

Correlation Between Insulin, Leptin and Polycystic Ovary Syndrome

Mohamed Nabih El-Gharib, Thoraya Elsayed Badawy¹

Departments of Obstetrics and Gynecology, ¹Clinical Pathology, Faculty of Medicine, Tanta University, Tanta, Egypt

ABSTRACT

Background: Polycystic ovary syndrome (PCOS) is the most common endocrine disorder among women of fertile age. Insulin can stimulate ovarian androgen production in normal women and in women with PCOS. Leptin levels were reduced among women with PCOS treated with insulin sensitizers. **Aim:** This study aims to investigate the serum levels of insulin and leptin in and their relationship with the endocrine and metabolic peculiarities of PCOS. **Subjects and Methods:** The study was carried out on 125 women during the reproductive age group, had primary infertility and diagnosed as having PCOS according to Rotterdam criteria. Patients were recruited from Tanta University Hospital, during the period May 2011 to December 2012. Cases were classified into two groups: Patient's group comprised 75 women having PCOS and control group entailing 50 normal fertile healthy women. All women were subjected to: History taking, clinical examination, assessment of body mass index (BMI), ultrasonographic examination and hormonal assay to estimation serum leptin, insulin, follicle stimulating hormone, luteinizing hormone, progesterone and free testosterone value. Statistical analysis was performed by Statistical Product and Service Solutions (IBM, USA) version 10.0. Student t-test was used to compare numerical variables while correlation was performed to determine the relationship between the variables. The value of $P < 0.05$ was taken as significant. **Results:** We found a significant positive correlation between leptin, BMI, age and a trivial upbeat correlation between leptin and insulin. The most interesting result is the significant positive correlation between insulin and leptin. **Conclusion:** There is a link between elevated serum leptin and insulin levels to obesity in PCOS suggesting that most probably they are responsible for the complicated picture of PCOS in obese patients.

KEY WORDS: Hyperinsulinemia, insulin, leptin, obesity, polycystic ovary syndrome

INTRODUCTION

Polycystic ovary syndrome (PCOS) is the most common endocrine disorder among women of fertile age. The prevalence of PCOS varies between 2.5% and 7.5%.^[1] Its clinical manifestations include menstrual dysfunction and hyperandrogenic symptoms and it can be associated with metabolic dysfunction in which hyperinsulinemia and peripheral insulin resistance are central features.^[1]

Insulin can stimulate ovarian androgen production in normal women and in women with PCOS. However, ovarian cells of women with PCOS display a higher responsiveness for insulin-stimulated androgen synthesis *in vitro*. Non-obese and normoinsulinemic women with PCOS significantly improve their serum testosterone levels after

serum insulin levels are reduced. It is therefore, probable that women develop PCOS because of a hypersensitivity of their intra-ovarian insulin androgen signaling pathway. The characterization of this potential defect could have significant implications for the development of specific and more efficient treatments of PCOS.^[2]

Leptin, an appetite suppressant hormone produced in fat tissue, delivers at least a partially overlapping message to the neurons that critically control energy balance. Leptin plays a significant role in the regulation of body-fat mass in animals and in humans.^[3]

Leptin deficiency and PCOS appear to have a lot of similarities between their clinical, metabolically and biochemical features. However, in previous studies that have examined the relationship between leptin and PCOS, inconsistent findings have been reported. Although those with PCOS generally have significant insulin resistance, several investigators have demonstrated that leptin levels

Access this article online

Quick Response Code	Website: www.jbcrcs.org
	DOI: ***

Address for correspondence

Prof. Mohamed Nabih El-Gharib,
Departments of Obstetrics and Gynecology,
Faculty of Medicine, Tanta University, Tanta, Egypt.
E-mail: mohgharib@hotmail.com

are higher rather than lower in women with PCOS compared with weight-matched control women, which leads to the conclusion of that insulin and leptin levels are positively correlated, implying a scenario that includes both insulin and leptin resistance.^[4]

Leptin levels were shown to be reduced among women with PCOS treated with insulin sensitizers, Diazoxide and Metformin, suggesting that improved insulin sensitivity with associated decreased circulating insulin levels diminishes the insulin-mediated stimulation of leptin production among affected women. Conversely, others have reported that independently of obesity, leptin levels do not differ significantly between women with PCOS and controls.^[5]

The aim of this work was to investigate the serum levels of insulin and leptin in and their correlation with the endocrine and metabolic peculiarities of PCOS.

SUBJECTS AND METHODS

Subjects

This study was carried out on 125 females attending the out-patient clinic of Tanta University Hospital seeking conception assistance. They were informed about the aim of the work and a written consent was taken from them. All women were in the reproductive age group, had primary infertility and diagnosed as having PCOS according to Rotterdam criteria. The most important exclusion criteria were: Women with other infertility factors, age over 30 years, receiving medicaments for any other diseases, women who had had induction of ovulation in previous cycles within 6 months, evidence of systemic diseases, women on hormonal contraception, women on dietary food regimen and women with other causes of hirsutism and oligomenorrhea. Patients included within this study were classified under three groups Patient's group: Included 75 women having PCOS.

Control group: This group included 50 normal fertile healthy women, received no hormonal medication during the last 6 months preceding the study (control group).

Methods

All women included within this study were subjected to: History taking, clinical examination and assessment of body mass index (BMI), ultrasonographic examination.

Informed consent was obtained from all patients according to local procedures in accordance the declaration of Helsinki. The ethical committees at the department approved the protocol.

A total volume of 5 ml of the venous blood was taken from each case in the morning of the 3rd day throughout the cycle to measure fasting serum concentrations of follicle stimulating hormone (FSH), luteinizing hormone (LH) and free testosterone values. The hormonal assays were

carried out by enzyme-linked immunosorbent assay (ELISA) technique. ELISA kits for leptin and insulin were supplied by DRG laboratories Ltd. ELISA kits for progesterone and testosterone were supplied by Dia Metra Laboratory Limited. Estimations of serum levels of FSH and LH were performed by radio-immunoassays.

Serum concentrations of leptin, insulin and progesterone were measured in the morning, fasting, of all cases during the mid-cycle period.

Statistical analysis was performed by Statistical Product and Service Solutions version 10.0. (IBM, USA) Student *t*-test was used to compare numerical variables while correlation was performed to determine the relationship between the variables. The value of *P* < 0.05 was taken as significant.

RESULTS

The clinical data of the studied cases are depicted in Table 1. It is evident that members of both the control and PCOS were well-matched concerning age and BMI.

The hormonal profiles of the studied cases clarify that women having PCOS had significantly higher serum concentrations of LH, free testosterone, insulin and leptin than the corresponding values of the control cases. On the contrary, serum progesterone was appreciably lower for PCOS cases than in the controls [Table 2].

Serum leptin level rises in both studied groups (control and PCOS) with raised BMI as demonstrated in Table 3. However, the increase in serum leptin was much higher in PCOS group than the controls, particularly among overweight and obese cases suggesting a sort of relationship with PCOS.

Serum insulin was insignificantly higher in PCOS than in the control cases. The rise in insulin value is in direct proportion to the body weight. These facts are shown in Table 4.

Statistical analysis on the results, using Pearson's correlation matrix, as depicted in Table 5, the presence of significant positive correlation between leptin and age, leptin and BMI. On the other side, we found substantial negative correlation between insulin and age and between BMI and LH:FSH ratio.

Table 1: The clinical data of the studied patients

The symptoms and ovarian U/S picture	Patient's group (mean±SD)	Control group (mean±SD)	P value
Age (years)	25.5 (3.9)	24 (3.7)	0.034
BMI (kg/m ²)	24.3 (2.6)	25.7 (2.5)	0.025
Infertility duration (in years)	3.6 (1.6)		
Cycle pattern		50	
Regular	26		
Oligomenorrhea	49		
Amenorrhea	0		
Hirsutism			
No	40		
Mild	20		
Moderate	15		
Severe			

NS – Non-significant; BMI – Body mass index; SD – Standard deviation

DISCUSSION

Women with PCOS are hyperandrogenic and most of them are obese. The impact of obesity is usually considered to operate through the associated insulin resistance.^[6]

It has become apparent that insulin resistance and hyperinsulinemia play a critical role in PCOS pathogenesis. Despite advances over the past decade, many of the questions remain regarding both the nature of insulin resistance in PCOS and the mechanism by which insulin resistance or insulin produces hyperandrogenemia.^[2]

The present study was designed to assess serum levels of insulin and leptin in and their correlation with the endocrine and metabolic peculiarities of PCOS. In this study, we found that the mean patient's age was $25.2 \pm (3.8)$ years. We found

that leptin levels alter significantly with age. This does not agree with the results of other previous studies that found that serum leptin is not affected by changes in age.^[7]

The mean BMI of women with PCOS was $24.9 (2.6)$ kg/m². The mean duration of infertility (in the PCOS groups I and II) was $3.8 (1.7)$ years. Women of the control group were in the obese side as their mean BMI was slightly higher than 25 kg/m² as PCOS cases that are to remove bias of obesity.

Women included within this study represent a homogenous group of the population, as regards age, BMI and infertility duration with insignificant differences. When comparing the clinical characteristics of the PCOS women in both groups, there were more women with hirsutism in the clomiphene citrate (CC) non-respondent group than in the CC respondent women, but this was statistically insignificant.

In the present study, we noticed that serum leptin level was significantly elevated in women with PCOS when compared to women with normal menstrual cycles and matched BMI. Serum leptin concentrations in women with PCOS have been reported to be higher than^[8-10] or similar to^[11,12] those in weight matched controls. On the contrary, Caro reported that leptin mean value was not different in PCOS patients compared with the normal controls.^[13]

Okasanen *et al.* concluded that PCOS is not a consequence of mutations of the leptin or leptin receptor genes, and they stated that the hypothesis that variations in the leptin receptor gene locus affect insulin regulation and this hypothesis was further supported by our study.^[14]

By comparing serum fasting insulin concentration in different groups of our present study, we found that there was a significant hyperinsulinemia in both PCOS groups when compared with the control group ($P < 0.5$). In addition, the results showed more significance in the second group (CC resistant group) with high significant difference ($P < 0.01$). Our findings agree with those reported by Ehrmann *et al.*,^[15] who found significant higher fasting insulin in women with PCOS than normal controls. Our results also go with that found by Tarkun *et al.*^[16]

On the contrary, our results do not agree with those who found a non-significant difference between PCOS patients and control group in regard to serum insulin; although it was higher among the PCOS group, this can be explained by the fact that most of their studied PCOS women were not obese.^[17]

Numerous studies have demonstrated that any treatment aimed at improving insulin resistance in women with PCOS results in lower androgen levels and improves ovulatory

Table 2: The serum hormonal profile of the studied cases

Hormone	Control mean (SD)	Patients mean (SD)	P value
FSH (mIU/ml)	5.1 (1.2)	5.0 (1.2)	0.40
LH (mIU/ml)	5.5 (1.4)	8.5 (2.5)	0.04
LH:FSH ratio	1.1 (0.2)	1.6 (0.7)	0.03
Free testosterone (pg/ml)	1.2 (0.1)	1.5 (0.9)	0.04
Progesterone (ng/ml)	5.5 (1.8)	2.5 (0.8)	0.02
Insulin (μ U/ml)	12.6 (3.3)	16.3 (3.5)	0.04
Leptin (ng/ml)	20.8 (2.7)	26.5 (7.0)	0.04

FSH – Follicle stimulating hormone; LH – Luteinizing hormone; SD – Standard deviation

Table 3: Serum leptin concentrations in relation to BMI in the studied groups

BMI (kg/m ²)	Control	Patients
Lean (<25)	19.8	24.1
Overweight (25-30)	20.82	28.8*
Obese (>30)	27.2	30.7*

*Statistically significant at $P < 0.05$ using spearman correlations test. Notice that serum leptin concentrations were higher in obese than in non-obese women ($P < 0.05$). BMI – Body mass index

Table 4: Serum insulin concentration in relation to BMI in the studied groups

BMI (kg/m ²)	Control	Patients
Lean (<25)	11.2	15.1
Overweight (25-30)	14.2	17.3
Obese (>30)	11.6	20.1

N.B. There is no significant differences between PCOS and control cases. PCOS – Polycystic ovary syndrome; BMI – Body mass index

Table 5: Pearson's correlation matrix among cases with PCOS

	Age	BMI	LH:FSH	Leptin	Insulin
Age	-	0.17	-0.21	0.04*	-0.02*
BMI	0.17	-	-0.04*	0.02*	0.20
LH:FSH	-0.21	-0.04*	-	0.20	0.16
Leptin	0.04*	0.02*	0.2	-	0.09
Insulin	-0.02*	0.2	0.16	0.09	-

*Statistically significant at $P < 0.05$ using spearman correlations test. N.B. It is clear that in women with PCOS, there is a significant positive correlation between serum leptin, BMI and patient's age. PCOS – Polycystic ovary syndrome; BMI – Body mass index; FSH – Follicle stimulating hormone; LH – Luteinizing hormone

function.^[4] Yet, this hypothesis was supported by other results, either concerned with a normal insulin level in PCOS patients^[18] or demonstrated a similar degree of insulin resistance in ovulatory and anovulatory obese subjects.^[19,20]

In the present investigation, we found a positive correlation between BMI and serum leptin in different groups of this study with a more positive correlation in the CC non-responders group. Furthermore, there was a positive correlation between BMI and serum fasting insulin in different groups within the study with a more positive correlation in PCOS and CC responders group. These results support the previously published data demonstrating a positive correlation between serum leptin, levels and BMI.^[11,21] The relationship between BMI and leptin concentrations was present in women with PCOS as well; these results came in agreement with other studies.^[16,22,23] However, Kale-Gurbuz *et al.* reported that among obese adolescents with PCOS, adiponectin and leptin levels do not seem to be determined by the existence of PCOS.^[24]

It can be seen that there was a positive correlation between leptin, BMI and age and this was statistically significant. Although a positive correlation was found between leptin and insulin but couldn't reach a statistically significant result. Yet, a positive correlation was found between age and insulin and this was statistically significant. These results suggest that it may be not the insulin that plays the major role in the pathophysiology of the syndrome and that increased insulin is probably an effect rather than a cause in obese PCOS patients and this in agreement with Conn.^[25]

The most outstanding finding of our current work was the significant positive correlation that detected between serum insulin and leptin levels. This agrees with that stated by Bloomgarden^[26] who found that insulin acutely increases plasma leptin and reduces adiponectin, suggesting a role of hyperinsulinemia in insulin resistance, which is associated to an increase in adipocyte inflammatory factors.

As PCOS is a well-characterized state of insulin resistance with compensatory hyperinsulinemia,^[27] it was not surprising that the hyperinsulinemia in obese women with PCOS results in a highly significant increase of leptin levels within this group compared with age and weight-matched control in the present study.

It seems that elevated serum leptin level seems to be the responsible mediator for the pathology of PCOS, most probably through dual mechanism. The first mediated at the central level through rising LH secretion, which in turn increase's androgen production, and the second mechanics, is mediated at the peripheral level by direct inhibitory

effect of leptin on the ovaries causing an increase in the androgenic media.^[28,29] Hence, there is a significant role of serum leptin in the determination of insulin resistance in PCOS patients.^[30]

On the other hand, the present study links the elevated serum leptin and insulin levels to obesity suggesting that most probably they are responsible for the complicated picture of the syndrome in obese patients. A hypothesis that can be easily explained by the direct effect of insulin on leptin expression causing more inhibitory action of leptin on the ovary and also the insulin and LH effect on the androgen level.

REFERENCES

1. Taylor AE. Insulin-lowering medications in polycystic ovary syndrome. *Obstet Gynecol Clin North Am* 2000;27:583-95.
2. Baillargeon J, Nestler J. Polycystic ovary syndrome: A syndrome of ovarian, hypersensitivity to insulin? *J Clin Endocrinol Metab* 2005;10:1804.
3. Halaas JL, Gajiwala KS, Maffei M, Cohen SL, Chait BT, Rabinowitz D, *et al.* Weight-reducing effects of the plasma protein encoded by the obese gene. *Science* 1995;269:543-6.
4. Spritzer PM, Poy M, Wiltgen D, Mylius LS, Capp E. Leptin concentrations in hirsute women with polycystic ovary syndrome or idiopathic hirsutism: Influence on LH and relationship with hormonal, metabolic, and anthropometric measurements. *Hum Reprod* 2001;16:1340-6.
5. Fedorcsák P, Storeng R, Dale PO, Tanbo T, Torjesen P, Urbancsek J, *et al.* Leptin and leptin binding activity in the preovulatory follicle of polycystic ovary syndrome patients. *Scand J Clin Lab Invest* 2000;60:649-55.
6. Pasquali R, Gambineri A, Biscotti D, Vicennati V, Gagliardi L, Colitta D, *et al.* Effect of long-term treatment with metformin added to hypocaloric diet on body composition, fat distribution, and androgen and insulin levels in abdominally obese women with and without the polycystic ovary syndrome. *J Clin Endocrinol Metab* 2000;85:2767-74.
7. Considine RV, Caro JF. Protein kinase C: Mediator or inhibitor of insulin action? *J Cell Biochem* 1993;52:8-13.
8. Vicennati V, Gambineri A, Calzoni F, Casimirri F, Macor C, Vettor R, *et al.* Serum leptin in obese women with polycystic ovary syndrome is correlated with body weight and fat distribution but not with androgen and insulin levels. *Metabolism* 1998;47:988-92.
9. El Orabi H, Ghalia AA, Khalifa A, Mahfouz H, El Shalkani A, Shoieb N. Serum leptin as an additional possible pathogenic factor in polycystic ovary syndrome. *Clin Biochem* 1999;32:71-5.
10. Mantzoros CS, Cramer DW, Liberman RF, Barbieri RL. Predictive value of serum and follicular fluid leptin concentrations during assisted reproductive cycles in normal women and in women with the polycystic ovarian syndrome. *Hum Reprod* 2000;15:539-44.
11. Rouru J, Anttila L, Koskinen P, Penttilä TA, Irjala K, Huupponen R, *et al.* Serum leptin concentrations in women with polycystic ovary syndrome. *J Clin Endocrinol Metab* 1997;82:1697-700.
12. Takeuchi T, Tsutsumi O. Basal leptin concentrations in women with normal and dysfunctional ovarian conditions. *Int J Gynaecol Obstet* 2000; 69:127-33.
13. Caro JF. Leptin is normal in PCOS, an editorial about three "negative" papers. *J Clin Endocrinol Metab* 1997;82:1685-6.
14. Okasanen L, Tiitinen A, Kaprio J, Koistinen HA, Karonen S. No evidence for mutations of the leptin receptor genes in women with polycystic ovary syndrome. *Fertil Steril* 2000;66:942-8.
15. Ehrmann D, Liljenquist DR, Kasza K. Prevalence and predictors of the metabolic syndrome in women with Polycystic Ovary Syndrome (PCOS). *J Clin Endocrinol Metab* 2005;10:1210.

16. Tarkun I, Cantürk Z, Arslan BC, Türemen E, Tarkun P. The plasminogen activator system in young and lean women with polycystic ovary syndrome. *Endocr J* 2004;51:467-72.
17. Mallique M, Perg-Bravo F, Calvihon M, Piwonka V. Relationship between leptin and insulin sensitivity in patients with PCOS. *Med Clin Brac* 1999;133:526-30.
18. van Hooff MH, Voorhorst FJ, Kaptein MB, Hirasig RA, Koppelaar C, Schoemaker J. Polycystic ovaries in adolescents and the relationship with menstrual cycle patterns, luteinizing hormone, androgens, and insulin. *Fertil Steril* 2000;74:49-58.
19. Jakubowicz DJ, Luorno MJ, Jakubowicz S, Roberts KA, Nestler JE. Effects of metformin on early pregnancy loss in the polycystic ovary syndrome. *J Clin Endocrinol Metab* 2002;87:524-9.
20. Dunaif A, Graf M, Mandeli J, Laumas V, Dobrjansky A. Characterization of groups of hyperandrogenic women with acanthosis nigricans, impaired glucose tolerance, and/or hyperinsulinemia. *J Clin Endocrinol Metab* 1987;65:499-507.
21. Jeon YE, Lee KE, Jung JA, Yim SY, Kim H, Seo SK, *et al.* Kisspeptin, leptin, and retinol-binding protein 4 in women with polycystic ovary syndrome. *Gynecol Obstet Invest* 2013;75:268-74.
22. Gennarelli G, Holte J, Wide L, Berne C, Lithell H. Is there a role for leptin in the endocrine and metabolic aberrations of polycystic ovary syndrome? *Hum Reprod* 1998;13:535-41.
23. Kedikova SE, Sirakov MM, Boyadzhieva MV. Leptin levels and adipose tissue percentage in adolescents with polycystic ovary syndrome. *Gynecol Endocrinol* 2013;29:384-7.
24. Kale-Gurbuz T, Akhan SE, Bastu E, Telci A, Iyibozkurt AC, Topuz S. Adiponectin, leptin and ghrelin levels in obese adolescent girls with polycystic ovary syndrome. *J Pediatr Adolesc Gynecol* 2013;26:27-30.
25. Conn JJ, Jacobs HS, Conway GS. The prevalence of polycystic ovaries in women with type 2 diabetes mellitus. *Clin Endocrinol (Oxf)* 2000;52:81-6.
26. Bloomgarden ZT. Second World Congress on the Insulin Resistance Syndrome: Mediators, pediatric insulin resistance, the polycystic ovary syndrome, and malignancy. *Diabetes Care* 2005;28:1821-30.
27. Book CB, Dunaif A. Selective insulin resistance in the polycystic ovary syndrome. *J Clin Endocrinol Metab* 1999;84:3110-6.
28. Blüher S, Mantzoros CS. The role of leptin in regulating neuroendocrine function in humans. *J Nutr* 2004;134:2469S-74.
29. Fekete C, Singru S, Sanchez E. Differential effects of central leptin, insulin or glucose administration during fasting on the Hypothalamic-Pituitary-Thyroid (HPT) axis and feeding-related neurons in the arcuate nucleus. *Endocrinology* 2005;10:1210-40.
30. Lichnovská R, Gwozdziwiczová S, Chlup R, Hřebíček J. Serum leptin in the development of insulin resistance and other disorders in the metabolic syndrome. *Biomed Pap Med Fac Univ Palacky Olomouc Czech Repub* 2005;149:119-26.

How to cite this article:***

Source of Support: Nil, **Conflict of Interest:** None declared