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The Effect of Myo-inositol Pretreatment on ICSI/IVF Outcome for Infertile Patients with Polycystic Ovary Syndrome

Abstract

Successful pregnancy and childbirth are among the most critical periods of life for women, and any factor affecting their fertility can have mental and social challenges for their life. Thus, examining the diseases that affect the physical and mental characteristics of women as sensitive and vulnerable members of society is very important. Polycystic Ovary Syndrome (PCOS) is one of these diseases .The current study examines myo-inositol pretreatment's effect on ICSI/IVF outcomes for infertile patients with polycystic ovary syndrome. Fifty-four patients with IVF with polycystic ovary diagnostic criteria were included in the study. They were divided into control and intervention groups based on basic experiments with block randomization methods. Data was analyzed by using descriptive and analytic statistics in Strata software. The significance level was designed at 0.05. The mean ovule at metaphase II, inoculation, number of zygote 2PN, freezing period, and infertility in the intervention group were significantly higher than in the control group. A significant decrease in side effects of Myo-inositol has made it desirable for patients with polycystic ovary syndrome.

Keywords: *Polycystic ovary syndrome, Intracytoplasmic sperm injection, Myo-inositol, Outcome, Infertility.*

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Introduction

Successful pregnancy and childbirth are among the most critical periods of life for women, and any factor affecting their fertility can have mental and social challenges for their life. Thus, examining the diseases that affect the physical and mental characteristics of women as sensitive and vulnerable members of society is very important. Polycystic Ovary Syndrome (PCOS) is one of these diseases [1-3]. PCOS is a set of disorders diagnosed through sonogram by symptoms including lack of ovulation or irregular ovulation, clinical or biochemical symptoms of hyperandrogenism (i.e., hirsutism, acne, male pattern of baldness, increased serum androgens), and polycystic ovules [4].

Polycystic ovary symptoms are mostly emerging immediately after the first menstruation. In some cases, gradual weight gain occurs in the next pregnancy years. Symptoms are different for different persons (both in terms of severity and type). The corresponding physician should search for at least two symptoms of irregular ovulation, polycystic ovules, and increased androgens or male hormones [5].

Polycystic ovary syndrome can increase the chance of diseases such as abnormal vaginal bleeding, pregnancy Diabetes or pregnancy hypertension [6, 7], and ondometer cancer caused by repeated contact with high amounts of estrogen. Polycystic ovary syndrome treatment is generally based on controlling personal concerns about pregnancy, hypertrichosis, acne, and obesity. Various studies have been conducted in this regard. Patients with polycystic ovary syndrome with gonadotropins and other treatments for stimulating ovulation and pregnancy are unsuccessful [8]. Patients taking In-Vitro Fertilization (IVF) with Intracytoplasmic Sperm Injection (ICSI) enter this phase. In IVF, gonadotropins are prescribed for stimulating the growth of follicles at the ovule level, ovule harvest, and fetus production under experimental conditions and then transferring them into the womb [9]. The pregnancy to IVF ratio is about 40-50%. Successful ART periods are affected by age, ovule quality, fetus quality, and other factors. Side effects, including the risk of hyper-stimulation of ovules and sometimes a low number of ovules or low-quality ovules despite aspiration of a high number of follicles, are frequently seen in these patients [10, 11].

In Artini et al.'s (2013) study on 50 patients with weight gain and polycystic ovary syndrome, patients were divided into two groups. One group received myo-inositol and folic acid; the other only received folic acid. After 12 weeks, parameters such as plasma LH, prolactin, insulin level, and LH to FSH ratio were significantly decreased. It was determined that myo-inositol affects the reproductive performance of patients with polycystic ovary syndrome and decreases hyperinsulinemia [12]. In a study by Govindarajan et al. at the Pharmacology Department of Chennai University in India in 2015, the effect of myo-inositol supplements on the treatment of a patient with polycystic ovary syndrome was examined. Myo-inositol was reported as a safe and effective drug for ovules' performance and metabolic and hormone parameters of women with polycystic ovary syndrome based on analyses conducted [13, 14]. Faccinetti et al., in a study in 2015, examined the medical effects of inositol compounds on women with polycystic ovary

syndrome and metabolic syndrome. They reported that myo-inositol and D-chiroIns intervene in several biologic routes, including insulin signals. Patients with polycystic ovary syndrome can benefit from receiving inositol compounds in the form of 2g 2 times a day. Moreover, they reported that combined treatment with myo-inositol and D-chiroIns can be more effective and efficient than a single use of each one [15]. Hyper-stimulated ovary syndrome, among the prevalent outcomes of ovulation induction in patients with polycystic ovary syndrome, can be accompanied by very high prevalence and sometimes even death. This syndrome is accompanied by decreased recycled ovules quality in the IVF period despite the high number of aspirated follicles. This resulted in a reduced probability of fetus transfer during the first period and decreased chance of pregnancy even during freezing periods, which is the ultimate goal for these patients. Thus, the current study aims to examine the effect of myo-inositol pretreatment on ICSI/IVF outcomes for infertile patients with polycystic ovary syndrome.

Methodology

The current study is a clinical trial with the double-blind, randomized method. The statistical population includes all infertile women with polycystic ovary syndrome taking IVF at Hazrat Zahra Infertility Center of Shahrekord.

Using similar studies [16], the sample size was determined to be 54, which was divided into an intervention group (24) and a control group (30). Therefore, 54 patients for IVF younger than 35 and had basic FSH relevant to their age had polycystic ovary syndrome diagnostic criteria, their body mass index was lower than 30, and inclusion criteria were selected. They were divided into two groups through block randomization by computer after basic experiments for the third day of menstruation, FSH, LH, estrogen, prolactin, anti-Mullerian, and insulin. One group received 400 mg/day of folic acid for three months, and the other group received 2000 mg/day of myo-inositol plus 400 mg/day of folic acid for three months. Then, both groups entered the ovulation stimulation period using the antagonist protocol, in which a 150-unit dose of rFSH was considered for the patient based on conditions until achieving an 18 mm follicle. Moreover, after the mature follicle (14 mm) emerged at the ovule level, an antagonist was prescribed as daily subcutaneous cetrotide. After observing 18-22 mm follicles at the ovule level, 10000-unit HCG was injected, and ovulation was observed and examined after 36 hours.

The period outcome was examined in terms of hyper-stimulation of ovule, quantitative and qualitative assessment of ovules and fetus, and clinical pregnancy resulting from fetus transfer. In this regard, IVF ovules were examined qualitatively and quantitatively after aspirating pre-ovulating follicles. After determining the rate of immature and atretic ovules (GV, metaphase I, degenerated ovules) to mature ovules (metaphase II) in experimental groups, zona pellucida quality, and PVS quality were examined and compared. Then, mature ovules were transferred to IVF 50 λ drops and were placed near the determined number of sperms (100.103 sperm/ml). After 18-20 h, the pronuclear formation process, inoculation, and fetus share percent were determined. Fetuses were qualitatively graded. Then, fetuses were transferred to the womb on days 3-5, and chemical and clinical pregnancy were examined and compared. Finally, Strata software analyzed data obtained through descriptive statistical tests such as dispersion indices and analytical statistics, including T-test and chi-square.

Findings

Parameters

Table 1 shows the results of studied parameters for infertile women with polycystic ovary syndrome taking IVF. The mean age of the study patient is 29.04 ± 3.62 for the myo-inositol group and 29.31 ± 4.05 for the control group. Moreover, the body mass index is 27.37 ± 3.66 and 27.4 ± 3.63 kg/m^2, respectively. LH density was 11.5 ± 3.23 µmol/dl for the intervention group and 12.10 ± 2.87 µmol/dl for the control group. FSH density was 5.04 ± 1.47 µmol/dl for the intervention group and 4.90 ± 1.39 µmol/dl for the control group. Anti-Mullerian Hormone (AMH) density was 9.26 ± 2.64 µmol/dl for the intervention group and 9.76 ± 2.39 µmol/dl for the control group. Moreover, estrogen density was 59.5 ± 9.90 µmol/dl for the intervention group and 53.54 ± 8.89 µmol/dl for the control group. The mean cetrotide used was 4.20 ± 0.65 µg for the intervention group and 5.96 ± 1.12 µg for the control group. Moreover, the mean rFSH used was 11.25 ± 0.73 µg for the intervention group and 12.96 ± 1.12 µg for the control group. Mean mature ovules were 9.12 ± 1.22 for the intervention group and 8.06 ± 1.36 for the control group. The number of MI-GV ovules was 5.83 ± 1.88 for the intervention group and 8.1 ± 2.89 for the control group. Inoculation was 82.33 ± 9.54 for the intervention group and 67.89 ± 13.14 for the control group. Share was 78.12 ± 10.44 for the intervention group and 71.49 ± 10.16 for the control group. The mean number of zygote 2PN was 9.79 ± 1.02 for the intervention group and 8.06 ± 1.77 for the control group. The mean ovules grade was 2.57 ± 0.51 for the intervention group and 2.6 ± 0.50 for the control group. The mean freezing period was 6.12 ± 1.87 for the intervention group and 4.32± 1.84 for the control group. Mean fertility was 0.29 ± 0.46 for the intervention group and 0.26 ± 0.44 for the control group.

Variable	Group	Frequency	SD ± Mean

Age	Intervention	24	27.04 ± 3.62
	Control	29	29.31 ± 4.05
BMI	Intervention	24	27.37 ± 3.66
	Control	30	27.4 ± 3.63
LH	Intervention	24	11.5 ± 3.23
	Control	30	12.10 ± 2.87
FSH	Intervention	24	5.04 ± 1.47
	Control	30	4.90 ± 1.39
AMH	Intervention	24	9.26 ± 2.64
	Control	30	9.76 ± 2.39
E2	Intervention	24	59.5 ± 9.90
	Control	30	53.54 ± 8.89

Table 1. Demographic characteristics of infertile patients with polycystic ovary syndrome participated in the study

Fertility

Table 2 shows the fertility of study patients. Of 54 subjects, 72.22% were infertile, and 27.78% had positive fertility. Infertility and positive fertility for the intervention group were 70.83% and 29.17%, respectively. Infertility and positive fertility for the control group were 73.33% and 26.67%, respectively. The results show no significant difference between the intervention and control groups regarding fertility (p = 0.83).

Group	Fertility Number/percent		Total	p
	Negative	Positive		
Intervention	17 (70.83)	7 (29.17)	24	0.83
Control	22 (73.33)	8 (26.67)	30	
Total	39 (72.22)	15 (27.78)	54	

Table 2. Fertility

Fetus grade

As seen in table 3, there is no significant difference between the intervention group and control group regarding fetus grade. The quality of fetuses obtained is similar for the intervention and control groups.

Group	Number	SD ± Mean	p
Intervention	14	2.57 ± 0.51	0.4
Control	15	2.6 ± 0.50	
Total	29	2.58 ± 0.50	

Table 3. Comparison of fetus

Hospitalization

Ten of 24 polycystic ovary syndrome patients in the intervention group were hospitalized due to hyper-stimulated ovary syndrome. Of 30 patients in the control group, 15 were hospitalized due to hyper-stimulated ovary syndrome. As can be seen in table 4, there is no significant difference between the intervention group and control group in terms of hospitalization due to hyper-stimulated ovary syndrome (p = 0.46).

Group	hyper-stimulated ovary syndrome			p
	Non-hospitalized	Hospitalized	Total	
Intervention	14	10	24	0.46
Control	15	15	30	

Total	29	25	54	

Table 4. Results of hyper-stimulated ovary syndrome

Discussion

The current study aims to examine the effect of myo-inositol pretreatment on ICSI/IVF outcomes for infertile patients with polycystic ovary syndrome. Results revealed that the mean number of ovules at metaphase II, inoculation, number of zygote 2PN, freezing period, and infertility in the intervention group were significantly higher than in the control group.

Bizzari & Bevilacqua (2016) examined the effect of myo-inositol on patients with polycystic ovary syndrome. In the literature review, they referred to various studies on the effect of myo-inositol as the secondary messenger of insulin in insulin-dependent patients with polycystic ovary syndrome. They reported that myo-inositol is effective across multiple signal routes at the ovule and non-ovule levels and plays an important role. D-ChiroIns, another isomer of myo-inositol, is not effective in promoting the performance of ovule cells, and its positive effects are generally limited to non-ovule textures. These isomers significantly prevent the negative cellular effects of hyperinsulinemia in non-ovule textures [17, 18].

Various studies have examined the effect of myo-inositol in IVF protocols on patients with polycystic ovary syndrome. Results reported increased mature ovules of metaphase II, decreased use of FSH to achieve follicle maturity, and a significant increase in pregnancy. Myo-inositol is effective in preventing hyper-stimulated ovary syndrome [19]. Moreover, various studies show that compared to metformin, prescribing myo-inositol before IVF treatment helps stimulate ovulation [17, 20]. These studies confirm the current research results revealing the positive effects of myo-inositol on the increased number of mature ovules, decreased need for FSH for follicle maturity, higher pregnancy rate, and decreased hyper-stimulated ovary syndrome. There is no significant difference between the intervention and control groups regarding hyper-stimulated ovary syndrome, despite the decreased chance of hyper-stimulated ovary syndrome in treated patients in both medical protocols.

Despite contradictory hypotheses and evidence on differences among clinical characteristics of polycystic ovary syndrome, it has become clear that insulin resistance plays a vital pathogen role in patients with polycystic ovary syndrome. Insulin stimulates cells in the ovule and directs or indirectly produces and releases androgens by regulating carbohydrate levels. High glucose levels in the blood prevent the synthesis of sexual hormones attached to globulin by the liver, resulting in increased free androgen. When insulin is attached to its receiver, two types of inositol phosphoglycans (IPG) containing myo-inositol and D-chiroIns are released through hydrolysis of glycosyl-phosphatidyl lipids of inositol at the outer level of the cell membrane. IPG affects intracellular metabolic processes by activating key enzymes controlling oxidative and non-oxidative metabolism of glucose [21]. In women with polycystic ovary syndrome, serum levels of D-chiroIns decrease, and D-chiroIns level increases in urine. These changes show the significant relationship between insulin resistance and inositol deficiency in these patients. Studies show that treating women with polycystic ovary syndrome with low doses of D-chiroIns (1.2 g/day) decreases lipid biomarkers, increases insulin sensitivity, decreases serum levels of androgens, and increases ovulation frequency. However, prescribing higher doses of D-chiroIns will negatively affect ovule textures. This negative effect reveals that D-chiroIns may worsen ovule response in patients with noninsulin-dependent polycystic ovary syndrome. It should be mentioned that D-chiroIns have an essential role in normalizing insulin resistance parameters [22]. Studies show that prescribing anti-Diabetes drugs improves clinical symptoms of polycystic ovary syndrome in some patients but decreases ovule quality. This indicates that insulin resistance is probably not the primary pathogen of polycystic ovary syndrome.

Moreover, some studies show that D-chiroIns can adversely affect ovule textures. Releasing high levels of D-chiroIns as the result of insulin stimulation increases the biosynthesis of testosterone by ovule cells, resulting in increased serum androgen levels. D-chiroIns may also significantly prevent myo-inositol absorption in mammals and result in an imbalance of their rate in ovule [23,26]. So, it was mentioned that myo-inositol is the most prevalent isomer of inositol in ovules and approximately shapes 99% of the total mass of inositol. This compound is converted to D-chiroIns through NAD-dependent epimerase and under the effect of insulin. Decreased epimerase activity in the muscular texture of patients with type 2 Diabetes and its decreased concentration in other non-ovule textures have been seen in patients with insulin resistance. A significant decrease in myo-inositol to D-chiroIns ratio in follicle liquid has been seen in patients with polycystic ovary syndrome [24].

Positive effects of prescribing myo-inositol for patients with polycystic ovary syndrome have been shown in various studies. Myo-inositol supplements improve ovule performance, ovule quality, and ovulation frequency; increase pregnancy and decrease the number of required FSH treatments for ovulation in patients with polycystic ovary syndrome. It should be mentioned that ovulation frequency and pregnancy increase through treatment with myo-inositol. Therefore, myo-inositol improves ovule performance and increases fertility parameters related to follicle maturity and

ovule, number and quality of fetus, and pregnancy in patients with polycystic ovary syndrome [25].

Conclusions

It became apparent in the current study that the number of unsuccessful periods, the number of freezing periods, and the chance of hyper-stimulated ovary syndrome significantly improved in patients treated with myo-inositol. Concerning the lack of side effects of this drug and its advantageous medical effects, using myo-inositol is recommended to increase the success of IVF and ICSI periods in infertile women with polycystic ovary syndrome.

Conflicts of interest

This article was derived from the MD thesis of dr. Shadrooz Moazzam whose protocol was approved by the Deputy of Research and Technology of Shahrekord
University of Medical Sciences.

Hereby, the financial support of the Deputy of Research and Technology of Shahrekord University and the collaboration of the Hazrat Zahra Infertility Center affiliated with the University are acknowledged.

The ethics code of this research was
IR.SKUMS.REC.1395.116.

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