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Assessment of Anti-Mullerian hormone, Fertility Hormones (LH, TSH, prolactin and testosterone) and Leptin Hormone Among obese with Poly cystic ovary Syndrome.

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Abstract

Polycystic ovarian syndrome (PCOS) is a complex endocrine disorder and is the leading cause of infertility, affecting approximately six to eight percent of reproductive-age women and is associated with obesity. Anti-Müllerian hormone (AMH) has been a marker tool for diagnosis of PCOS and fertility beside fertility hormones. So this study concerned about PCOS, fertility hormones among obese infertile women compared with obese fertile ones. They were at reproductive age, recruited in Khartoum state fertility centers. Whole blood samples were collected and assessed for AMH, luteinizing hormone (LH), follicle-stimulating hormone (FSH), testosterone and prolactin as they were obese. Comparing measured parameters between PCOS obese women and control group contained fertile obese women, revealed increased levels of all parameters among PCOS obese women, except FSH. Anti-müllerian hormone (AMH) levels showed increased levels among women with PCOS was to be 9.20 Pmol and 2.99P mol/l for obese control A significant difference (p=0.000) was found between obese and non-obese groups. The average concentration for leptin was 24.8ng/ml for obese control, 34.5ng/ml for obese PCOS. A significant difference (p=0.000) in the average concentration was found between obese and non-obese groups.

Keyword: leptin. Poly cystic ovary syndrome, Anti-Müllerian hormone, Prolactin.

Introduction

Anti-mullerian hormone (AMH) is a glycoprotein which is involved in tissue growth and differentiation (Durlinger, *et al*, 2002), it has been predominantly known for its role in male sexual differentiation (Jost, 1947). In women, AMH expression is restricted to one cell type: the granulosa cells of the ovary. It starts around the 25th week of gestation continuing until menopause (Rajpert-De Meyts, *et al*, 1999) (Kuiri-Hanninen, *et al*, 2011). AMH is expressed at all steps of folliculogenesis. It is initiated as soon as primordial follicles are recruited to grow into small preantral follicles and its highest expression is observed in pre antral and small antral follicles. AMH expression then decreases with the selection of follicles for dominance and is no longer expressed during the FSH

dependent stages of follicular growth (except in the cumulus cells of pre ovulatory follicles), or in atretic follicles (Salmon, *et al*, 2004) (Durlinger, *et al*, 2002). Its levels reflect the size of the ovarian follicular pool. The main function of AMH seems to be the inhibition of the early stages of follicular development and of the FSH-dependent selection process (Köninger, *et al*, 2014).
Polycystic Ovary Syndrome (PCOS) is the most common cause of chorionic anovulation and anovulatory infertility (Wood, *et al*, 2007). PCOS is mentioned as a common endocrinopathy in women who are at reproductive age and it is associated with metabolic disorder and reproductive dysfunction (Azziz, *et al*, 2004). Ovarian dysfunction continues to be the main feature which makes this syndrome the major cause of anovulatory associated with infertility (Baker, *et al*, 2007). PCOS is a disorder affecting up to 6%-10% of women in reproductive age (Rackow, 2012).

Recently the prevalence of obesity has increased globally and reached pandemic proportions. In high-income countries, approximately one-third of adults are obese and one-third are overweight (Ng, *et al*, 2014). This rise in the prevalence of obesity has important health-care implications, since obesity is an important risk factor for cardiovascular disease and all-cause mortality (Lu Y, *et al*, 2014) (Di Angelantonio, *et al*, 2016).

A critical body mass of adipose tissue is essential for the normal development of female reproductive functions (Tataranni, *et al*, 1997). Reproductive anomalies in young pre-menopausal women seem to occur as a consequence of the linear decline in adiposity. Obesity, on the other hand, has been shown to produce derangement of female reproductive functions and infertility (Miller, *et al*, 1998).

A number of studies evaluated the association between obesity and the outcome of assisted reproduction in patients with PCOS (KonstantinosTziomalos1 and KonstantinosDinas, 2018).

Leptin, an adipocyte-derived hormone been proposed as the peripheral signal indicating the adequacy of nutritional status for reproductive functions (Almog, *et al*, 2001). But contradictory reports are available pertaining to how leptin modulates or is being modulated by pituitary gonadotropins or sex steroids. Polycystic ovarian syndrome (PCOS) is often associated with obesity and insulin resistance, both of which are features that are linked to leptin and its receptors. Serum level of leptin is higher in obese women. Obesity modifies insulin sensitivity and gonadotropin dynamics and is associated with disorders of ovulation. But, the relationship between leptin and insulin sensitivity, sex steroids, and insulin concentration in derangement of ovarian function still remain elusive and controversial (Maliqueom, *et al*, 1999).

Women with PCOS are characterized by hyperandrogenemia, increased leutinizing hormone (LH) concentrations and obesity (Christine, et al, 2010). The aim of this study to evaluate if women with PCOS differ from women in general regarding levels of AMH, leptin and reproductive hormones (Testosterone, LH, FSH and prolactin), and to compare changes in these variables in the same control.

Materials and Methods

This case control study was conducted among 55 obese infertile women diagnosed with poly cystic ovary syndrome set as case group, their age's mean 29.9 years, with BMI 31.4 kg/m^2, and 28 obese fertile healthy women with BMI 28.7 kg/m^2 set as control group, their age's mean 30.6 years, whole blood samples were collected under

hygienic conditions, serum collected and measured for anti-mullrien hormone, fertility hormones (LH, FSH, testosterone and prolactin) and leptin hormone by means of enzyme linked immune sorbent assay (ELISA). The reagent used in the estimation of sex hormones Bio vision 155 s. Milpitas Blvd., Milpitas, CA 95035 USA. The reagent used in the estimation of (AMH)Beckman Coulter, Inc. 250 S. Kraemer Blvd. Brea, CA 92821 U.S.A. Made in U.S.A. Revised March 2018. Bio vision's Human Leptin ELISA (Enzyme-Linked Immunosorbent Assay) kit is an in vitro enzyme-linked immunosorbent assay for the quantitative measurement of human Leptin. This assay employs an antibody specific for human Leptin coated on a 96-well plate.

Statistical analysis was performed using SPSSl7.0 statistical software. Measurement data were expressed as mean ± standard deviation (M ± S). Data were tested for normality and homogeneity of variance and compared using either t test (equal variances) or t' test (unequal variances). Significance level $\alpha = 0.05$, $P < 0.05$ was considered statistically significant.

Results

This case control group study involved obese infertile women diagnosed with PCOS and **obese** fertile women set as control. AMH, LH, FSH, testosterone and prolactin hormones were assessed among both groups. The measurement revealed that assessed hormones were increased among obese PCOS women than obese fertile women giving significant difference, except FSH which didn't produce any different as in table (1). Correlation of leptin hormone among obese PCOS and body mass index, showed positive correlation (Figure 1).

Table (1): Mean concentration comparison of fertility hormones among obese control and obese PCOS patients.

Parameters	Obese control (Mean±SD)	Obese PCOs (Mean±SD)	*P-value*
LH (mlU/ml)	4.40±2.72	12.90±9.86	0.000
FSH (mlU/ml)	9.08±5.17	8.30±7.60	0.627
TES (nmol/L)	1.28±0.81	6.53±1.87	0.000
AMH (Pmol/l)	2.99±0.91	9.20±4.10	0.000
PRL (mlU/L)	174.29±120	506±244	0.000
leptin ng/ml	24.8	34.5	0.000

Results expressed as Mean ± SD. P-value ≤ 0.05 consider as significant difference

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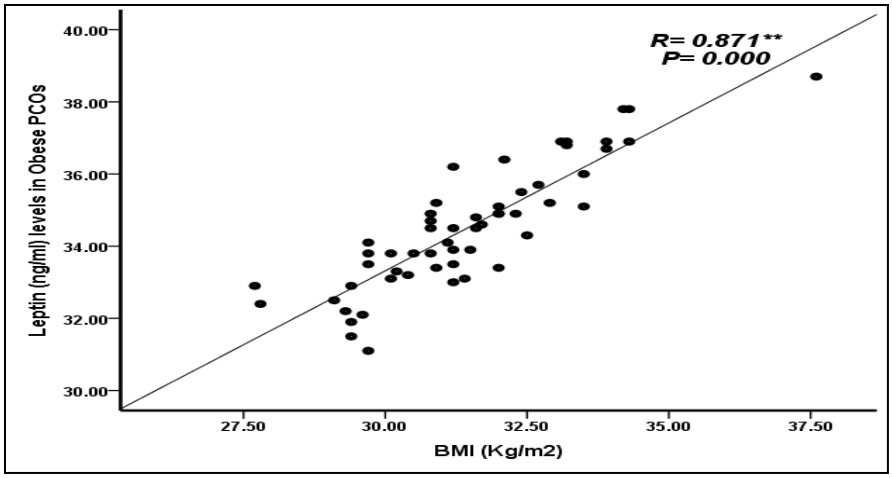

Figure1: correlation between Leptin levels and BMI among obese PCOs patients

Discussion

Polycystic ovary syndrome is a multifactorial, complex genetic, endocrine and metabolic disorder. Characteristics are chronic anovulation, polycystic ovaries and hyperandrogenism (Witchel, 2006). PCOS has a tremendous negative impact on the physiology and metabolism of the body such as metabolic syndrome, insulin resistance, dyslipidemia, hypertension, abdominal obesity and type 2 diabetes mellitus (Moran, *et al*, 2011).

In this study almost total coverage of fertility status was obtained by measurement of AMH, fertility hormones and leptin hormone among obese women diagnosed with PCOS and obese women, who were fertile and no PCOS for them. The assessment showed mean±SD of hormones among obese PCOS and obese non-PCOS respectively AMH (9.20±4.10) and (2.99±0.91), LH (12.90±9.86) and (4.40±2.72), prolactin (506±244) and (174.29±120), testosterone (6.53±1.87) and (1.28±0.81) and leptin (34.5) and (24.8), all gave significant difference while FSH was decreased among obese PCOS women (8.30±7.60) than obese fertile ones (9.08±5.17) with no significant difference. Correlation of leptin hormone and BMI revealed positive correlation among PCOS obese women.

On other study, it conducted through measuring AMH levels for 489 women. Of these, 104 were diagnosed with PCOS. in the women with PCOS, there was a significant association between BMI and AMH ($r -0.31$, $p < 0.01$) (Maya Kriseman, *et al*, 2015).

A study concerned also about body weight and fertility of women with PCOS. The study conducted through medical records of 350 women (159 Caucasian, 99 African-American, 58 Hispanic, 34 Asian with ages 16–46) evaluated for infertility at an academic-affiliated center and who had AMH levels measured as part of their evaluation were reviewed. Age, AMH, body mass index (BMI), self-reported race, etiology of infertility, smoking history, maximum serum early follicular follicle-stimulating hormone (FSH) levels, antral follicle count (AFC), and history of ovarian surgery, chemotherapy, or radiotherapy were recorded. Age correlated negatively with AMH and antral follicle count across all races ($p < 0.05$). After adjusting for age, polycystic ovary syndrome diagnosis, and smoking, elevated BMI had a negative correlation with AMH in Caucasian women

($\beta = 0.17$, $p = 0.01$) but not in African-American, Hispanic, or Asian women (Vicky Moy , *et al*, 2015).

Other study bound obesity with development of PCOS it reviewed the recent literature regarding the mechanisms linking the development of PCOS and obesity in adolescent girls and found that excess abdominal adipose tissue (AT) initiates' metabolic and endocrine aberrations that are central in the progression of PCOS. As an example, abdominal AT impairs insulin action, which interacts with the progression of hyperandrogenism. In addition, excessive androgen levels lead to impaired glucose uptake, which also contributes to insulin resistance, which again increases the deposition of visceral fat. The body composition is influenced by testosterone, which decreases subcutaneous fat lipolysis and influences adipocyte distribution. These mechanisms may explain why PCOS girls have an increased visceral adipose mass independent of body mass index (Vilmann, *et al*, 2012).

Hyun evaluated the leptin and AMH levels, but no PCOS women. To explore the relationship of insulin resistance (IR) and adipokine (leptin) to anti-Mullerian hormone (AMH) levels in women without polycystic ovary syndrome (PCOS), recruited 120 healthy, reproductive age women without PCOS. An overnight fasting blood draw, anthropometric measurements, analyses of serum levels of AMH, leptin and total testosterone, serum AMH levels negatively correlated with insulin, fasting glucose and a positive correlation was identified between serum AMH and adiponectin. A final multiple stepwise linear regression demonstrated that homeostasis model assessment for insulin resistance was independently associated with AMH, possibly due to the effect of abnormal insulin action on AMH secretion by granulosa cells (Hyun, *et al*, 2009).

An Indian study agrees with this study as it involved thirty-six female subjects of reproductive age, the inclusion criteria was the presence of acne (irrespective of grade) with one or more of the following characteristics:, weight gain, menstrual disturbances. The typical age of the study participants ranged from 16 to 39 years. FSH, LH, serum testosterone (total), anti-Müllerian hormone (AMH), thyroid stimulating hormone (TSH), and serum prolactin were measured. Of the 33 study participants 14 (45%) showed the presence of PCOS. It has been shown that the development of acne was a symptom of hyperandrogenism, as increased levels of prolactin and testosterone. High LH and low FSH was found as well (Sujata Mehta-Ambalal, 2017).

Conclusion

PCOS is related to increased levels of, testosterone prolactin, LH and presence of high AMH to diagnosis poly cystic ovary syndrome.

Lifestyle in general and specific for females should be adjusted to gain healthy long life and pre-assessment of fertilization issues and treatment programs should be applied.

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