann, 2005).

The association between PCOS and hyperinsulinemia has been documented for almost 29 years (Burghen et al., 1980). Later studies confirmed that thirty to forty percent of women with the polycystic ovary syndrome have impaired glucose tolerance, and as many as ten percent have type 2 diabetes by their fourth decade. These prevalence rates are among the highest known among women of similar Age (Krosnick, 2000).

Insulin receptor substrate (IRS) proteins are critical to signal transduction in insulin target tissue. Once insulin bind to the insulin receptor situated on the surface of most of the cells of the body, the intrinsic phosphotransferase function of the insulin receptor (IR) beta-subunit is activated, resulting in the tyrosine

phosphorylation of a number of intracellular proteins, including insulin receptor substrate (IRS)-1,2,3 and 4 (Bernier et al.,2000).

IRS-1 is a major cytoplasmic substrate of the insulin receptor. Following the insulin binding to the insulin receptor, the IRS-1 protein is phosphorylated, its phosphorylation allows it to associate and activate the PI3K, leading to an increase in glucose uptake (Bernier et al., 2000) and a cascade of cellular events leading to mitogenesis (Waters et al., 1993). Disruption of IRS-1 in mice causes growth retardation and insulin resistance (Araki et al., 1994) and islets from knockout mice lacking IRS-1 exhibit a marked secretory defect in response to glucose (Kulkarni et al., 1999).

The insulin receptor substrate-1 (IRS-1) which plays a central role in insulin sensitivity has been extensively studied in this respect (Sentinelli et al., 2006).

Many polymorphisms have been described in IRS-1 gene; the role of IRS genes polymorphisms was suggested by the identification of several allelic variants that are more prevalent in type 2 diabetes (Almind et al., 1999).

While many sequence variants within the IRS-1 gene have been identified, the main focus has been on two nonsynonymous variants, Ala512Pro and Gly972Arg. Most attention has been concentrated on the Gly972Arg variant, which is more common than Ala512Pro, and has stronger evidence supporting direct consequences on gene product function (Hribal et al., 2004).

The importance of these polymorphisms in PCOS lies in their proximity to the phosphatidyl inositol kinase-3 (PI3K) motif, which is an important step in the insulin signaling cascade (Lin et al., 2006).

PCOS is a good model to study influent genes, because of the complexity of disease and the variety of factors influencing its phenotype (Urbanek, 2008).

Aim of work

The aim of this study is to elucidate the association between polycystic ovary syndrome and the allelic variants Gly972Arg and Ala512Pro of insulin receptor substrate-1 gene in Egyptian PCOS patients as a possible genetic mechanism in the etiology of PCOS.

Chapter 1

POLYCYSTIC OVARY SYNDROME

Polycystic ovary syndrome (PCOS) is a genetically complex disorder that is characterized by hyperandrogenemia and amenorrhea/oligomenorrhea, resulting in the most frequent cause of infertility in females. It is also the most common endocrinopathy among women (Diamanti-Kandarakis et al., 1999) affecting approximately 105 million reproductive age women worldwide (Azziz et al., 2005). In addition to its reproductive features, PCOS is associated with an increased risk of developing obesity, insulin resistance , and type 2 diabetes mellitus (T2DM) (Urbanek, 2008).

FEATURES OF PCOS:

In an attempt to standardize the definition of PCOS, guidelines for the designation of PCOS were established, first in 1990, then in 2003 and most recently in 2006 (Table 1).

The National institute of health criteria:

The 1990 National Institute of health (NIH) Criteria requires that two criteria be fulfilled for the diagnosis of PCOS: (1) clinical (acne or hirsutism) or/and biochemical hyperandrogenemia (measured elevated androgen levels) and (2) menstrual irregularity (Zawadski et al., 1992).