Six-month treatment with low-dose dexamethasone further reduces androgen levels in PCOS women treated with diet and lifestyle advice, and metformin

E.Vanky1,3, K.Å.Salvesen1 and S.M.Carlsen2

1Department of Obstetrics and Gynecology and 2Section of Endocrinology, Department of Medicine, St Olavs Hospital, University Hospital of Trondheim, Trondheim, Norway.

3To whom correspondence should be addressed. E-mail: eszter.vanky@medisin.ntnu.no

BACKGROUND: The purpose of this study was to investigate the effect of low-dose dexamethasone on androgen levels in women with polycystic ovary syndrome (PCOS) treated with diet and lifestyle counselling, and metformin.

METHODS: A prospective, randomized, double blind, placebo-controlled study was carried out. Thirty-eight women with PCOS were randomized to either dexamethasone 0.25 mg daily or placebo for 26 weeks. All received diet and lifestyle counselling at inclusion and metformin 850 mg three times daily during the whole study. Main outcome measures were: androgen levels, body mass index (BMI), insulin c-peptide, fasting glucose and serum lipids. Two-tailed t-tests and Pearson’s statistics were used. RESULTS: Compared with the placebo, dexamethasone reduced testosterone by 27%, androstenedione by 21%, dehydroepiandrosterone sulphate by 46% and free testosterone index by 50% in women with PCOS treated with diet and lifestyle advice, and metformin. BMI, fasting glucose, insulin c-peptide and serum lipid levels were unaffected. CONCLUSIONS: Six-month, low-dose dexamethasone treatment further reduces androgen levels in metformin-treated PCOS women.

Key words: androgens/dexamethasone/metformin/PCOS

Introduction

Polycystic ovary syndrome (PCOS) is characterized by polycystic ovaries, oligo-amenorrhoea and hyperandrogenism and is the most common endocrine disorder in women of fertile age. Prevalence estimates vary between 3 and 20% depending on diagnostic criteria used and the population studied (Franks, 1995). In Caucasians, a prevalence of 5–7% has been reported (Diamanti-Kandarakis et al., 1999; Asuncion et al., 2000). In PCOS women, ovarian androgen synthesis is increased, and in 50–70% a concomitant increase in adrenal androgen synthesis has been observed, (Martikainen et al., 1996). While the aetiology of PCOS is unknown, the condition is probably of multifactorial origin. Accumulating evidence supports the view that insulin resistance and hyperinsulinaemia are of major importance. Insulin stimulates androgen synthesis in the ovaries and to some extent in the adrenal glands by enhancing androgen production through CYP 450 17α-related enzyme systems (Nestler, 1997; Franks et al., 1999; la Marca et al., 1999).

The use of the anti-diabetic drug metformin in the treatment of PCOS is quite well established (Nestler and Jakubowicz, 1997; Velazquez et al., 1997; Morin-Papunen et al., 1998; Moghetti et al., 2000; Pasquali et al., 2000). Metformin reduces insulin resistance in muscle, fat and liver tissue, and thereby reduces circulating insulin levels. By reducing insulin levels, metformin reduces both the insulin-enhanced adrenal and ovarian androgen synthesis and the insulin-mediated inhibition of hepatic sex hormone-binding globulin (SHBG) synthesis (la Marca et al, 1999).

PCOS women have increased adrenal androgen synthesis in response to adrenocortictrophic hormone, demonstrating increased activity in the pituitary–adrenal axis (Loughlin et al., 1986). The use of corticosteroids to treat ovulatory dysfunction was first reported by Jones et al. (1953). Ovulatory menses were achieved in 11 out of 14 anovulatory, oligo-amenorrhoeic women treated with cortisone 50 mg daily. Another study reported that 60% of 29 hyperandrogenic women with abnormal menses, receiving dexamethasone 0.25–1.0 mg daily for 6–15 months, improved their menstrual pattern (Abraham et al., 1981). In a study of PCOS women, 10 out of 15 women achieved regular menses after 3 months of treatment with dexamethasone 0.5 mg daily (Loughlin et al., 1986). To our knowledge, there are no prospective randomized controlled studies of the effect of glucocorticoids in a well-defined population of PCOS women.

We regard diet and lifestyle advice and treatment with metformin as the standard treatment of both lean and obese women with PCOS (Kiddy et al., 1989; Kowler et al., 2002). However, in most cases, such treatment is not sufficient to relieve all the signs and symptoms of PCOS. In a previous
Materials and methods

From March to November 2001, 50 women with PCOS were recruited either from our university hospital or gynaecological out-patient clinic, or by advertisement in the local newspaper. Inclusion criteria were polycystic ovaries (≥9 subcapsular follicles with a diameter of 3–8 mm), verified by transvaginal ultrasonography and age between 18 and 40 years. In addition, at least one of the following criteria had to be fulfilled: testosterone >2.5 nmol/l; SHBG <30 nmol/l; fasting insulin c-peptide >1.0 nmol/l; oligo-menorrhoea (length of menstrual cycle >35 days or <10 periods per year); or hirsutism, judged clinically as male pattern growth of body hair. When evaluating inclusion criteria according to ‘Rotterdam 2003’ criteria for PCOS, we found that all patients fulfilled them, i.e. at least two of the three criteria: polycystic ovaries, hyperandrogenism and oligo-menorrhoea.

Exclusion criteria included pregnancy, breast-feeding, known liver disease, alanine aminotransferase >60 IU/l, creatinine >130 μmol/l, known alcohol abuse, diabetes mellitus and treatment with oral glucocorticoids or hormonal contraceptives. Women who had discontinued hormonal contraception at least 1 month prior to inclusion were allowed to enter the study. Congenital adrenal hyperplasia (CAH) was excluded by 17-hydroxprogesterone measurements, and all participants had normal prolactin levels (<784 mIE/l). One of the authors (E.V.) enrolled and assigned all the participants.

The study design is shown in Figure 1. Fifty patients were included, and 38 patients completed the study. Four patients became pregnant, despite being instructed to use non-hormonal contraception during the study period (three in the dexamethasone group and one in the placebo group). Three patients withdrew because of gastrointestinal side effects of metformin (nausea or frequent diarrhoea lasting >3 weeks). In one woman, early ovarian failure had been overlooked. Two patients withdrew from the study due to lack of motivation and another two patients left the study without giving any reason. Of the 38 women completing the whole study, 18 were randomized to dexamethasone 0.25 mg and 20 to placebo. The randomization and encapsulation of dexamethasone and placebo to identical capsules were performed at the pharmacy of our hospital. Block randomization was performed in groups of six according to three categories of body mass index (BMI) <30 kg/m², 30–37 kg/m² and >37 kg/m².

Women were randomized to the dexamethasone or control groups by sealed envelopes, half containing the name of dexamethasone and half the name of placebo. The envelopes were mixed in a random manner and given a randomization number by a pharmacist without connection to the research group. The participants registered their menstrual pattern and were instructed to use non-hormonal contraception during the study period.

At randomization, after 8 weeks and at the end of the study (26 weeks after inclusion), venous blood samples were drawn from an antecubital vein, at between 8 and 10 a.m. after an overnight fast. Blood samples were centrifuged at room temperature within 30 min and stored at −70°C until analysis (1–9 months) as described below. The blood pressure was measured while the patient was in the sitting position after at least 15 min of rest with a digital blood pressure monitor (Fuzzy Logic UA-779, Scan.Med. Norway). The blood pressure was measured three times, at least 2 min apart. The mean of the second and third measurement was calculated.

Study protocol

All participants received individual, written and verbal diet and lifestyle counselling at inclusion. Treatment with metformin 850 mg (metformin hydrochloride, Metformin®, Weifa A/S, Oslo, Norway) was initiated at inclusion. All women used metformin once daily during the first week, twice daily during the second week, and thereafter three times daily for the rest of the study period. At inclusion (week 0), the participants were randomized to additional treatment with either dexamethasone 0.25 mg (dexamethasone natriumphosphate, Decadron®, MSD, Drammen, Norway) or placebo one capsule at bedtime. We hypothesized that dexamethasone would induce a further reduction of androgen levels in PCOS patients treated with metformin, diet and lifestyle counselling.

Primary outcome measures were testosterone, androstenedione, SHBG, free testosterone index (FTI) and dehydroepiandrosterone sulphate (DHEA-S).

Secondary outcome measures were BMI, blood pressure, serum lipids, insulin c-peptide, fasting glucose and menstrual pattern.

The Committee for Medical Research Ethics of Health Region IV, Norway, and The Norwegian Medicines Agency approved the study. A written informed consent was obtained from each patient before inclusion, and the Declaration of Helsinki was followed throughout the study.

Assays

Testosterone and androstenedione were measured by a double antibody technique on an Elecsys 2010 analyser (Roche Diagnostics GmbH, Mannheim, Germany) using reagents and calibrators supplied by the manufacturer. 17-Hydroxyprogesterone was measured using a radioimmunoassay technique with reagents and calibrators supplied by Orion Diagnostica, Espoo, Finland. SHBG and DHEA-S were measured using a competitive immunoassay on an Immulite 2000 analyser using the reagents and calibrators supplied by the manufacturer (Diagnostic Products Corporation, Los Angeles, CA).

The lower detection limits for testosterone, androstenedione, 17-hydroxyprogesterone, SHBG and progesterone were 0.1, 0.1, 0.2, 0.02 and 0.6 nmol/l, respectively. FTI was calculated as total testosterone divided by SHBG and multiplied by a factor of 10.

Reference values were: testosterone 0.1–2.9 nmol/l; androstenedione 0.7–11.0 nmol/l; DHEA-S 0.9–11.7 μmol/l; and 17-hydro-
Effects of dexamethasone in metformin-treated PCOS women

Table I. Patient characteristics at inclusion

<table>
<thead>
<tr>
<th></th>
<th>Dexamethasone group (n = 18)</th>
<th>Placebo group (n = 20)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>26.4 ± 3.8</td>
<td>30.6 ± 5.9*</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>33.5 ± 7.0</td>
<td>32.9 ± 7.7</td>
</tr>
<tr>
<td>No. of additional criteria met for PCOS diagnosis:</td>
<td></td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>2 patients</td>
<td>3 patients</td>
</tr>
<tr>
<td>4</td>
<td>6 patients</td>
<td>6 patients</td>
</tr>
<tr>
<td>3</td>
<td>8 patients</td>
<td>7 patients</td>
</tr>
<tr>
<td>2</td>
<td>2 patients</td>
<td>4 patients</td>
</tr>
<tr>
<td>1</td>
<td>0 patient</td>
<td>0 patient</td>
</tr>
<tr>
<td>Mean no. of criteria met</td>
<td>3.4 ± 0.9</td>
<td>3.4 ± 1.0</td>
</tr>
<tr>
<td>Bleedings per year</td>
<td>4.9 ± 3.2</td>
<td>4.4 ± 3.9</td>
</tr>
</tbody>
</table>

a/P value <0.05.

Table II. Baseline values and the effect of dexamethasone on PCOS women already treated with diet, lifestyle counselling and metformin

<table>
<thead>
<tr>
<th></th>
<th>Placebo group</th>
<th>Dexamethasone</th>
<th>Placebo group</th>
<th>Dexamethasone</th>
</tr>
</thead>
<tbody>
<tr>
<td>Testosterone (nmol/l)</td>
<td>Placebo</td>
<td>20 ± 1.2</td>
<td>Placebo</td>
<td>20 ± 1.2</td>
</tr>
<tr>
<td>SHBG (nmol/l)</td>
<td>Placebo</td>
<td>20 ± 1.2</td>
<td>Placebo</td>
<td>20 ± 1.2</td>
</tr>
<tr>
<td>Free testosterone index</td>
<td>Placebo</td>
<td>20 ± 1.2</td>
<td>Placebo</td>
<td>20 ± 1.2</td>
</tr>
<tr>
<td>Androstenedion (nmol/l)</td>
<td>Placebo</td>
<td>20 ± 1.2</td>
<td>Placebo</td>
<td>20 ± 1.2</td>
</tr>
<tr>
<td>DHEA-S (nmol/l)</td>
<td>Placebo</td>
<td>20 ± 1.2</td>
<td>Placebo</td>
<td>20 ± 1.2</td>
</tr>
<tr>
<td>17-OH progesterone (nmol/l)</td>
<td>Placebo</td>
<td>20 ± 1.2</td>
<td>Placebo</td>
<td>20 ± 1.2</td>
</tr>
<tr>
<td>Prolactin (mIE/l)</td>
<td>Placebo</td>
<td>20 ± 1.2</td>
<td>Placebo</td>
<td>20 ± 1.2</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SD.

xyprogesterone 1.5–12.8 nmol/l. Blood glucose, total cholesterol, high-density lipoprotein (HDL)-cholesterol and triglycerides were analysed on the day of the blood sampling. Serum glucose was analysed by the glucose dehydrogenase method after protein precipitation with perchloric acid using the Merck Granatest 250 reagent kit (E. Merck, Darmstadt, Germany). For serum lipid analyses, the routine method of our laboratory was used. Insulin c-peptide was analysed on an Immulite 2000 analyser using reagents, methods and calibrator obtained from the instrument supplier.

Statistical analysis

All statistical procedures were performed using the Statistical Package for the Social Sciences (SPSS) version 10.0 for Windows SPSS Inc., Chicago, IL.

Sample size calculations, assuming 90% power to detect 1.0 nmol/l change in testosterone between groups indicated the need for 23 patients in each group. SDs were estimated to 0.6 nmol/l. As we anticipated a 5–10% possible ‘drop out’, we included 25 patients in each group.

To evaluate treatment effects, the changes from week 0 to week 8 and week 26 were calculated for each participant. The differences in change between the study and control groups were compared with two-tailed t-tests for independent samples. Values are reported as means and SD. Pearson’s statistics were used for correlation analyses. P-values <0.05 were considered significant. No adjustments for multiple comparisons were performed. Data were analysed according to the ‘intention to treat’ principle.

Results

The study population

The mean age of the participants was 28.6 ± 5.4 years, with older patients in the placebo group than in the dexamethasone group (30.6 ± 5.9 versus 26.4 ± 3.8 years). The mean age of menarche was 12.7 ± 1.6 years and mean BMI was 33.2 kg/m², with no differences between the study groups (data not shown). Five patients met all five of the additional criteria described in Materials and methods, and 12 met four of the criteria. Fifteen and six patients met three and two of the criteria, respectively. None of the participants met only one criterion (Table I).

Twenty-eight (74%) patients met the testosterone criterion (testosterone ≥ 2.5 nmol/l), 24 (63%) met the SHBG criterion (SHBG ≥ 30 nmol/l) and 26 (63%) met the insulin c-peptide criterion (insulin c-peptide ≥ 1.0 nmol/l). Thirty-four (90%) were oligo-amenorrhoeic and 31 (82%) were hirsute. All patients were normoprolactinaemic. Sixteen of the 38 patients completing the study had been pregnant, with 22 pregnancies altogether. These pregnancies resulted in three (13%) spontaneous abortions, two (9%) legal abortions and 17 (77%) live born infants with a mean birth weight of 2960 g. None of the study participants was treated with drugs known to interfere with blood pressure. After completing the study, three patients
were referred for further evaluation and treatment of hypertension.

After 8 weeks of dexamethasone treatment, testosterone was reduced by 45%, androstenedione by 34%, DHEA-S by 32% and FTI by 50% (Table II). Compared with the placebo group, the reduction was 35% for testosterone, 28% for androstenedione, 52% for DHEA-S and 45% for FTI. After 26 weeks, the reduction in the dexamethasone group was 38% for testosterone, 28% for androstenedione, 30% for DHEA-S and 46% for FTI. Compared with the placebo group, the reduction was 27% for testosterone, 21% for androstenedione, 46% for DHEA-S and 50% for FTI. Changes in estradiol \((P = 0.054)\) and SHBG levels \((P = 0.063)\) did not reach statistical significance (Table II).

In the dexamethasone group, there was a positive correlation between the DHEA-S level at inclusion and the change of testosterone \((r = 0.58, P < 0.05)\), androstenedione \((r = 0.70, P < 0.005)\) and FTI \((r = 0.52, P < 0.05)\) at the end of the study.

Fasting glucose, insulin c-peptide, BMI, serum lipid levels and blood pressure were unaffected by dexamethasone treatment (Table III). In the placebo group, BMI at inclusion correlated negatively with change in FTI, i.e. the higher the BMI, the lower the reduction in FTI from week 0 to week 26. In the dexamethasone group, no such correlation was observed.

During the last 12 months prior to the study, the women in the placebo and dexamethasone groups had a mean of 4.4 and 4.9 bleedings per year, respectively. During the study, the frequency of the bleedings increased in both groups to 7.5 and 8.6 per year in the placebo and dexamethasone group, respectively. There was no difference between groups.

**Discussion**

We consider treatment with diet, lifestyle counselling and metformin as the first line treatment in PCOS. We therefore investigated the effect of dexamethasone in PCOS women already treated with this regime. In accordance with our hypothesis, the present study shows that 26 weeks of treatment with a low dose of dexamethasone further reduces circulating androgen levels in PCOS patients.

The dominating view is that hyperandrogenism in PCOS women is caused mainly by ovarian androgen synthesis. The fact that an increased activity in the pituitary–adrenal axis has been reported in PCOS women might be important (Arslanian et al., 2002).

Dexamethasone is not known to exert effects in the ovaries. In the present study, DHEA-S, an androgen precursor produced only by the adrenals, was reduced by 46% in the dexamethasone group. This strongly indicates that dexamethasone suppresses adrenal androgen synthesis and that in metformin-treated patients the adrenals are major contributors to circulating androgen levels. This is in accordance with a study of obese hyperinsulinaemic women (Martikainen et al., 1996). Notably, the suppression of adrenal androgen synthesis achieved by low-dose dexamethasone was only partial, as DHEA-S was within the reference range in all participants throughout the study. Nevertheless, FTI was reduced by 50% compared with the control group.

Dexamethasone is a diabetogenic drug and often promotes weight increase. However, in the present study, no adverse effects of dexamethasone 0.25 mg daily were observed on glucose homeostasis or serum lipid levels. Patients in both groups lost weight during the study, and there was no difference in the reduction of BMI between the groups. Diet, lifestyle advice and metformin might have prevented or counteracted adverse effects of dexamethasone commonly reported in previous studies. We believe, however, that the low dexamethasone dose used (0.25 mg daily) explains why no adverse effects were observed.

By and large, long-term, low-dose treatment with dexamethasone appears to be safe when used in PCOS women.
Unfortunately, we did not measure progesterone levels consecutively to detect ovulations. The frequency of menstruations during the 6-month study period almost doubled, with 7.5 versus 8.6 bleedings per year in the placebo and dexamethasone group, respectively. This difference between the groups did not reach statistical significance. The present study was not designed to and does not clarify whether the major reduction in circulating androgen levels leads to improved menstrual cyclicity. However, in women with CAH, an increased incidence of menstrual irregularities and PCOS was observed. Probably this phenomenon is secondary to increased adrenal androgen synthesis and elevated circulating androgen levels. This indicates that increased adrenal androgen synthesis may have a negative impact on ovarian function (New, 1993).

The reason for improved menstrual pattern with metformin treatment is not clear. In the largest prospective placebo-controlled trial to date, Fleming et al. (2002) showed that metformin improved ovulation in the absence of significant changes in androgen levels and insulin sensitivity. Hence, the improved menstrual pattern induced by metformin may have alternative explanations. In the same study, they showed that the least androgenic women responded best to metformin treatment. It is possible that reduction of androgen levels with dexamethasone may lead to an improved response to metformin in the PCOS women with the highest androgen levels. As could be expected, DHEA-S levels at inclusion correlated with reduction in androgen levels in the dexamethasone group. Hence, PCOS women with the highest adrenal androgen production, manifested by high levels of DHEA-S, achieve the greatest reductions in circulating androgen levels with additional dexamethasone. Both a longer period of observation and a larger study population will be needed to clarify if dexamethasone and metformin act synergistically in improving ovulatory dysfunction in PCOS patients.

To our knowledge, this is the first controlled study of dexamethasone treatment in a well-defined population of PCOS women. Low-dose dexamethasone treatment further decreased androgen levels in PCOS women treated with diet, lifestyle counselling and metformin. The large reduction in bioavailable testosterone and DHEA-S levels in the dexamethasone group indicates a role for adrenal androgen synthesis in the pathogenesis of PCOS. The adverse effects of dexamethasone reported in previous studies were not found in the present study, probably due to lower dosage of the drug. This study suggests that the combination of metformin and low-dose dexamethasone may be beneficial in the treatment of PCOS. Larger controlled studies are needed to address this question.

Acknowledgements

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References


