

# Use of Metformin for Ovulation Induction in Women Who Have Polycystic Ovary Syndrome With or Without Evidence of Insulin Resistance

Kimberly E. Liu, MD, Ivanna Viola Tataryn, MD, Margaret Sagle, MSc, MD

Department of Obstetrics and Gynaecology, University of Alberta, Edmonton AB

## Abstract

**Objective:** To determine whether women with polycystic ovary syndrome (PCOS) and abnormal insulin levels treated with metformin had different rates of ovulation and pregnancy from women with PCOS and normal insulin levels.

**Methods:** The outcomes of treatment with metformin in 146 infertile women with PCOS were analyzed using a retrospective cohort study design. Baseline characteristics and initial blood work results were recorded. The follow-up period was three months, and the primary outcome was ovulation.

**Results:** Of the 146 women with PCOS, one third had elevated fasting insulin levels. After treatment with metformin, cumulative rates of ovulation were similar in women with elevated fasting serum insulin levels (48.8%) and those with normal levels (44.7%). Rates of ovulation were also similar in women with normal and abnormal glucose to insulin ratios. There was no difference in cumulative pregnancy rates based on fasting insulin levels. A fasting insulin level above 20 mU/L correlated with an abnormal glucose to insulin ratio (98%).

**Conclusion:** In anovulatory women with PCOS, fasting insulin levels and glucose to insulin ratios do not predict the ovulatory response to metformin.

## Résumé

**Objectif :** Déterminer si les femmes présentant un syndrome des ovaires polykystiques (SOPK) et des taux d'insuline anormaux, et ayant été traitées à la metformine présentaient des taux différents d'ovulation et de grossesse, par comparaison avec les femmes présentant un SOPK et des taux d'insuline normaux.

**Méthodes :** Les issues de traitement à la metformine chez 146 femmes infertiles présentant un SOPK ont été analysées au moyen d'une étude de cohorte rétrospective. Les caractéristiques de départ et les résultats de l'hémogramme initial ont été consignés. La période de suivi a été de trois mois et le critère d'évaluation principal a été l'ovulation.

**Résultats :** On a constaté des taux élevés d'insuline à jeun chez le tiers des 146 femmes présentant un SOPK. À la suite du traitement à la metformine, les taux cumulatifs d'ovulation étaient

similaires chez les femmes qui présentaient des taux sériques élevés d'insuline à jeun (48,8 %) et chez celles qui en présentaient des taux normaux (44,7 %). Les taux d'ovulation étaient également semblables tant chez les femmes qui présentaient un rapport glucose-insuline normal que chez celles qui présentaient un rapport glucose-insuline anormal. Aucune différence n'a été constatée en ce qui concerne les taux cumulatifs de grossesse en fonction des taux d'insuline à jeun. Un taux d'insuline à jeun supérieur à 20 mU/l était en corrélation avec un rapport glucose-insuline anormal (98 %).

**Conclusion :** Chez les femmes anovulatoires présentant un SOPK, les taux d'insuline à jeun et les rapports glucose-insuline ne permettent pas de prédire la réaction ovulatoire à la metformine.

J Obstet Gynaecol Can 2006;28(7):595-599

## INTRODUCTION

Polycystic ovary syndrome (PCOS), first described in 1935 by Stein and Leventhal, has been associated with a variety of clinical and laboratory findings, including chronic anovulation or oligomenorrhea, infertility, hyperandrogenism, and insulin resistance. The 2003 Rotterdam criteria required two of the following characteristics for the diagnosis of PCOS: oligo-ovulation or anovulation, clinical and/or biochemical signs of hyperandrogenism, and polycystic ovaries with the exclusion of other etiologies (congenital adrenal hyperplasia, androgen-secreting tumours, Cushing's syndrome).<sup>1</sup>

Many women with PCOS have been treated with clomiphene for infertility, resulting in rates of ovulation of 70% to 85%, and pregnancy rates of 33% to 45%.<sup>2-4</sup> More recently, the recognition of an association between insulin resistance and PCOS has led to the use of metformin, a biguanide insulin-sensitizing agent, for ovulation induction. Several large studies and a meta-analysis have shown that metformin is effective for ovulation induction in women with PCOS.<sup>5-7</sup> Metformin is effective both in thin women with PCOS<sup>8,9</sup> and in women with PCOS who are resistant to clomiphene,<sup>10-14</sup> and treatment with metformin and clomiphene has been shown to be more effective than use

**Key Words:** Metformin, polycystic ovary syndrome, ovulation induction

Competing Interests: None declared.

Received on February 2, 2006

Accepted on March 14, 2006

**Table 1. Mean variables of patients with normal and abnormal fasting insulin levels**

Characteristic	Fasting insulin < 20 mU/L (86)	Fasting insulin ≥ 20 mU/L (38)	<i>P</i> *
Age	30.5	30.4	NS
Gravidity	0.6	0.2	NS
Parity	0.2	0.2	NS
BMI (kg/m <sup>2</sup> )	33.6	38.3	< 0.01
Previous clomiphene use	69.8%	67.8%	NS**
Fasting plasma glucose (mmol/L)	5.0	5.4	0.02
Serum testosterone (nmol/L)	2.0	2.7	< 0.01
Serum DHEA-S (μmol/L)	5.4	4.3	NS
Serum insulin (mU/L)	12.5	32.7	< 0.01
Metformin dose (mg)	1078	1232	0.01
Concurrent clomiphene use	39.5%	42.1%	NS**
Clomiphene dose (mg)	104.5	112.5	NS

\*Student *t* test unless specified

\*\*Two-tailed Fisher exact test

NS: not significant; BMI: body mass index; DHEA-S: dehydroepiandrosterone sulphate.

of clomiphene alone for inducing ovulation and subsequent pregnancy.<sup>6</sup> Treatment with metformin is as effective as laparoscopic ovarian drilling in inducing ovulation in clomiphene-resistant women.<sup>13,14</sup> There is some evidence that use of metformin improves hyperandrogenism and insulin resistance.<sup>15–17</sup>

Insulin resistance is more common in both thin and obese women with PCOS than in women without PCOS.<sup>18</sup> There are currently no validated clinical tests for the diagnosis of insulin resistance. Invasive tests such as the euglycemic clamp, the gold standard in research settings, are not feasible for clinical use. Other methods described include the homeostasis model assessment of insulin resistance (HOMA-IR) and the glucose to insulin ratio or area under the curve (AUC) following a 2-hour oral glucose tolerance test.<sup>19–21</sup> One small study has shown serum insulin levels may be an independent predictor of treatment success with metformin.<sup>22</sup> Currently, screening for insulin resistance is not routinely recommended for women with PCOS or prior to treatment with insulin-sensitizing agents. However, screening for the metabolic syndrome and impaired glucose tolerance may help identify a high-risk population and allow a reduction in long-term health concerns.<sup>1</sup>

In most studies, an unselected population of women with PCOS has been treated effectively with metformin.<sup>23</sup> Metformin is generally effective in inducing ovulation in 50% of women with PCOS,<sup>6</sup> but there is little information about clinical or laboratory measurements that may help

predict success. We completed a retrospective study of women with PCOS who were treated with metformin for ovulation induction. The purpose of our study was to determine whether the rate of ovulation with metformin was similar in patients with normal and abnormal fasting indices of insulin resistance.

## MATERIALS AND METHODS

For this retrospective cohort study, we reviewed the medical records of all patients who began treatment with metformin for ovulation induction at the Regional Fertility and Women's Endocrine Clinic (RFWEC) at the Royal Alexandra Hospital between January 2000 and December 2004. Inclusion criteria for the study were (1) a history of infertility, (2) a diagnosis of PCOS according to the 2003 Rotterdam criteria, and (3) normal serum thyroid-stimulating hormone (TSH) and prolactin levels. Exclusion criteria were (1) documented ovulatory cycles prior to treatment, (2) concurrent treatment with gonadotropins, and (3) diabetes mellitus. Women who were undergoing concurrent treatment with clomiphene were included if they had failed to respond to clomiphene at the same dosage prior to metformin treatment.

During this period, patients were seen by one of four reproductive endocrinologists. Patients were managed according to the individual physician's practice with regard to metformin dosing, although most patients began treatment at a dose of 500 mg twice daily. Initial blood work, including fasting insulin and glucose levels and a hormone profile,

**Table 2. Occurrence of ovulation and pregnancy in women with normal and abnormal fasting insulin levels**

	Fasting insulin < 20 mU/L (n = 86)	Fasting insulin ≥ 20 mU/L (n = 38)	Normal fasting glucose/ insulin ratio (n = 74)	Abnormal fasting glucose/insulin ratio (n = 37)
Ovulation	42 (48.8%)	17 (44.7%)	38 (51.4%)	18 (48.6%)
	<i>P</i> = 0.7*		<i>P</i> = 0.84*	
Pregnancy	11 (12.8%)	5 (13.2%)	8 (10.8%)	5 (13.5%)
	<i>P</i> = 1.0*		<i>P</i> = 0.76*	

\*Two-tailed Fisher exact test.

was completed prior to initiation of treatment. Patients were reviewed three months after beginning metformin. Fasting insulin levels were considered elevated if equal to or greater than 20 mU/L. Fasting glucose/insulin ratios were defined as abnormal if less than 4.5 mg/10<sup>-4</sup> U.<sup>20</sup> All blood work was done at provincial laboratories through a centralized laboratory agency. Serum glucose levels were converted from mmol/L to mg/dL prior to calculating the fasting glucose/insulin ratio. Baseline characteristics of patients with normal and abnormal fasting insulin and glucose/insulin ratios were recorded.

The primary outcome of the study was the occurrence of ovulation within three months of beginning metformin therapy. Ovulation was confirmed by measuring a serum progesterone level of greater than 15 ng/mL. Progesterone levels were measured on cycle day 21, or seven days before the anticipated next menses, then every seven days until day 35 if not shown to be ovulatory. The ovulation rate was calculated by dividing the number of women who ovulated by the total number of women in that group. Assuming a rate of ovulation in women with elevated insulin levels of 50%, we calculated that 40 patients would be required in each group to detect a 35% difference between groups. All charts were reviewed for baseline characteristics, diagnosis of PCOS, and outcome by one investigator. Ethics approval was received through the Health Research Ethics Board at the University of Alberta.

Statistical analysis for ovulation and pregnancy outcomes was performed with two-tailed Fisher exact test and chi-square test. Patient characteristics were analyzed with the Student *t* test and Fisher exact test where appropriate.

## RESULTS

Between January 2000 and December 2004, 3732 women were seen at the RFWEC because of infertility. A total of 146 women with PCOS who were treated with metformin met the study criteria. The average age of these women was

30 years, the overall rate of ovulation was 48%, and the pregnancy rate was 12%. Of the 146 women, 124 had documented fasting insulin levels prior to metformin treatment, and of these 30.6% had a fasting insulin level of 20 mU/L or greater. The mean fasting insulin level was 18.7 mU/L. Women with elevated fasting insulin levels had a higher body mass index (BMI), higher fasting glucose levels, and higher serum testosterone levels than women with normal fasting insulin levels (Table 1). The rate of concurrent clomiphene usage and dosage was similar in each group. One-third (33.3%) of women had an abnormal fasting glucose/insulin ratio.

There was no difference in ovulation or pregnancy rates in women with normal or abnormal fasting insulin levels or glucose to insulin ratios (Table 2). In women without documented insulin levels prior to metformin treatment (22), 50% ovulated, and one woman became pregnant.

In women with normal fasting insulin levels (< 20 mU/L), 3.5% had an abnormal glucose/insulin ratio, but almost all women with fasting insulin levels of 20 mU/L or greater had an abnormal glucose/insulin ratio (98%).

The average BMI in our study population was 35 kg/m<sup>2</sup>; 62 women had a BMI of greater than 35 kg/m<sup>2</sup>. In these women, there was a trend towards a lower rate of ovulation (41.9% vs. 55.2%, *P* = 0.15) and a lower rate of pregnancy (4.8% vs. 17.9%, *P* = 0.03). Of the nine women with normal weight (BMI 18.5–25 kg/m<sup>2</sup>), one had an elevated fasting insulin level.

More than half (56%) of the women in the study had an elevated serum testosterone level. There was a trend towards a lower rate of ovulation in women with an elevated testosterone level that was not statistically significant (40% vs. 57.5%, *P* = 0.08), but there was no difference in pregnancy rates. More than one third of women with elevated testosterone levels had elevated fasting insulin levels (38.3% vs. 12.8%, *P* = 0.004).

## DISCUSSION

In this study, pre-treatment fasting insulin levels or fasting glucose/insulin ratios did not predict the outcome of treatment with metformin for ovulation induction in women with PCOS. Metformin was effective in women with both normal and abnormal fasting insulin levels; however, women with elevated fasting insulin levels were treated with higher doses of metformin than those with normal levels (1232 mg vs. 1078 mg). The clinical significance of this difference in dosage is unknown.

Our study population was an unselected population of women with PCOS, and most of them were overweight or obese. Current consensus guidelines state that insulin levels do not need to be measured as part of the diagnosis of PCOS, or prior to treatment with metformin for ovulation induction.<sup>1</sup> Our study confirms the lack of usefulness of fasting insulin levels in deciding whether to treat women with metformin.

Currently the standard for first-line therapy for inducing ovulation in women with PCOS is clomiphene citrate, but recent evidence suggests that metformin can also be effective as first-line therapy.<sup>6,24</sup> The suggestion to use metformin is based on theories that hyperandrogenism in women with PCOS is caused by hyperinsulinemia.<sup>25,26</sup> Metformin therapy reduces insulin levels and increases insulin-like growth factor binding protein (IGFBP)-1.<sup>16,17,27</sup> Because IGF-1 stimulates ovarian androgen production, metformin therapy may decrease intra-ovarian androgen levels through its effect of decreasing the IGF-1/IGFBP-1 ratio.<sup>27</sup> Hyperinsulinemia may also stimulate intra-ovarian androgen production through stimulation of cytochrome P450c17 $\alpha$ , an enzyme involved in androgen synthesis.<sup>28</sup> Metformin has been shown to decrease serum insulin levels, P450c17 $\alpha$  activity, and serum androgen levels in both obese and thin women with PCOS.<sup>28,29</sup>

Approximately 20% of women with PCOS are resistant to clomiphene, and hyperinsulinemia and obesity appear to correlate with clomiphene failure.<sup>30</sup> In our study population, there did not appear to be a correlation between a woman's BMI and her ovulatory response to treatment. Although both severe obesity and elevated serum testosterone levels predicted lower rates of ovulation, a type II error in this conclusion cannot be excluded. The average daily dosage of metformin used in our study was 1100 mg, and women with a higher BMI may require a higher dosage for a clinical effect. Given that women with PCOS and insulin resistance are more likely not to respond to treatment with clomiphene, metformin may be a useful adjunct in this population.

PCOS is associated with peripheral insulin resistance (decreased insulin-mediated glucose utilization) and hyperinsulinemia.<sup>18</sup> Intrinsic alterations in insulin secretion, insulin receptors, and genetic susceptibility may contribute to insulin resistance in thin women with PCOS.<sup>31–34</sup> In obese women with PCOS, insulin resistance is increased. There are no currently validated clinical tests for detecting insulin resistance. Almost one third of our study population had an elevated fasting insulin level or an abnormal fasting glucose/insulin ratio. These screening tests are not routinely recommended because they do not predict clinical events.<sup>35</sup> In our study, an elevated fasting insulin level correlated with an abnormal glucose to insulin ratio. Women with PCOS are at increased risk of developing diabetes mellitus type 2, dyslipidemia and long term cardiovascular complications.<sup>1,36</sup> Although PCOS guidelines do not recommend screening for insulin resistance for diagnosis or treatment,<sup>1</sup> screening women with PCOS for impaired glucose tolerance or the metabolic syndrome is recommended if they have risk factors such as obesity and family history. Recent evidence shows that lifestyle modification or treatment with metformin can reduce the incidence of diabetes mellitus type 2.<sup>37</sup>

## CONCLUSION

We found metformin to be effective for inducing ovulation within three months of initiation in approximately 50% of women with PCOS. Metformin therapy was effective in patients with normal and abnormal insulin levels in our population; however, higher doses of metformin may be needed in patients with elevated fasting insulin levels and higher BMI. Further studies are needed in order to determine if there are clinical, biological, or laboratory markers that can help individualize the best treatment option for each woman with PCOS.

## REFERENCES

1. The Rotterdam ESHRE/ASRM-Sponsored PCOS Consensus Workshop Group. Revised 2003 consensus on diagnostic criteria and long-term health risks related to polycystic ovary syndrome. *Fertil Steril* 2004;81:19–25.
2. Female infertility: treatment options for complicated cases. The ESHRE Capri Workshop. *European Society for Human Reproduction and Embryology. Hum Reprod* 1997;12:1191–6.
3. Infertility revisited: the state of the art today and tomorrow. The ESHRE Capri Workshop. *European Society for Human Reproduction and Embryology. Hum Reprod* 1996;11:1779–807.
4. Nasser S, Ledger WL. Clomiphene citrate in the twenty-first century. *Hum Fertil (Camb)* 2001;4:145–51.
5. Lord JM, Flight IH, Norman RJ. Metformin in polycystic ovary syndrome: systematic review and meta-analysis. *BMJ* 25–10–2003;327:951–3.
6. Kashyap S, Wells GA, Rosenwaks Z. Insulin-sensitizing agents as primary therapy for patients with polycystic ovarian syndrome. *Hum Reprod* 2004;19:2474–83.
7. Palomba S, Orio F, Jr., Falbo A, Manguso F, Russo T, Cascella T, et al. Prospective parallel randomized, double-blind, double-dummy controlled

- clinical trial comparing clomiphene citrate and metformin as the first-line treatment for ovulation induction in nonobese anovulatory women with polycystic ovary syndrome. *J Clin Endocrinol Metab* 2005;90:4068–74.
8. Maciel GA, Soares Jnr JM, Alves da Motta EL, Abi HM, de Lima GR, Baracat EC. Nonobese women with polycystic ovary syndrome respond better than obese women to treatment with metformin. *Fertil Steril* 2004;81:355–60.
  9. Kumari AS, Haq A, Jayasundaram R, Abdel-Wareth LO, Al Haija SA, Alvares M. Metformin monotherapy in lean women with polycystic ovary syndrome. *Reprod Biomed Online* 2005;10:100–4.
  10. Vandermolen DT, Ratts VS, Evans WS, Stovall DW, Kauma SW, Nestler JE. Metformin increases the ovulatory rate and pregnancy rate from clomiphene citrate in patients with polycystic ovary syndrome who are resistant to clomiphene citrate alone. *Fertil Steril* 2001;75:310–5.
  11. Kocak M, Caliskan E, Simsir C, Haberal A. Metformin therapy improves ovulatory rates, cervical scores, and pregnancy rates in clomiphene citrate-resistant women with polycystic ovary syndrome. *Fertil Steril* 2002;77:101–6.
  12. George SS, George K, Irwin C, Job V, Selvakumar R, Jeyaseelan V, et al. Sequential treatment of metformin and clomiphene citrate in clomiphene-resistant women with polycystic ovary syndrome: a randomized, controlled trial. *Hum Reprod* 2003;18:299–304.
  13. Palomba S, Orio F, Jr., Nardo LG, Falbo A, Russo T, Corea D, et al. Metformin administration versus laparoscopic ovarian diathermy in clomiphene citrate-resistant women with polycystic ovary syndrome: a prospective parallel randomized double-blind placebo-controlled trial. *J Clin Endocrinol Metab* 2004;89:4801–9.
  14. Malkawi HY, Qublan HS, Hamaideh AH. Medical vs. surgical treatment for clomiphene citrate-resistant women with polycystic ovary syndrome. *J Obstet Gynaecol* 2003;23:289–3.
  15. Lord JM, Flight IH, Norman RJ. Insulin-sensitising drugs (metformin, troglitazone, rosiglitazone, pioglitazone, D-chiro-inositol) for polycystic ovary syndrome. *Cochrane Database Syst Rev* 2003;CD003053.
  16. Nestler JE, Jakubowicz DJ, Evans WS, Pasquali R. Effects of metformin on spontaneous and clomiphene-induced ovulation in the polycystic ovary syndrome. *NEJM* 1998;338:1876–80.
  17. Sahin Y, Yirmibes U, Kelestimur F, Aygen E. The effects of metformin on insulin resistance, clomiphene-induced ovulation and pregnancy rates in women with polycystic ovary syndrome. *Eur J Obstet Gynecol Reprod Biol* 15–4–2004;113:214–20.
  18. Dunaif A, Segal KR, Futterweit W, Dobrjansky A. Profound peripheral insulin resistance, independent of obesity, in polycystic ovary syndrome. *Diabetes* 1989;38:1165–74.
  19. Deugarte CM, Bartolucci AA, Azziz R. Prevalence of insulin resistance in the polycystic ovary syndrome using the homeostasis model assessment. *Fertil Steril* 2005;83:1454–60.
  20. Legro RS, Finegood D, Dunaif A. A fasting glucose to insulin ratio is a useful measure of insulin sensitivity in women with polycystic ovary syndrome. *J Clin Endocrinol Metab* 1998;83:2694–8.
  21. Ciampelli M, Leoni F, Cucinelli F, Mancuso S, Panunzi S, De Gaetano A, et al. Assessment of insulin sensitivity from measurements in the fasting state and during an oral glucose tolerance test in polycystic ovary syndrome and menopausal patients. *J Clin Endocrinol Metab* 2005;90:1398–406.
  22. Moghetti P, Castello R, Negri C, Tosi F, Perrone F, Caputo M, et al. Metformin effects on clinical features, endocrine and metabolic profiles, and insulin sensitivity in polycystic ovary syndrome: a randomized, double-blind, placebo-controlled 6 month trial, followed by open, long term clinical evaluation. *J Clin Endocrinol Metab* 2000;85:139–46.
  23. Costello MF, Eden JA. A systematic review of the reproductive system effects of metformin in patients with polycystic ovary syndrome. *Fertil Steril* 2003;79:1–13.
  24. Sills ES, Perloe M, Palermo GD. Correction of hyperinsulinemia in oligoovulatory women with clomiphene-resistant polycystic ovary syndrome: a review of therapeutic rationale and reproductive outcomes. *Eur J Obstet Gynecol Reprod Biol* 2000;91:135–41.
  25. Nardo LG, Rai R. Metformin therapy in the management of polycystic ovary syndrome: endocrine, metabolic and reproductive effects. *Gynecol Endocrinol* 2001;15:373–80.
  26. La Marca A, Morgante G, Paglia T, Ciotta L, Cianci A, De L, V. Effects of metformin on adrenal steroidogenesis in women with polycystic ovary syndrome. *Fertil Steril* 1999;72:985–9.
  27. De Leo V, La Marca A, Orvieto R, and Morgante G. Effect of metformin on insulin-like growth factor (IGF) I and IGF-binding protein I in polycystic ovary syndrome. *J Clin Endocrinol Metab* 2000;85:1598–600.
  28. Nestler JE, Jakubowicz DJ. Decreases in ovarian cytochrome P450c17 alpha activity and serum free testosterone after reduction of insulin secretion in polycystic ovary syndrome. *N Engl J Med* 29–8–1996;335:617–23.
  29. Nestler JE, Jakubowicz DJ. Lean women with polycystic ovary syndrome respond to insulin reduction with decreases in ovarian P450c17 alpha activity and serum androgens. *J Clin Endocrinol Metab* 1997;82:4075–9.
  30. Ben Haroush A, Yogev Y, Fisch B. Insulin resistance and metformin in polycystic ovary syndrome. *Eur J Obstet Gynecol Reprod Biol* 10–8–2004;115:125–33.
  31. Ehrmann DA, Breda E, Cavaghan MK, Bajramovic S, Imperial J, Toffolo G, et al. Insulin secretory responses to rising and falling glucose concentrations are delayed in subjects with impaired glucose tolerance. *Diabetologia* 2002;45:509–17.
  32. El Mkaed SA, Lautier C, Macari F, Molinari N, Lefebvre P, Renard E, et al. Role of allelic variants Gly972Arg of IRS-1 and Gly1057Asp of IRS-2 in moderate-to-severe insulin resistance of women with polycystic ovary syndrome. *Diabetes* 2001;50:2164–68.
  33. Dunaif A, Xia J, Book CB, Schenker E, Tang Z. Excessive insulin receptor serine phosphorylation in cultured fibroblasts and in skeletal muscle. A potential mechanism for insulin resistance in the polycystic ovary syndrome. *J Clin Invest* 1995;96:801–10.
  34. Ciaraldi TP, el Roeiy A, Madar Z, Reichart D, Olefsky JM, Yen SS. Cellular mechanisms of insulin resistance in polycystic ovarian syndrome. *J Clin Endocrinol Metab* 1992;75:577–83.
  35. Consensus Development Conference on Insulin Resistance. 5–6 November 1997. American Diabetes Association. *Diabetes Care* 1998;21:310–4.
  36. Sheehan MT. Polycystic Ovarian Syndrome: Diagnosis and Management. *Clin Med Res* 2004;2:13–27.
  37. Knowler WC, Barrett-Connor E, Fowler SE, Hamman RF, Lachin JM, Walker EA, et al. Reduction in the incidence of type 2 diabetes with lifestyle intervention or metformin. *N Engl J Med* 7–2–2002;346:393–403.