Original Article

A prospective, randomized trial comparing the effects of letrozole versus clomiphene citrate for induction of ovulation and pregnancy rate in women with polycystic ovary syndrome

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Abstract

Objective: To compare the effects of letrozole and clomiphene citrate (CC) for ovulation induction in women with polycystic ovary syndrome (PCOS).

Design: Prospective, randomized, not blinded, controlled trial.

Materials and Methods: This prospective, randomized clinical trial included 127 patients of infertile women with PCOS. The first group comprised 66 patients who received letrozole (2.5–5 mg) daily and the second group 61 patients who received (50–100 mg) CC daily for 5 days starting on day 3 of menses. Both the groups were followed by ultrasound until the dominant follicle reached a diameter ≥18 mm, human chorionic gonadotropin (hCG) 10,000 IU was given, and timed intercourse was advised. The treatment continued for three cycles in both the groups.

Main Outcome Measures: Occurrence of ovulation, endometrial thickness, and pregnancy rate.

Results: The mean age, duration of infertility, body weight, body mass index, and endocrine status in both the groups were similar at baseline. The total number of follicles during stimulation was statistically significantly greater in the letrozole group (6.81 ± 1.0 vs. 6.1 ± 1.5; $P = 0.002$). The number of follicles ≥18 mm was statistically significantly higher in the letrozole group compared with the CC group. There was no statistically significant difference in pretreatment endometrial thickness between the two groups, but endometrial thickness at the time of hCG administration was statistically significantly greater in the letrozole group (9.82 ± 0.7 vs. 8.13 ± 0.56; <0.0001). Ovulation occurred in 25 subjects (37.87%) in the letrozole group and 13 (19.67%) in the CC group, with a statistically significant difference between the two groups ($P = 0.024$). Serum E2 concentrations were statistically significantly lower in the letrozole group ($P = 0.001$).

Conclusion: The effect of letrozole showed a better endometrial response and ovulation rate compared with CC. Letrozole may have a role as a first-line treatment for anovulatory patients with PCOS.

Keywords: Clomiphene citrate, letrozole, PCOS, pregnancy

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INTRODUCTION

It is now well known that polycystic ovary syndrome (PCOS) is among the most common endocrine disorders in women of reproductive age and has a strong genetic component. It is characterized by ovarian dysfunction, and its clinical manifestations may include obesity, increased insulin resistance and compensatory hyperinsulinemia, oligo-/anovulation, and infertility.[1] It has been recognized that PCOS has an extremely heterogeneous clinical picture and is multifactorial in etiology. PCOS may represent the largest underappreciated segment of the female population at risk of infertility. The pathophysiology is complex involving the hypothalamo–pituitary–ovarian axis, ovarian theca cell hyperplasia, hyperinsulinemia, and a multitude of other cytokine and adipocyte-driven factors. The diagnosis of PCOS is based on the Rotterdam criteria for the presence of any two of the following conditions: (i) chronic anovulation, (ii) clinical/biochemical parameters for hyperandrogenism, and (iii) polycystic ovaries on ultrasonography.[2] Infertility is a unique medical condition, in that it is a disorder that often involves a couple, not an individual. An infertile couple is one that has been unable to conceive in one year of unprotected intercourse. It is subdivided into primary and secondary.[3] Primary infertility applies to those who have never conceived while secondary infertility is designated to those who have conceived at some time in the past. Clomiphene citrate (CC) is still the standard drug for inducing or augmenting ovulation. It is not, however, equally successful in all situations. Clomiphene resistance, which refers to the persistence of anovulation after standard CC therapy, occurs in 15–20% of the patients.[4] In addition, CC may have a negative effect on the cervical mucus and endometrium. Treatment with CC is associated with a discrepancy between ovulation and conception rates, and the incidence of miscarriage has been claimed to be higher than in the general population.[4,5] Alternative treatments to CC with insulin-sensitizing drugs such as letrozole have attracted attention. Letrozole is an aromatase inhibitor that has been widely used in women with breast cancer.[6] It works by suppressing estrogen production, and has been used to induce ovulation. But does letrozole add anything to the art of ovulation induction and represent a real alternative to CC?

The aim of the study is to compare between the use of letrozole and CC in women with polycystic ovaries undergoing ovarian stimulation and to evaluate the pregnancy rate (PR) between the two groups.

MATERIALS AND METHODS

This prospective clinical trial was conducted at the infertility center of MAGS Medical & Research Center, Kolkata, India. A written informed consent was obtained from all the participants. One hundred and twenty-seven infertile women were selected from those who were attending the infertility center with primary infertility.

All patients were diagnosed as having anovulation due to PCOS. PCOS were diagnosed when the ultrasonography findings of the ovaries were more than 10 follicles 2–8 mm in diameter scattered either around or through an echo-dense thickened central stroma were present plus one or more of the following: oligomenorrhea, positive progesterone, withdrawal bleeding, hirsutism/acne, obesity, and raised luteinizing hormone (LH)/follicle-stimulating hormone (FSH) ratio more than two or raised circulating androgen. All patients have documented at least one patent fallopian tube by either hysterosalpingogram or laparoscopy, and history of pelvic surgery with tubal blockage was excluded from the study. The male partners had to have a normal seminal analysis by World Health Organization (WHO) (2010) criteria. The patients were examined clinically. Their weight, height, and body mass index (BMI) were estimated. Transvaginal ultrasonography (U/S) examination was performed to exclude any pelvic pathology before treatment.

On the basis of previous studies to achieve a statistically valid comparison of PRs in the two groups, with a type I error of 0.05 and a power of 80%, a sample size of at least 40 women in each arm was required.

The randomization of recruited women was performed using online software (www.randomization.com) to generate a random number table. The patients were randomly divided into two groups. Sixty-six patients received letrozole with a starting dose of 2.5 mg, increasing up to 5 mg daily for 5 days starting from the third day of a menstrual bleeding. Sixty-one patients had received CC with a starting dose of 50 mg, increasing up to 100 mg daily for 5 days beginning on day 3 of the menstrual cycle. All women continued to receive 500 mg metformin t.i.d. Follicular development was monitored using transvaginal U/S from day 10 onward. When at least one mature follicle (with a mean diameter ≥18 mm) was observed, 10,000 IU of human chorionic gonadotropin (hCG) were given subcutaneously to trigger ovulation. The second

transvaginal U/S was performed after 48 h of injection of (hCG) to observe the release of ova. Ovulation was ascertained by observing the rupture of the follicle by transvaginal U/S and elevated serum progesterone level. Each woman was asked to have timed intercourse/intrauterine insemination 24–36 h after the ovulatory dose of hCG. Women in both the groups without the evidence of ovulation and with negative pregnancy tests were asked to follow the respective schedule of treatment in subsequent cycles. Chemical pregnancy was assessed by the serum level of beta-hCG measurement once the patient missed her period. The work was approved by the local ethical committee.

Statistical analysis

The analysis was performed using the Statistical Package for the Social Sciences version 20.0 software (SPSS Inc., Chicago, IL, USA) statistical software. Data were expressed as the mean ± standard deviation (SD). A Student's *t*-test was performed. Proportions were analyzed using the chi-square test. $P < 0.05$ was considered a statistically significant difference.

RESULTS

During the study period, a total of 162 patients were analyzed for recruitment. Twenty-one patients did not meet inclusion criteria and 14 patients were lost to follow-up in between the study, and therefore, 127 patients entered and completed the study. The flow of participants is shown in Figure 1. There were no statistically significant differences between the two groups in age, duration of infertility, body weight, BMI, and endocrine status at baseline level [Table 1]. The total number of follicles during stimulation was statistically significantly greater in the letrozole group (6.81 ± 1.0 vs. 6.1 ± 1.5; $P = 0.002$). The number of follicles ≥18 mm was statistically significantly higher in the letrozole group compared with the CC group. There was no statistically significant difference in pretreatment endometrial thickness between the two groups, but endometrial thickness at the time of hCG administration was statistically significantly greater in the letrozole group (9.82 ± 0.7 vs. 8.13 ± 0.56; <0.0001). Ovulation occurred in 25 subjects (37.87%) in the letrozole group and 13 subjects (19.67%) in the CC group, with a statistically significant difference between the two groups ($P = 0.024$). Serum E2 concentrations were statistically significantly lowered in the letrozole group ($P = 0.001$). There is no significant difference in PR between the two groups [Table 2]. There was no occurrence of ovarian hyperstimulation syndrome in the letrozole group compared with the CC group.

DISCUSSION

The first line of ovulation inducing agent CC is not equally effective in all situations for induction of ovulation or superovulation. Clomiphene resistance occurs in 15–20% of the patients. The use of CC may be associated with poor cervical mucous and endometrial thinning in 15–50% of the patients due to prolonged estrogen-receptor depletion in the endometrium and possibly in the cervix.[7-11] CC, which is the most commonly prescribed medication, initiates ovulation by blocking the negative feedback on endogenous estrogen at the level of hypothalamus–pituitary promoting on the increase in the pulsatile release of LH and FSH in

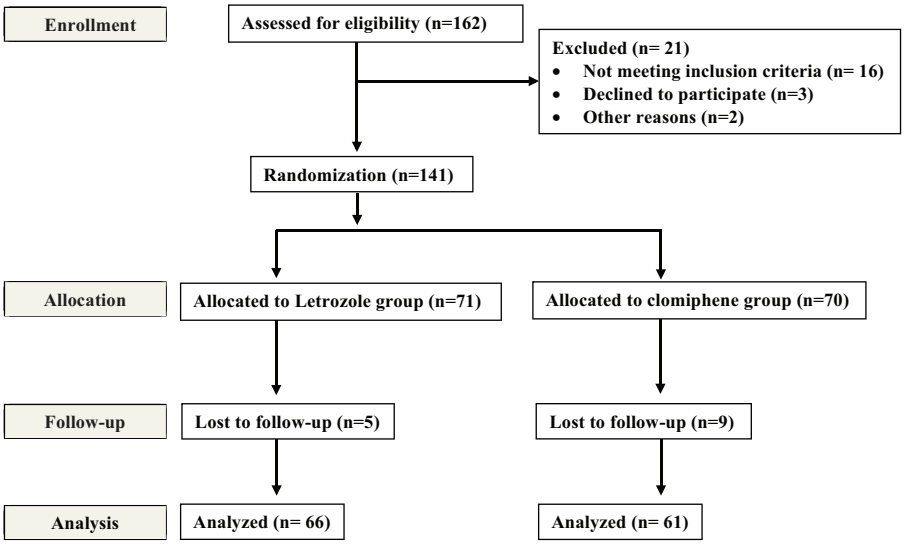

Figure 1: Flow of participants through the study

Table 1: Baseline characteristic of the patients with primary infertility in both the groups

Parameters	Letrozole $N = 66$	CC $N = 61$	P value
Age (years)	27.3 ± 1.9	26.9 ± 2.1	0.261
Duration of mean infertility period (years)	2.1 ± 0.8	2.3 ± 0.6	0.115
Height (cm)	162.03 ± 3.9	163.3 ± 4.2	0.079
Weight (kg)	74.8 ± 2.9	75.1 ± 4.3	0.643
Body mass index (kg/m^2)	27.8 ± 2.7	28.3 ± 2.3	0.265
FSH (IU/mL) on day 2/3 of cycle	7.7 ± 2.7	8.1 ± 2.3	0.372
LH (IU/mL) on day 2/3 of cycle	6.34 ± 1.9	5.97 ± 2.1	0.299
E2 (pg/mL) on day 2/3 of cycle	63.41 ± 1.6	64.12 ± 2.7	0.071
TSH (mIU/L) on day 2/3 of cycle	3.3 ± 1.9	2.92 ± 1.77	0.246
Prolactin (ng/dL) on day 2/3 of cycle	27.39 ± 6.72	26.93 ± 8.1	0.727

Data are expressed as mean ± SD.

Table 2: Response to ovarian stimulation in the patients with letrozole versus clomiphene citrate therapy

Parameters	Letrozole $N = 66$	CC $N = 61$	P value
Total number of follicles	6.81 ± 1.0	6.1 ± 1.5	0.002
Follicular development by day 14 (mm)	23.4 ± 0.7	22.7 ± 1.6	0.001
No. of follicles ≥ 18 mm on day of hCG administration	3.62 ± 0.5	2.8 ± 0.9	<0.0001
Serum E2 on day of hCG (pg/mL)	287.78 ± 75.17	324.7 ± 52.9	0.001
Pretreatment endometrial thickness (mm)	4.2 ± 0.7	4.5 ± 1.1	0.067
Endometrial thickness (mm) at day 14	9.82 ± 0.7	8.13 ± 0.56	<0.0001
Serum progesterone (ng/mL)	10.13 ± 1.01	9.17 ± 0.92	<0.0001
Ovulation	25 (37.87%)	13 (19.67%)	0.024
Pregnancy	6 (24%)	2 (15.38%)	0.541

Data are expressed as mean ± SD.

anovulatory patients with POCS.[12] For many years, the first treatment of choice for ovulation induction in POCS was CC,[13] but up to 58% of such patients are resistant to it and do not ovulate.[14] The PR per cycle remains relatively low. It has also been demonstrated that CC has an antagonistic effect on the endometrium and may reduce endometrial thickness.[15] The inappropriate development of endometrium is associated with a low implantation rate and early pregnancy loss caused by luteal phase defect.[16] Some patients (20–28%) do not respond to CC in spite of high dose, because the antiestrogenic effect is dose-dependent, and a daily dose of CC >150 mg is not recommended.[4]

Letrozole (4,40-[1H-1,2,4-triazol-1-ylmethylene]-bis-benzo nitrile) is a type IIa third-generation aromatase inhibitor. It was postulated that blocking estrogen production by inhibiting aromatization, the conversion of andro-stenedione and testosterone to estrogen in the ovary would release the hypothalamic/pituitary axis from estrogenic negative feedback. As a result, the FSH secretion increases, stimulating the development of ovarian follicles. Preliminary studies reported that aromatase inhibitors were useful for inducing ovulation and in superovulation. Mitwally and Casper[14] described the use of 2.5 mg of letrozole on days 3–7 of menses in 12 patients with PCOS. Ovulation occurred in nine patients (75%), and pregnancy was achieved in three (25%). Sammour *et al.*[17] found in their double-blind randomized trial comparing the use of an aromatase inhibitor with CC for stimulation in 49 women with infertility that increased endometrial thickness compared with those receiving CC, because threefold increase in the PR was observed in the patients who received aromatase inhibitor compared with CC treatment (16.7% vs. 5.6%, respectively).

Compared with CC, the use of letrozole led to a statistically significant increase in the number of developing and mature follicles (18 mm follicles). Al-Fozan *et al.*[18] reported better results in the letrozole group for the number of developing and mature follicles than in the CC group. The endometrium was, astoundingly, statistically significantly thicker in the letrozole group. Information on the teratogenic capacity of letrozole in humans is lacking, but animal studies have shown that the low doses of letrozole are effective in inducing noxious effects on the developing conceptus. CC has been reported to elicit various ocular side effects in 1.5–10.0% of the patients taking CC.[19] Mitwally *et al.*[20] reported favorable pregnancy outcomes and low multiple gestation rates with aromatase inhibitors for ovarian stimulation. Bedaiwy *et al.*[21] have suggested that a letrozole-FSH combination could be more cost-effective than FSH alone for ovarian stimulation in intrauterine insemination cycles.

CONCLUSION

Although there are some limitations to the present study, our study found advantages to using letrozole rather than

CC as a first-line treatment for inducing ovulation in women with PCOS. The strength of our study is that we have been able to preliminarily explore the effects of letrozole and CC for ovulation induction in women with PCOS.

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Conflicts of interest

There are no conflicts of interest.

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