Evaluating the Role of High Molecular Weight Adiponectin in Women with Polycystic Ovary Syndrome Treated with Omega-3 Fatty Acids

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Abstract

Polycystic ovary syndrome (PCOS) is a significant health problem affecting women during the reproductive age and characterized by hormonal disturbance resulting in irregular menstrual periods and infertility. Adiponectin is a protein produced by adipocytes and involved in glucose and lipid metabolism. This study included the evaluation of two doses of omega-3 (300 and 600mg/day for 14 weeks). Age, body mass index (BMI), fasting blood glucose level, homeostatic model assessment (HOMA), lipid profile, luteinizing hormone (LH), follicle-stimulating hormone (FSH), prolactin, estrogen and HMW adiponectin levels were evaluated. Our data showed no significant differences between control group and treatment groups in BMI, age, sex hormones, fasting blood glucose, and HOMA levels. However, the level of HMW adiponectin was significantly higher in women treated with 600mg/day for 14 weeks ($p < 0.0001$) but not 300mg / day of Omega-3 fatty acids for the same period.

Keywords: *Polycystic Ovary Syndrome (PCOS), Omega-3 fatty acids, High Molecular Weight (HMW) Adiponectin*

Introduction

Polycystic ovary syndrome (PCOS) is an endocrine metabolic disorder leading to anovulation in female causing infertility. It is responsible for about 50-70% of infertility in women. The clinical features of PCOS include hirsutism and oligomenorrhea or amenorrhea.[1]

Obesity and insulin resistance as the most common causes of PCOS.[2,3] Adipokines are among peptides that are synthesized by adipose tissue, which act as autocrine and paracrine controlling the adipose tissue and as endocrine to control other organs such as adrenal gland, CNS and skeletal muscle. In addition, they mediate insulin action.[4]

Many studies have shown that the HMW adiponectin can regulate insulin sensitivity and involved obesity and cardiovascular diseases. In addition, it has been shown that the other forms of low and intermediate molecular weight are less effective.[5,6] It has been reported that metabolic disorders such as PCOS is associated with hypoadiponectinemia.[7,8,9] In addition, it has been reported by many studies related to nutrition and PCOS that energy management and weight loss in overweight patients can modify biochemical parameters.[8]

Omega-3 is one of the supplements that regulate obesity, inflammation and insulin resistance.[9] The relationship between adiponectin level and omega-3 supplement has been reported at the experimental level.[10] However, at the clinical level it is still controversial which may be due to different doses and periods of omega -3 treatment recoded.[11] Nevertheless, omega-3 has shown a role in metabolic equilibrium and some diseases.[12,13]

This study investigates the effect of two different doses of omega-3 fatty acids (300mg and 600mg) with period of treatment for 14 weeks on the plasma of adiponectin for women PCOS.

Materials and Methods

Study design

Our study included 270 Iraqi females suffering from PCOS. They were divided into three groups, 90 participants in each group: Group A (control; women with PCOS with no treatment), group B (women with PCOS treated with 300mg /day omega-3 fatty acids for 14 weeks), and group C (women with PCOS treated with 600mg /day omega-3 fatty acids for 14 weeks).

Sample preparation

Samples were collected during the period between December 2018 to December 2019. The biochemical analysis was performed in the laboratories of the Department of Biochemistry in the AL-Sader Teaching Hospital in Al Najaf Al Ashraf Province.

The three groups were diagnosed clinically depending on the inclusion criteria: [14] oligomenorrhea, amenorrhea (i.e the absence of period for 6 months), hirsutism, elevated LH/FSH ratio, increased testosterone and ultrasound (ovaries contain multiple cyst).

The exclusion criteria of the study include the following: Cushing's syndrome, hyperprolactinemia, adrenal gland dysfunction, congenital adrenal hyperplasia, thyroid gland diseases, androgen secreting tumors, female treated with oral contraceptives, antiandrogen, body agents and weight restriction drugs, smokers, those patients with insulin sensitivity, diabetes mellitus, renal diseases, hepatic disorders and/or gastrointestinal disturbances.

The study protocol was accepted by Ethical Committee of College of Pharmacy, Jabir ibn Hayyan Medical University. The individuals in the study were asked for regulation of life style and diet management.

The (BMI) body mass index was calculated as follows:

$$BMI = \frac{\text{Weight in kg}}{(\text{Height in m})^2}$$

The collection of samples under fasting conditions about (8-10 hr).[15] after that the blood samples were centrifuged (2000xg for 10 minutes) and stored the serum in freezing condition at −70°C.

The HMW adiponectin was estimated according to the commercial ELISA kit (catalog number: MBS702778). Hormones, insulin, testosterone, LH and FSH were estimated by Promega® (U.S.A). The HOMA-IR was calculated as follows:[16]

$$\text{HOMA-IR} = \frac{\text{Insulin } (\mu U/ml) \times \text{Glucose } (mg/dl)}{405}$$

When HOMA-IR is more than 3.5, there would be insulin resistance.

The total cholesterol (TC) was estimated according to BIOLABO (France), high density lipoprotein cholesterol (HDLc) was estimated by DRG Gmb (Germany) and the low density lipoprotein cholesterol (LDLc) was calculated according to Fridewal *et al*., 1972.[18]

The statistical analysis was performed using SPSS program and data were presented Mean±SD. Student's t-test was used to test the significance level. Data with p value less than 0.05 was considered significantly different.

Results

Before treatment, there was a screening for the study parameter in all the three groups. Our findings showed no significant differences in age, BMI, fasting insulin, HOMA-IR, LH/ FSH ratio, total cholesterol, triglycerides, LDL, TG, VLDL, HDL, estradiol, progesterone, free and total testosterone, prolactin, dehydroepiandrosterone sulfate, TSH and HMW adiponectin between group A and group B before treatment or group A and group C (before treatment). However, there was a significantly higher level of fasting blood glucose in B (before treatment) when compared to A (p= 0.002) as shown in Table 1.

Table 1: Biochemical screening for the study parameters in subjects in group A (control, n=90), group B (before treatment with 300mg omega-3, n=90) and group C (before treatment with 600mg omega-3, n=90).

Parameter	Group A Control	Group B Before treatment	P value	Group C Before treatment	P value
Age	25.4 ± 4.3	24.2 ± 5.3	0.105	24.9 ± 4.3	0.44
BMI (kg/m2)	28.5± 1.69	28.3 ± 2.84	0.56	28.1 ± 1.98	0.14
Fasting insulin (μU/ml)	18.4± 2.9	17.7± 3.3	0.132	18.1± 3.1	0.5
Fasting blood glucose (mg/dl)	72.63± 11.8	77.79± 10.5	0.002	80.89± 12.5	0.3
HOMA-IR	3.29 ± 0.7	3.39 ± 0.8	0.37	3.61 ± 0.69	0.84
LH/ FSH ratio	2.1± 0.79	2.2± 0.8	0.39	2.05± 0.69	0.65
Total Cholesterol (mg/dl)	181.5 ± 20.1	181.1 ± 21.2	0.89	180.9 ± 22.1	0.84
Triglycerides (mg/dl)	107.8 ± 21.1	108.2 ± 25.1	0.9	109.1 ± 22.1	0.70
VLDL (mg/dl)	21.56 ± 4.2	21.64 ± 5.02	0.9	21.82 ± 4.42	0.68
LDL (mg/dl)	110.84 ± 11.98	110.16± 11.78	0.7	110.13± 13.58	0.71
HDL (mg/dl)	49.1± 3.9	49.3 ± 4.4	0.74	48.95 ± 4.1	0.80
Estradiol, pg/ml	56.4± 29.2	57.2± 26.2	0.84	55.9 ± 27.2	0.98
Progesterone, ng/ml	0.88 ± 0.3	0.91 ± 0.22	0.44	0.81± 0.69	0.071
Free Testosterone	3.9 ± 0.8	4.1 ± 0.75	0.07	3.4 ± 0.8	1
Total testosterone, ng/ml	0.79± 0.6	0.82± 0.71	0.75	0.81± 0.69	0.76
Prolactin ng/ml	14.6 ± 2.9	14.2 ± 3.6	0.41	14.3 ± 3.1	1
Dehydroepiandrosterone sulfate, pg/dl	314.2± 64.2	312.8± 65.4	0.88	313.1 ± 64.8	0.90
TSH mIU/l	3.6 ± 0.6	3.75 ± 0.7	0.12	3.76± 0.59	0.072
HMW Adiponectin ng/ml	3.9± 2.3	3.8± 2.1	0.76	3.75± 2.2	0.65

The study parameter in group B after treatment were compared to those in group A. We found no statistically significant differences between groups in all parameters except the fasting insulin which was significantly lower and the fasting blood glucose which was higher in group B when compared to group A (Table 2). On the other hand, in group C (after treatment) we found that BMI, fasting insulin, LH/ FSH ratio, total cholesterol, triglycerides, LDL, TG, VLDL, HDL, estradiol, progesterone, free and total testosterone, prolactin, dehydroepiandrosterone sulfate, TSH and HMW adiponectin were statistically different.

Table 2: Biochemical screening for the study parameters in subjects in group A (control, n=90), group B (after treatment with 300mg omega-3, n=90) and group C (after treatment with 600mg omega-3, n=90).

parameter	Control group A	Group B After treatment	P value	Group C After treatment	P value
BMI (kg/m2)	28.5± 1.69	28.4 ± 2.79	0.77	25.9± 1.5	0.000
Fasting insulin(μU/ml)	18.4± 2.9	16.61± 4.05	0.000	19.5 ± 2.96	0.012
Fasting blood glucose (mg/dl)	72.63± 11.8	78.01± 11.03	0.001	65.2± 8.5	0.000
HOMA-IR	3.29 ± 0.7	3.2± 0.75	0.406	3.13± 0.5	0.07
Total Cholesterol (mg/dl)	181.5 ± 20.1	181.02± 20.9	0.89	170.5 ± 18.2	0.000
Triglycerides (mg/dl)	107.8 ± 21.1	109.1± 24.9	0.9	101.2 ± 18.4	0.02
VLDL (mg/dl)	21.56 ± 4.2	21.82 ± 4.98	0.705	20.24 ± 3.68	0.026
LDL (mg/dl)	110.84 ± 11.98	109.19 ± 11.97	0.35	99.36 ± 12.42	0.000
HDL (mg/dl)	49.1± 3.9	50.01± 3.95	0.74	50.9 ± 2.1	0.000
HMW Adiponectin ng/ml	3.9± 2.3	3.95 ± 2.95	0.76	5.61 ± 1.89	0.000

The study parameters in group B after treatment were measured and compared with those in group B before treatment. No significant differences in the study parameters were reported between measurements before and after treatment.

Table 3: Biochemical screening for the study parameters in subjects in group B before and after treatment, n=90.

Parameter	Group B (Before treatment)	Group B after treatment	P value
BMI (kg/m2)	28.3 ± 2.84	28.4 ± 2.79	0.81
Fasting insulin, μU/ml	17.7± 3.3	16.61± 4.05	0.048
Fasting blood glucose(mg/dl)	77.79± 10.5	78.01± 11.03	0.89

Cont... **Table 3: Biochemical screening for the study parameters in subjects in group B before and after treatment,**

HOMA-IR	3.39 ± 0.8	3.2± 0.75	0.08
Total Cholesterol (mg/dl)	181.1 ± 21.2	181.02± 20.9	0.97
Triglycerides (mg/dl)	108.2 ± 25.1	109.1± 24.9	0.80
VLDL (mg/dl)	21.64 ± 5.02	21.82 ± 4.98	0.8
LDL (mg/dl)	110.16± 11.78	109.19 ± 11.97	0.58
HDL (mg/dl)	49.3 ± 4.4	50.01± 3.95	0.25
HMW Adiponectin ng/ml	3.8± 2.1	3.95 ± 2.95	0.96

We found that BMI, fasting insulin, HOMA-IR, LH/ FSH ratio, total cholesterol, triglycerides, LDL, TG, VLDL, HDL, estradiol, progesterone, free and total testosterone, prolactin, dehydroepiandrosterone sulfate, TSH and HMW adiponectin, BMI, fasting glucose, total cholesterol, triglycerides and LDL were in group C after treatment were significantly different when compared to those in the same group before treatment (Table 4).

Table 4: Biochemical screening for the study parameters in subjects in group C before and after treatment, n=90.

parameter	**Group C Before treatment**	**Group C after treatment**	**P value**
BMI (kg/m2)	28.1 ± 1.98	25.9± 1.5	0.000
Fasting insulin, µIU/ml	18.1± 3.1	19.5 ± 2.96	0.002
Fasting glucose (mg/dl)	80.89± 12.5	65.2± 8.5	0.000
HOMA-IR	3.61 ± 0.69	3.13± 0.5	0.000
Total Cholesterol (mg/dl)	180.9 ± 22.1	170.5 ± 18.2	0.000
Triglycerides (mg/dl)	109.1 ± 22.1	101.2 ± 18.4	0.01
VLDL (mg/dl)	21.82 ± 4.42	20.24 ± 3.68	0.009
LDL (mg/dl)	110.13± 13.58	99.36 ± 12.42	0.000
HDL (mg/dl)	48.95 ± 4.1	50.9 ± 2.1	0.000
HMW Adiponectin ng/ml	3.75± 2.2	5.61 ± 1.89	0.000

Discussion

PCOS is a multifactorial disease in which the pathogenesis could be related to proinflammatory state and changes in adipokines.[19] The present study involved the evaluation of adiponectin levels in patients with PCOS treated with omega-3 using two different doses. While using a dose of 300 mg/day omega-3 induced no significant difference in the level of adiponectin between the control and treatment, increasing the dose to a double resulted in significantly higher level of adiponectin.

A previous study showed that treatment with omega-3 for 8 weeks resulted in increased level of adiponectin.[20,21] In addition, Higuchi and colleagues reported an elevated level of adiponectin in animals received fish oil.[22] A further study has shown consistent findings with ours when Mohammadi et al.[23] recorded that taking the omega-3 for 8 weeks lead to increase level of adiponectin. It has been shown that the longer the period of administrating omega-3 the higher the level of adiponectin. Poreba and colleagues found that 6 weeks of omega-3 tratment had no effect on adiponectin in women with PCOS.[24]

The present study that show significantly decreased in total cholesterol, triglycerides, triglycerides, LDL-c as well as significantly increased HDL-c with omega-3 either treatment (300mg or 600mg for 7 or 14 weeks). Similar findings were shown by several study .[25]

The study showed that a significantly elevated level of adiponectin when doubled the dose of omega-3 fatty acids from 300mg /day to 600 mg/day dose for 14 weeks after compared with control group. This means that the dose and time duration can significantly improve the effectiveness of omega-3.

We found that 600mg omega-3 for 14 weeks significantly affected the BMI and 600mg omega-3 for 14 weeks, findings that agree with a previous study. [26] The significant changes in lipid profile may be due to the activation of enzyme AMP-activated protein kinase which plays an important role in lipolysis and lipogenesis .[27]

By using two different doses of omega-3, we found that it can modulate adiponectin levels in women with PCOS in a dose-dependent manner.

Ethical Clearance: The Research Ethical Committee at scientific research by ethical approval of both MOH and MOHSER in Iraq

Conflict of Interest: Non

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