Provisionally accepted for publication

ORIGINAL ARTICLE

Comparative efficacy of letrozole versus letrozole-metformin in the ovulation and clinical pregnancy rate in polycystic ovary syndrome: a prospective randomized controlled study

Efficacy of letrozole versus letrozole-metformin in ovulation induction in PCOS

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DOI: 10.36129/jog.2026.261

ABSTRACT

Objectives. The study provides an insight on the effect of letrozole and letrozole-metformin combination on the formation of mature follicle, ovulation, endometrial thickness and pregnancy rates in anovulatory PCOS infertile women.

Materials and Methods. A prospective randomised controlled study conducted at Ridge IVF centre, Delhi from May 2024- April 2025.120 women of ages 21-40 years affected by PCOS and trying naturally for conception were included. Patients were randomly selected into 2 groups: Group A -letrozole and metformin and Group B- letrozole only.

Metformin 500mg twice daily was added from day 1 of cycle. Letrozole 2.5mg BD was started from day 2/3 of menstrual cycle. Follicular monitoring was done from day 10 by transvaginal sonography and once a mature follicle of size >18mm is seen on TVS, inj. HCG 10,000 IU was given. Once ovulation was documented, couple was advised for alternate day intercourse and luteal phase support was given for 14 days. Pregnancy confirmation was done by S. ß hcg after 14 days.

Results. The study showed that the formation of mature follicle was more in letrozole-metformin group and ovulation rate was higher in letrozole only group. The endometrial thickness on the day of the trigger was comparable in both groups. The clinical pregnancy was slightly higher in combination group, but not statistically significant.

Conclusions. Adding metformin to letrozole does insignificant improvement in ovulation patterns in PCOS anovulatory women.

Key words

PCOS; letrozole; metformin; ovulation

INTRODUCTION

Polycystic ovary syndrome (PCOS) is the most common endocrinological disorder affecting 4–12% of women [1-2]. It has been the most controversial medical condition affecting women of reproductive age group. It is one of the causes for anovulatory female infertility. PCOS is a heterogeneous disorder with a combination of genetic, reproductive and metabolic characteristics. These women are at risk of Type 2 DM and cardiovascular disease.

According to 2003 Rotterdam Consensus revised diagnostic criteria, at least two of the following is required for the diagnosis of PCOS:

1. oligo or anovulation, or both, that is, menstrual disturbance

2. clinical or biochemical signs, or both, of hyperandrogenism

3. PCO on ultrasound and exclusion of other aetiologies of menstrual disturbance and hyperandrogenism (such as congenital adrenal hyperplasia, androgen-secreting tumours, Cushing's syndrome) [3]. A recent update to guidelines in view of advancement in ultrasound technology and resolution, state that the diagnostic criteria for ultrasound PCO morphology is either 20 or more follicles per ovary or increased ovarian volume, over 10 mL, when using a transvaginal ultrasound scan [ESHRE 2023] [4].

Insulin resistance is a very common feature of PCOS, owing to either genetic propensity or obesity [5]. It is not a diagnostic feature of this disorder. In PCOS, hyperinsulinemia can induce

androgen production and decrease the levels of sex hormone binding globulin. A high level of androgen levels affects the oocytes growth and causes atresia of follicles. PCOS is responsible for about 80% of anovulatory infertility [6]. Women with insulin resistance have increased prevalence of hirsutism than women with non-insulin-resistant PCOS [7]. These women are more likely to develop resistance to ovulation induction treatment.

Lifestyle modification including weight loss and exercise reduces central fat, improves insulin sensitivity, reduces testosterone levels and restoring ovulation in overweight, infertile women with PCOS [7].

Metformin is the member of the biguanide family that has been used for the treatment of T2DM. Metformin works by improving the insulin sensitivity of peripheral tissues [8], which results in a reduction of circulating insulin levels. The main side effects associated with metformin treatment are the gastrointestinal symptoms of nausea, diarrhoea, flatulence, bloating, anorexia, metallic taste and abdominal pain, causing reluctance in its use over a prolong period.

Within the ovary, metformin has a direct impact on cells to reduce excessive steroidogenesis and folliculogenesis [9]. It reduces theca cell proliferation, decreases androgen production, reduces pituitary luteinizing hormone and increase sex hormone binding globulin by the liver and promotes ovulation.

Letrozole is a nonsteroidal reversible and competitive aromatase inhibitor. Letrozole has a half-life of about 45 hours [10]. It has a central effect on the pituitary–hypothalamic axis by releasing it from estrogen negative feedback and a local ovarian effect by blocking androgen conversion to estrogen, with the concomitant accumulation of androgens inside the ovary, augmenting the follicular FSH receptor expression, and promoting folliculogenesis [11].

This study provides an insight on the effect of letrozole and letrozole-metformin combination on anovulatory PCOS infertile women in ovulation and pregnancy.

MATERIALS AND METHODS

Study site

The study was a prospective randomised controlled study conducted in Department of Assisted Reproduction at Ridge IVF centre, New Delhi from May 2024- April 2025. The approval was taken from the ethics committee on board for the study.

Inclusion criteria

Women of all ages (21-40 years) who were affected by PCOS according to Rotterdam criteria with normal hysterosalpingogram and were trying naturally for conception were included.

Exclusion criteria

Patients with hormonal disorder like hyperprolactinemia, hypo or hyperthyroidism or diabetes, endometriosis, uterine fibroid, male factor infertility were excluded from study.

All patients were admitted through OPD after complete evaluation with history, examination, investigation and verbal informed consent for inclusion in the study. Blood for Insulin Postprandial is sent 1 ½ hours after meals on first visit along with other investigations.

Patients were selected randomly into 2 groups: Group A -letrozole and metformin and Group B- letrozole only. Letrozole and metformin was administered as oral preparations.

In Group A tablet metformin 500mg twice daily with meals starting from day 1 of cycle was added and continued for three months and continued even after conception till first 3 months of pregnancy. Patients were followed for 3 months to evaluate for pregnancy in both the groups. Oral ovulogen letrozole 2.5mg BD was started from day 2/3 of menstrual cycle after spontaneous or withdrawal bleeding in both groups. Follicular monitoring was done from day 10 and follicle growth was monitored by transvaginal sonography (TVS). Once a mature follicle of size >18mm was seen, inj. HCG 10,000 IU was given as a trigger. Once follicular rupture was documented, couple was advised for alternate day intercourse and luteal phase

support was given with dydrogesterone 10mg BD for 14 days. Pregnancy confirmation was done by estimating the S. ß hcg concentration in the blood after 14 days and doubling of the S.ßhcg after 48 hours. The clinical pregnancy was established by the presence of gestational sac by ultrasonography performed at 6-7 weeks of pregnancy.

Treatment was repeated for 3 months if the patient fails to ovulate or achieve pregnancy.

The primary outcomes were assessed as documentation of mature follicle (>18mm) and documentation of ovulation (ovulation rate)

The secondary outcomes were assessed as measurement of ovarian volume, Hormonal profile on day 2 (S.FSH, S. LH, S.E2), Endometrial thickness on day of HCG trigger and Clinical Pregnancy rate.

Statistical analysis

The collected data was entered in Microsoft Excel and then analysed and statistically evaluated using SPSS-25 version. Normality of each variable was assessed by using the Kolmogorov- Smirnov test. Quantitative data was expressed by mean, standard deviation and difference between means of two group were tested by Unpaired t test or Mann Whitney U test. Qualitative data were expressed in percentage and difference between percentage of two group were tested by chi square test or Fisher exact test. 'P' value less than 0.05 was considered statistically significant.

RESULTS

The baseline characteristics of participants in both groups were largely comparable (Table 1). The mean age was similar in Group A (30.13 ± 3.21 years) and Group B (30.22 ± 3.25 years). The mean BMI was slightly higher in Group A (27.68 ± 4.78 kg/m²) compared to Group B (26.22 ± 4.12 kg/m²), though this difference was not statistically significant (P=0.07). In terms of type of infertility, primary infertility was predominant in both groups—76.7% in Group A and 80.0% in Group B.

However, a significant difference was observed in menstrual cycle regularity. 56.7% of Group A had irregular cycles compared to only 18.3% in Group B and regular cycles were more common in Group B with statistical difference <0.001(Table 1).

In Table 2, comparison of hormonal and ultrasonographic parameters were done. Right ovarian volume was significantly higher in Group A (9.83 ± 1.48 ml) compared to Group B (6.76 ± 1.94 ml), and similarly, the left ovarian volume was also greater in Group A (9.24 ± 1.07 ml vs. 6.86 ± 2.01 ml) with statistically significant (P < 0.001) (FIG 1).

Serum AMH levels were high in Group A (8.12 ± 3.42 ng/ml) as compared to Group B (6.13 ± 2.44 ng/ml). Baseline Serum FSH was marginally higher in Group A (6.50 ± 1.29) than in Group B (6.02 ± 1.61), with borderline significance (P = 0.05). Serum LH levels were significantly higher in Group A (7.40 ± 3.19) compared to Group B (6.31 ± 4.17) (P = 0.01) (FIG 2) and serum estradiol (E2) levels were also higher in Group A (43.27 ± 17.42 vs. 36.65 ± 13.07).

A striking difference was observed in postprandial insulin levels, which were markedly elevated in Group A (67.98 ± 27.23 µU/ml) compared to Group B (26.92 ± 15.62 µU/ml), showing a highly significant difference (P < 0.001) (FIG 3).

Endometrial thickness on the trigger day was comparable between both groups (8.83 ± 1.65 mm in Group A vs. 8.80 ± 1.21 mm in Group B (FIG 4).

The comparison of clinical hirsutism between Group A and Group B in table 3 showed a higher proportion of individuals in Group A (41.7%) experienced clinical hirsutism, compared to just 16.7% in Group B.

In Table 4, comparison of clinical outcomes to ovulation induction was compared. A higher proportion of individuals in Group A (88.3%) had mature follicles compared to Group B (100%). Still, it was found that Group B (93.3%) achieved higher ovulation compared to Group A (70%), with a **P value of 0.002**. In terms of pregnancy, Group A had a higher percentage of positive S.βHCG results (40.0%) compared to Group B (25.0%) and higher clinical pregnancy rate (40%vs 23.3%), not statistically significant.

DISCUSSION

PCOS is the most prevalent endocrine disorder in the reproductive age of women. In these patients, medications such as clomiphene citrate, letrozole and metformin are used to induce ovulation and overcome infertility [12].

Clomiphene citrate binds to estrogen receptors for prolonged periods i.e. weeks (2 weeks) rather than hours as with natural estrogen. This extended binding ultimately depletes estrogen receptors' concentrations by interfering with the normal process estrogen receptors' replenishment and may be responsible for the peripheral antiestrogenic effect of clomiphene citrate on the endometrium and cervix [13].

Compared to CC, letrozole is cleared from circulation more rapidly due to shorter half-life; associated with monofollicular growth and thicker endometrium. Letrozole is devoid of any antiestrogenic peripheral actions and has no impact on the endometrial receptivity and cervical mucous quality [14].

Metformin, an insulin sensitizer may be used alone or in concert with other medications such as clomiphene citrate, letrozole it has been shown to increase the ovulatory response to clomiphene citrate in patients who were previously clomiphene resistant [15].

In our study, demographic parameters i.e. age, BMI, type of infertility was comparable in both groups. The only significant demographic character was menstrual cycle regularity. As women were selected randomly in the study, cycles were more irregular in letrozole-metformin group, but cycle regularity is not the sole criterion for metformin use [4]. A study by Hamada et al also has reported similar age, BMI and duration of infertility distribution in both groups [16].

In Group A, patients had higher insulin PP, day 2 S.FSH, S. LH, S.E2 and S.AMH levels. The clinical hirsutism was also higher in metformin-letrozole group.

When ovulation parameters were compared it was found that group A participants had higher achievement of formation of mature follicle than Group B, but ovulation rate was higher in group B. A study by Hamada et al found no significant differences between the letrozole and the metformin-letrozole groups regarding ovulation rate, number of patients with growing follicle, number of follicles ≥ 18 at day 12, number of ruptured follicles after 48 hours of injection of HCG, serum E2, Parity, clinical presentation, Period of infertility, FSH, LH [16].

In our study, clinical pregnancy rate was slightly higher in Group A, but not statistically significant. A retrospective study on 268 women undergoing ovulation induction/IUI in 2012-2016 has found that the addition of metformin to Letrozole does not improve follicular recruitment or pregnancy rates in OI/IUI cycles in infertile PCOS women [17].

Liu et al revealed a pregnancy rate of 57.9% in letrozole in addition to metformin gathering and just 46.8% in patients who got letrozole alone [18].

In our study the endometrial thickness was comparable in both groups (8.83 vs 8.80). A study by Ahmad et al found superior endometrial thickness with letrozole than clomiphene citrate showing CC antiestrogenic effects [19]. Hamada et al found a significant improvement in endometrial thickness in metformin-letrozole group [16]. On other hand, EL-Gharib et al found nonsignificant change between endometrial thickness in both letrozole and letrozole metformin groups, like our study results [20].

The risk of Gestational diabetes in pregnancy is high in PCOS especially androgenic PCOS. The hormonal and metabolic profile of hyperandrogenic PCOS patients includes insulin resistance, and an elevated androgen levels both of which are risk factors for the development of GDM. Women with PCOS are at further risk of obstetric complications including hypertensive disorders, preterm birth, induction of labor and cesarean delivery [21]. Similarly in IVF conceived pregnancies risk of GDM is high as many females have significant risk factors for GDM, such as advanced maternal age, obesity, multiple pregnancies and

polycystic ovary syndrome (PCOS), suggesting a potential relationship between GDM and ART [22].

Limitation

The study involved a smaller number of participants. In clinical situations, individual treatment dosages varied, and it was not fully considered in our study. The study's limited duration prevented a thorough examination of longer-term outcomes.

Conclusions

Polycystic Ovary Syndrome (PCOS) is a complex disorder that requires a comprehensive approach to management, addressing both reproductive and metabolic concerns. While multiple treatment options are available for PCOS, a combination of lifestyle changes, medication, and, in some cases, surgical interventions, may be necessary to address the diverse manifestations of the disorder. Only letrozole 2.5mg BD given from day 2 for ovulation induction and performing follicular monitoring is the standard protocol in PCOS from our research as adding metformin to letrozole does insignificant improvement in ovulation patterns in PCOS anovulatory women. The ovulation rate was slightly higher in letrozole group. Similarly, pregnancy rates were also insignificant in both groups.

COMPLIANCE WITH ETHICAL STANDARDS

Authors contribution

SC: Conceptualization, manuscript writing, editing, RH: manuscript review and editing, MGD: critical inputs into the manuscript.

Funding

NONE

Disclosure of interest

None

Study registration

N/A

Ethical approval

This study was approved by the Ethics Committee of Ridge IVF, Gouri Hospital (No. RIDGE/HEC/001/2025 date: 29/4/2024).

Informed consent

Informed consent was obtained from all participants.

Data sharing

Data are available under reasonable request to the corresponding author.

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Table 1: Data comparing the baseline characteristics between Group A and Group B

	Group A (letrozole-metformin)(n=60)	Group B (letrozole) (n=60)	P value
Mean age in years	30.13±3.21	30.22±3.25	0.88
Mean BMI (kg/m^2)	27.68±4.78	26.22±4.12	0.07
Type of infertility			
PRIMARY	46(76.7%)	48(80.0%)	0.65
SECONDARY	14(23.3%)	12(20.0%)	
Cycle			
IRREGULAR	34 (56.7%)	11 (18.3%)	<0.001
REGULAR	26 (43.3%)	49 (81.7%)	<0.001

Table 2: Data comparing the other parameters between Group A and Group B

	Group A (letrozole-metformin)(n=60)	Group B (letrozole)(n=60)	P value
RT OVARY VOL (ml)	9.83±1.48	6.76±1.94	**<0.001**
LT OVARY VOL (ml)	9.24±1.07	6.86±2.01	**<0.001**
S.AMH (ng/ml)	8.12±3.42	6.13±2.44	**0.001**
S.FSH	6.50±1.29	6.02±1.61	0.05
S.LH	7.40±3.19	6.31±4.17	**0.01**
S.E2	43.27±17.42	36.65±13.07	**0.02**
Insulin PP (µU/ml)	67.98±27.23	26.92±15.62	**<0.001**
Endometrial thickness (mm)	8.83±1.65	8.80±1.21	0.92

Table 3: Data comparing clinical hirsutism between Group A and Group B

CLINICAL HIRSUITISM	Group A (letrozole-metformin) (n=60)	Group B(letrozole) (n=60)	P value
ABSENT	35(58.3%)	50(83.3%)	**<0.01**
PRESENT	25(41.7%)	10(16.7%)	

Table 4: Data comparing different outcome between Group A and Group B

	Group A(letrozole-metformin) (n=60)	Group B (letrozole)(n=60)	P value
Mature follicle			
NO	7(11.7%)	0	**0.01**
YES	53(88.3%)	60	
Ovulation			
NO	18(30.0%)	4(6.7%)	**0.002**
YES	42(70.0%)	56(93.3%)	
S.βHCG			
NEGATIVE	36(60.0%)	45(75.0%)	0.07
POSITIVE	24(40.0%)	15(25.0%)	
Clinical pregnancy			
NO	36(60.0%)	46(76.7%)	0.07
YES	24(40.0%)	14(23.3%)	

FIG1: OVARIAN VOLUME OF RIGHT AND LEFT OVARY

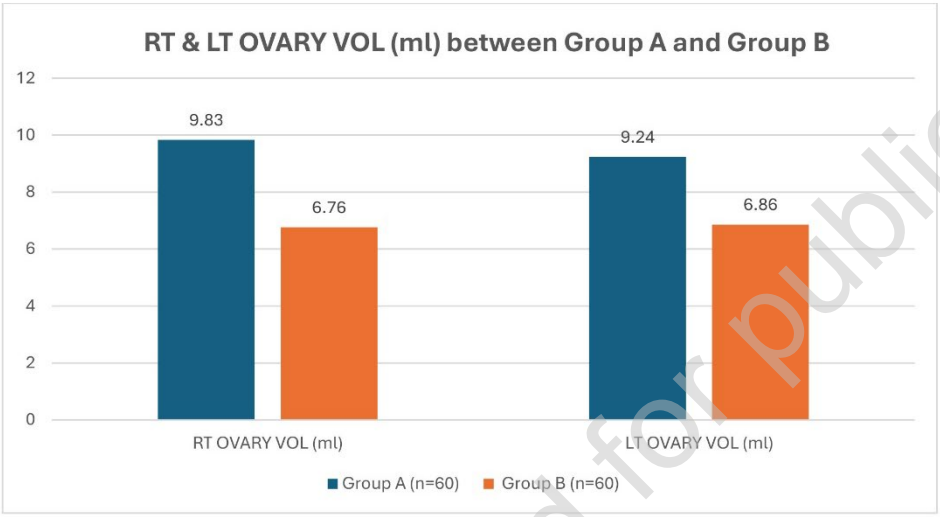

FIG 2: SERUM LH ON DAY 2 OF CYCLE

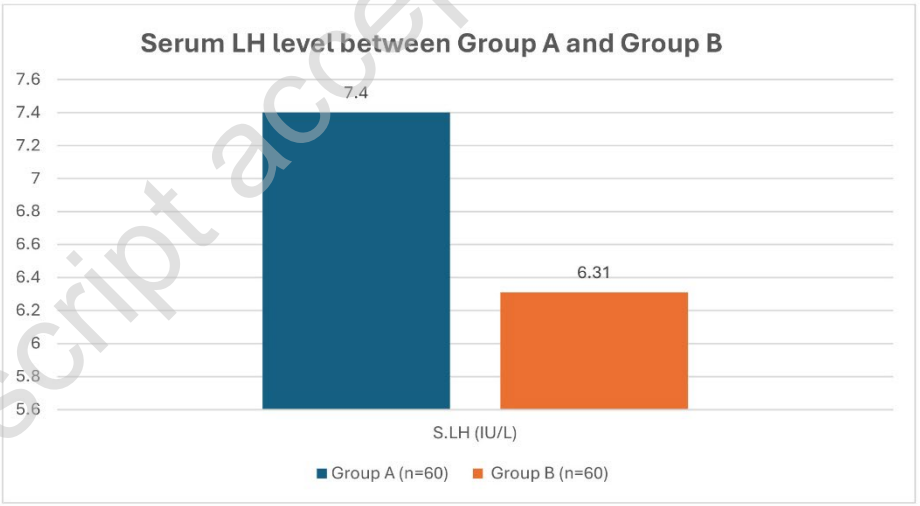

FIG 3: INSULIN POST PRANDIAL

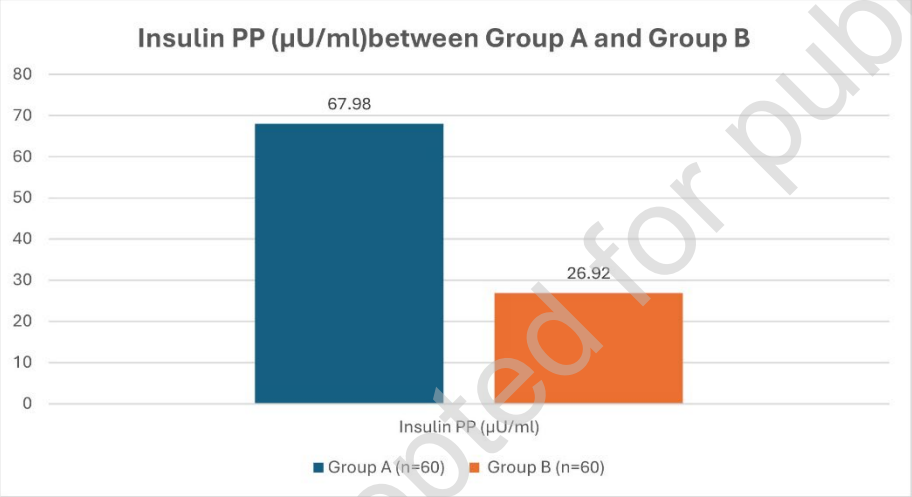

FIG. 4: ENDOMETRIAL THICKNESS ON THE TRIGGER DAY