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Metabolic and hormonal effects of myo-inositol in women with polycystic ovary syndrome: a double-blind trial

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Abstract. – To investigate the effects of treatment with Myo-inositol (an insulin sensitizing drug), on circulating insulin, glucose tolerance, ovulation and serum androgens concentrations in women with the Polycystic Ovary Syndrome (PCOS). Forty-two women with PCOS were treated in a double-blind trial with Myo-inositol plus folic acid or folic acid alone as placebo. In the group treated with Myo-inositol the serum total testosterone decreased from 99.5±7 to 34.8±4.3 ng/dl (placebo group: from 116.8±15 to 109±7.5 ng/dl; *P*=0.003), and serum free testosterone from 0.85±0.1 to 0.24±0.33 ng/dl (placebo group: from 0.89±0.12 to 0.85±0.13 ng/dl; *P*=0.01). Plasma triglycerides decreased from 195±20 to 95±17 mg/dl (placebo group: from 166±21 to 148±19 mg/dl; *P*=0.001). Systolic blood pressure decreased from 131±2 to 127±2 mmHg (placebo group: from 128±1 to 130±1 mmHg; *P*=0.002). Diastolic blood pressure decreased from 88±1 to 82±3 mmHg (placebo group: from 86±1 to 90±1 mmHg; *P*=0.001). The area under the plasma insulin curve after oral administration of glucose decreased from 8.54±1.149 to 5.535±1.792 μU/ml/min (placebo group: from 8.903±1.276 to 9.1±1.162 μU/ml/min; *P*=0.03). The index of composite whole body insulin sensitivity (ISIcomp) increased from 2.80±0.35 to 5.05±0.59 mg-2/dl-2 (placebo group: from 3.23±0.48 to 2.81±0.54 mg-2/dl-2; *P*<0.002). 16 out of 23 women of Myo-inositol group ovulated (4 out of 19 in placebo group). Treatment of PCOS patients with Myo-inositol provided a decreasing of circulating insulin and serum total testosterone as well as an improvement in metabolic factors.

Key Words:

Polycystic Ovary Syndrome, PCOS, Myo-inositol, Metabolic syndrome.

Introduction

Polycystic ovary syndrome (PCOS) is one of the most common endocrine disorders in women of reproductive age[1-3]; its aetiology remains unknown[4]. Ovulatory disorders represent a major cause of infertility, and the oligoovulation and anovulation with polycystic ovary syndrome (PCOS) are common cause of infertility, which is the most common endocrinopathy of reproductive aged women affecting 6-10% of the population.

The current definition of PCOS requires the presence of two of the following three conditions: (i) oligo- and/or anovulation; (ii) clinical and/or biochemical signs of hyperandrogenism that may be associated with hirsutism; and (iii) polycystic ovaries – and the exclusion of other aetiologies. Other features of PCOS are acne, seborrhea, obesity, insulin resistance, impaired glucose tolerance and type 2 diabetes mellitus, dyslipidaemia, cardiovascular disease and infertility. Furthermore, endocrine and metabolic alterations as elevated serum concentrations of testosterone, insulin, luteinizing hormone (LH) and prolactin are prevalent in PCOS population. These last may have a profound implications for the long-term health of patients.

In young women with PCOS, insulin resistance may occur with high frequency. In fact many studies revealed that it is intrinsic to the syndrome and affects 30 to 40% of patients with PCOS[5]. Some studies showed that insulin resistance in the PCOS may be linked to abnormal ovarian steroidogenesis by means of altered insulin signal transduction[6,7].

Inositol phosphoglycan molecule is known to have a role in activating enzymes that control

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glucose metabolism[8-10]. A defect in tissue availability or altered metabolism of inositol or inositol phosphoglycan mediators, as in PCOS women, may contribute to insulin resistance[11,12]. Isoform of inositol belongs to the vitamin B complex. Epimerization of the six hydroxyl-groups of inositol leads to the formation of up to nine stereo isomers, including Myo-inositol (MYO) and D-chiro-inositol (DCI). Elevated concentrations of MYO in follicular fluid appear to play a role in follicular maturity and provide a marker of good quality oocytes[13-15]. Furthermore, experiments on mouse oocytes showed that an adding of MI to the culture increases the meiotic progression of germinal vesicle by enhancing the intracellular Ca^{2+} oscillation[16].

Women with the PCOS could be respond favourably to treatment with insulin-sensitizing drugs. Previous studies have shown that the use of metformin, troglitazone or Myo-inositol reduces serum androgens, and improves ovulation in women with the PCOS. In those studies, administration of metformin to patients showed a reduction in circulating and a decrement serum total and free testosterone concentrations[17].

A recent study outlines a deficiency of Myo-inositol in insulin resistance in women with the PCOS and the administration of Myo-inositol reduces serum insulin, decreases serum testosterone, and enhances ovulation[18].

The aim of this study was to investigate the metabolic and hormonal effects of MI in PCOS patients.

Material and Methods

This study was a double-blind trial (subjects and investigators).

42 patients, 18 to 40 years of age, were selected to study. They were PCOS affected with oligomenorrhea, high serum free testosterone level and/or hirsutism.

Women were observed by pelvic, ultrasonography and PCOS was found[19].

13 of the 42 women were taking some drugs (oral contraceptives, insulin-sensitizing agents and others) during two months before the study.

After randomization, 23 women received 4 gr of Myo-inositol plus 400 mcg of folic acid (Inofolic®) and 19 women received 400 mcg folic acid (Fertifol®) alone as placebo. The treatment was made for 12-16 weeks.

Seven women had impaired glucose tolerance (plasma glucose concentration >140 mg/dl, <200 mg/dl two hours after oral ingestion of 75 g of dextrose). Four of them was assigned to receive Myo-inositol (Inofolic®) and three were assigned to receive placebo (Folic acid only).

The study was approved by the Institutional Review Boards and each woman gave written informed consent.

When we started the study, the patients were in the follicular phase of the menstrual cycle (serum progesterone concentration lower than 2.5 ng/ml). On the first day blood pressure, weight, height, waist to hip ratio were measured. In the morning, 8:30, 8:45 and 9:00 am., sex hormone binding globulin and serum steroids were obtained. At 9:00 a.m., 75 g of dextrose were administered and plasma glucose and insulin were measured after 30, 60, 90, 120 minutes.

How to take drugs (Myo-inositol or placebo orally once a day) was explained to the patients as well as not to change usual habits both for food, sport and lifestyle.

The serum progesterone was measured weekly and if the relevant results were over 8 ng/ml the ovulation was supposed

After 6 weeks of drugs (day 49), women in the follicular phase (serum progesterone concentration <2.5 ng/ml) repeated all the baseline measurement.

Statistical Analysis

The results are reported as mean values ± SE.

The areas under the response curves by the trapezoidal rule were used to evaluate the plasma glucose and insulin concentration after the oral administration of glucose.

The oral glucose tolerance test (OGTT), by the use of the index of composite whole-body insulin sensitivity (ISIcomp), was used to determine the insulin sensitivity. This methodology was developed by Matsuda and De Fronzo[20]: ISIcomp=10.000/square root of ([fasting glucose × fasting insulin] × [mean glucose × mean during OGTT]).

To analyze the difference in ovulation rates between the women who received the Myo-inositol and those who received placebo the Fisher exact test was used. The results of the other variables were obtained by comparing the changes from to baseline to the end of the study in both the groups. The distribution of the changes in the two groups was first tested for normality with use of the Wilks-Shapiro test, and then the distri-

Table I. Baseline characteristics.

Variable	Myo-inositol N = 23	Placebo N = 19
Age	28.8 ± 1.5	27.1 ± 1.4
Waist to hip ratio	0.88 ± 0.02	0.87 ± 0.02
BMI (kg/m^2)	22.8 ± 0.3	22.5 ± 0.3
Menstrual period/yr	3 ± 1	3 ± 1
Free testosterone (ng/dl)	0.85 ± 0.11	0.89 ± 0.12
Androstenedione (ng/dl)	267 ± 19	271 ± 21
DHEAS (µg/dl)	366 ± 47	384 ± 63
Total testosterone (ng/dl)	99.5 ± 6.9	116.8 ± 14.7
17 beta estradiol (pg/ml)	45 ± 2.5	70 ± 6.7
Sex hormone binding globulin (nmol/L)	144.4 ± 18.6	147 ± 14.5
Total cholesterol (mg/dl)	210 ± 10.4	195 ±7.35
Triglycerides (mg/dl)	195 ± 20.2	166 ± 20.6
ISIcomp (mg-2/dl-2)	2.80 ± 0.35	3.23 ± 0.48
Glucose AUC (mg/dl/min)	12.409 ± 686	12.970 ± 802
Insulin AUC (µU/ml/min)	8.549 ± 1.149	8.903 ± 1.276
Fasting insulin (µU/ml)	32.5 ± 4.1	30.8 ± 7.3
Fasting glucose (mg/dl)	87.6 ± 3.5	84.9 ± 5.8
Systolic blood pressure (mmHg)	131 ± 2.3	128 ± 1.3
Diastolic blood pressure (mmHg)	88 ± 1.0	86 ± 7.0

DHEAS = dehydroepiandrosterone; AUC = area under the curve during 2 hours, 75 g oral glucose tolerance test; ISIcomp = index of composite whole body insulin sensitivity.

butions were compared with each other by using the Student to-tailed unpaired or the Wilcoxon rank sum test.

P values <0.05 were considered significant.

Results

The women of the two groups were similar for baseline characteristics (age, BMI, waist to hip ratio, plasma lipids, and other) (Table I). No significant differences were recorded in the two groups for fasting plasma insulin, plasma glucose, areas under the curve for insulin and glucose during the OGTT, and frequency of glucose tolerance.

There was a slight change in BMI in both study groups (Table II).

There was not a statistically significant modification of waist to hip ratio in both groups.

There was a decrement in systolic pressure in Myo-inositol group (from 131±2 to 127±2 mmHg) while an increment in placebo group (from 128±1 to 130±1 mmHg; *P*=0.002); similarly about the diastolic blood pressure, with

Table II. Anthropomorphic and lipid characteristics.

Characteristic	Myo inositol group N = 23		Placebo group N = 19		*P* value for change comparison
	Baseline	*After treatment*	*Baseline*	*After treatment*	
Systolic blood pressure (mmHg)	131 ± 2	127 ± 2	128 ± 1	130 ± 1	0.002
Diastolic blood pressure (mmHg)	88 ± 1	82 ± 3	86 ± 7	90 ± 1	0.001
Triglycerides (mg/dl)	195 ± 20	95 ± 17	166 ± 21	148 ± 19	0.001
Total cholesterol (mg/dl)	210 ± 10	171 ± 11	195 ± 7	204 ± 9	0.001
BMI (kg/m^2)	22.8 ± 0.3	22.9 ± 0.3	22.5 ± 0.3	22.4 ± 0.1	NS
Waist to hip ratio	0.88 ± 0.02	0.87 ± 0.02	0.87 ± 0.02	0.89 ± 0.01	NS

NS: not significant.

Table III. Plasma glucose and insulin sensitivity index measurements (for 6 to 8 weeks).

Characteristic	Myo inositol group N = 23		Placebo group N = 19		*P* value for change comparison
	Baseline	*After treatment*	*Baseline*	*After treatment*	
Fasting insulin (μU/ml)	32 ± 4	26 ± 8	30.8 ± 7	38 ±7	0.20
Fasting glucose (mg/dl)	87.6 ± 4	81.6 ± 4	84.9 ± 6	88 ± 4	0.12
Glucose AUC (mg/dl/min)	12.409 ± 686	10.452 ± 414	12.970 ± 802	12.992 ± 793	0.04
Insulin AUC (μg/ml/min)	8.54 ± 1.149	5.535 ± 1.792	8.903 ± 1.276	9.1 ± 1.162	0.03
ISIcomp (mg^{-2}/dl^{-2})	2.80 ± 0.35	5.05 ± 0.59	3.23 ± 0.48	2.81 ± 0.54	< 0.002

AUC = Area under the curve during 2 hours, 75 g oral glucose tolerance test; ISIcomp = index of composite whole body insulin sensitivity.

decrement (from 88±1 to 82±3 mmHg) in Myo-inositol group and increment (from 86±7 to 90±1 mmHg) in placebo group respectively (*P*=0.001).

In the Myo-inositol group plasma triglycerides decreased by 52% (from 195±20 to 95±17 mg/dl) and total cholesterol decreased significantly (from 210±10 to 171±11 mg/dl).

The fasting plasma insulin concentration did not change significantly in either study group (Table III). The area under the plasma insulin curve decreased by 36% (from 8.54±1.149 to 5.535±1.792 μU/ml/min) while the same was not in the placebo group (from 8.903±1.276 to 9.1±1.162 μU/ml/min; *P*=0.03).

Likely was for the fasting plasma glucose concentration (from 87.6±4 to 81.6±4 mg/dl). The area under the plasma glucose curve during OGTT decreased in Myo-inositol group (from 12.409±686 to 10.452±414 mg/dl/min) while a slight increment was in placebo group (from 12.970±802 to 12.992±793 mg/dl/min; *P*=0.04).

The composite whole body insulin sensitivity index (ISIcomp) increased by 84% (from 2.80±0.35 to 5.05±0.59 mg^{-2}/dl^{-2}) in the Myo-inositol group and did not change in the placebo group (from 3.23± 0.48 to 2.81±0.54 mg^{-2}/dl^{-2}) (Table III). The change between two groups was significant (*P*<0.002).

Sixteen (69,5%) and four (21%) women ovulated in the Myo-inositol group and the placebo group respectively. The different is statistically significant (*P*=0.001).

The progesterone peak value was higher in the Myo-inositol group (15.1±2.2 ng/ml).

In the Myo-inositol group there was a decrement of serum total testosterone (from 99.5±7 to 34.8±4.3 ng/dl) and free testosterone concentrations (from 0.85±0.11 to 0.24±0.03 ng/dl) (Table IV).

An increase of serum sex hormone binding globulin was revealed for each groups (*P*=0.40).

There was an important decrement of the serum dehydroepiandrosterone sulphate in the Myo-inositol group (from 366±47 to 188±24 μg/dl; *P*=0.003) while it wasn't significant in the placebo group (from 384± 63 to 320±35 μg/dl; *P*=0.06).

The other serum sex steroid concentration did not change between two groups.

Table IV. Serum sex hormone (for 6 to 8 weeks).

Characteristic	Myo inositol group N = 23		Placebo group N = 19		*P* value for change comparison
	Baseline	*After treatment*	*Baseline*	*After treatment*	
Total testosterone (ng/dl)	99.5 ± 7	34.8 ± 4.3	116.8 ± 15	109 ± 7.5	0.003
Free testosterone (ng/dl)	0.85 ± 0.11	0.24 ± 0.03	0.89 ± 0.12	0.85 ± 0.13	0.01
DHEAS (μg/dl)	366 ± 47	188 ± 24	384 ± 63	320 ± 35	0.06
SHBG (nmol/l)	144.4 ± 19	198 ± 24	147 ± 4	163 ± 26	0.40
Androstenedione (ng/dl)	267 ± 19	196 ± 26	271 ± 21	306 ± 41	0.09
Progesterone peak value (ng/ml)*	–	15.1 ± 2.2	–	6.6 ± 1.3	0.003

DHEAS= Dehydroepiandroserone; SHBG= Sex Hormone binding globulin; *the highest progesterone concentration measured for an individual subject during the study.

Discussion

Insulin-sensitizing agents have been recently suggested as the therapy of choice for polycystic ovary syndrome (PCOS), since insulin resistance and associated hyperinsulinemia are recognized as important pathogenetic factors of the syndrome. In fact, almost all obese PCOS women and more than half of those of normal weight are insulin resistant, and therefore present some degree of hyperinsulinemia. For this reason the use of insulin sensitizers had been suggested in most patients with PCOS, as a treatment useful in the reduction of serum androgen levels and gonadotropins, and in the improvement in serum lipids. and prothrombotic factor plasminogen-activator inhibitor type 1. These therapies have also been associated with a decrease in hirsutism and acne, and with a regulation of menses and an improvement of ovulation and fertility.

Recently a defect in the insulin signal pathway (inositol-containing phosphoglycan mediators) had been discovered to be implicated in the pathogenesis of insulin resistance[8,12]. As consequence, the administration of different isoforms of inositol as D-Chiro-inositol (DCI) or myo-inositol (MYO) is newly demonstrated improving the physiological insulin-receptor activity, restoring spontaneous ovulatory function in most of PCOS women[14,15,18,21].

Aim of our study was to better focus on metabolic implication of a chronic treatment with MYO in PCOS patients.

We analyzed a total of 42 patients treated by Myo-inositol (N° 23) or placebo (N° 19). Myo-inositol increased insulin sensitivity, improved glucose tolerance and decreased glucose stimulated insulin release. In these patients there was a 66% decrement of serum total testosterone and 73% decrement of serum free testosterone concentrations. In addition there was a decrement in systolic and diastolic blood pressure. Plasma triglycerides and total cholesterol concentration decreased.

In women with the PCOS, insulin resistance may be related to a deficiency in Myo-inositol containing mediator of insulin action and the administration of the Myo-inositol improves insulin sensitivity.

In conclusion, Myo-inositol decreases serum androgen concentrations, reduces circulating insulin and improves glucose tolerance and other metabolic values altered associated with insulin resistance in women affected by Polycystic ovary syndrome.

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