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Level of Chemerin and Anti-Müllerian Hormone in Egyptian Obese Women with Polycystic Ovarian Syndrome

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ABSTRACT

Background: Chemerin is a newly discovered adipokine that regulates adipocyte development, metabolic function as well as immune function. Anti-Müllerian hormone (AMH) is a dimeric glycoprotein that belongs to the transforming growth factor-beta family.

Aim of the study: was to evaluate the serum levels of Chemerin and AMH in one trial to show their possible involvement in the pathogenesis of PCOS and their association with obesity.

Subjects and methods: This study was performed on eighty subjects divided into 4 groups: Group I: 20 non-obese control (BMI< 30), Group II: 20 non-obese patient with polycystic ovarian syndrome (BMI< 30), Group III: 20 obese Control (BMI ≥ 30) and Group IV: 20 obese patient with PCOS (BMI ≥ 30). Serum levels of Chemerin and AMH were estimated by ELISA.

Results: Results showed a significant increase in serum levels of ***Chemerin and AMH*** in (non-obese and obese) PCOS groups when compared to their controls. Results showed also a significant increase in the serum level of ***Chemerin*** in obese PCOS group when compared to non-obese PCOS group. While a significant decrease in serum level of ***AMH*** in obese PCOS group when compared to non-obese PCOS group. There were a significant positive correlation between ***Chemerin and AMH*** in PCOS groups.

Conclusion: Chemerin together with AMH may be used as a biomarker for early detection of PCO and Clinical parameters as Infertility and Hyper and rogenism in women with PCOS.

Keywords: Polycystic ovary syndrome, Anti-Müllerian hormone, Chemerin.

INTRODUCTION

Polycystic ovary syndrome (PCOS) is a complex condition characterized by elevated androgen levels, menstrual irregularities, and/or small cysts on one or both ovaries **(1)**.It affects 5-10% of reproductive-age women **(2)**.Polycystic Ovary Syndrome can be described as an oligogenic disorder in which the interaction of a number of genetic and environmental factors determine the heterogeneous, clinical, and biochemical phenotype **(1)**.The relationship between PCOS and obesity is complex, not well understood, and most likely involves interaction of genetic and environmental factors **(3)**.Obesity leads to several co-morbidities, such as diabetes, dyslipidaemia, hypertension, sleep apnea, osteoarthritis, menstrual disorders, infertility, gout, stroke, ischemic heart disease, congestive heart failure, deep vein thrombosis and pulmonary embolism (4). Although not required for diagnosis, the presence of insulin resistance and hyperinsulinemiais common and places those affected at increased risk of diabetes and cardiovascular disease. Thus, PCOS adversely affects endocrine, metabolic, and cardiovascular health **(5)**. Insulin resistance and concomitant hyperinsulinemia are frequently found in obese PCOS women and occurs in around 50% to 80% of women with PCOS. The cause of insulin resistance is likewise complex and multifactorial with genetic and environmental contributors **(6)**.Women with PCOS have defects in both peripheral, which reflects primarily skeletal muscle, and hepatic insulin action, as well as pancreatic β-cell dysfunction **(7)**.Chemerin is a newly discovered adipokine that regulates adipocyte development and metabolic function as well as immune function **(8)**.Chemerin may be one of the most important links between adiposity and insulin resistance, and thus a good clinical marker for metabolic dysfunction **(9)**.Anti-Müllerian hormone (AMH) is a dimeric glycoprotein that belongs to the transforming growth factor-beta family. It is involved in the regression of the Mullerian ducts during male fetal development **(10)**.

So, this study was aimed to evaluate serum levels of chemerin and AMH innormal weight and obese patients with PCOS in one trial to show their possible involvement in the pathogenesis and progression of PCOS and analyze their association with obesity.

SUBJECTS AND METHODS

This study was performed on eighty subjects, their ages ranged between 22-37 years, selected

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from outpatients Clinic of Obstetrics and Gynecology Department, Al-Hussien Hospital, Faculty of medicine, Al-Azhar University, Cairo, Egypt in the period between December 2015to July 2016.

The venous Blood samples were collected during the early follicular phase. Polycystic Ovarian Syndrome was diagnosed according to the Rotterdam Criteria (2003), in which two of the following three features were present:

(1) Oligo ovulation and/or anovulation,

(2) Clinical and/or biochemical signs of hyper and rogenism, and

(3) Polycystic ovaries on ultrasound examination (the presence of 12 or more follicles measuring 2–9mm in diameter and/or ovarian volume greater than $10mm^3$).

These subjects were classified into four groups:

Group I: 20 non –obese healthy women their BMI<30**.**

Group II: 20 non-obese patient with polycystic ovarian syndrome their BMI<30.

Group III: 20 obese healthy women their BMI ≥30**.**

Group IV: 20 patient with Polycystic ovarian syndrome their BMI ≥30.

Patients with congenital adrenal hyperplasia, thyroid disorders, Cushing's disease, hypertension, a history of neoplasm, those using medication (e.g. insulin-sensitizing drugs, oral contraceptives, antiandrogens, statins, aspirin, corticosteroids and GnRH agonist and antagonists) during the period of 90 days prior to enrollment and Pregnant females should excluded.

♦ Weight was recorded using Seca electronic weighing scale (SecaGmBH and Co Kg, Hamburg, Germany). Height was recorded using the anthropometric height board. The body mass index was calculated as following:

BMI= Body weight in kg/ (height in m^2) = kg/m^2.

♦ Waist and hip circumferences were measured to the nearest centimeter (standing position) at the umbilicus and at the maximum circumference over the buttocks. Waist to hip ratio (WHR) was calculated by dividing the waist circumference by the hip circumference.

Collection of samples

10 ml of peripheral venous blood were drawn by vein puncture from each participant within 2 and 4 days of menstrual period after an overnight fasting. The samples were divided into two tubes. 2ml were put on fluoride/ oxalate for estimating fasting plasma glucose. The remaining blood without additives, allowed to clot for 30 minutes at room temperature and centrifuged at 3000 rpm for 10 minutes, to obtain serum supernatant (Serum sample) which was labeled and stored at -20°c until analyzed.

Available commercial kits were used for determination of fasting plasma glucose (FPG) by endpoint technique), Insulin measured by ELISA using (Bios ELISA ®), lipid profile (total cholesterol, triacylglycerol, HDL-cholesterol and LDL-cholesterol) measured by endpoint technique using (Seimens®health care diagnostics), LH measured by ELISA using (Bio Tina GmbH ELISA ®), FSH measured by ELISA using (Bios ELISA ®), Progesterone and Total testosterone measured by ELISA using (NovaTecimmundiagnostica GMBH ®), AMH measured by ELISA using (CUSABIO®) and finally Chemerin measured by ELISA using (Bio-Vender Research and diagnostic Products Company®).

The study was approved by the Ethics Board of Al-Azhar University.

Statistical Methods

Data were analyzed and entered to the computer using SPSS (Statistical Package for Social Science) program for statistical analysisVersion16 (SPSS Inc., 2007, Chicago, IL, USA) and Sigma Plot version13.

Data was summarized as mean± Standard Error of the Mean (SEM).

Differences between groups were analyzed by standard Student's T-test.

Simple linear correlation (Pearson correlation coefficient test) (r) was also done to test for linear relations between AMH, Chemerin and other variables.

P-value is considered significant if $P \leq 0.05$.

n-significant result is considered if $P > 0.05$.

RESULTS

Eighty subjectswere enrolled in this study, to show Chemerin and AMH possible involvement in the pathogenesis and progression of PCOS and analyze their association with obesity, their ages ranged between 22-37 years classified into four groups: Group I: 20 non-obese healthy women their BMI<30. Group II: 20non-obese patient with polycystic ovarian syndrome their BMI<30.Group III: 20 obese healthy women their BMI ≥30. Group IV: 20 patients with polycystic ovarian syndrome their BMI ≥30.There were a significance differences in some parameters among groups (Table 1 and Table 2).

(A) Comparison between descriptive statistics

Table (1): Comparison between statistics of all study groups regarding clinical, biochemical and Hormonal data

Parameters	Group I (Non-Obese Control) N=20	Group II (Non-Obese PCOS) N=20	P-Value	Group III (Obese Control) N=20	Group IV (Obese PCOS) N=20	P-Value
	Mean ± SEM	Mean ± SEM		Mean ± SEM	Mean ± SEM	
BMI (kg/m2)	25.11 ± 0.64	25.99 ± 0.36	(NS)	37.37 ± 0.97	36.23 ± 0.88	(NS)
W/H ratio	0.79 ± 0.01	0.80 ± 0.01	(NS)	0.90 ± 0.01	0.88 ± 0.01	(NS)
FPG (mg/dl)	91.10 ± 3.34	110.65 ± 2.65	[a]p < 0.0001	93.80 ± 2.91	112.05 ± 2.53	[b]p < 0.0001
Fasting Insulin (uIU/ml)	8.20 ± 0.27	10.25 ± 0.54	[g]p < 0.01	8.47 ± 0.23	15.45 ± 0.29	[b]p < 0.0001
HOMA-IR	1.82 ± 0.03	2.75 ± 0.13	[a]p < 0.0001	1.95 ± 0.05	4.29 ± 0.55	[b]p < 0.0001
TC (mg/dl)	176.10 ± 2.58	177.85 ± 2.78	(NS)	169.35 ± 3.39	258.70 ± 38.19	[b]p < 0.0001
TAG (mg/dl)	88.90 ± 6.59	119.55 ± 6.33	[g]p < 0.01	91.25 ± 3.96	207.00 ± 5.58	[b]p < 0.0001
HDL-C (mg/dl)	77.05 ± 2.67	37.85 ± 0.98	[a]p < 0.0001	53.20 ± 2.50	27.65 ± 1.51	[b]p < 0.0001
LDL-C (mg/dl)	107.00 ± 1.71	118.40 ± 3.82	(NS)	113.75 ± 5.25	195.85 ± 6.75	[b]p < 0.0001
LH (mIU/ml)	4.32 ± 0.25	15.49 ± 0.69	[a]p < 0.0001	5.05 ± 0.20	13.00 ± 0.55	[b]p < 0.0001
FSH (mIU/ml)	8.27 ± 0.41	8.82 ± 0.43	(NS)	8.73 ± 0.42	8.40 ± 0.38	(NS)
Progesterone (ng/ml)	0.95 ± 0.05	0.94 ± 0.05	(NS)	0.91 ± 0.04	0.98 ± 0.05	(NS)
TT (pg/ml)	0.25 ± 0.02	0.62 ± 0.05	[a]p < 0.0001	0.25 ± 0.02	0.63 ± 0.04	[b]p < 0.0001
AMH (ng/ml)	5.86 ± 0.26	9.09 ± 0.21	[a]p < 0.0001	5.49 ± 0.29	8.32 ± 0.20	[b]p < 0.0001
Chemerin (ng/ml)	131.35 ± 5.61	425.60 ± 4.78	[a]p < 0.0001	278.63 ± 4.61	559.31 ± 15.79	[b]p < 0.0001

(NS): Non significant (P-Value > 0.05) [a] p < 0.0001 group II versus group I, [g]p < 0.01 group II versus group I, [b]p < 0.0001 group IV versus group III.

Table (2): Comparison between statistics of PCOS groups regarding clinical, biochemical and Hormonal data

Parameters	Group II (Non-Obese PCOS) N=20	Group IV (Obese PCOS) N=20	P-Value
	Mean ± SEM	Mean ± SEM	
BMI (kg/m2)	25.99 ± 0.36	36.23 ± 0.88	[f]p < 0.001
W/H ratio	0.80 ± 0.01	0.88 ± 0.01	[c]p < 0.0001
FPG (mg/dl)	110.65 ± 2.65	112.05 ± 2.53	(NS)
Fasting Insulin (uIU/ml)	10.25 ± 0.54	15.45 ± 0.29	[c]p < 0.0001
HOMA-IR	2.75 ± 0.13	4.29 ± 0.55	[c]p < 0.0001
TC (mg/dl)	177.85 ± 2.78	258.70 ± 38.19	[c]p < 0.0001
TAG (mg/dl)	119.55 ± 6.33	207.00 ± 5.58	[c]p < 0.0001
HDL-C (mg/dl)	37.85 ± 0.98	27.65 ± 1.51	[f]p < 0.001
LDL-C (mg/dl)	118.40 ± 3.82	195.85 ± 6.75	[c]p < 0.0001
LH (mIU/ml)	15.49 ± 0.69	13.00 ± 0.55	[f] p < 0.01
FSH (mIU/ml)	8.82 ± 0.43	8.40 ± 0.38	(NS)
Progesterone (ng/ml)	0.94 ± 0.05	0.98 ± 0.05	(NS)
TT (pg/ml)	0.62 ± 0.05	0.63 ± 0.04	(NS)
AMH (ng/ml)	9.09 ± 0.21	8.32 ± 0.20	[i] p < 0.01
Chemerin (ng/ml)	425.60 ± 4.78	559.31 ± 15.79	[c]p < 0.0001

(NS): Non significant (P-Value > 0.05) [c] p < 0.0001 group IV versus group II, [f]p < 0.001 group IV versus group II [i] p< 0.01 group IV versus group II.

As expected from appropriate matching ***BMI*** and ***W/H ratio***was virtually identical in the two obese and non-obese study groups for both comparisons. Obese women with PCOS demonstrated significantly higher ***BMI*** and ***W/H ratio***compared to non-obese women with PCOS.

According to Table (1, 2) the mean values of ***Fasting plasma Glucose (FPG)***, ***Fasting insulin,HOMA-IR*** and***Triacylglycerol (TAG)*** showed significant increase in non-obese PCOS patients when compared to the non-obese controls and significant increase in obese PCOS patients when compared to the obese controls.

The mean serum level of ***Total Cholesterol (TC)*** and ***Low density lipoprotein-Cholesterol*****(LDL-C)** showed statistically significant increase observed only in obese PCOS patients when compared to the obese controls, while non-obese PCOS patients showed no significant differences with non-obese controls.

PCOS groups showed significant increase in obese women when compared to the non-obese women in***Fasting Insulin***, ***HOMA-IR, Total Cholesterol (TC)***, ***Triacylglycerol (TAG)*** and ***Low density lipoprotein-Cholesterol*****(LDL-C)** level.

A statistically significant decrease was detected in the mean serum level of ***High density lipoprotein-Cholesterol*** **(HDL-C)** between non-obese PCOS patients when compared to the non-obese control and significant increase in obese PCOS patients when compared to the obese control. PCOS groups showed also significant decrease in obese women when compared to the non-obese women.

The mean serum levels of ***LH andTotal Testosterone (TT)*** showed statistically significant increase between non-obese PCOS patients when compared to the non-obese controls and significant increase in obese PCOS patients when compared to the obese controls.

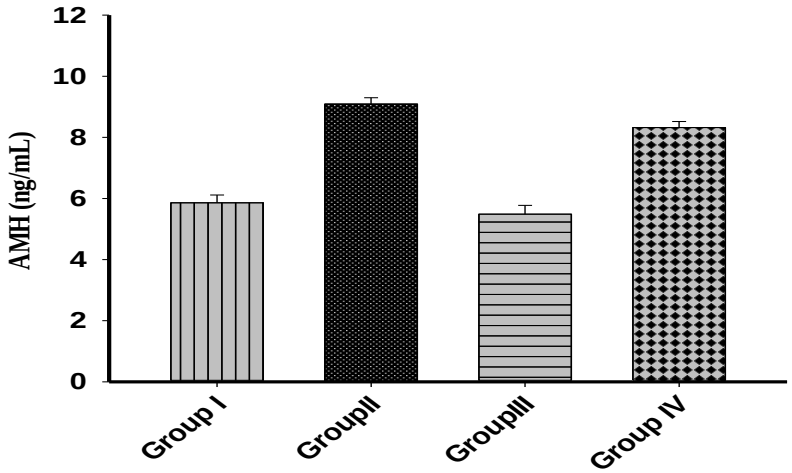
A significant negative correlation between***LH*** and obesity was deticted where non-obese women with PCOS demonstrated significantly higher levels of their concentrations compared to obese women with PCOS.

PCOS groups showed no significant differences in ***Total Testosterone (TT)*** between obese women when compared to the non-obese women.

Finaly, The mean serum levels of ***AMH*** showed statistically significant increase (at $p < 0.0001$) in non-obese PCOS patients (9.09 ± 0.21 ng/ml) compared to non-obese women without PCOS (5.86 ± 1.14 ng/ml) also obese women with PCOS demonstrated significantly (at $p < 0.0001$) higher ***AMH*** concentrations (8.322 ± 0.20 ng/ml) compared to obese women without PCOS (5.49 ± 1.29 ng/ml).

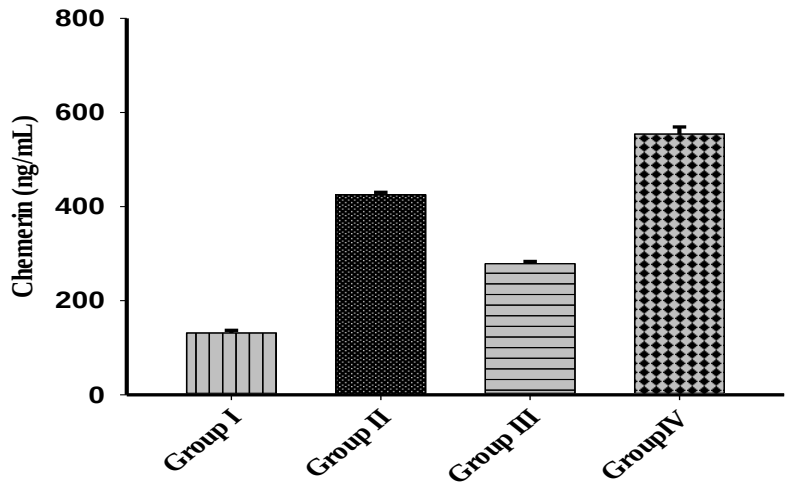
In this study there werea significant negative correlation between ***AMH*** and obesity where non-obese women with PCOS demonstrated significantly ($p < 0.01$) higher ***AMH*** concentrations (9.09 ± 0.206 ng/ml) compared to obese women with PCOS (8.32 ± 0.198 ng/ml) (Figure 1). Also, non-obese women with PCOS demonstrated significantly (at $p < 0.0001$) higher ***Chemerin*** concentrations (425.60 ± 4.78 ng/ml) compared to non-obese women without PCOS (131.35 ± 5.61ng/ml) also obese women with PCOS demonstrated significantly (at $p < 0.0001$) higher ***Chemerin***concentrations (559.31 ± 15.79 ng/ml) compared to obese women without PCOS (278.63 ± 4.61ng/ml).

In this study there werea significant positive correlation between ***Chemerin*** and obesity where obese women with PCOS demonstrated significantly (at $p < 0.0001$) higher ***Chemerin*** concentrations (559.31 ± 15.79 ng/ml) compared to non-obese women with PCOS (425.60 ± 4.78 ng/ml) (Figure 2).



***P** < 0.01, *****P** < 0.0001: Statisticaly Significant
[a]p< 0.0001 group II versus group I, [b]p < 0.0001 group IV versus group III,
[f]p< 0.01 group IV versus group II.

Figure 1: Mean ± SEM of AMH of all studied groups



*****P** <0.0001:Statisticaly Significant [a]p< 0.0001 group II versus group I,
[b]p < 0.0001 group IV versus group III, [c]p< 0.0001 group IV versus group II

Figure 2: Mean ± SEM of Chemerin of all studied groups

(B) Correlations

(I)- Table (3): Correlation between serum AMH and other data of PCOS groups (group II and IV):

Correlation of AMH with	Group II		Group IV	
	R	P-value	r	P-value
BMI (kg/m2)	0.323	P > 0.05 (0.164)	0.395	P > 0.05 (0.085)
W/H ratio	-0.267	P > 0.05(0.256)	0.289	P > 0.05(0.216)
FPG (mg/dl)	0.120	P > 0.05(0.615)	0.100	P > 0.05(0.674)
FastingInsulin (uIU/ml)	0.102	P > 0.05(0.670)	0.346	P > 0.05(0.135)
HOMA-IR	0.199	P > 0.05(0.400)	0.292	P > 0.05(0.211)
TC (mg/dl)	0.301	P > 0.05(0.198)	0.084	P > 0.05(0.725)
TAG (mg/dl)	0.346	P > 0.05(0.135)	0.140	P > 0.05(0.555)
HDL-C (mg/dl)	-0.323	P > 0.05(0.165)	- 0.160	P > 0.05(0.500)
LDL-C (mg/dl)	0.287	P > 0.05(0.220)	0.313	P > 0.05(0.179)
LH (mIU/ml)	0.448*	P < 0.05(0.048)	0.497*	P < 0.05(0.026)
FSH (mIU/ml)	0.135	P > 0.05(0.571)	-0.166	P > 0.05(0.484)
Progesterone (ng/ml)	0.241	P > 0.05(0.307)	0.003	P > 0.05(0.991)
TT (pg/ml)	0.462*	P < 0.05(0.040)	0.560*	P < 0.05 (0.010)

*Correlation is significant at P ≤ 0.05.
**Correlation is significant at P < 0.01.

(II)- Table (4): Correlation between serum Chemerin and other data of PCOS groups (group II and IV)

Correlation of Chemerin with	Group II		Group IV	
	R	P-value	r	P-value
BMI (kg/m2)	0.699**	P < 0.01(0.001)	0.874**	P < 0.01(0.001)
W/H ratio	0. 270	P > 0.05(0.249)	0.295	P > 0.05 (0.207)
FPG (mg/dl)	0.088	P > 0.05(0.713)	0.313	P > 0.05 (0.180)
FastingInsulin (uIU/ml)	0.464*	P < 0.05(0.039)	0.710**	P < 0.01(0.001)
HOMA-IR	0.541*	P < 0.05(0.014)	0.732**	P < 0.01(0.001)
TC (mg/dl)	0.557*	P < 0.05(0.011)	0.616**	P < 0.01(0.004)
TAG (mg/dl)	0.178	P > 0.05(0.452)	0.084	P > 0.05(0.724)
HDL-C (mg/dl)	- 0.444*	P < 0.05(0.048)	- 0.555*	P < 0.05(0.011)
LDL-C (mg/dl)	0.369	P > 0.05(0.109)	0.339	P > 0.05(0.143)
LH (mIU/ml)	0.246	P > 0.05(0.296)	0.300	P > 0.05(0.199)
FSH (mIU/ml)	-0.101	P > 0.05(0.671)	- 0.033	P > 0.05(0.891)
Progesterone (ng/ml)	0.057	P > 0.05(0.812)	0.266	P > 0.05(0.258)
TT (pg/ml)	0.228	P > 0.05(0.333)	0.335	P > 0.05(0.148)
AMH (ng/ml)	0.622**	P < 0.01(0.003)	0.558*	P < 0.01(0.011)

*Correlation is significant at P ≤ 0.05
**Correlation is significant at P < 0.01

According to Table (3) there were a significant positive correlations between ***AMH and LH*** where (r =0.448 at p=0.048) (r =0.497 at p= 0.026) in non-obese and obese PCOS groupsrespectively.Also, there were a significant positive correlations between ***AMH and TT*** where (r =0.462 at p=0.040) (r =0.560 at p= 0.010) in non-obese and obese PCOS groups respectively.

According to Table (4) there were a significant positive correlation between ***Chemerin and BMI***

(r =0.699 at p = 0.001), (r = 0.874 at p = 0.001) in non-obese and obese PCOS groups respectively. There were also a significant positive correlations between ***Chemerin and Fasting insulin*** (r =0.464 at p = 0.039) (r = 0.710 at p = 0.001) in non-obese and obese PCOS groups respectively.

There were significant positive correlations between ***Chemerin and HOMA-IR*** in PCOS groups (r = 0.541 at p = 0.014), (r =0.732 at p = 0.001) in (non-obese and obese PCOS) groups. Also, There were significant positive correlations between ***Chemerin and Total Cholesterol (TC)in PCOS*** groups (r =0.557 at p = 0.011), (r = 0.616 at p = 0.004) in (non-obese and obese PCOS) groups respectively. While, there were significant negative correlations between ***Chemerin and HDL-C*** (r = -0.444 at p = 0.048), (r = -0.555 at p = 0.011) in (non-obese and obese PCOS) groups respectively.

There were significant positive correlations between ***Chemerin and AMH in PCOS*** groups (r =0.622 at p= 0.003) (r = 0.558 at p= 0.011) in (non-obese and obese PCOS) groups respectively (figure 3,4).

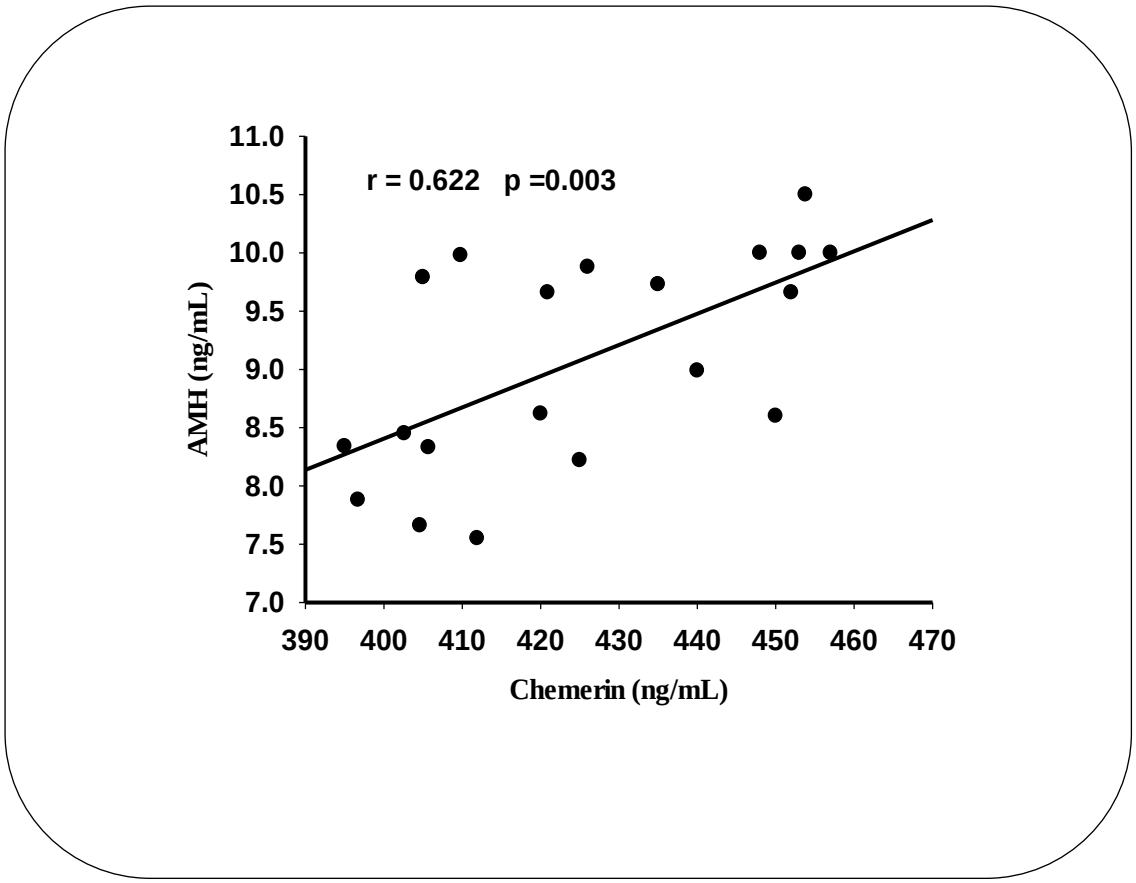


Figure 3: Correlation between Chemerin and AMH in Non-obese PCOS (group II)

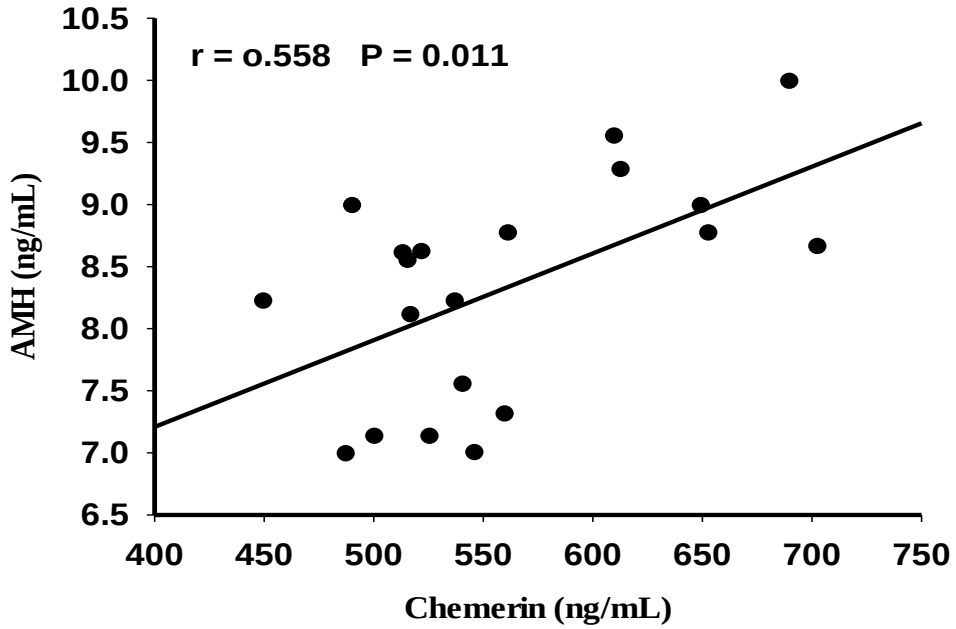


Figure 4: Correlation between Chemerin and AMH in Obese PCOS (group IV)

(C) Receiver operating characteristics (ROC) curves:

ROC curve were carried out to assess the diagnostic performance of Chemerin whether it is more sensitive and specific than AMH or not. A cut-off serum Chemerin value is 350ng/ml was determined for discriminating between women with and without PCOS, Serum Chemerin level of > 350 ng/ml predicted the presence of PCOS with 100% sensitivity and 100% specificity (Table 5) (Figure 5).While a cut-off serum AMH 7ng/ml was determined for discriminating between women with and without PCOS, Serum AMH of > 7 ng/ml predicted the presence of PCOS with 98% sensitivity and 88% specificity (Table 6) (Figure 6), Serum Chemerinismore sensitive than Serum AMH(Table 7)(Figure 7).

Table (5): Characters of Receiver operating characteristic (ROC) curve for Chemerin levels in the diagnosis of women with and without PCOS:

Variable	Cutoff	sensitivity	Specificity	PPV	NPV	Accuracy
Chemerin	350(ng/ml)	100%	100%	100%	100%	100%

PPV = Positive predictive value, NPV = negative predictive value

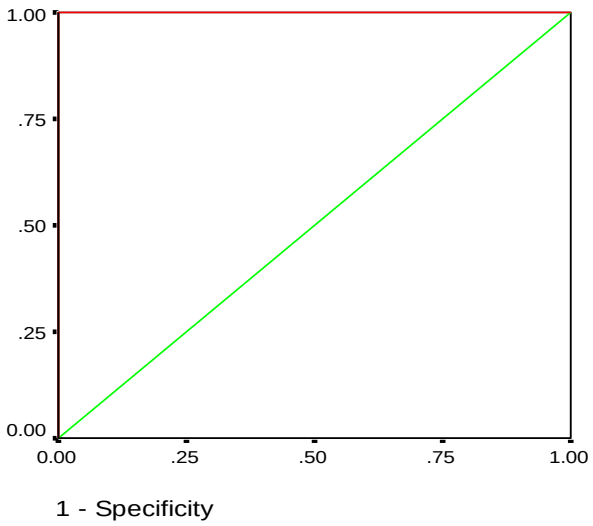


Figure 5: Receiver operating characteristic curveforChemerin levels in the diagnosis of PCOS.

Table (6): Characters of receiver operating characteristic (ROC) curve for AMH levels in the diagnosis of women with and without PCOS:

Variable	Cutoff	sensitivity	Specificity	PPV	NPV	Accuracy
AMH	7 (ng/ml)	98%	88%	89%	97%	93%

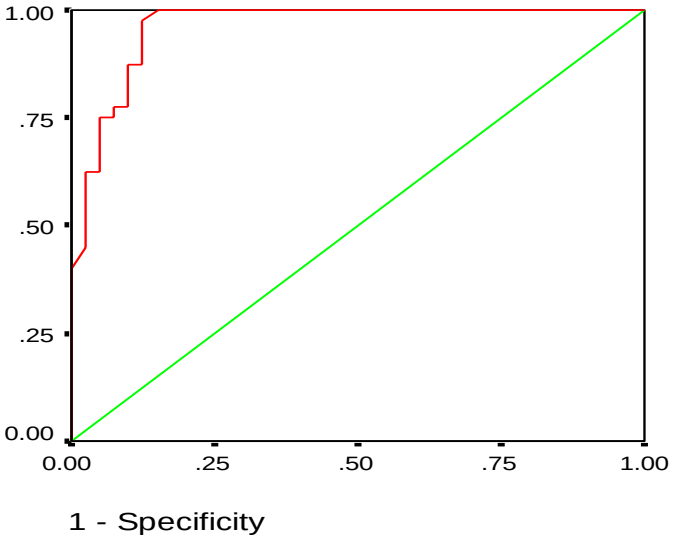


Figure 6: Receiver operating characteristic curve for AMH levels in the diagnosis of PCOS

Table (7): Comparison between characters of receiver operating characteristic curves for serum Chemerin and serum AMH in women with and without PCOS:

Differences between areas	Standard error	95% Confidence interval	P- value
0.039	0.012	0.013 – 0.078	0.027*

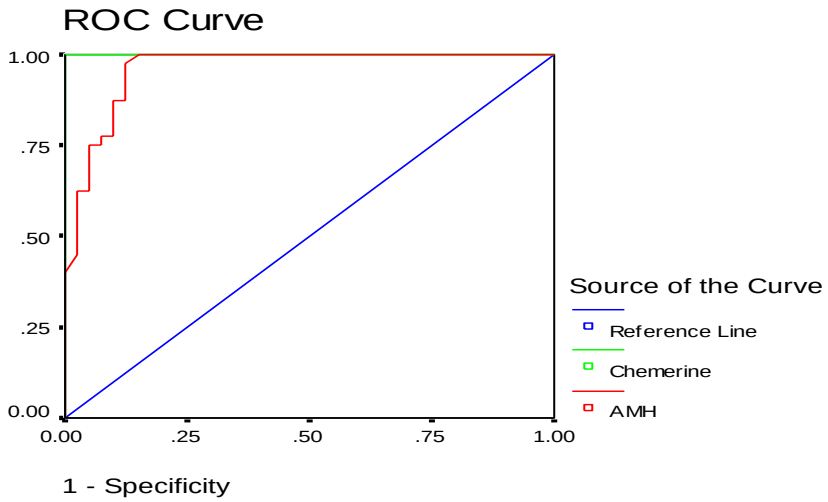


Figure 7: Comparison between receiver operating characteristic curvesfor serum Chemerin and serum AMH in women with and without PCOS

DISCUSSION

Regarding Insulin resistance assessment, this study showed a statistically significant higher levels of Fasting plasma Glucose (FPG), Fasting insulin and HOMA-IR in non-obese and obese PCOS patients in contrast to non-obese and obese control subjects respectively.Once compared non-obese PCOS women with obese PCOS women, a statistically significant reduction in Fasting insulin and HOMA-IR was found in non-obese group. These results agreed with ***Kyrou et al.*** [(3)].where theysuggested that this insulin resistance and hyperinsulinemia noticed in PCOS subjects was resulted from overconsumption of carbohydrate diet. ***Agarwal et al.*** [(11)].suggested that hyperinsulinemia occurs because of insulin resistance and increase ovarian androgen over production and it may also contribute to the development of diabetes and dyslipidemia in PCOS patients.Also, ***Majid et al.*** [(12)].stated that the increased insulin resistance is a prominent feature of polycystic ovary syndrome (PCOS) and HOMA-IR performed better than the hyperinsulinemia for diagnosing IR.

Regarding lipid profile, this study showed a statistically significant increase in the mean serum levels of Total Cholesterol (TC), Triacylglycerol (TAG) and low density lipoprotein-Cholesterol (LDL-C) in PCOS-obese patients when compared to the obese controls and ccccin the serum level of (TAG) only in PCOS non-obese patients when compared to the non-obese controls. While, a statistically significant decrease was detected in the serum level of High density lipoprotein-Cholesterol (HDL-C) in both groups of PCOS (non-obese and obese) when compared to the controls (non-obese and obese) respectively.

PCOS groups showed significant increase in TC, TAG, and LDL-C and significant decrease in HDL-C in obese women when compared to the non-obese women.

Similar results were found in a study done by ***Shoaib et al.*** [(13)].***Mohamadin et al.*** [(14)].and***Cengiz et al.*** [(15)].

These results also agreed with***Guzel et al.*** [(16)].Theystated that The PCOS women had higher LDL cholesterol and lower serum HDL cholesterol levels Therefore, the control of obesity in patients with PCOS might help to control IR and improve metabolic parameters.

On the other hand, these results were not concurrent with the study done buy***Abdelgadir et al.*** [(17)].who demonstrated that there were no significant differences in lipid or lipoprotein concentrations between patients with PCOS and weight-matched controls.

Also ***Valkenburg et al.*** [(18)] showed that when lipid changes occur in PCOS women, this may get affected by obesity and hyperandrogenism.

Regarding the Hormonal profile the present results revealed significant increase in serum LH and total testosterone level and no significant differences in serum FSH and progesterone level between PCOS patients compared with control subjects.

These results also agree with those of ***Mohamadin et al.*** [(14)] and ***Lewandowski et al.*** [(19)] who postulated that this phenomenon potentially useful in the diagnosis of PCOS.

On the other hand, these results were not concurrent with the study done by ***Samy et al.*** [(20)]

where they found that LH level was higher in obese women with PCOS when compared with non-obese PCOS patients and normal control subjects.

Also ***Cengiz et al.*** [15] found no difference in the mean serum level of LH in patients with PCOS when compared to the control women.

In this study, like ***Aigner et al.*** [21] the mean levels of total testosterone were significantly higher in dependent way with BMI in PCOS patients.

In these study women with PCOS havehigher levels of Anti-Mullerian hormone (AMH) compared with healthy women. These results agreed with***Matsuzaki et al.*** [22] and***Bernardi et al.*** [23]

In this study there were a significant negative association between AMH level and obesity. Like this result study done by***Bernardi et al.*** [23] where they stated that there was a significant inverse association between obesity and AMH. AMH concentrations declined as current BMI increased, and AMH was significantly lower in PCOS women with obesity compared with those who were normal weight suggesting that folliculogenesis impaired as BMI increases.

Nybacka et al. [24] found that AMH concentrations decreased during dieting in 57 overweight/obese women with PCOS.

A number of theories have been proposed to explain the relationship between obesity and AMH. ***Park et al.*** [25] proposed that insulin resistance in individuals with obesity impacts granulosa cells and consequently alters AMH concentration.Also, ***Chen et al.*** [26] proposed that not only age-related variables, but also obesity and IR, have been reported to have a negative effect on ovarian granulosa cell function. Another possibility is that AMH may be metabolized, stored, and cleared differently in individuals with obesity [27].

A significant positive association between LH and AMH levels has been observed in this study. Higher AMH levels observed in non-obese women with PCOS compared to obese women with the syndrome could be attributed to the higher LH levels. Thus, the lower LH concentrations observed in obese women may be attributed to the increased aromatization of androgens to estrogens which takes place in the peripheral fat tissue, resulting in the suppression of LH [28].

This result agreed with ***Catteau-Jonard et al.*** [29] and as discussed before by ***Silfen et al.*** [30] where they postulated that the hyperinsulinemia in obese PCOS may be responsible for the diminished LH levels.

This study revealed that circulating levels of Chemerin were higher in PCOS patients when compared with controls. Serum Chemerin concentrations were elevated in obese women compared to normal weight women.This resultsare in agreement with the findings of ***Tan et al.*** [31] who demonstrated an increase of chemerin expression in women with PCOS which may involve in the pathogenesis of PCOS, regardless of obesity.

This finding also was in harmony with results of ***Wang et al.*** [32] who have shown a higher level of chemein expression in the ovary of dihydrotestosterone induced PCOS rats.

Also, several experiments have demonstrated that chemerin may play a role in pathophysiology of PCOS in animal or human by direct action on ovary ***Tang et al.*** [33] and ***Kim et al.*** [34].

Chemerin is a recently identified adipokine via its known effects; it may be thought to act in different pathways leading to insulin resistance and inflammatory events regarding metabolic disturbances that may take place during PCOS [35].

In the current study, there were appositive correlation between chemerin and BMI. ***Goraleski et al.*** [36] supposed that increased release of adipokines as well as free fatty acids from triglyceride-overloaded adipocytes stimulates macrophage infiltration and activation of a local inflammatory response and impair adipocyte sensitivity to insulin also, CMKLR1 is highly expressed in the stromal vascular compartment of white adipose tissue and suggesting that adipocyte-derived chemerin could act as a paracrine regulator of recruitment of CMKLR1-expressing of immune cells to white adipose tissue as part of the local inflammatory response that coincides with the development of obesity.

Li et al. [37] proposed that, the mechanisms of chemerin in obesity might be as follows. Firstly, chemerin could promote the differentiation of adipocytes and metabolism. Secondly, chemerin could inhibit the decomposition of fat to a certain extent. Thirdly, chemerin might participate in the inflammatory immune response.

Schultz and Beck-Sickinger [38] stated that Chemerin significantly decreased after bariatric surgery in obese patients, accompanied by pronounced weight loss and improvements in parameters of lipid and glucose metabolism. So chemerin may be a promising new target for the treatment of obesity, metabolic syndrome and type 2 diabetes.

This study showed thatchemerin level wassignificantly positively correlated with HOMA-IR in PCOS women; It seems most probably that HOMA-IR is a determinant of chemerin levels because in this studythere wasa significant positive correlation between Chemerin and HOMA-IR in PCOS patients agreeing with ***Kort et al.*** [(9)] who stated that Chemerin may contribute to the pathogenesis of IR, However, the exact regulation of chemerin on morbidity of IR and glucose metabolism is poorly understood, which may involve in reduced glucose uptake and insulin sensitivity chemerin receptors signaling pathways, or chronic inflammation they also stated that Chemerin was the only adipokine tested to be positively associated with all three measures of adiposity and insulin resistance in PCOS (BMI, SQ fat and HOMA-IR), As chemerin is a chemo-attractant adipokine with inflammatory properties, chemerin may be one of the most important links between adiposity and insulin resistance, and thus a good clinical marker for metabolic dysfunction. However, serum levels may not always reflect these changes as levels are determined by both the type and the abundance of adipose tissue, which by nature can be heterogeneous.

Guzel et al. [(16)] also stated that ex-vivo treatment showed that chemerin synthesis and production was increased by insulin and decreased by metformin, suggesting that chemerin may be a link between hyperinsulinemia and PCOS. Their study also elevated the possibility that strategies that can modulate concentrations of omentin and chemerin would improve glucose tolerance and metabolic disarrangements in women with PCOS. Recently it is well known that formation of polycystic ovaries is critically associated with abnormal steroidogenesis. It has been found that chemerin decreases estradiol secretion and suppressed FSH-induced progesterone and estradiol secretion in prenatal follicles and granulosa cells [(34)].

Some studies suggested that chemerin is a novel negative regulator of FSH-induced follicular steroidogenesis and that it may contribute to the pathogenesis of PCOS [(32)].

This resultshowthat Chemerin concentration positively correlated with AMH.

Nardo et al. [(39)] found a positive correlation between AMH concentrations and insulin levels as well as HOMA-IR and mentioned that in light of these findings, higher adipokine levels may be implicated in the higher AMH concentrations in PCOS patients.

For testing the power of Chemerin as a predictive parameter for Polycystic Ovary syndrome we constructed a ROC curve for Chemerin and AMH at different cut off levels. The ROC curve showed 100% sensitivity for the prediction of PCOS at a cut off value 350 ng/ml. Therefore, estimation of Chemerin level helps in early prediction and prevention of PCOS.

CONCLUSION

The present results suggest that the serum chemerin and AMH are directly correlate with PCOS. There is a significant positive correlation between Chemerin and obesity in women with PCOS. While AMH is directly correlates with PCOS; there is a negative correlation between AMH and obesity.A significant positive correlation between Chemerin and AMH observedin this study and Serum chemerin can be used as biomarker for early detection of PCOS. Additional large scale studies should be conducted on patients with Polycystic Ovary Syndrome to assess the disease progression relative to the role of chemerin and AMH. Further studies are needed to explain the pathophysiological roles of the increased serum chemerin and AMH observed in PCOS and their relationship with obesity.

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