# Distribution of Adiponectin Multimeric Forms In Chinese Women With Polycystic Ovary Syndrome and Their Relation To Insulin Resistance

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## Abstract

**Objective**—Adiponectin, an abundant adipokine with insulin sensitizing properties, exists different multimeric forms, including low molecular weight (LMW), medium molecular weight (MMW), and high molecular weight (HMW) species. Alterations in the distribution of adiponectin multimers and the relationship between adiponectin multimers and insulin resistance (IR) in women with the polycystic ovary syndrome (PCOS) remain unclear. To compare adiponectin multimerization status and estimate of insulin sensitivity in Chinese women with PCOS compared with age and body mass index (BMI)-matched controls.

**Methods**—Cross-sectional study involving 64 Chinese women with PCOS and 59 normal women. Circulating total adiponectin and its multimeric forms were determined by ELISA and insulin resistance was estimated using the homeostasis assessment insulin resistance index (HOMA-IR).

**Results**—After controlling for BMI status, levels of both total and HMW adiponectin were significantly lower in women with PCOS compared with normal women ($P<0.05$). Furthermore, HMW adiponectin provided a stronger contribution to models predicting insulin resistance than total adiponectin. Lastly, decreased HMW adiponectin was associated with increased HOMA-IR in both normal and PCOS women, and this association was independent of both overall adiposity and visceral adiposity.

**Conclusion**—Levels of both total and HMW adiponectin are decreased in Chinese women with PCOS compared with normal control women and the differences in HMW adiponectin persists after controlling for BMI. Furthermore, HMW adiponectin is a stronger predictor of insulin resistance in both women with PCOS and normal women than total adiponectin.

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## Introduction

The polycystic ovary syndrome (PCOS) is one of the most common endocrine diseases in reproductive age women and is a common cause of menstrual dysfunction, infertility, and hirsutism [1]. Insulin resistance and obesity play a central role in the pathogenesis of PCOS [2–3]. In addition to the burden of insulin resistance attributed to obesity, women with PCOS demonstrate a form of insulin resistance intrinsic to the syndrome [4]. However, the mechanism responsible for the insulin resistance intrinsic to the syndrome is unclear [5]. Therefore, an important consideration is whether adipocytokines such as adiponectin, a potential mediator of insulin sensitivity [6], are also implicated in the pathogenesis of PCOS. Levels of adiponectin, an abundant adipocyte-derived cytokine, are strongly correlated with measures of insulin sensitivity [7–8] and low levels of adiponectin have been associated with future risk of cardiovascular disease and type 2 diabetes [9–10]. In recent years, the relationship between the physiological action and structure of adiponectin has attracted wide attention.

Adiponectin circulates in different multimer complexes, specifically classified as high molecular weight (HMW) multimers, medium molecular weight hexamers (MMW), and low molecular weight (LMW) trimers [11]. Recent clinical studies have revealed that the high molecular weight complex is the most biologically potent form and plays a key role in the regulation of insulin resistance [12–13]. Although the relationship between alterations in adiponectin multimerization and insulin resistance in women with PCOS compared with normal women remains unclear, we hypothesized that the distribution of adiponectin multimeric forms, in particular the HMW form, is decreased in women with PCOS compared with normal control women. We further hypothesized, that this difference is independent of obesity, and that alterations in HMW adiponectin contribute to the insulin resistance intrinsic to the syndrome.

Thus, in the present study we evaluated adiponectin multimerization status and estimates of insulin sensitivity in Chinese women with PCOS compared with age and body mass index (BMI)-matched controls.

## Materials and Methods

Sixty-four women with PCOS (43 obese, 21 normal weight) from Shanghai, China and fifty-nine normal cycling, non-hirsute women (38 obese, 21 normal weight) between 16–35 years of age were enrolled in the study. PCOS was defined using the 1990 National Institutes of Health criteria [14]. Pregnancy was excluded by a urine pregnancy test. All women had normal thyroid function and prolactin levels, and late-onset nonclassic congenital hyperplasia was excluded by a basal 17-α hydroxyprogesterone value of less than 300 ng/dL [15]. Women receiving glucocorticoids, antiandrogens, or oral contraceptives within the previous 30 days, or ovulation induction agents, antiobesity medications, or insulin sensitizing agents within the previous 60 days were excluded. All women were evaluated by transvaginal ultrasonography to define ovarian morphology [16]. Control subjects were screened by medical history, physical examination, laboratory evaluation and transvaginal ultrasound. Normal weight women were defined as those with a body mass index (BMI) <25 kg/m$^2$ and obesity was defined as those with a BMI of 25 kg/m$^2$ according to the 2000 WHO-WPR criteria [17]. Women with PCOS and control women were matched for age,

BMI and fat mass. All study evaluations and procedures were conducted in accordance with the guidelines of Helsinki Declaration on human experimentation. The study was approved by the ethics committee of Shanghai Renji hospital and all subjects provided written informed consent.

## Anthropometric measurements

The height and weight of each subject wearing light clothing were measured to the nearest 0.1 cm and 0.1 kg, respectively, using a digital scale and stadiometer. BMI was calculated as body weight (kg) divided by height (m) squared. Waist circumference (WC) and hip circumference (HC) were measured by a single individual. WC was determined by measuring the circumference at the narrowest point between the lower border of the rib cage and the iliac crest. HC was determined by measuring the circumference at the level of the symphysis pubis and the greatest gluteal protuberance. The waist-to-hip ratio (WHR) was then calculated by dividing the WC by the HC. Percent body fat was assessed by foot-to-foot measures of bioelectrical impedance obtained using a TBF-300 body composition analyzer (TANITA, U.K. Ltd., Middlesex, UK, www.tanita.com).

## Laboratory assays

All laboratory evaluations were performed at 0800 h after an overnight fast during the early follicular phase (days 2–5) of a spontaneous menstrual cycle, except in subjects with amenorrhoea >3 months who were examined randomly. Fasting glucose and insulin samples were stored at 4° C and analyzed the day of sampling. All serum samples for total adiponectin and adiponectin multimeric forms were stored at −70° C until assayed.

Competitive electrochemiluminescence immunoassays on the Elecsys autoanalyzer 2010 (Roche Diagnostics, Indianapolis, IN) were used to quantify serum total testosterone. Sex-hormone binding globulin (SHBG) levels were measured by chemiluminescent immunoassay (Elecsys autoanalyzer 2010, Roche Diagnostics) validated for plasma SHBG [18]. The coefficient of variation for SHBG using this methodology was 6%. Free testosterone values were calculated based on total testosterone and SHBG levels according to the method outlined by Vermeulen et al [19] assuming an albumin concentration of 4g/dl (http://www.issam.ch/freetesto.htm). Plasma glucose was determined using the glucose oxidase methodology. All measurements were performed with Roche reagents (D 2400 and E 170 Modular Analytics modules with Roche/Hitachi analyzers; Roche Diagnostics, Indianapolis, USA). Insulin levels were measured by radioimmunoassay (RIA). The intra-assay CV of insulin and steroid hormone assays were 5.5% and <10%, respectively. To estimate insulin resistance, the homeostasis assessment insulin resistance index (HOMA-IR) was calculated according to fasting serum insulin ($\mu$UI ml-1) fasting plasma glucose (mmol/l)/22.5 [20].

Serum total adiponectin, HMW adiponectin, and combined HMW+MMW adiponectin levels were assayed directly in the same plate using a double monoclonal sandwich enzyme-linked immunosorbent assay (ELISA) method (Daiichi pure chemicals, Tokyo, Japan, distributed by ALPCO diagnostics). To detect HMW adiponectin, serum samples were pre-treated with a protease that selectively digested MMW and LMW adiponectin species. The combined HMW and MMW adiponectin concentrations were determined by pre-treating the samples with a protease that specifically digested LMW adiponectin. MMW adiponectin concentrations were obtained by subtracting the HMW adiponectin value from the combined HMW+MMW value. Finally, the LMW adiponectin value was computed by subtracting HMW and MMW adiponectin values from the total adiponectin values. This specific assay for adiponectin multimers demonstrates a sensitivity of 0.04 ng/ml, an inter-assay CV <15%

and an intra-assay CV of 5.3%, 3.3% and 4.1% for total, HMW-adiponectin and MMW +HMW adiponectin, respectively [21].

## Statistical analysis

Results not normally distributed, based on the normal quartile plot, were log-transformed for all statistical analyses and reported back-transformed in their original units. All results were reported as means, or geometric means for log-transformed variables, with 95% CIs. *P*-values <0.05 were considered significant. All statistical analyses were performed using JMP 8.0 software (SAS Institute, Cary, NC).

For continuous variables, subgroup means were compared with one-way analysis of variance (ANOVA) testing considering four subgroups of women: lean control (LC), obese control (OC), lean PCOS (LP), and obese PCOS (OP). Tukey's honestly significantly difference (HSD) test was performed when the result from the ANOVA was statistically significant. The relationships between levels of total and HMW adiponectin and hormonal and metabolic variables were evaluated by Spearman's correlation tests. Two forward, stepwise, multiple linear regression analyses were performed. The first model was performed to determine the best model to predict insulin resistance as estimated by HOMA-IR considering biologically plausible predictors, i.e. PCOS status, age BMI, WHR, total adiponectin, HMW adiponectin, and LMW adiponectin. These independent variables were selected based on literature and because their correlations with insulin sensitivity were significant in the univariate analyses. Based on the results of the first model, a second model was created to determine the best model to predict serum levels of HMW adiponectin considering PCOS status, age, BMI, WHR, and free testosterone as plausible independent variables.

## Results

The clinical characteristics and biochemical variables for the 4 groups of women according to PCOS and BMI status are summarized in Table 1. As expected, mean BMI ($F_{(3,119)}$ =59.50, $P<0.0001$) and percent body fat ($F_{(3,119)}$=83.61, $P<0.0001$), were significantly different between groups, with higher mean values in both groups of obese women compared with groups of lean women. However, there were no significant differences in BMI or percent body fat in LC compared with the LP group or in OC compared with the OP group. Similarly, WC ($F_{(3,119)}$=57.77, $P<0.0001$), HC ($F_{(3,119)}$=43.38, $P<0.0001$), and WHR ($F_{(3,119)}$=64.12, $P<0.0001$) were significantly different between groups. Specifically, mean WC, HC, and WHR values where greater in the obese groups (OC and OP) compared with the lean groups (LC and LP) of women. As with BMI, there were no significant differences in WC, HC, or WHR between control and PCOS women according to BMI status. Mean total and free testosterone values were also significantly different between groups (F $_{(3,119)}$ =4.81, $P=0.003$; F $_{(3,119)}$=10.45, $P<0.0001$, respectively). Total testosterone values where significantly higher in the LP group compared with the other three groups. Free testosterone values where highest in the OP and LP groups and lowest in the LC groups. Significant differences in fasting glucose levels were noted between groups (F $_{(3,119)}$ =9.74, $P<0.001$) with higher mean glucose values in the OP group and lower glucose values in the LC group. Mean HOMA-IR values were significantly different between groups (F $_{(3,119)}$=61.72, $P<0.0001$), with higher HOMA-IR values in women in the obese groups (OC and OP) compared with the lean groups (LC and LP). However, significant differences in HOMA-IR values between OC and OP women or LC and LP women were not observed.

## Levels of total and three oligomeric forms of adiponectin

Figure 1 illustrates the mean values of total adiponectin and the three multimeric forms of adiponectin for each group. Mean levels of total adiponectin were significantly different among groups ($F(3,119)=57.02$, $P<0.001$) with the highest mean value observed in LC women (22.11, 95% CI [18.29–26.73]) and the lowest mean value observed in OP women (4.81[4.22–5.50]). Total adiponectin levels were similar between women in the OC (8.59 [7.46–9.90]) and LP (8.56 [7.08–10.35]) groups. Mean HMW adiponectin values were also significantly different between groups ($F(3,119)=43.51$ $P<0.0001$) with differences between groups similar to the pattern observed in total adiponectin (LC 8.52 [6.69–10.85], LP 4.58 [3.60–5.83], OC 4.05 [3.38–4.85], OP 1.72 [1.45–2.03]). Mean levels of MMW ($F(3,119)=17.32$, $P<0.0001$) and LMW adiponectin ($F(3,119)=12.40$, $P<0.0001$) also differed significantly between groups. Specifically, MMW and LMW adiponectin were highest in the LC women and lowest in the OP women.

When women with PCOS (n=59) and normal control women (n=64) were considered as whole groups, both levels of total ($F(1,121)=49.46$, $P<0.001$) and HMW adiponectin ($F(1,121)=40.95$, P<0.001) remained significantly decreased in women with PCOS compared with normal women. However, ratios of HMW to total adiponectin were not significantly different between women with PCOS and normal control women ($P=0.47$).

## Correlation between adiponectin levels and clinical/biochemical characteristics

Given the association between hypoadiponectinemia and increased adiposity, correlations between total and HMW adiponectin and anthropometric/metabolic parameters were preformed (Table 2). Considering all women (n=123), both total and HMW adiponectin levels had inverse linear relationships with BMI, percent body fat, and WHR ($P<0.0001$). Although neither total or HMW adiponectin levels correlated with total testosterone levels, both total and HMW adiponectin levels demonstrated inverse linear relationships with estimated free testosterone levels ($P<0.001$ and $<0.01$, respectively.) Furthermore, strong positive linear correlation were observed between SHBG levels and both total and HMW adiponectin (P<0.0001 for both). Significant inverse linear relationships were also demonstrated between both total and HMW adiponectin and total cholesterol ($P<0.0001$ for both), LDL cholesterol ($P<0.001$ for both), and triglycerides ($P=0.003$ and 0.02, respectively). However, neither total nor HMW adiponectin levels correlated significantly with levels of HDL cholesterol. Finally, both total and HMW adiponectin levels were found to have strong inverse linear relationships with fasting glucose, fasting insulin, and HOMA-IR values ($P<0.0001$ for all).

## Determinants of insulin resistance

To investigate the independent determinants of insulin resistance among all women studied, we performed a forward, stepwise, multiple regression analysis with HOMA-IR as the dependent variable and PCOS status, age, BMI, WHR, total adiponectin, HMW adiponectin, and LMW adiponectin as potential contributors to insulin resistance. In the analysis, HMW adiponectin, BMI, and WHR, but not PCOS status or total adiponectin, were selected as independent variables for the final model of HOMA-IR. In the final model for HOMA-IR, the adjusted $R^2$ was 0.48 (p<0.0001) and the regression coefficients ($\beta$) were−0.12 ($P=0.02$) for HMW adiponectin, 0.04 ($P<0.0001$) for BMI, and 1.64 ($P=0.02$) for WHR. Thus, HMW adiponectin, BMI, and WHR were all independent predictors of insulin resistance as estimated by HOMA-IR. However, PCOS status and levels of total adiponectin did not appear to contribute significantly to the model for HOMA-IR after controlling for the other potential independent variables.

Since percent body fat estimated by bioimpedence was strongly correlated with BMI ($r$=0.93, $P$<0.001) both variables were not included in the model for insulin resistance to avoid multcollinearity. However, similar results were obtained if percent body fat was included as a dependent variable in the model in place of BMI (data not shown).

### Determinants of HMW adiponectin

After HMW adiponectin was selected as a better predictor of HOMA-IR than total adiponectin in the first linear regression analysis, a second model was created, again using forward, stepwise, linear regression analysis, with HMW adiponectin as the dependent variable and age, PCOS status, free testosterone, BMI, and WHR as potential independent variables. Considering these variables, only free testosterone was not selected as an independent variable for the final model predicting HMW adiponectin. The adjusted $R^2$ for the model predicting HMW adiponectin was 0.40 ($P$<0.0001), and age (β=0.016, $P$=0.05), PCOS status (β= −0.40, $P$<0.0001), and BMI (β= −0.04, $P$<0.01) where all independent contributors to levels of HMW adiponectin in the final model.

## Discussion

In this study, we investigated the distribution of circulating adiponectin oligomers in PCOS women compared with normal control women, and determined which multimeric forms of adiponectin correlated with insulin resistance. There are three principal findings in our study: 1) After controlling for BMI status, levels of both total and HMW adiponectin levels were significantly lower in women with PCOS compared with those in normal women; 2) HMW adiponectin provided a stronger contribution to models predicting insulin resistance than total adiponectin among all women; and 3) decreased HMW adiponectin was associated with increased insulin resistance in both normal and PCOS women, and this association is independent of PCOS status, overall adiposity (BMI), and visceral adiposity (WHR).

Previous studies that assessed only total adiponectin in women with PCOS have yielded conflicting results [22–23]. However, it is noteworthy that adipocytes secret adiponectin as multimeric complexes, and different adiponectin isoforms appear to have unique regulatory effects on metabolic and inflammatory pathways [24–25]. In particular, the HMW form appears to have the most potent insulin sensitizing effects. Recent clinical studies suggest that the HMW form of adiponectin is the primary mediator of the effects of adiponectin on insulin-stimulated glucose metabolism [12,13, 26,27]. In these studies, only HMW adiponectin showed a similar close association with progression to type 2 diabetes, the degree of insulin resistance, and the presence of the metabolic syndrome. Therefore, we suggest that some of the inconsistencies in prior studies investigating alterations in adiponectin in PCOS women may be explained by the failure to assess levels of HMW adiponectin.

In this study, we demonstrated that women with PCOS have decreased levels of both total and HWM adiponectin and that these alterations are independent of BMI. Furthermore, levels of HMW adiponectin demonstrated a strong inverse linear relationship with estimated levels of free testosterone. In light of previous evidence demonstrating that testosterone inhibits the secretion of HMW adiponectin from rat adipocytes [28], elevated serum androgen levels may represent a plausible explanation for the observed differences in HMW adiponectin between PCOS and normal women. However, given the cross-sectional design of our study, causality cannot be established. Additionally, in a forward stepwise, linear regression analysis performed to determine predictors of HMW adiponectin in PCOS and normal women, free testosterone was no longer an independent predictor of HMW adiponectin after controlling for PCOS status, BMI, and age.

Furthermore, our results suggested that HMW adiponectin may be more important than total adiponectin in predicting insulin resistance in PCOS women and control women. The observed association between HMW adiponectin and insulin resistance is consistent with other studies suggesting that the HMW form of adiponectin has the most potent insulin sensitizing properties [12–13]. At the onset of the study, we hypothesized that alterations in adiponectin multimers in women with PCOS may contribute to the insulin resistance intrinsic to the syndrome. Although we demonstrated that levels of HMW adiponectin were decreased in both lean and obese women with PCOS compared with BMI-matched controls and that HMW adiponectin was a stronger predictor of insulin resistance than total adiponectin considering the entire study cohort, differences in insulin resistance as estimated by HOMA-IR were not observed between women with PCOS and control women in the unadjusted analysis. In light of the well-described differences in insulin resistance previously observed between PCOS and normal women after controlling for BMI [2,5], we suggest that using HOMA-IR as an estimate for insulin resistance, as opposed to other more robust methods, may have been unable to detect subtle differences in insulin resistance between groups and represents a potential limitation of the study.

Even though we did not observe significant differences in HOMA-IR between PCOS and normal women, SHBG levels were significantly lower in women with PCOS compared with normal women in this study. Although not a direct measure of insulin resistance, SHBG levels have been previously shown to correlate with insulin sensitivity measured by euglycemic clamp [29]. Additionally, in this study, strong positive linear correlations were observed between both total and HMW adiponectin and SHBG levels. Alternatively, the observation that total and HMW adiponectin were lower in women with PCOS compared with BMI- matched normal women despite similar estimates of insulin resistance may suggest that alterations in adiponectin multimer distribution in women with PCOS contribute to the phenotypic presentation of the syndrome via additional mechanisms independent of influences on insulin sensitivity.

While there is ample literature regarding total adiponectin concentrations in patients with PCOS, data concerning the relative distribution of the different multimeric complexes of adiponectin in women with PCOS are limited [30]. Furthermore, the studies reporting the relative distribution of adiponectin isoforms in patients with PCOS [31–33] have reported conflicting results. Aroda's [31] group previously reported that women with PCOS had lower total adiponectin levels compared with control women and that the high molecular weight species accounted for a smaller proportion of circulating adiponectin in women with PCOS. However, they did not examine the relationship between adiponectin multimer status and insulin resistance. Although Glintborg's group [32] also demonstrated lower total adiponectin levels in obese women with PCOS compared with age and BMI-matched control women, they failed to show significant differences in either HMW adiponectin levels or the ratio of HMW adiponectin to total adiponectin between the two groups of women. However, Glintborg and colleagues did report a significant negative correlations between the HMW form of adiponectin and both WHR and insulin sensitivity. They also described changes in total and HMW adiponectin among PCOS patient randomized to receive pioglitazone or placebo for 14 weeks [32]. Both total and HMW-adiponectin levels increased with pioglitazone treatment and the magnitude of change correlated with changes of insulin sensitivity. Barber et al [33] compared adiponectin multimers between PCOS women and female controls, including a subset of 22 BMI- and fat mass-matched pairs, and discovered that both total and HMW adiponectin levels are lower in women diagnosed with PCOS. However, the differences in total and HMW adiponectin were no longer significant after adjusting for age and fat mass. As a result, they concluded that the differences in adiponectin mutlimers between the groups of women were explained by differences in fat

mass and not by PCOS status. Consequently, they suggest that adiponectin does not play a central role in the pathophysiology of PCOS independent of adiposity.

The findings of the present study contrast with the results of two of these reports [32–33]. Several explanations for the alleged disparity in these findings can be hypothesized, including differences in study subject demographics (specifically ethnicity, different PCOS criteria) and clinical characteristics (i.e. higher prevalence of obesity in the present study). In our study, we used the 1990 NIH PCOS criteria, but the studies by Glintborg and Barber based the diagnosis of PCOS on the Rotterdam diagnostic criteria and thus represent a more heterogenous group of women. Furthermore, inclusion criteria for the control group and the methodology used to quantify adiponectin multimers and estimate insulin resistance also differed among the studies.

Several limitations of our study should be considered. The hyperinsulinemic-euglycemic clamp technique is the most scientifically sound technique for measuring insulin sensitivity [34] and is the "gold standard" by which other assessments are compared. Although HOMA-IR has been demonstrated to correlate well with clamp techniques and been frequently used in clinical applications [35], this index, calculated using fasting glucose and insulin levels, may be less discriminating than other measures of insulin sensitivity. Additionally, we quantified the multimeric forms of adiponectin via an ELISA that required pretreatment with one of several proteases in order to selectively digest specific forms of adiponectin. The results obtained by this method for measuring adiponectin species have previously been shown to correlate with results obtained by quantitative Western blot analysis [21]. Consequently, this commercially available ELISA has also been used by other groups as well [36–37]. Although additional ELISA assays have been developed that measure HMW-adiponectin directly [38–39]; results obtained from the various the ELISA systems have not been directly compared.

In conclusion, the present study demonstrates that levels of both total and HMW adiponectin are decreased in Chinese women with PCOS compared with normal control women, and the differences in HMW adiponectin persist after controlling for BMI. Furthermore, HMW adiponectin is a stronger predictor of insulin resistance as estimated by HOMA-IR in both women with PCOS and normal women than total adiponectin as indicated by forward, stepwise, linear regression analysis. Although differences in insulin resistance estimated by HOMA-IR were not readily apparent between PCOS and normal groups of women in this study, levels of SHBG, a potential alternative index of insulin resistance, were significantly decreased in women with PCOS.

Furthermore, total and HMW adiponectin levels demonstrated strong inverse correlates with SHBG levels. Additionally, it is possible that alterations in adiponectin multimers may contribute to the phenotypic presentation of PCOS via metabolic pathways independent of insulin resistance. Further investigation is warranted to determine both the mechanisms by which HMW adiponectin is altered in PCOS and the potential contributions of alterations in adiponectin multimers on ovulatory dysfunction in PCOS women.

## Acknowledgments

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**A**



**B**
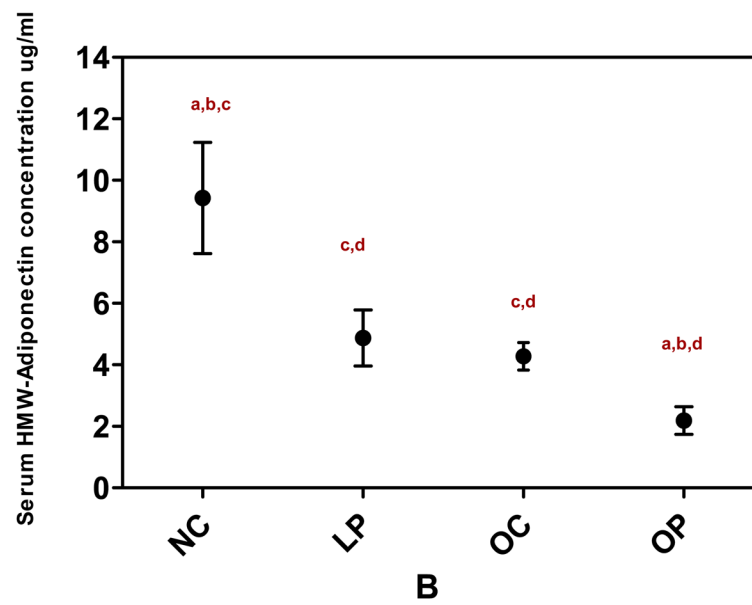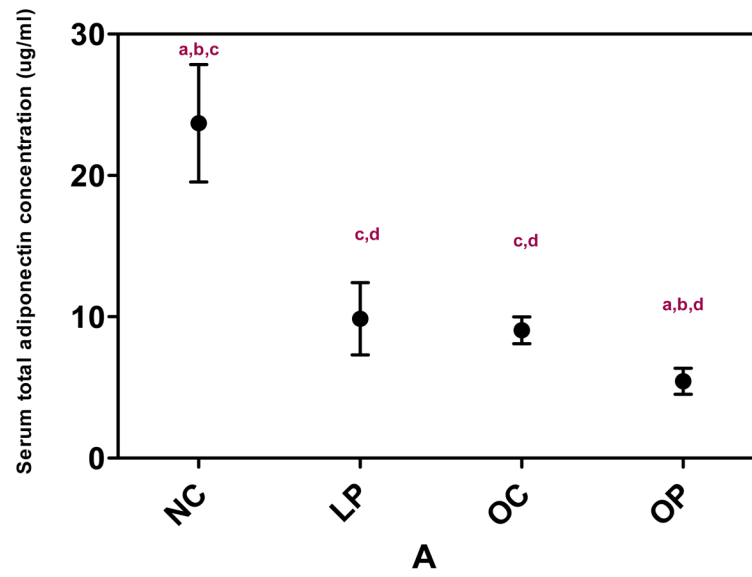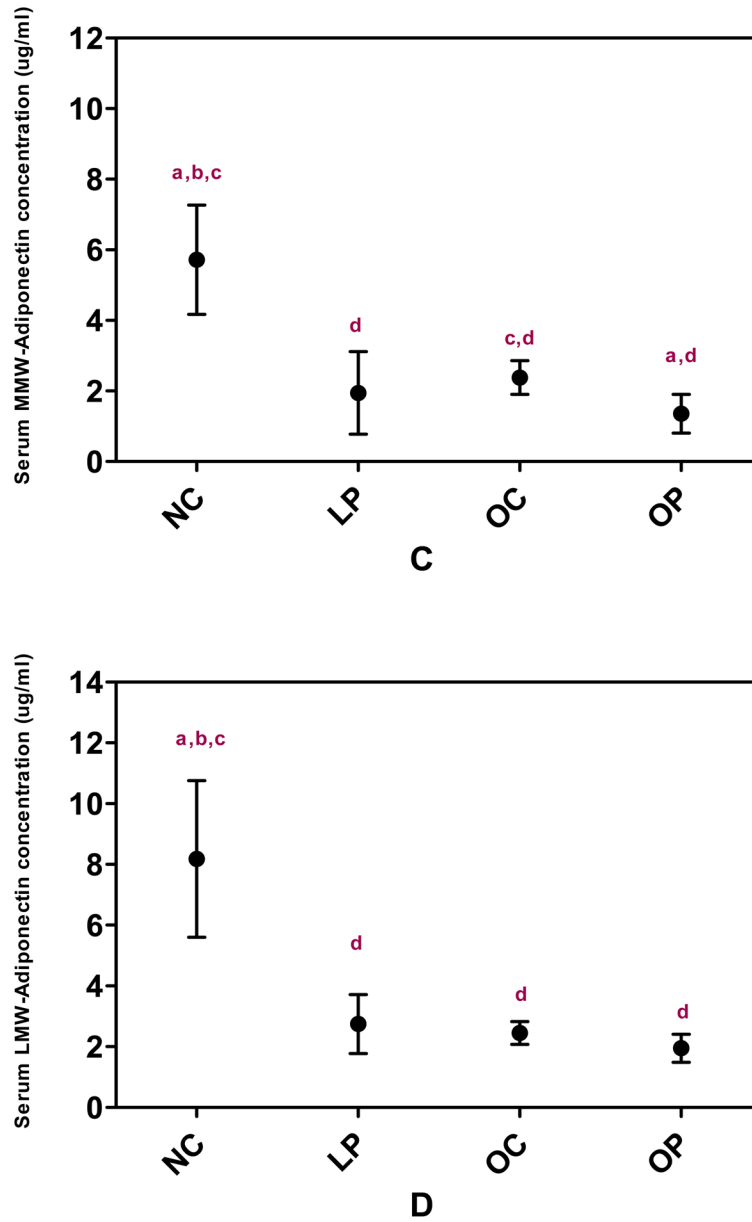
**Figure 1.**

Serum total adiponectin (A), HMW adiponectin (B), MMW adiponectin (C) and LMW adiponectin (D) concentrations in women with and without PCOS according to BMI category. Data are geometric means (95% CI). LC – lean control women (n=21); LP – lean PCOS women (n=21), OC – obese control women (n=38); OP – obese PCOS women (n=43). HMW – high molecular weight; LMW – low molecular weight; MMW – middle molecular weight.

P-value for ANOVA for difference between groups <0.0001 for total, HMW, MMW, and LMW adiponectin.

a- <0.05 compared to obese control (OC), b- <0.05 compared to lean PCOS (LP)

c- <0.05 compared to obese PCOS (OP), d-<0.05 compared to lean control (LC)

**Table 1**

Clinical and biochemical variables in women with PCOS and normal, control women according to BMI staus.

| Variable | Lean control | Obese control | Lean PCOS | Obese PCOS | P-value |
|---|---|---|---|---|---|
| n | 21 | 38 | 21 | 43 | N/A |
| Age (yrs) | 21.9 (18.7–25.0) | 26.0 (23.7–28.3) | 22.9 (19.7–26.0) | 25.9 (23.7–28.0) | 0.08 |
| BMI (kg/m$^2$) | 20.5 (18.8–22.2) a,c | 30.9 (29.6–32.2) b,d | 20.0 (18.3–21.7) a,c | 29.8 (28.6–31.0) b,d | <0.0001 |
| Percent body fat (%) | 23.1 (20.4–25.9) a,c | 42.7 (40.7–44.8) b,d | 24.8 (22.0–27.5) a,c | 43.3 (41.4–45.2) b,d | <0.0001 |
| Waist circumference (cm) | 66.1 (61.8–70.3) a,c | 92.4 (89.2–95.6) b,d | 67.8 (63.6–72.1) a,c | 90.3 (87.4–93.3) b,d | <0.0001 |
| Hip circumference (cm) | 86.7 (83.3–90.1) a,c | 105.8 (103.3) b,d | 86.9 (83.5–90.3) a,c | 102.3 (99.9–104.7) b,d | <0.0001 |
| WHR | 0.76 (0.74–0.79) a,c | 0.87 (0.85–0.89) b,d | 0.78 (0.76–0.80) a,c | 0.88 (0.87–0.90) b,d | <0.0001 |
| Total cholesterol (mmol/L) | 3.56 (3.27–3.85) a,b,c | 4.16 (3.95–4.38) c,d | 4.44 (4.15–4.74) d | 4.69 (4.49–4.89) a,d | <0.0001 |
| HDL cholesterol (mmol/L) | 1.24 (1.10–1.38) | 1.21 (1.10–1.31) | 1.43 (1.29–1.57) | 1.31 (1.21–1.40) | 0.08 |
| LDL cholesterol (mmol/L) | 2.29 (2.21–2.38) a,c | 2.84 (2.63–3.04) d | 2.68 (2.38–2.97) | 2.97 (2.70–3.24) d | 0.003 |
| Triglycerides (mmol/L) | 0.96 (0.68–1.23) c | 1.32 (1.13–1.52) | 1.09 (0.82–1.35) c | 1.65 (1.47–1.84) b,d | 0.0001 |
| Fasting insulin * (mIU/L) | 9.98 (8.37–11.90) a,c | 20.14 (17.67–22.95) b,d | 10.01 (8.39–11.93) a,c | 18.84 (16.67–21.31) b,d | <0.0001 |
| Fasting glucose (mmol/L) | 4.33 (4.20–4.47) a,c | 4.77 (4.61–4.94) c,d | 4.69 (4.53–4.85) c | 5.09 (4.87–5.31) a,b,d | <0.0001 |
| HOMA-IR * | 1.92 (1.59–2.32) a,c | 4.25 (3.69–4.89) b,d | 2.03 (1.38–2.45) a,c | 4.22 (3.70–4.83) b,d | <0.0001 |
| Total testosterone (nmol/L) | 1.97 (1.49–2.46) b | 2.30 (1.94–2.66) b | 3.21 (2.73–3.70) a,c,d | 2.38 (2.04–2.72) b | 0.003 |
| SHBG (nmol/L) | 63.34 (59.06–67.62) a,b,c | 41.92 (38.02–45.81) c,d | 46.92 (42.04–51.80) c,d | 31.55 (28.84–34.18) a,b,d | <0.0001 |
| Free testosterone * (pmol/L) | 22.82 (18.52–28.12) b,c | 31.99 (27.39–37.36) b | 47.48 (38.54–58.50) a,d | 41.42 (35.80–47.92) d | <0.0001 |

Data are means (95% CI) unless noted.

*
Geometric means.

BMI - body mass index, HDL - high density lipoprotein, HMW - high molecular weight, HOMA-IR - homeostasis model assessment - insulin resistance index, P-value for ANOVA for difference between groups.

[a]
<0.05 compared to obese control,

[b]
<0.05 compared with lean PCOS,

[c]
<0.05 compared with obese PCOS,

[d]
<0.05 compared to lean control

To convert cholesterol to mg/dL, divide by 0.02586; to convert triglycerides to mg/dL, divide by 0.01129; to covert glucose to mg/dL.; divide by 0.05551; to convert total testosterone to ng/ml, divide by 3.467; to convert free testosterone to pg/ml, divide by 3.467.

**Table 2**

Correlation between metabolic parameters and total and HMW adiponectin in normal control and PCOS women

| Variable | Total adiponectin* | | HMW adiponectin* | |
| --- | --- | --- | --- | --- |
| | Correlation | P-value | Correlation | P-value |
| BMI | −0.39 | <0.0001 | −0.35 | <0.0001 |
| Percent body fat | −0.46 | <0.0001 | −0.40 | <0.0001 |
| WHR | −0.44 | <0.0001 | −0.37 | <0.0001 |
| Total testosterone | −0.09 | 0.35 | −0.06 | 0.51 |
| SHBG | 0.60 | <0.0001 | 0.59 | <0.0001 |
| Free testosterone * | −0.30 | <0.001 | −0.28 | 0.002 |
| Total cholesterol | −0.48 | <0.0001 | −0.45 | <0.0001 |
| HDL cholesterol | −0.07 | 0.44 | −0.09 | 0.31 |
| LDL cholesterol | −0.34 | 0.0001 | −0.31 | <0.001 |
| Triglycerides | −0.26 | 0.003 | −0.22 | 0.02 |
| Fasting glucose | −0.42 | <0.0001 | −0.39 | <0.0001 |
| Fasting insulin * | −0.38 | <0.0001 | −0.37 | <0.0001 |
| HOMA-IR * | −0.42 | <0.0001 | −0.40 | <0.0001 |

*
Log transformed prior to analysis

BMI - body mass index, HDL - high density lipoprotein, HMW - high molecular weight,

HOMA-IR - homeostasis model assessment - insulin resistance index, LDL - low density lipoprotein,

SHBG - sex hormone binding globulin, WHR - waist-to-hip ratio,