Effects of Metformin on serum Insulin, Anti-Mullerian Hormone and on Androgen Levels in patients with Polycystic Ovarian Syndrome

Thesis submitted for partial fulfillment of Doctorate Degree by:

Zamzam Ahmed Dawoud, M.B.B.Ch, MSc

Assistant Lecturer of Obstetrics and Gynecology at Faculty of Medicine, Cairo University

Under supervision of:

Prof. Dr. Omar Abdel Aziz Mohamed
Prof. of Obstetrics and Gynecology
Faculty of Medicine
Cairo University

Prof. Dr. Manal Mohamed Kamal
Prof. of Chemical Pathology
Faculty of Medicine
Cairo University

Dr. Sherif Adel Dahab
Lecturer of Obstetrics and Gynecology
Faculty of Medicine
Cairo University

2015

Acknowledgement

First and before all, All praises are due to the almighty ALLAH for every step I take in my life.

I'd like to thank Prof. Dr. **Omar Abdel Aziz** for his guidance, patience and support.

I'm very grateful to Prof. Dr **Manal Kamal** for her advice and ideas that helped make this work the way it is now.

As for Dr. **Sherif Dahab**, I owe him a lot for his continuous support, follow up, encouragement and even direct help, guiding me with patience throughout this whole work.

Finally, I'd like to thank my family for their patience and support.

Table of Contents

List of Tables

List of Figures

Abstract

Objective: Evaluate the effect of metformin on serum insulin (fasting and 2hr postprandial), androgens (total and free testosterone, DHEAS and SHBG) and AMH in PCOS patients at 3 and 6 months interval.

Design: Prospective study

Setting: Kasr Al Aini University Hospital, Obstetrics and Gynecology Department.

Patients: 50 hyperandrogenic PCOS patients.

Interventions: 1700 mg metformin was given for a period of 6 months; serum insulin, previously mentioned androgens and AMH were measured at baseline, 3 months and 6 months of treatment.

Main outcome measure: serum fasting and 2 hrs. postprandial insulin and glucose levels, insulin resistance, serum total and free testosterone, DHEAS and SHBG and serum AMH. The impact of treatment and biochemical changes on menstrual regularity, clinical manifestations of hyperandrogenism, ovarian morphology and fertility.

Results: Metformin treatment produced a significant reduction in insulin, serum androgens and AMH levels. 62% of the patients had improvement in their cycle regularity and 2 out of 50 patients had spontaneous pregnancy. However, there were no improvement in hyperandrogenic manifestations or change in ovarian morphology.

Conclusion: Metformin as an insulin sensitizing agent improved insulin resistance in PCOS patients and improved biochemical but not clinical hyperandrogenism. Metformin produced significant reduction in AMH levels and modest improvement in cycle regularity; but no change in the ovarian morphology. Metformin could not be used for the treatment of hirsutism or in improving fertility in PCOS.

Keyword

PCOS-AMH-ACTH-DHEA- AE-PCOS

List of Abbreviations

3 B HSD 3 Beta Hydroysteroid Dehydrogenase enzyme

ACTH Adrenocorticotrophic Hormone

AE-PCOS Androgen Excess and PCOS Society

AMH Anti- Mullerian Hormone

AMHR2 Anti mullerian Hormone receptor type 2

ASRM American Society of Reproductive Medicine

AT Androstenedione

BMI Body Mass Index

BMP Bone Morphogenic Proteins

DHEA DiHihudroepiandrostenedione

DHEA-S Dihyrdroepiandrosterone Sulphate

ESHRE European Society for Human Reproduction and Embryology

FSH Follicle Stimulating Hormone

GCs Granolosa Cells

GH Growth Hormone

GLCs Granulosa Luteal Cells

GLP-1 Glucagon like Peptide 1

GLUT Glucose Transporter

hCG Human Chorionic Gonadotropin

HOMA Homeostatic Model Assessment

HPO Hypothalamo-pituitatry ovarian

IFT Impaired Fasting Glucose

IDT Impaired Glucoe Tolerance

INSR Insulin resistance

IGF-1 Insulin like Growth Factor- 1

IRS Insulin Recptor Substrate

ITT Insulin Tolerance Test

IVF In Vitro Fertilization

LDL-p Low Density Lipoprotein Particles

LH Leutinising Hormone

PAI-1 Plasminogen Activator Inhibitor 1

PMAT Plasma Membrane monoamine Transporter

SHBG Sex Hormone Binding Globulin

SOGC Society of Obstetricians and Gynaecologists of Canada

StAR Steroid Acute Regulatory Protein

TGF- β Transforming Growth Factor β

NICHD National Institute of Child Health and Human Disease

NIH National Institute of Health

NIDM Non Insulin Dependent Diabetes Mellitus

OCPs Oral Contraceptive Pills

PCOS Polycystic Ovarian Syndrome

QUICKI Quantitative insulin sensitivity check index

Introduction

Polycystic Ovary Syndrome (PCOS) is the most common endocrinological disorder affecting 4-12% of reproductive aged women ***(Diamanti-Kandarakis et al 1999, Farah et al, 1999, Knochenhauer et al, 1998).*** It has also been the most medical condition that caused a lot of controversy in its every aspect ***(Lashen 2010).***

PCOS is a disorder with a combination of reproductive and metabolic characteristics ***(Lashen 2010)***. Diagnosis of PCOS should include at least two of the following features: oligo/anovulation, hyperandrogenism and enlarged ovaries with multiple follicles ranging in size from 2-10 mm by ultrasound scan (ESHRE/ASRM, 2004). However the Androgen Excess Society recommends that androgen excess should remain a constant feature of PCOS, regardless of the morphological appearance of the ovaries and their ovulatory function ***(Azziz et al, 2006***). PCOS is a life course disease, that in addition to its reproductive impact, it has long term effects with the risk of developing Type II Diabetes mellitus and the metabolic syndrome ***(Apridonidze et al, 2005)*** as well as cardiovascular disease (***Grundy, 2002).***

Insulin resistance is a condition characterized by hyperinsulinemia secondary to failure of the target cells to respond to normal or ordinary levels of insulin ***(Le Roith and Zick, 2001).*** IR has been linked to the pathogenesis of PCOS through the effect of hyperinsulinemia on increasing serum androgen levels via a central role ***(Barbieri et al, 1986)*** or by decreasing the circulating levels of SHBG ***(Nestler et al, 1991).***

Hyperinsulinemia has also been attributed to increased AFC and ovarian volume in PCOS women (***Pache et al, 1993, Carmina et al, 2005)***. It is unclear if PCOS related hyperinsulinemia state could induce the development of antral follicles by increasing the sensitivity of granulosa cells to FSH, thus resulting in increased ovarian volume and higher follicular count seen by ultrasound in such patients ***(Loucks et al, 2000, Fulghesu et al, 1997).***

Though IR is not considered a diagnostic criteria in PCOS ***(ESHRE, ASRM, 2004)***, yet it is recognized by many as a common feature in PCOS independent of obesity ***(Burghen et al, 1980, Chang et al, 1983, Dunaif et al, 1987)***. The prevalence of IR has been estimated to be 60-70% ***(De- Urgarte et al, 2005).***

PCOS is also characterized by elevated AMH levels. The relationship between elevated serum AMH levels and pre-antral follicular number and PCOS patients has been a matter of controversy **(Piltonen et al, 2005, Chen et al, 2008, Catteau-Jonard et al, 2008).** Therefore, it is still unknown if AMH excess in PCOS is secondary to an intrinsic increase in AMH production by the granulose cells ***(Angela- Talbu et al, 2010).***

Therapies directed for treatment of PCOS, had arrived at reducing hyperinsulinemia as a contributing factor to hyperandrogenism (***Lashen, 2010).*** The biguanide metformin is an insulin sensitizing drug routinely used for its antidiabetic effect in NIDDM ***(Bailey, Turner 1996)***. Metformin as antihyperglcemic action and does not cause clinical hypoglycaemia, as it acts by decreasing basal hepatic glucose production and increases glucose uptake in insulin stimulated state after meals ***(Bailey 1992, Dunn, Peters 1995).***

Metformin action on insulin resistance had been investigated in non-diabetics, where improvements in insulin sensitivity have been reported in first

degree relatives of diabetics ***(Widen 1992),*** subjects with upper body obesity (***Schen et al, 1995)*** and hyperinsulinemic hypertensive men (***Burella et al, 1986).***

The effect of metformin on androgen production has been controversial ***(Arif et al, 2001, Bailey and Puah, 1986).*** ***Attia et al*** demonstrated in invitro experiments in 2001, that metformin significantly inhibited both androstenedione and testosterone production by the cells. In addition***, Bailey and Turner*** suggested in their review in 1986, that metformin reduces hyperandrogenism through suppression of adrenal production of both the ovary and adrenal gland. In addition, metformin also reduces pituitary LH and increases the production of SHBG by the liver ***(Bailey and Turner, 1996).*** On the other hand, Harborne and colleagues reported no significant change in SHBG in hirsute patients ***(Harborne et al, 2003).*** They assigned that improvement of symptoms was due to reduction of insulin levels.

Concerning the effect of metformin on ovarian morphology and volume, the available data are scarce ***(De Leo V et al 1999, Stadtmauer et al, 2001 and Elter et al, 2002)***. Similarly, the effect of metformin on serum AMH. Some authors demonstrated a reduction in serum AMH levels during metformin treatment ***(Terhi et al, 2005)***. Others demonstrated no effect on serum AMH ***(Nasciemento et al, 2013).***

Aim of Work

- Evaluate the effect of metformin treatment on serum insulin, andorgens and AMH in PCOS patients.
- The role of metformin in improving cycle regularity in PCOS patients.
- The role of metformin in improving fertility in PCOS patients and affecting their response to ovulation induction.