# The role of serum adiponectin levels in women with polycystic ovarian syndrome

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**Summary**

*Purpose of investigation:* The aim of this study was to measure serum adiponectin concentrations in women with polycystic ovarian syndrome (PCOS) and to assess possible correlations between adiponectin and the hormonal or metabolic parameters of this syndrome. *Materials and Methods:* Serum adiponectin levels were evaluated in 20 women with PCOS and 22 women without PCOS whose age and body mass index (BMI) matched the patients. The levels of fasting blood glucose, fasting insulin, gonadotropin, and sex steroid hormones were evaluated in both groups. The homeostasis model assessment (HOMA) score was also calculated. The serum adiponectin levels were assayed by enzyme-linked immunoabsorbent assay (ELISA). *Results:* Serum adiponectin levels were significantly lower in obese women than in normal-weight women, and they were also significantly lower in PCOS patients with HOMA scores greater than 1.7 compared with those with HOMA scores lower than 1.7. When the subjects were divided in two groups based on serum adiponectin levels (> 40 μg/ml, < 40 μg/ml), 65% of patients with PCOS were included in the lower adiponectin group ($p < 0.05$). In addition, gonadotropin levels were increased, dependent on the adiponectin levels in women with PCOS. *Conclusion:* Adiponectin is regarded as a possible link between adiposity and insulin resistance (IR). From this data, the secretions of gonadotropin are implicated in the levels of adiponectin in women with PCOS. It is suggested that adiponectin may play an important role in the pathogenesis of PCOS.

*Key words:* PCOS; Adiponectin; Insulin resistance; Obesity.

## Introduction

Polycystic ovarian syndrome (PCOS) has been shown to be identified by oligomenorrhea or amenorrhea as menstruation disorders, hyperandrogenism, and small multiple cystic follicles in the ovary on ultrasonography, and is usually found as a complex and heterogenous endocrine disorder [1]. It occurs in about ten percent of women around reproductive age. In addition, it is associated with obesity in approximately 16% to 80% with PCOS. Recent work has identified that PCOS is often complicated with insulin resistance (IR) accompanied by compensatory hyperinsulinemia [2]. IR is suggested to be enhanced by the interaction between obesity and this syndrome [3].

These facts that both lean and obese PCOS patients show reduced insulin sensitivity and resultant hyperinsulinemia to some degree [4], suggest that hyperinsulinemia caused an increase in androgen biosynthesis [5] and a decrease in the levels of sex hormone-binding globulin (SHBG) [6]. These findings could possibly indicate the pathogenesis of hyperandrogenism. In addition to reproductive disorder, IR and hyperinsulinemia are recognized to increase the risk of long-term metabolic diseases, not only impaired glucose tolerance and type 2 diabetes [7], but also as cardiovascular disease [8].

Several studies have been reported to measure the circulating levels of adiponectin because of the importance of IR and obesity in PCOS [9, 10]. In recent years, it has been shown that adipocytes are secretory cells which produce various proteins with hormonal-type functions called adipocytokines. It is demonstrated that adiponectin is a 244-amino-acid protein, which is produced exclusively by adipose cells, and may have a role in preventing or counteracting the development of insulin resistance [11, 12]. In contrast to other adipocytokines, such as leptin, the production of adiponectin is decreased in obese subjects [12, 13].

The aim of this study was to clarify the determinants of adiponectin levels and to investigate the potential role of adiponectin in IR in women with PCOS. Furthermore, another objective of this study was also to clarify whether adiponectin is a marker of some degree in PCOS patients.

## Materials and Methods

Twenty-seven consecutive reproductive-aged, amenorrheic women with PCOS were recruited at the Infertility and Endocrinology Clinic, Oita University Hospital, between January 2002 and December 2004. Exclusion criteria were excess alcohol consumption (n = 1), cigarette smoking (n = 2), previous or current oral contraceptive use (n = 3), and endurance physical training (n = 1).

Criteria for PCOS were chronic anovulation (fewer than six cycles in 12 months) or amenorrhea, elevated serum levels of luteinizing hormone (LH), with normal follicle-stimulating hormone (FSH), and LH/FSH of at least 1.5, and polycystic appearance of the ovaries on ultrasound, defined by ten or more follicles two to eight mm in diameter, with a tendency toward peripheral distribution and bright echodense stroma. Baseline characteristics included age, height, weight, body mass index (BMI), and hirsutism status. BMI was calculated as weight (kg) divided by height squared ($m^2$). Subjects with Ferriman-Gallwey scores exceeding ten were defined as hirsute [14]. None of the PCOS patients had evidence of an androgen-secreting neoplasm, pituitary adenoma, homozygous adrenal hyperplasia,

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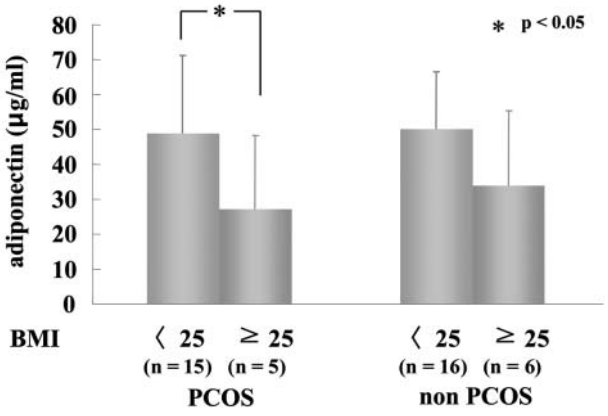


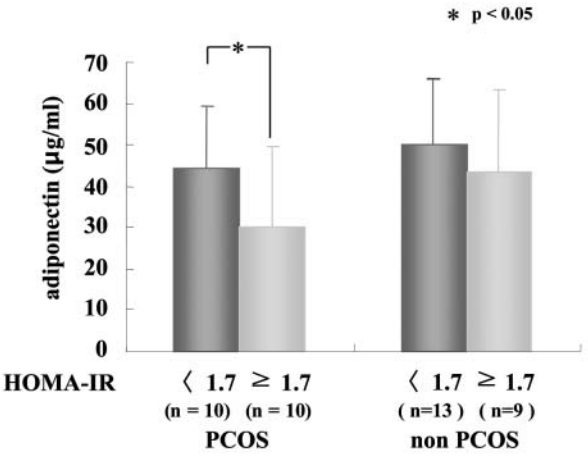


Figure 1. — The concentrations of serum adiponectin levels in PCOS patients and controls, with classification based on body weight. The figures below the X-axis indicate the number of subjects, in each subgroup. A statistically significant interaction between PCOS and body weight was observed.
* $p < 0.05$ vs controls. The data are expressed as means ± SD.

Figure 2. — The concentrations of serum adiponectin levels in PCOS patients and controls, with classification based on the degree of insulin resistance (HOMA-IR). The figures below the X-axis indicate the number of subjects, in each subgroup. A statistically significant interaction between PCOS and the degree of HOMA-IR was observed.
* $p < 0.05$ vs controls. The data are expressed as means ± SD.

acromegaly, or Cushing syndrome in accordance with National Institutes of Health criteria. None of the subjects were taking any medication likely to affect muscle size, muscle strength, or body fat distribution. All women in the control group had normal ovulating cycles and no signs of hyperandrogenism.

In all women, the basal serum levels of serum gonadotropin (FSH, LH), estradiol 17β, testosterone, dehydroepiandrosterone sulfate (DHEAS), and androstenedione were measured using commercially available radioimmunoassays (RIAs). Serum levels of prolactin (PRL), glucose, and insulin were also measured.

Serum adiponectin was measured using a commercially available enzyme linked immunosorbent assay (ELISA). The intra-assay and inter-assay coefficients of variation for these RIAs and ELISA were 3%-5% and 8% to l0%, respectively.

IR in the fasting state was evaluated by using homeostasis model assessment (HOMA) and was calculated with the following formula: fasting plasma glucose (mg/dl)×fasting serum insulin (μU/ml) divided by 405. High HOMA scores denote IR [15]. The subjects were allocated to four groups on the basis of the adiponectin value and a diagnosis of PCOS. Hence, group 1 (n = 35) women had PCOS + adiponectin < 40 μg/ml; group 2 (n = 35) had PCOS + adiponectin > 40 μg/ml; group 3 (controls; n = 15) were ovulating without PCOS + adiponectin < 40 μg/ml; and group 4 (controls; n = 15) were ovulating without PCOS + adiponectin > 40 μg/ml.

Informed consent was obtained from each subject, and the study was approved by the Institutional Review Board, and was conducted in accordance with institutional guidelines and the Declaration of Helsinki.

*Statistical analysis*

Data are presented as means ± SD, and were analyzed using the Mann-Whitney U test, chi-square ($\chi^2$) test, and Bonferroni/Dunn test for multiple comparisons. A $p$ value < 0.05 was considered to be statistically significant.

Table 1. — *Clinical and endocrine features of PCOS patients and controls.*

| | PCOS | Controls |
|---|---|---|
| No. of patients | 20 | 22 |
| Age | 31.3 ± 4.7 | 30.3 ± 4.8 |
| Height (cm) | 157.5 ± 5.5 | 157.6 ± 4.4 |
| Weight (kg) | 56.6 ± 11.0 | 55.8 ± 10.0 |
| BMI (kg/m$^2$) | 22.9 ± 4.8 | 22.5 ± 4.3 |
| LH (mIU/ml) | 9.3 ± 5.8* | 4.7 ± 1.6 |
| FSH (mIU/ml) | 5.7 ± 1.5** | 7.2 ± 1.5 |
| LH/FSH | 1.7 ± 0.8* | 0.7 ± 0.2 |
| $E_2$ (pg/ml) | 43.0 ± 24.0 | 36.6 ± 21.8 |
| PRL (ng/ml) | 11.0 ± 6.1 | 12.4 ± 9.2 |
| T (ng/ml) | 34.2 ± 21.3 | 32.2 ± 13.7 |
| FBS (mg/dl) | 93.0 ± 7.6 | 92.5 ± 7.2 |
| IRI (pmol/l) | 13.2 ± 12.1 | 7.9 ± 5.6 |
| HOMA-IR | 3.1 ± 3.0 | 1.9 ± 1.5 |

BMI = body mass index; LH = luteinizing hormone; FSH = follicle-stimulating hormone; $E_2$ = estradiol; PRL = prolactin; T = testosterone; FBS = fasting blood glucose; IRI = insulin resistance index; HOMA-IR = homeostasis model assessment-insulin resistance; *$p < 0.01$, **$p < 0.05$ for differences between PCOS and controls by the Mann-Whitney U test. Data represent mean ± SD.

Table 2. — *The number of subjects on the basis of adiponectin levels in PCOS and controls.*

| | Adiponectin (μg/ml) | |
|---|---|---|
| | < 40 | ≥ 40 |
| PCOS (n = 20) | 13 (65%) | 7 (35%)* |
| Controls (n = 22) | 6 (27%) | 16 (73%)* |

*$p < 0.05$ for differences between PCOS with lower adiponectin levels and controls with higher adiponectin levels by the $\chi^2$-test.

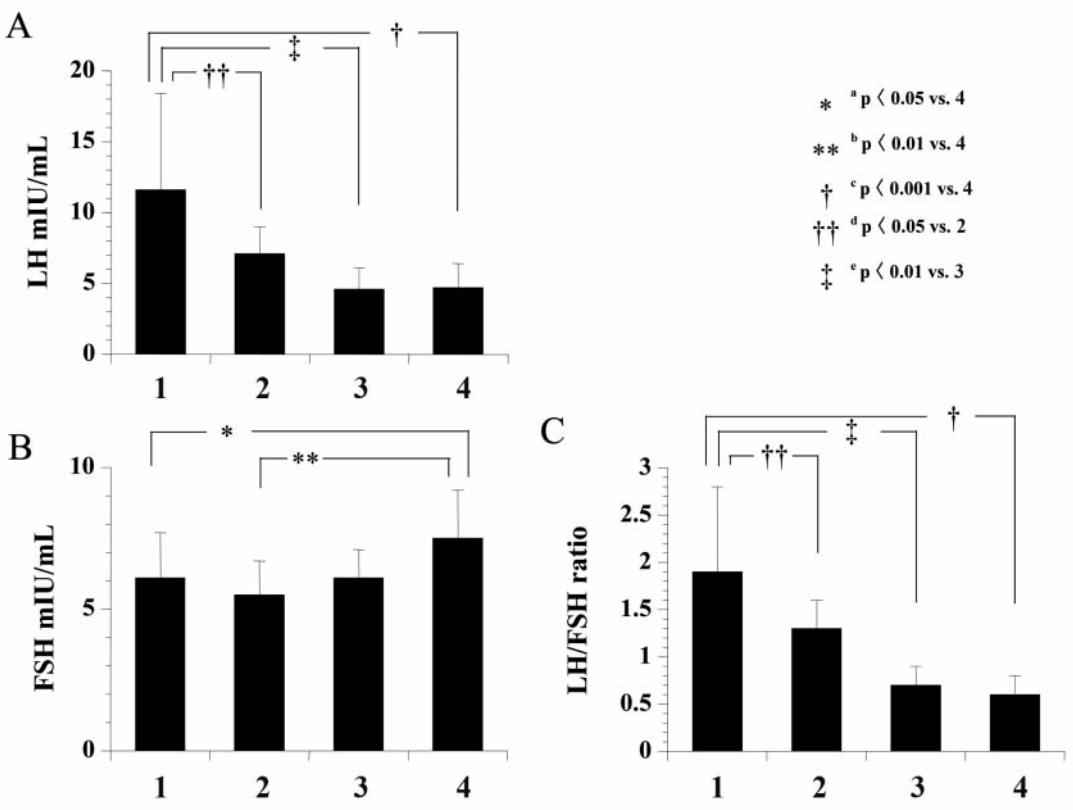


Figure 3. — The concentrations of serum LH and FSH levels in PCOS patients and controls, with classification based on serum adiponectin levels. The figures below the X-axis indicate the number of each subgroup. * [a] $p < 0.05$ between groups 1 and 4 in FSH levels, ** [b] $p < 0.01$ between groups 2 and 4 in FSH levels, † [c] $p < 0.001$ between groups 1 and 4 in LH levels and LH/FSH ratio, †† [d] $p < 0.05$ between groups 1 and 2 in LH levels and LH/FSH ratio, ‡ [e] $p < 0.01$ between groups 1 and 3 in LH levels, and LH/FSH ratio. The data are expressed as means ± SD.

Table 3. — *Baseline characteristcs and hormonal features in PCOS and controls.*

| Adiponectin | 1 PCOS < 40 µg/ml | 2 PCOS ≥ 40 µg/ml | 3 Controls < 40 µg/ml | 4 Controls ≥ 40 µg/ml |
|---|---|---|---|---|
| No. of patients | 13 | 7 | 6 | 16 |
| Age | 29.4 ± 4.8[d] | 33.9 ± 3.3 | 30.0 ± 4.6 | 30.5 ± 5.1 |
| BMI (kg/m$^2$) | 24.1 ± 5.5[a,d] | 20.3 ± 1.7 | 26.7 ± 3.5[b,d] | 21.0 ± 3.3 |
| HOMA-IR | 3.0 ± 2.3[a] | 3.0 ± 4.2 | 3.4 ± 2.1 | 1.3 ± 0.6 |
| PRL (ng/ml) | 9.5 ± 6.5 | 12.3 ± 5.4 | 11.6 ± 8.0 | 12.7 ± 9.9 |
| E2 (pg/ml) | 43.5 ± 20.9 | 47.0 ± 31.3 | 23.8 ± 5.8 | 41.4 ± 23.7 |
| T (pg/ml) | 40.4 ± 49.5 | 22.7 ± 19.2 | 36.2 ± 8.2 | 30.7 ± 15.3 |

BMI = body mass index; HOMA-IR = homeostasis model assessment; PRL = prolactin; $E_2$ = estradiol; T = testosterone.
PCOS patients and controls were classified according to serum diponectin levels as described in Methods. [a]$p < 0.05$ vs group 4 [b]$p < 0.01$ vs group 4 [c]$p < 0.001$ vs group 4 [d]$p < 0.05$ vs group 2 [e]$p < 0.01$ vs group 3 for differencies between four groups by Bonferroni-Dunn test. Data represent mean ± SD.

## Results

Patients and controls were equally distributed according to age, BMI, and degree of obesity (Table 1). LH and LH/FSH ratio were significantly higher in patients with PCOS compared with controls. However, no significant differences were observed between the BMI-matched groups.

The results of the univariate analysis of the effects of PCOS or of control status and of the degree of obesity are shown in Figure 1. Serum adiponectin levels were significantly lower in the ≥ 25 kg/m$^2$ BMI group than among normal-weight (BMI < 25 kg/m$^2$) women among PCOS patients; however, these levels were not affected by obesity in controls.

These levels were also significantly lower in women with a HOMA score greater than 1.7, compared with those with an HOMA score less than 1.7 among PCOS patients. No difference was found in adiponectin levels among controls as shown in Figure 2.

Women with PCOS (subjects) were classified according to serum adiponectin levels as described in Materials and Methods. When PCOS patients and controls were divided into two groups by serum adiponectin level (< 40 µg/ml, > 40 µg/ml), 65% of patients with PCOS were included in the lower adiponectin group (Table 2). LH and LH/FSH ratio were significantly increased in lower adiponectin group (group 1) compared with higher adiponectin group (group 2) among PCOS patients shown in Figure 3. By contrast, there were no significant differences between two groups in other hormone levels (Table 3).

## Discussion

In the present study, the authors investigated the relationship between endocrine parameters and adiponectin levels in PCOS patients. Adiponectin is thought to be almost exclusively produced in adipose tissue. It was demonstrated that obesity, IR, and type 2 diabetes were associated with low plasma adiponectin levels in previous study [13]. In this data, obese women (BMI $\geq$ 25 kg/m$^2$) showed significantly decreased fasting serum concentrations of adiponectin as compared with those of matched lean women (BMI < 25 kg/m$^2$) with PCOS.

It has been reported that serum adiponectin levels are decreased in PCOS patients [10, 16, 17]. Thus, this result may be particularly important in the context of the concurrence of obesity (9), IR [18, 19] and/or impaired glucose tolerance [20] in these women. It is well-recognized that IR is frequently observed and has been linked to the clinical and endocrine alterations, such as hyperandrogenism and reproductive disorders in PCOS patients [21, 22]. Likewise, hyperinsulinaemia associated with IR might be physiological roles of not only impaired glucose tolerance and type 2 diabetes mellitus, but also atherosclerosis and cardiovascular disease observed in women with PCOS [7, 23].

Overall, these findings are based on the previous studies, in which significant lower adiponectin levels were evident, in obese women with PCOS [15]. On the other hand, lean women with PCOS did not show significant decreases in adiponectin levels as compared with the corresponding lean women in control group.

It is demonstrated that adiponectin is highly-expressed in white adipose tissue, and is by far the most abundant circulating specific protein derived from adipose tissues in humans [13]. The evidence that adiponectin has the potential to enhance insulin sensitivity and to improve glucose metabolism [11, 12, 24, 25] has been demonstrated *in vitro* and *in vivo* studies using rodents as a model. The mechanisms of improvement of IR and glucose metabolism by adiponectin are currently under investigation, although it is well-recognized that the effects of insulin-sensitizing agents have been implicated both in the liver and muscle [24].

Consistent with findings in a rodents' model, the adiponectin levels were involved in obesity, type 2 diabetes mellitus, and cardiovascular disease [12, 26]. In this way, circulating low adiponectin levels in PCOS may not only determine the degree of IR, but could also provide a link to a higher risk of type 2 diabetes mellitus and cardiovascular disease [21].

The decreasing of adiponectin levels may contribute to IR in women with PCOS, because adiponectin is considered to reduce the triglyceride content of muscle, enhancing insulin signaling, and activates peroxisomal proliferator-activated receptor alpha (PPAR$\alpha$), resulting to increase energy combustion. Adiponectin also up-regulates fat oxidation and transport of muscle and inhibits the expression of enzymes with gluconeogenesis, reducing hepatic glucose production by phosphorylation of AMP-activated protein kinase [27].

Overall, one interesting point that arises from these results is that serum adiponectin levels are observed in hormonal differences (elevated LH and LH/FSH ratio) in PCOS, but not observed in controls. The fact that gonadotropin secretion is associated with adiponectin concentrations suggests that it may represent the role of adiponectin on the endocrine condition directly or indirectly in women with PCOS.

In conclusion, these data have shown that compared with controls of similar body weight, PCOS patients have altered adiponectin secretion. These differences may be caused by the result of altered adipose tissue function. Likewise, altered adiponectin secretion may still be involved in the characteristic IR of PCOS. Further studies will be needed to elucidate this issue.

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## References

[1] Frank S.: "Polycystic ovary syndrome". *N. Engl. J. Med.* 1995, *333*, 853.
[2] Burghen G.A., Givens J.R., Kitabchi A.E.: "Correlation of hyperandrogenism with hyperinsulinism in polycystic ovarian disease". *J. Clin. Endocrinol. Metab.* 1980, *50*, 113.
[3] Dunaif A.: "Molecular mechanisms of insulin resistance in polycystic ovary syndrome". *Semin. Reprod. Endocrinol.* 1994, *12*, 15.
[4] Dunaif A.: "Insulin resistance and the polycystic ovary syndrome: mechanism and implications for pathogenesis". *Endocr. Rev.* 1997, *18*, 774.
[5] Adashi E.Y., Resnick C.E., D'Ercole A.J., Svoboda M.E., Van Wyk J.J.: "Insulin-like growth factors as intraovarian regulators of granulosa cell growth and function". *Endocr. Rev.* 1985, *6*, 400.
[6] Nestler J.E., Powers L.P., Matt D.W., Steingold K.A., Plymate S.R., Rittmaster R.S., Clore J.N., Blackard W.G.: "A direct effect of hyperinsulinemia on serum sex hormone- binding globulin levels in obese women with the polycystic ovary syndrome". *J. Clin. Endocrinol. Metab.* 1991, *72*, 83.
[7] Ehrmann D.A., Barnes R.B., Rosenfield R.L., Cavaghan M.K., Imperial J.: "Prevalence of impaired glucose tolerance and diabetes in women with polycystic ovary syndrome". *Diabetes Care.* 1999, *22*, 141.
[8] Futterweit W.: "Polycystic ovary syndrome: clinical perspectives and management". *Obstet. Gynecol. Surv.* 1999, *54*, 403.
[9] Orio F., Palomba S., Cascella T., Milan G., Mioni R., Pagano C. *et al.*: "Adiponectin levels in women with polycystic ovary syndrome". *J. Clin. Endocrinol. Metab.* 2003, *88*, 2619.
[10] Panidis D., Kourtis A., Farmakiotis D., Mouslech T., Rousso D., Koliakos G.: "Serum adiponectin levels in women with polycystic ovary syndrome". *Hum. Reprod.* 2003, *18*, 1790.
[11] Kubota N., Terauchi Y., Yamauchi T., Kubota T., Moroi M., Matsui J. *et al.*: "Disruption of adiponectin causes insulin resistance and neointimal formation". *J. Biol. Chem.* 2002, *277*, 25863.
[12] Weyer C., Funahashi T., Tanaka S., Hotta K., Matsuzawa Y., Pratley R.E., *et al.*: "Hypoadiponectinemia in obesity and type 2 diabetes: close association with insulin resistance and hyperinsulinemia". *J. Clin. Endocrinol. Metab.* 2001, *86*, 1930.
[13] Arita Y., Kihara S., Ouchi N., Takahashi M., Maeda K., Miyagawa J., *et al.*: "Paradoxical decrease of an adipose-specific protein, adiponectin, in obesity". *Biochem. Biophys. Res. Commun.* 1999, *257*, 79.
[14] Ferriman D., Gallwey J.D.: "Clinical assessment of body hair growth in women". *J. Clin. Endocrinol. Metab.,* 1961, *21*, 1440.

[15] Matthews D.R., Hosker J.P., Rudenski A.S., Naylor B.A., Treacher D.F., Turner R.C.: "Homeostasis model assessment: insulin resistance and β-cell function from fasting plasma glucose and insulin concentration in man". *Diabetologia.* 1985, *28*, 412.
[16] Ardawi M.S., Rouzi A.A.: "Plasma adiponectin and insulin resistance in women with polycystic ovary syndrome". *Fertil. Steril.* 2005, *83*, 1708.
[17] Carmina E., Orio F., Palomba S., Cascella T., Longo R.A., Colao A.M., *et al*.: "Evidence for altered adipocyte function in polycystic ovary syndrome". *Eur. J. Endocrinol.* 2005, *152*, 389.
[18] Spranger J., Möhlig M., Wegewitz U., Ristow M., Pfeiffer A.F., Schill T. *et al*.: "Adiponectin is independently associated with insulin sensitivity in women with polycystic ovary syndrome". *Clin. Endocrinol. (Oxf).* 2004, *61*, 738.
[19] Sepilian V., Nagamani M.: "Adiponectin levels in women with polycystic ovary syndrome and severe insulin resistance". *J. Soc. Gynecol. Investig.* 2005, *12*, 129.
[20] Sieminska L., Marek B., Kos-Kudla B., Niedziolka D., Kajdaniuk D., Nowak M, *et al*.: "Serum adiponectin in women with polycystic ovarian syndrome and its relation to clinical, metabolic and endocrine parameters". *J. Endocrinol. Invest*., 2004, *27*, 528.
[21] Robinson S., Kiddy D., Gelding S.V., Willis D., Niththyananthan R., Bush A., *et al*.: "The relationship of insulin insensitivity to menstrual pattern in women with hyperandrogenism and polycystic ovaries". *Clin. Endocrinol. (Oxf).* 1993, *39*, 351.
[22] De Leo V., La Marca A., Petraglia F.: "Insulin-lowering agents in the management of polycystic ovary syndrome". *Endocr. Rev.* 2003, *24*, 633.
[23] Lobo R.A., Carmina E.: "The importance of diagnosing the polycystic ovary syndrome". *Ann. Intern. Med.* 2000, *132*, 989.
[24] Yamauchi T., Kamon J., Waki H., Terauchi Y., Kubota N., Hara K., *et al*.: "The fat-derived hormone adiponectin reverses insulin resistance associated with both lipoatrophy and obesity". *Nat. Med.,* 2001, *7*, 941.
[25] Maeda N., Shimomura I., Kishida K., Nishizawa H., Matsuda M., Nagaretani H., *et al*.:" Diet-induced insulin resistance in mice lacking adiponectin /ACRP30". *Nat. Med.,* 2002, *8*, 731.
[26] Ouchi N., Kihara S., Arita Y., Maeda K., Kuriyama H., Okamoto Y., *et al*.: "Novel modulator for endothelial adhesion molecules: adipocyte-derived plasma protein adiponectin". *Circulation,* 1999, *100*, 2473.
[27] Kadowaki T., Yamauchi T.: "Adiponectin and adiponectin receptors". *Endocr. Rev.,* 2005, *26*, 439.

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