Preconception Dieting regimen and Aerobic Exercise can reduce incidence of Early Pregnancy Loss in PCOS women through normalization Kisspeptin levels

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Abstract
Objectives: To evaluate the ability of preconception weight reduction and aerobic exercise intervention to improve early pregnancy loss (EPL) rate among women with polycystic ovary syndrome and affect pre-interventional (Pre-I) serum levels of kisspeptin1 (KP1) and tumor necrosis factor-α (TNF-α).

Patients & Methods: 531 PCOS women had previous EPL underwent 12-wk intervention composed of high-fiber/low calories dieting regimen plus supervised regularly increasing intensity aerobic exercise. Treatment protocol included clomiphine citrate (50 mg) twice daily since day-2 till day-6 of menstrual period and metformin oral tablets (0.5 gm three times daily). Pre-I data included determination of body mass index (BMI), estimation of fasting blood glucose (FBG), serum insulin and calculation of homeostasis model assessment for insulin resistance (HOMA-IR) and gave blood samples for later ELISA estimation of serum TNF-α and KP1 and all parameters are re-evaluated at end of intervention (Post-I). Women who got positive chemical pregnancy and negative TUV were collected as group A, women who had positive TUV signs continued their pregnancy till >12 GW were included as Group B, while women who developed EPL were grouped as Group C.

Results: Study group A-C included 56, 66 and 41 women, respectively. Post-I body weight, BMI, FBG, FSI and HOMA-IR score, and serum TNF-α and KP1 were significantly lower compared to Pre-I data with significant variance among studied groups, but in favor of women of group B. The percentage of decrease of BMI was positively correlated with the percentage of decrease of HOMA-IR and serum TNF-α and KP1 levels. Regression analysis defined high percentage of decrease in serum TNF-α as the most significant predictor for upcoming clinical pregnancy manifestations among women had positive chemical pregnancy followed by percentage of decrease in serum KP1 and BMI, while high percentages of decrease in serum KP1 and HOMA-IR were the significant predictors for continued pregnancy till 12th GW.

Conclusion: Dieting regimen and aerobic exercise allows breaking of the vicious circle of obesity, IR and high serum levels of KP1 and TNF-α mostly through reducing levels of TNF-α and KP1 in accordance with reduction of BMI and IR.

Keywords: Kisspeptin1, Tumor necrosis factor-α, Weight reduction, Aerobic exercise, Early pregnancy loss, PCOS
Introduction

Polycystic ovary syndrome (PCOS) is the most prevalent cause of anovulatory infertility and hyperandrogenism (1) with insulin resistance (IR) and compensatory hyper-insulinemia as the predominant defects in PCOS (2).

Obesity is a global epidemic which is a critical public health problem closely associated with the development of metabolic disease (3). Excess white adipose tissue, specifically visceral adipose tissue is an active endocrine organ producing hormones that control systemic metabolism (4). Obesity is a major factor in development of IR and metabolic features in PCOS patients (5). Lower GLUT4 expression (6) and/or disturbed coupling of insulin with its receptor (7) may underlie IR development with compensatory increase in insulin concentration despite of increased blood glucose levels (6).

The kisspeptin (KP) neuronal system consists of a neuropeptide kisspeptin and its receptor Gpr54 and is a key factor of reproductive regulation through the hypothalamic-pituitary-gonadal axis (8). KP is obligate for normal gonadotropin secretion (9) and its neurons are the principal stimulators of gonadotropin-releasing hormone neurons, so disruption of kiss signaling abrogates the LH surge (10). Moreover, KP signaling has a putative role in direct control of ovarian function, including follicular development, oocyte maturation, steroidogenesis, and ovulation (8). Dysregulation or naturally occurring mutations of the kisspeptin/KISS1R system may negatively affect the ovarian function, leading to reproductive pathology or female infertility (11).

Early pregnancy loss (EPL) is a disastrous state with depressing psychological impacts on affected women especially if recurrent. Infertile PCOS women are more vulnerable to EPL as documented by Nawaz & Rizvi (12), Banu et al. (13) and Al-Biate (14) who reported EPL rates of 36%, 49.4% and 75%, respectively, among pregnant women with PCOS. However, Joham et al. (15) documented that PCOS was not independently associated with EPL and multiple coincident factors and events may predispose to or precede the development of EPL.

Objectives

The current study targets to evaluate the ability of preconception weight reduction and aerobic exercise intervention to improve EPL rate among PCOS women and affect the pre-interventional KP1 and TNF-α serum levels.

Design

Prospective multicenter interventional study

Setting

University, Health Insurance and multiple private Hospitals, Benha and Tanta, Egypt
Patients & Methods

After approval of the study protocol by the Local Ethical Committee, all PCOS women attended the Gynecology Outpatient Clinics were eligible for evaluation. PCOS was diagnosed on fulfillment of at least two of the Rotterdam criteria for PCOS diagnosis including polycystic ovaries with >12 follicles in range of 2–9 mm and/or an ovarian volume >10 ml per ovary by vaginal ultrasound, hyper-androgenemia with serum total testosterone level >0.8 ng/ml and/or oligomenorrhea or amenorrhea (16, 17). Inclusion criteria included history of previous EPL even once with no male factor for infertility or inducing pregnancy loss and have regular intercourse especially at time of suspected ovulation. Exclusion criteria included obesity inducing endocrinopathy, current pregnancy, using hormonal contraception within the last 6 months prior to inclusion in the study, uterine anatomical anomalies or endometriosis.

The proposal and possible benefits of the preconception intervention protocol were fully described to the couples, fulfilled the inclusion criteria, by the participating dietician (Wahab SM) and physiotherapist (Said EA). Couple accepted to participate in the study were asked to sign fully informed written consent to undergo the intervention and attend regularly for follow-up, and then were clinically evaluated for determination body height and weight and body mass index (BMI) was calculated as weight (kg)/height (m²) (18). Obesity grades were defined as average weight (BMI<24.9 kg/m²), overweight (BMI=25–<30 kg/m²) and obese (BMI=≥30–<35kg/m²) and morbid obese (BMI≥35 kg/m²) (19). All women were asked to visit their dietician while being overnight fasting to give blood samples for routine and study investigations and according to estimated fasting blood glucose (FBG) and serum insulin (FSI) patients were evaluated for insulin resistance (IR) using homeostasis model assessment IR (HOMA-IR) score calculated as following fasting serum insulin (µU/ml) x [fasting plasma glucose (mg/ml)/18]/22.5 (20) and women had HOMA-IR score ≥2 were considered IR (21).

Intervention Protocol
A- Dietary regimen

All women asked to follow 12-wk dietary intervention based to provide ≈ 6000 kJ/day (22) which involves eating food containing more plant (>400 gm/day of vegetables and fruits), less animal food (≈100 gm meat) and low carbohydrate and fat (≤90 gm brown bread, ≤200 gm potatoes, rice, pasta or beans, ≤30 gm butter and oil, ≈1.5 slice of cheese and ≈450 ml of milk), as recommended in the guidelines of the Dutch Food Guide (23) that was revised and updated in 2015-2016 as documented in Bulletin of Food and Agriculture Organization of UN.
B- Exercise protocol

All study women undertook exercise training, supervised by the physiotherapist, started with 3-session/wk; each session extends 45–60 min on a motorized treadmill. Training protocol consisted of moderate-intensity exercise in the form of 20-60 minutes of walking with intervening high-intensity training for 6-8 minute and 2-min passive recovery periods \(^{24}\). The training course started with 3-session course which is progressed to achieve eight repetitions by the 4\(^{th}\) wk and during the next 4-wk duration the recovery time was reduced to 1-min by the 8\(^{th}\) wk and this regimen was continued till the end of the 12\(^{th}\) wk. Target exercise heart rates are 75–80% of maximal heart rate [HR\(_{\text{max}}\)] during walking and ~95–100% HR\(_{\text{max}}\) during high-intensity interval training \(^{25}\); average heart rate was assessed by heart rate monitors (Polar Electro Oy, Kempele, Finland). Exercise adherence was mandatory and any missed session was replaced to complete the 12-wk training course.

C- Treatment protocol

Doses of the used drugs were calculated and adjusted by hospital clinical pharmacist. Applied treatment protocol consisted of clomiphine citrate (CC) 50 mg at morning and evening starting on day-2 till day-6 of a menstrual period or after a progestogen withdrawal bleeding using medroxyprogesterone acetate. All women received metformin oral tablet in a dose of 0.5 gm three times daily, with the starting dose was 0.5 gm once daily to be gradually increased to the full dosage of 1.5 g/d.

D- Laboratory workup

Under complete aseptic conditions, two fasting venous blood samples (5 ml) were withdrawn from the antecubital vein, the 1\(^{st}\) at time of enrolment and the 2\(^{nd}\) at the end of intervention. The obtained blood sample was divided into two parts:

i- The 1\(^{st}\) part was collected in an EDTA containing tube for estimation of FBG level using glucose oxidase method \(^{26}\).

ii- The 2\(^{nd}\) part was kept in plane container, left to clot and then serum was separated by 5-min centrifugation at 3000 rpm and stored at –80°C. Serum insulin, TNF-\(\alpha\) and kisspeptin1 levels were measured using enzyme linked immunosorbent assay (ELISA) kits according to the manufacturer's instructions and were read using a 96 well microplate ELISA reader (Dynatech. MR 7000).

- Human insulin was measured with the enzyme linked immunoassay (ELISA) kit (catalogue no. ab200011, Abcam, Inc., Cambridge, Massachusetts, UK) by quantitative sandwich enzyme immunoassay technique \(^{27}\).

- Human TNF-\(\alpha\) was measured with the enzyme linked immmunoassay (ELISA) kit (catalogue no. ab46087, abcam Inc., San Francisco, USA) by quantitative sandwich enzyme immunoassay technique \(^{28}\).
- Human kisspeptin1 was measured with the enzyme linked immunoassay (ELISA) kit (catalogue no. MBS081321, MyBioSource, Inc., California, San Diego, USA) by quantitative sandwich enzyme immunoassay technique (29).

**Study Outcomes**

**a. Primary outcomes**

1. BMI and HOMA-IR scores were reevaluated for all enrolled women at the end of the intervention (Post-I) and addressed as the percentage of BMI loss (%BMIL) and percentage of HOMA-IR change (%HOMA-IR) in relation to pre-intervention (Pre-I) data.

2. The pregnancy rate among studied population; pregnancy was diagnosed depending on having positive serum pregnancy test (positive chemical pregnancy) and was assured using TVU (positive clinical pregnancy). Gestational age was defined as the number of gestational weeks (GW) since the last menstrual period.

3. The frequency of pregnancy loss within the 1st 12-GW gestation among women got pregnant (EPL rate).

4. The change of serum KP1 and TNF-α levels at end of the intervention, relation of the percentage of change and its predictability of to the resultant %BMIL, %HOMA-IR change, and pregnancy and EPL rate.

**Study grouping**

The study intended include PCOS women who got positive chemical pregnancy for evaluation of outcomes. Women who had negative TUV were collected as group A, while women who had positive TUV signs were followed up till 12 GW and those continued their pregnancy successfully for >12 GW were included as Group B, while women who developed EPL were grouped as Group C. Serum levels of KP1 and TNF-α were estimated in blood samples of women got positive chemical pregnancy, while blood samples of women failed to get pregnancy were discarded.

**Statistical analysis**

Recent findings supposed that the frequency of natural conceptions leading to ongoing pregnancies in obese infertile women ranged between 16% in those received no intervention and 26% in women undertook lifestyle intervention (30), so to get a study power of 80% with α value=0.05, β value=20%, the initial study population must be ≥518 women.

Obtained data were presented as mean±SD, numbers and percentages. Results were analyzed using one-way ANOVA Test for intragroup variance and paired t-test for inter-group variance. Receiver operating characteristic (ROC) curve analysis was used to evaluate the sensitivity and specificity of studied variables as predictors for changes as judged by the area under the
curve (AUC) compared versus the null hypothesis that AUC=0.05. Regression analysis (Stepwise Method) was used to define the persistently significant predictors for occurrence of pregnancy and EPL among PCOS pregnant women. Statistical analysis was conducted using the IBM SPSS (Version 23, 2015) for Windows statistical package. P value <0.05 was considered statistically significant.

Results
Throughout the study 610 PCOS women attended the OPC; 69 were excluded for not fulfilling inclusion criteria and 541 signed consent, but 10 were lost during the intervention period. Fortunately, 166 developed positive chemical pregnancy (30.7%), 110 women (20.3%) showed signs of clinical pregnancy, but 41 women (37.3%) developed EPL (Group C) and 69 women (Group B) completed uninterrupted 12-GW (Fig. 1).

Baseline age, BMI data and HOMA-IR scores (Pre-I) showed non-significant (p>0.05) difference between patients categorized according to outcome. The applied intervention did favorably for all patients got pregnant as manifested by significantly lower body weight (BW) and BMI in comparison to Pre-I data with mean BMIL% of 16.7 (±6.6). Moreover, all patients got significantly lower FBG, FSI and HOMA-IR score in comparison to Pre-I data with mean HOMA-IR-L% by 18.5 (±6.84). Despite of this significant improvement, patients categorized according to outcome showed significant variance as regards Post-I body weight (p=0.0002), BMIL% (p=0.0017), FSI...
(p=0.011), HOMA-IR score (p=0.009) and HOMA-IR-L% (p=0.0001) and this variance was in favor of women of group B (Table 1).

Table (1): Pre- and Post-intervention BMI and HOMA-IR score of studied chemically diagnosed pregnant PCOS women categorized according to pregnancy outcome at the 12th GW

<table>
<thead>
<tr>
<th>Data</th>
<th>Patients</th>
<th>Total pregnant women</th>
<th>Chemical Pregnancy only (Group A)</th>
<th>Uninterrupted 12-GW (Group B)</th>
<th>EPL (Group C)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number (%)</td>
<td>166 (30.7%)</td>
<td>56 (39.7)</td>
<td>69 (35.6%)</td>
<td>41 (24.7%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Age (years)</td>
<td>28.6±4.5</td>
<td>33.7±4.9</td>
<td>25.8±3.3</td>
<td>26.2±4.2</td>
<td>0.109</td>
<td></td>
</tr>
<tr>
<td>Height (cm)</td>
<td>166.8±3.2</td>
<td>168.5±1.8</td>
<td>166.1±3.3</td>
<td>166.6±4</td>
<td>0.065</td>
<td></td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>Pre-I: 90.1±7.4</td>
<td>91±8.6</td>
<td>89.5±5.1</td>
<td>89.8±8.8</td>
<td>0.526</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Post-I: 74.9±6.6*</td>
<td>77.8±7.1*</td>
<td>73.1±5.8*</td>
<td>74±6.1*</td>
<td>0.0002</td>
<td></td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>Pre-I: 32.4±2.2</td>
<td>31±2.7</td>
<td>32.4±1.6</td>
<td>32.7±2.4</td>
<td>0.318</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Post-I: 26.9±2.1*</td>
<td>27.4±2.3*</td>
<td>26.5±2.1*</td>
<td>27±1.9*</td>
<td>0.063</td>
<td></td>
</tr>
<tr>
<td>BMIL%</td>
<td>Pre-I: 16.7±6.6</td>
<td>14.1±7.3</td>
<td>18.2±6.1</td>
<td>17.4±5.5</td>
<td>0.0017</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Post-I: 14.2±4.5</td>
<td>12.6±6.1</td>
<td>14.2±4.6</td>
<td>13.6±6.1</td>
<td>0.029</td>
<td></td>
</tr>
<tr>
<td>FBG (mg/dl)</td>
<td>Pre-I: 127.8±10.9</td>
<td>129.5±11.7</td>
<td>125.8±10.4</td>
<td>128.8±10.5</td>
<td>0.132</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Post-I: 121.8±9.9*</td>
<td>123.3±9.7*</td>
<td>120±9.7*</td>
<td>123±10.1*</td>
<td>0.114</td>
<td></td>
</tr>
<tr>
<td>FSI (µU/ml)</td>
<td>Pre-I: 18.36±5.56</td>
<td>19.14±6.21</td>
<td>17.85±4.56</td>
<td>18.16±5.66</td>
<td>0.424</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Post-I: 15.75±5.22*</td>
<td>17±5.64*</td>
<td>14.35±4.09*</td>
<td>16.38±5.86*</td>
<td>0.031</td>
<td></td>
</tr>
<tr>
<td>HOMA-IR</td>
<td>Pre-I: 1.71±0.56</td>
<td>1.81±0.63</td>
<td>1.64±0.47</td>
<td>1.65±1.71</td>
<td>0.276</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Post-I: 1.4±0.5*</td>
<td>1.53±0.54*</td>
<td>1.26±0.42*</td>
<td>1.47±0.56*</td>
<td>0.009</td>
<td></td>
</tr>
<tr>
<td>HOMA-IR-L%</td>
<td>Pre-I: 18.5±6.84</td>
<td>15.39±3.7</td>
<td>23.55±6.42</td>
<td>14.35±5.27</td>
<td>0.0001</td>
<td></td>
</tr>
</tbody>
</table>

Data are presented as mean±SD; EPL: Early pregnancy loss; Pre-I: Pre-intervention; Post-I: Post-intervention; BMI: Body mass index; BMIL%: Percentage of change of post-intervention BMI; FBG: Fasting blood glucose; FSI: Fasting serum insulin; HOMA-IR: homeostasis model assessment-Insulin resistance; HOMA-IR-L%: Percentage of change of post-intervention HOMA-IR; P value indicates significance of variance among groups; *: indicates significance of difference between pre- and post-intervention estimated levels.

There was non-significant difference between Pre-I serum levels of TNF-α and KP1 among patients' groups. Post-I serum levels of both parameters were significantly lower compared to Pre-I levels in total patients and within each group. However, Post-I serum levels showed significant variance between patients of the three groups. Similarly, the percentage of decrease of serum levels of TNF-α and KP1 were significantly varied between the three groups (Table 2).

The percentage of decrease of BMI showed positive correlation with the percentage of decrease of serum levels of TNF-α (r=0.188, p=0.015) and KP1 (r=0.207, p=0.007). Moreover, the percentage of decreased HOMA-IR score showed positive significant correlation with BMIL% (r=0.202, p=0.009) and the percentage of decrease of serum levels of TNF-α (r=0.299, p=0.0009) and KP1 (r=0.426, p=0.0004).
Table (2): Pre- and Post-intervention serum TNF-α and KP1 of studied chemically diagnosed pregnant PCOS women categorized according to pregnancy outcome at the 12th GW

<table>
<thead>
<tr>
<th>Data</th>
<th>Patients</th>
<th>Total pregnant women</th>
<th>Chemical Pregnancy only (Group A)</th>
<th>Uninterrupted 12-GW (Group B)</th>
<th>EPL (Group C)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>TNF-α (ng/ml)</td>
<td>Pre-I</td>
<td>7.31±3.47</td>
<td>7.37±3.16</td>
<td>7.19±3.49</td>
<td>7.44±3.88</td>
<td>0.925</td>
</tr>
<tr>
<td></td>
<td>Post-I</td>
<td>4.81±2.11*</td>
<td>5.8±2.45*</td>
<td>4.07±1.47*</td>
<td>4.72±2.05*</td>
<td>0.0002</td>
</tr>
<tr>
<td></td>
<td>Change (%)</td>
<td>31.14±17.1</td>
<td>20.4±10.51</td>
<td>38.82±18.2</td>
<td>32.6±15</td>
<td>0.0001</td>
</tr>
<tr>
<td>KP1 (ng/ml)</td>
<td>Pre-I</td>
<td>1.67±0.09</td>
<td>1.68±0.09</td>
<td>1.66±0.09</td>
<td>1.66±0.1</td>
<td>0.418</td>
</tr>
<tr>
<td></td>
<td>Post-I</td>
<td>1.52±0.11*</td>
<td>1.58±0.11*</td>
<td>1.475±0.12*</td>
<td>1.54±0.1*</td>
<td>0.0001</td>
</tr>
<tr>
<td></td>
<td>Change (%)</td>
<td>8.45±4.84</td>
<td>6.17±3.22</td>
<td>11.1±5.77</td>
<td>7.08±2.2</td>
<td>0.0001</td>
</tr>
</tbody>
</table>

Data are presented as mean±SD; EPL: Early pregnancy loss; TNF-α: Tumor necrosis factor-α; KP1: Kisspeptin 1; P value indicates significance of variance among groups; *: indicates significance of difference between pre- and post-intervention estimated levels.

Regression analysis defined high percentage of decrease in serum TNF-α as the most significant predictor for upcoming clinical pregnancy manifestations among women had chemical diagnosis of pregnancy with β=0.326 (p=0.0006) followed by the percentage of decrease in serum KP1 (β=0.200, p=0.007) and lastly, the percentage of decrease of BMI (β=0.164, p=0.020). The high percentages of decrease in serum KP1 (β=0.535, p=0.0003) and HOMA-IR (β=0.168, p=0.043) were the significant predictors for continued pregnancy till 12th GW. ROC curve analysis assured the predictability of low percentages of change in serum KP1 (AUC=0.146, p=0.0002; 95%CI=0.071-0.221) and HOMA-IR (AUC=0.288, p=0.0006; 95%CI=0.194-0.382) as significant sensitive predictors for the upcoming EPL (Fig. 2).

Fig. (2): ROC curve analysis for predictors of upcoming EPL among pregnant PCOS women diagnosed clinically
Discussion

The applied intervention composed of high-fiber/low calories dieting regimen plus supervised regularly increasing intensity aerobic exercise assured its efficacy as manifested by the reported post-intervention (Post-I) significant weight loss and decreased BMI with significantly lower FBG, FSI and decreased HOMA-IR score compared to pre-intervention (Pre-I) data.

In line with the efficacy of the proposed intervention, Nybacka et al. (31) found dieting is effective to improve metabolic disturbances in overweight/obese PCOS women and increased fiber and reduced trans fatty acid intake are primary predictors of metabolic improvement and weight control. Thereafter, Kogure et al. (32) found resistance exercise programs for PCOS women promoted increases in muscle strength and lean body mass with decreased testosterone concentration, glycemia and body fat. Also, Costa et al. (33) reported that supervised aerobic exercise training improved cardiorespiratory fitness, and cardiometabolic profile of overweight/obese PCOS women and decreased BMI, waist circumference, blood pressure, and total cholesterol level. Moreover, Zhang et al. (34) recommended avoidance of long-term sedentary lifestyle habits and to add to the duration of, or enhancing the intensity of, physical activity for women to guard against development of PCOS.

Interestingly, Post-I serum TNF-α and kisspeptin1 (KP1) levels were significantly lower than Pre-I levels in all women. These findings allowed supposition that excess adipose tissue may be the source of both cytokines or at least spotted light on an association between excess adipose tissue and the elevated serum levels of these cytokines. In support of this assumption, the reported extent of decrease of BMI and HOMA-IR score showed positive significant correlation with the extent of decrease in serum TNF-α and KP1.

In accordance with these data, Diane et al. (35) using obese PCOS-prone animals reported that the combination of exercise and dietary energy restriction exerts beneficial effects on cardiometabolic and reproductive indices with normalization of the hypothalamic neuropeptide; Kisspeptin. Also, Wang et al. (36) experimentally reported that GPR54, Kisspeptin-1 receptor directly participates in obesity development by promoting adipocyte differentiation and fat accumulation. Clinically, Hofmann et al. (37) found KP was positively correlated with BMI and body fat mass and negatively associated with physical activity. Also, Kordestani et al. (38) detected higher TNF-α serum levels in PCOS women and these levels were significantly correlated with HOMA factor.

Current study reported positive chemical pregnancy rate of 30.7%; a finding which assures the relationship between PCOS-infertility and obesity with its subsequent sequelae; insulin resistance (IR). In support of this assumption, getting clinically diagnosed pregnancy showed positive significant correlation with BMIL% and percentage of decrease of HOMA-IR score and serum TNF-α and KP1. On the other hand, EPL rate showed negative significant correlation with the extent of change in BMI, HOMA-IR score and
serum TNF-α and KP1 levels, thus indicating a possible role of obesity, IR and both cytokines for PCOS-infertility and EPL.

These finding are coincident with Yang & Dhillo (39) who documented that there is an increasing appreciation that KP may act as a signal transmitter between metabolic status and reproductive function and exogenous KP can stimulate physiological gonadotropin responses in both healthy subjects and those with disorders of reproduction. Recently, Albalawi et al. (40) reported higher LH levels and BMI in PCOS women than controls with positive correlation between plasma KP and LH levels and detected significantly higher frequency of GG genotype of SNP rs4889 C/G in obese PCOS women who had higher KP and FSH levels. Also, Oróstica et al. (41) reported that in human endometrial stromal cells, high TNF-α levels negatively affect insulin action through decreased adiponectin signaling and GLUT-4 protein and increased NFκB nuclear content.

Moreover, Kaya et al. (42) detected significantly higher KP levels in PCOS infertile women than in infertile women secondary to male factor or unexplained infertility and considered higher KP levels as reliable marker for PCOS diagnosis and Umayal et al. (43) found serum KP levels are higher in women with PCOS manifesting from adolescence than controls and propose its estimation as a useful marker to recognize PCOS.

Multiple studies provided in vivo evidence for the relation between disturbed KP levels and PCOS where Aliabadi et al. (44) found letrozole induced POCS is associated with disturbed number of KP-positive cells in hypothalamic nuclei; increased in arcuate (Arc) and decreased in anteroventral periventricular (AVPV) nuclei compared to control animals and Hu et al. (45) found PCOS in mice was improved on corcetin treatment via re-adjusting hypothalamic KP expression levels which was increased in AVPV but decreased in Arc nuclei on corcetin therapy. Moreover, Iwasa et al. (46) supposed vicious cycle drawn by disturbed secretion of KP which is a potent regulator of gonadotropin-releasing hormone (GnRH) functions leading to disrupted GnRH levels and actions, so it in turn suppressed female reproductive functions.

Conclusion

Obesity and insulin resistance could be considered as major obstacles for fertility in PCOS women. Excess adipose tissue also acts as a source for inflammatory cytokines. PCOS was also associated with disturbed levels of hypothalamic neuropeptides affecting reproduction. Dieting regimen and aerobic exercise allows breaking of this vicious circle mostly through reducing levels of inflammatory cytokines and kisspeptin in accordance with reduction of BMI and IR.

Acknowledgment

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