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Letrozole in Treating Polycystic Ovary Syndrome-Associated Infertility

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Abstract

Letrozole was originally reported for the treatment of breast malignancy, however, it exhibited potential in treating infertility disorders linked with polycystic ovary syndrome (PCOS). The purpose of this research was to investigate the usefulness of letrozole in treating infertile females with PCOS. A randomized, prospective, clinical trial involving 105 females aged between 19 and 33 was performed. Over a couple of months, evaluations were completed on days two and twelve of the females who received letrozole tablets. Substantial statistical ovulatory increment, more endometrial thickness, and altered hormone levels were identified. These changes signify letrozole's potential as a useful choice for PCOS-associated infertility. To fully comprehend its therapeutic potential and optimize treatment regimens, additional future study is required.

الليتروزول في علاج العقم المرتبط بمتلازمة تكيس المبايض

رحاب كاظم، امال عمران موسى، حميدة هادي عبد الواحد

الخلاصة

تم الإبلاغ في الأصل عن الليتروزول لعلاج الأورام الخبيثة في الثدي، ومع ذلك، فقد أظهر إمكانات في علاج اضطرابات العقم المرتبطة بمتلازمة تكيس المبايض. كان الغرض من هذا البحث هو التحقيق في فائدة الليتروزول في علاج الإناث العقيمات المصابات بمتلازمة تكيس المبايض. تم إجراء تجربة سريرية عشوائية مستقبلية شملت 105 أنثى تتراوح أعمارهن بين 19 و33 عامًا. على مدار شهرين، تم الانتهاء من التقييمات في اليوم الثاني والثاني عشر للإناث المتقدمات اللاتي تلقين أقراص الليتروزول ومواضيع التحكم الصحية. تم تحديد زيادة كبيرة في التبويض الإحصائي، وزيادة سمك بطانة الرحم، ومستويات هرمون متغيرة. تشير هذه التغييرات إلى إمكانات الليتروزول كخيار مفيد للعقم المرتبط بمتلازمة تكيس المبايض. لفهم إمكاناته العلاجية بشكل كامل وتحسين أنظمة العلاج، هناك حاجة إلى دراسة مستقبلية إضافية.

1. Introduction

Midway through 1997, letrozole was initially registered for the treatment of breast cancer in France (Bhatnagar, 2007). Given that letrozole medicine is used to overcome PCOS-related infertility, its multifaceted role becomes even more imperative (Chen et al., 2024a). The principal form of estrogen, estradiol, is produced by the conversion of androgenic hormone synthesized by the ovaries (Eskew et al., 2019). Letrozole is a non-steroidal aromatase inhibitor that is rapidly and completely absorbed from the gastrointestinal tract, with its absorption unaffected by food intake (Rashdan et al., 2024). The drug has a large volume of distribution, approximately 1.9 L/kg, and is about 60% bound to plasma proteins, primarily albumin (Rashdan et al., 2024). Letrozole's terminal elimination half-life is approximately 42 hours, allowing for steady-state concentrations to be reached within 2 to 6 weeks of daily administration (Desai et al., 2024). The metabolism of letrozole primarily occurs through cytochrome P450 enzymes, particularly CYP3A4 and CYP2A6, resulting in the formation of an inactive carbinol metabolite (Desai et al., 2024, Rashdan et al., 2024). Approximately 90% of the administered dose is recovered in urine, with around 75% as the glucuronide conjugate of the carbinol metabolite (Kivrak et al., 2024). There are often substantial hormonal alterations in PCOS. By inhibiting this conversion, letrozole increases androgen levels in the blood. Growing follicles may increase almost as a result, possibly enhancing the quality of the discharged follicles (Chen et al., 2024a). A 2022 study by Gowri Vi. et al. confirm this idea more strongly by showing that the administration of letrozole is favorably connected with increased mean follicle size in PCOS individuals undergoing in-vitro fertilization (IVF) (Gowri et al., 2022). Letrozole has effects that go beyond the follicle quality. The imbalance between estrogen and androgens in females with PCOS can result in a thinning endometrium, leaving the uterus less conducive to implantation. Letrozole may trigger an increase in endometrial thickness by boosting the synthesis of growth-promoting factors through elevated testosterone ratios (Pritts et al., 2011). All these kinds of hormones work together to create a denser endometrial lining, which can increase the implantation process. Consistent with a recent study, letrozole administration might boost endometrial growth among ladies with PCOS, indicating it can be an appropriate substitute for cases suffering from thin endometrium (Alhibshi et al., 2021). The most prominent action of letrozole in PCOS cases is its physiological activity to trigger ovulation. In females, ovulation remains on hold by the ordinary hormonal path-ways hindered by the excess estradiol production in PCOS. This antagonistic feedback loop is suppressed by letrozole usage, which prevents the peripheral estradiol conversion (Chen et al., 2024a). Owing to the continuous hypophysial secretion of FSH, further released eggs become accessible for spermatic fertilization by promoting the evolution of plentiful follicles and over-ovulation (Chen et al., 2024a). Accordingly, letrozole treatment has been confirmed to raise PCOS-afflicted female's prospects of becoming pregnant (Yang et al., 2021a). Compared to clomiphene citrate letrozole is cheap, easier to acquire, and has minor adverse reactions, therefore it is seen as a valuable substitute for other medications (Yang et al., 2021b). (Reed BG). Letrozole lowers the odds of multiple pregnancies by promoting mono-follicular development, which results in singleton pregnancies. Furthermore, letrozole has demonstrated efficacy in increasing sperm qualities in infertile with low blood levels of testosterone and estrogen, which may improve fertility in men with oligospermia (AlJuboory et al., 2020). The wide range of manifestations that women with PCOS experience accounts for a large portion of the uncertainty surrounding the diagnosis of PCOS. Considering the large percentage of females who are suffering from PCOS and the substantial impact it has on patients, it is important to gain a greater awareness of the current burden in the vicinity of the Middle East (Liu et al., 2021). The majority of research investigations on PCOS have been carried out in developed countries, with limited data

available on the magnitude of the problem in developing countries, such as Iraq (Motlagh Asghari et al., 2022). A previous study estimated that a third of reproductive-aged females in Iraq have PCOS (Reed BG). Infertility is defined generally as the inability to conceive after one year or longer of unprotected sex. This condition can affect both men and women and may result from various factors affecting the process of conception, such as problems with ovulation, sperm quality, or the reproductive organs (Adnan A. H. Al-Bdairi, 2021, Adnan A. H. Al-Bdairi 2023). Infertility is a common issue, with about 1 in 5 women aged 15 to 49 in the United States being unable to have conception after one year of trying (Adnan A. H. Al-Bdairi 2022). Hence the current study aimed to fill the gap in the available data and to investigate the precise role of letrozole in Iraqi infertile females with PCOS.

2. Patients and Methods

2.1. Study Design

This was a prospective, single-center, randomized, controlled pragmatic clinical trial conducted at the Teba Center for Infertility and In-Vitro Fertilization, Babylon-Iraq, from September 2023 to February 2024.

2.2. Selection of Participants

A total of 105 female patients participated in the study, with an average age of 27.1 ± 4.9 years.

Inclusion Criteria: Females with a history of infertility (primary or secondary) and PCOS, were diagnosed at the Teba Center based on the modified Rotterdam criteria with an age range of 19-33 years old. The modified Rotterdam criteria for diagnosing Polycystic Ovary Syndrome (PCOS) were established to provide a standardized approach for clinicians. These criteria include the following components:

- Oligo- or Anovulation: This refers to irregular menstrual cycles or the absence of ovulation, which can lead to infertility.
- Clinical and/or Biochemical Signs of Hyperandrogenism: This includes symptoms such as hirsutism (excess hair growth), acne, and alopecia (hair loss), as well as elevated levels of androgens (male hormones) in the blood.
- Polycystic Ovaries: This is determined through ultrasound imaging, which reveals the presence of multiple small follicles (typically 12 or more) in one or both ovaries, often accompanied by an increased ovarian volume (Christ and Cedars, 2023).

To meet the diagnosis of PCOS using the modified Rotterdam criteria, a woman must present with at least two of the three components listed above. These criteria help in identifying PCOS effectively while considering its diverse manifestations.

Exclusion Criteria: any cases of tubal blockage or refusal to participate.

2.3. Data Collection

Baseline Assessment: On the second day of the menstrual cycle, the following data were collected for each participant: demographic information, clinical history, medical history, and ultrasound examinations by two separate specialist sonographers to assess egg size and endometrial thickness. Hormonal assessments for LH, AMH, and prolactin using Electro-Chemiluminescence Immunoassay (ECLIA) kits from Mindray® Medical International Limited, China. AMH assays (performed only on the second day of the cycle).

2.4. Intervention:

All the participants were randomly selected to receive letrozole treatment. Letrozole was administered as 2.5 mg tablets (Femara®, NOVARTIS®, Basel, Switzerland) once daily after a meal for five consecutive days, starting from the second day of the menstrual cycle, for two consecutive cycles (Reed BG).

2.5. Follow-up Assessment:

After two months of letrozole administration, on the 12th day of the cycle, the following assessments were repeated: ultrasound examination, and hormonal tests for LH and prolactin.

2.6. Sample Size Calculation Method

The sample size for this study was calculated using a power analysis method to ensure adequate statistical power for detecting differences in outcomes related to CYP3A4*18 genetic variations. The following steps were employed:

1. Effect Size: A medium effect size (Cohen's d = 0.5) was assumed based on previous studies.
2. Significance Level: A significance level (alpha) of 0.05 was established.
3. Desired Power: The power was set at 80% (0.8), indicating a 20% chance of Type II error.
4. Statistical Test: The Kruskal-Wallis test was selected for comparing multiple groups.
5. Sample Size Formula: The formula used for sample size estimation was: $n = ((Z\alpha/2 + Z\beta)^2 \times k) / d^2$, (Here, n is the sample size per group, $Z\alpha/2$ is the Z-value for the significance level, $Z\beta$ is the Z-value for power, k is the number of groups, and d is the effect size).
6. Calculation: For three genotype groups (TT, TC, CC), the calculation yielded a minimum requirement of approximately 95 participants.

2.7. Statistical Analysis

Statistical studies were completed using SPSS (V-27) and JASP (V- 0.18.3.0). Continuous variables were resented as mean and standard deviation (SD). *The* categorical variables were examined using Chi-square tests to assess associations. The normality of the data was tested and the data were normally distributed. Comparison of means was completed using independent samples t-tests to compare means between two study groups, and ANOVA was utilized to compare means between more than two groups. Pearson correlation coefficients were used to assess relationships between continuous data. A significance level below 0.05 was considered statistically significant.

3. Results

Important details on the clinical and demographic traits of the individuals under study are provided in Table1. An improved understanding of the age distribution within the population under study is made possible by the table's additional division of the age distribution into three categories: < 20 years (16%), 20 – 29 years (55%), and > 30 years (29%). The average age was 27.11 (4.9) years, and in the group over 30, the mean BMI rises with age. The patients' mean ± SD BMI and marriage length are 29.7±4.9 and 6.0±3.5 years, respectively. The patients in the study had 0.8 children and 0.6 miscarriages. The ultrasound measures on U/S Day 2 and the endometrial thickness on Days 2 and 12 are displayed in the table. The table also includes the amounts of other hormones, such as LH, Prolactin, and AMH.

Table 1: Basal Demographic Features of the Studied Patients

Variables	Mean	SD
Age	27.1	4.9
< 20 y	18.8	0.7
20 – 29 y	24.9	2.1
> 30 y	33.1	0.9
Marriage span/years	7.1	0.9
BMI kg/m^2	28.9	2.7
No. of miscarriage	0.7	0.9
No. of children	0.8	0.8
Endometrial thickness Day 2	3.9	5.1
Follicular size on U/S Day 2	3.9	1.2
Endometrial thickness Day 12	7.7	0.8
Follicular size on U/S Day 12	18.1	3.9
LH (mlU/mL)	8.8	3.3
Prolactin (ng/mL)	16.1	9.7
AMH (ng/mL)	4.4	1.9

Table2 displays the variations in important study variables among the age groups of the patients. Remarkably, the p-values for the majority of the variables are in proximity to the traditional significance threshold ($p < 0.05$), suggesting possible correlations between age groups and these factors. Nonetheless, p-values for a few variables are somewhat higher than this cutoff. Age-related increases in BMI seem to be modest, however they are not statistically significant ($p = 0.071$). The duration of marriage varies significantly ($p = 0.001$) among age groups, with older age groups showing longer marriages. There are substantial age-group variations in both the number of children and miscarriages ($p = 0.002$ and $p = 0.062$, respectively), with older age groups showing higher mean values. Follicular size and endometrial thickness vary slightly between age groups, but not to a statistically significant degree ($p > 0.05$).

Table2: Differences in the Study Variables According to the Studied Age Groups

Variables	Age classes/y	Mean± SD	P-value
BMI	< 20	29.4 ±5.1	0.073
	20 – 29	29.1 ±4.9	
	> 30	30.8 ±5.1	
Marriage span	< 20	2.9 ±3.2	0.001
	20 – 29	6.1 ±3.3	
	> 30	7.6 ±4.1	
No. of children	< 20	0.3 ±0.5	0.002
	20 – 29	0.8 ±0.6	
	> 30	1.2 ±0.9	
No. of miscarriage	< 20	0.4 ±0.8	0.068
	20 – 29	0.5 ±1.1	
	> 30	0.9 ±1.2	
Follicular size on U/S Day 2	< 20 y	4.0 ±1.3	0.92
	20 – 29 y	4.0 ±1.1	
	> 30 y	3.9 ±1.2	
Follicular size on U/S Day 12	< 20 y	18.8 ±4.1	0.91
	20 – 29 y	17.7 ±4.4	
	> 30 y	18.2 ±3.8	
Endometrial thickness Day 2	< 20 y	3.9 ± 0.7	0.079
	20 – 29 y	7.9 ± 0.8	
	> 30 y	3.7 ± 0.7	
Endometrial thickness Day 12	< 20 y	7.8 ±0.6	0.079
	20 – 29 y	7.8 ±0.3	
	> 30 y	7.7 ±0.8	

The ultrasonography results for endometrial thickness and egg size on days 2 and 12 are shown in Fig.1, which showed a highly significant increase on day 12 of the menstrual cycle

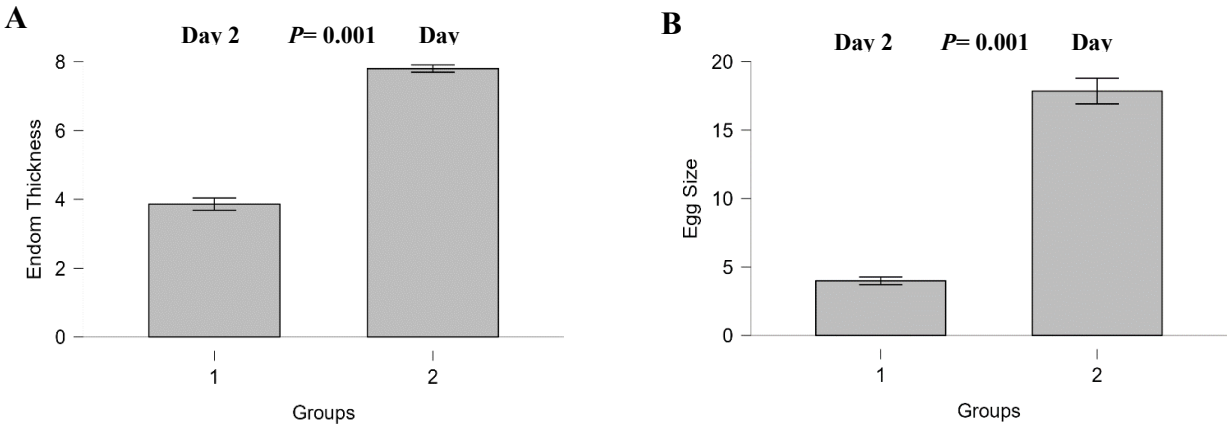


Figure1: Ultrasonography Results for Endometrial Thickness and Egg Size on Day 2 And Day 12 of the Menstrual Cycle

The left panel shows endometrial thickness, and the right panel shows follicular (egg) size in Groups 1 and 2. Both parameters were significantly higher in Group 2 compared to Group 1 on Day 12 (P = 0.001). Error bars represent standard error of the mean (SEM).

Hormonal results for LH and prolactin on days 2 and 12 of the menstrual cycle are shown in Fig.2A & Fig.2B. Between Day 2 and Day 12 of the menstrual cycle, there are significant differences in LH levels (p=0.001), but there are no significant alterations in prolactin levels ($p > 0.05$).

The variation in study parameters based on menstrual cycle regularity [regular (N=64) and irregular (N=41)] is seen in Fig.3A-R. The data suggests that although women with irregular menstrual cycles and those with regular cycles do not vary significantly in age, BMI, or most hormonal parameters, they do vary significantly in the duration of their marriages, with the latter group having a shorter average duration.

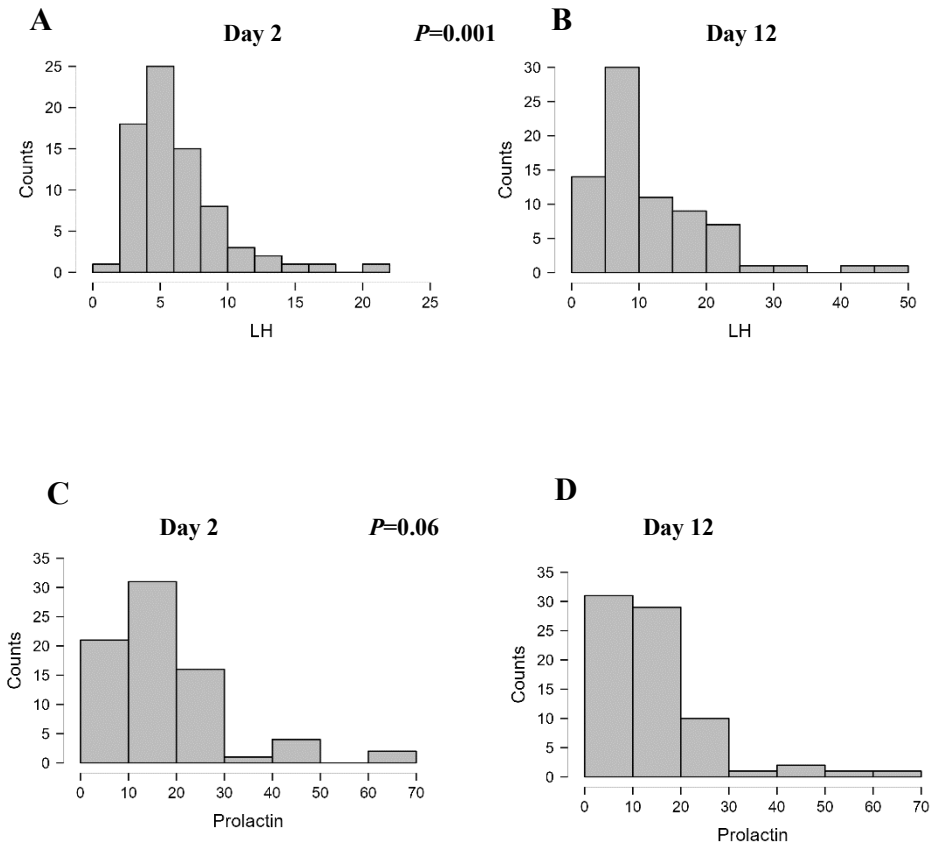
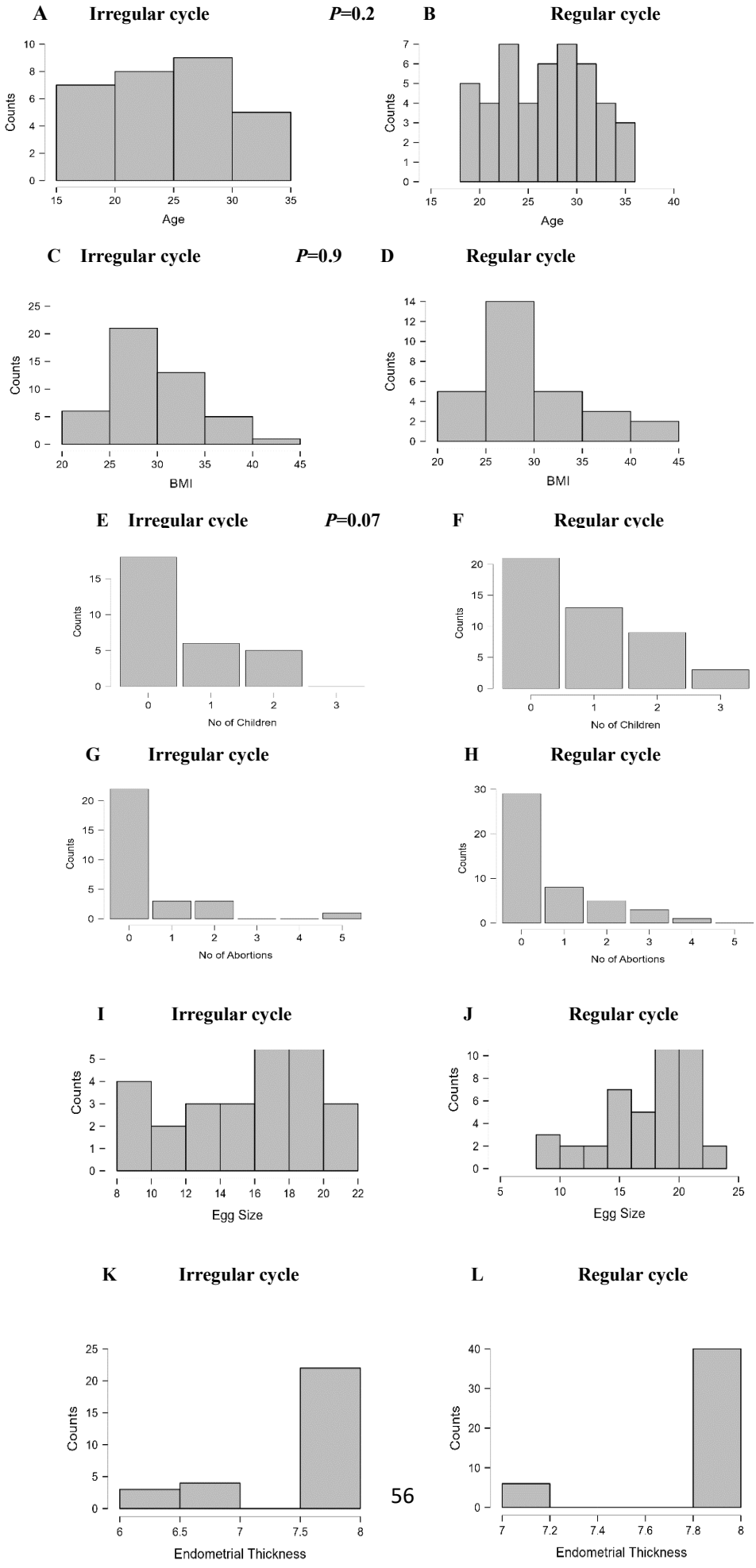


Figure 2. Hormonal Measurements on Day 2 And Day 12 of The Menstrual Cycle
A and **B** show the distribution of luteinizing hormone (LH) levels on day 2 and day 12, respectively, with a significant increase observed on day 12 (**P = 0.001**), consistent with the expected pre-ovulatory LH surge. **C** and **D** illustrate the distribution of prolactin levels on day 2 and day 12, showing a slight but non-significant decrease in prolactin levels on day 12 (**P = 0.06**).


A Irregular cycle
P=0.2
B Regular cycle
Counts
Age
C Irregular cycle
P=0.9
D Regular cycle
BMI
E Irregular cycle
P=0.07
F Regular cycle
No of Children
G Irregular cycle
H Regular cycle
No of Abortions
I Irregular cycle
J Regular cycle
Egg Size
K Irregular cycle
L Regular cycle
Endometrial Thickness

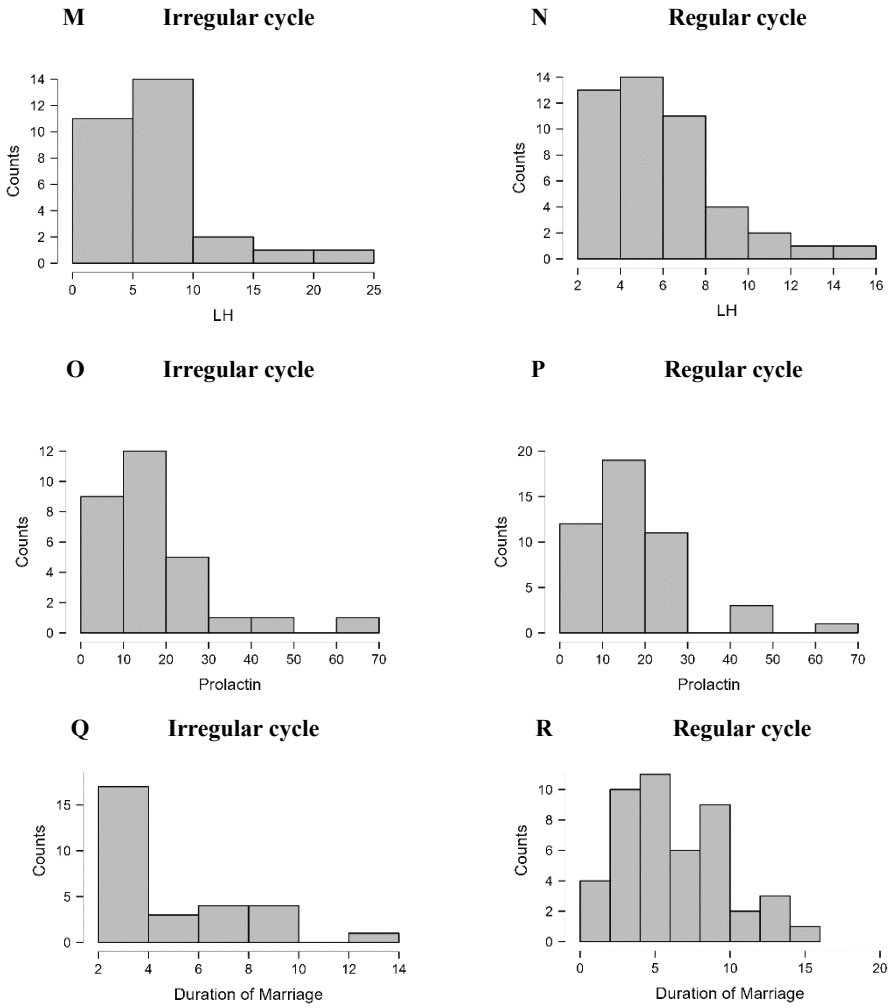


Figure3: Distribution of Demographic, Reproductive, Hormonal, and Clinical Variables Among Women with Irregular and Regular Menstrual Cycles

(A–B) Age distribution in women with irregular and regular cycles, respectively (**P = 0.2**). **(C–D)** Body Mass Index (BMI) distribution across both groups (**P = 0.9**). **(E–F)** Number of children, showing a near-significant difference (**P = 0.07**). **(G–H)** Number of abortions, with no significant variation. **(I–J)** Distribution of follicular (egg) size. **(K–L)** Distribution of endometrial thickness measurements. **(M–N)** Luteinizing hormone (LH) levels. **(O–P)** Prolactin levels. **(Q–R)** Duration of marriage in both groups. Each pair of histograms compares the respective variable between women with irregular cycles (left) and regular cycles (right), highlighting patterns and differences in reproductive and endocrine profiles.

The capacity of many factors, such as follicular size, prolactin, LH, and endometrial thickness, to differentiate between hormonal and ultrasonic changes on days 2 and 12 of the cycle, is shown in Table3 and Table4 and Fig.4. For each variable, the table gives the sensitivity, specificity, 95% confidence intervals, p-values, and area under the receiver operating characteristic (AUC) curve. Endometrial thickness and follicle size are highly effective markers for distinguishing changes, making them valuable for monitoring menstrual cycle-related changes after letrozole administration. With an AUC value of 0.515 and a non-significant p-value of 0.752, LH demonstrates low discriminative capacity. Additionally, the moderate sensitivity and specificity imply little value in differentiating between acoustic and hormonal changes. With the highest AUC value of 0.777, prolactin stands out as having good discriminative capacity. The statistical significance of the low p-value of 0.001 indicates that prolactin levels are useful in differentiating between changes that are hormonal and ultrasonic. Prolactin's usefulness in this situation is further supported by the comparatively high sensitivity and specificity values.

Table3: Ability of Endometrial Thickness, Follicular Size, LH, and Prolactin to Distinguish Between Hormonal and Ultrasonic Changes on Days 2 And 12 of The Cycle

Variables	AUC	P-value	Sensitivity	Specificity	95% Confidence Interval	
Endometrial thickness	1.0	0.001	1.00	1.00	1.00	1.00
Folicular size	1.0	0.001	1.00	1.00	1.00	1.00
LH	0.515	0.752	0.578	0.508	0.422	0.608
Prolactin	0.777	0.001	0.613	0.514	0.699	0.854

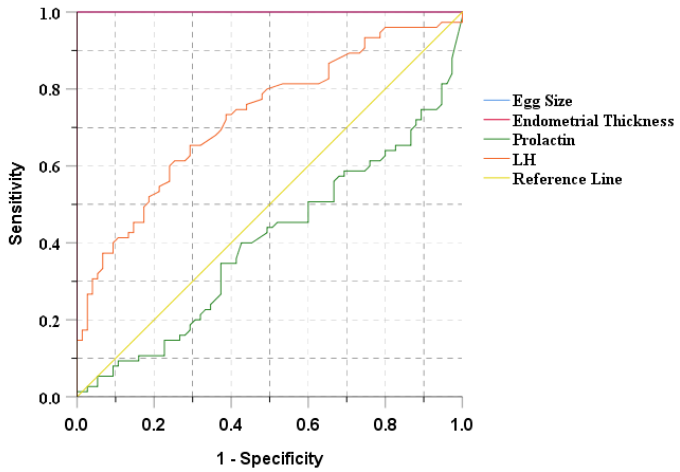


Figure4: Receiver Operating Characteristic (ROC) Curve Analysis of Egg Size, Endometrial Thickness, Prolactin, and Luteinizing Hormone (LH) As Predictors of Menstrual Cycle Regularity

The ROC curves illustrate the diagnostic performance of four parameters: egg size (blue), endometrial thickness (pink), prolactin (orange), and LH (green). The yellow diagonal line represents the reference line (no discrimination). Among the tested parameters, endometrial thickness shows the highest sensitivity and specificity, followed by prolactin. Egg size and LH display comparatively lower diagnostic accuracy. The analysis supports the potential clinical utility of endometrial thickness and prolactin as strong indicators for distinguishing between regular and irregular

Spearman's correlation analysis Table4 reveals numerous important relationships between the study variables, providing insights into the interplay between various factors associated with fertility and reproductive health.

Table4: Spearman's Correlations Evaluations of the Study Variables Among Each Other's

Parameters (means)	Analyses	Age	BMI	Follicular Size	Endometrial Thickness	LH	Prolactin	AMH
Age	Correlation	—						
	Significance	—						
BMI	Correlation	0.251**	—					
	Significance	0.033	—					
Follicular Size	Correlation	-0.046	-0.051	—				
	Significance	0.881	0.811	—				
Endometrial Thickness	Correlation	-0.110	-0.251	.898**	—			
	Significance	0.299	0.169	.000	—			
LH	Correlation	-0.181	-0.323	.375**	0.323**	—		
	Significance	0.452	0.445	.000	.000	—		
Prolactin	Correlation	-0.211	-0.201	-.152	-.149	-.030	—	
	Significance	0.318	0.105	.063	.069	.711	—	
AMH	Correlation	-0.309	-0.100	.027	-.108	.173	-.077	—
	Significance	0.071	0.560	.817	.355	.138	.509	—

** **p < .01**

4. Discussion

The study aimed to evaluate the effectiveness of letrozole in ovulation induction outcomes in infertile Iraqi females. The study found that letrozole was an effective ovarian stimulant, and achieved a significantly higher endometrial thickness. Letrozole, which is largely applied to treat breast tumors, has drawn attention due to its likely benefits in treating PCOS-related infertility. The existing investigation contributes to the growing body of trials supporting letrozole as a first-line choice for infertility in PCOS females. Letrozole's capacity to stimulate ovulation and increase gestation rates sheds light on its potential in the management of PCOS-related infertility (Yang et al., 2021b, Chen et al., 2024a, Franik et al., 2022a). To inhibit the negative feedback on the hypothalamic-pituitary-gonadal axis and encourage the development of new follicles, letrozole works by blocking the synthesis of estrogen (Eskew et al., 2019, Franik et al., 2022a, Pritts et al., 2011, Yang et al., 2021b).

Simultaneous modulation of other hormonal biomarkers, like LH and prolactin, was also detected. These hormonal shifts are crucial for regulating the complex processes of follicular growth, ovulation, and synthesis of corpus luteum—all of which must occur for a fruitful pregnancy (Duncan, 2021, Przygrodzka et al., 2021, Reed BG). Additionally, after letrozole was orally administered, favorable increases in endometrial thickness and morphology were exposed by sonography assessment. The observed rise in endometrial thickness, which reflects appropriate endometrial proliferation and responsiveness, was particularly significant. From this, it can be inferred that letrozole may improve endometrial dynamics and increase the chances of successful embryo implantation and subsequent pregnancy. While the current study indicates that letrozole is an effective ovarian stimulant, it is essential to consider that the evidence base for its use in ovulation induction is still developing. A recent systematic review included 41 randomized controlled trials involving 6,522 women, demonstrating that letrozole improves live birth rates compared to clomiphene citrate (CC) for ovulation induction (Franik et al., 2022b). However, the variability in study designs and populations underscores the need for caution in generalizing these findings. For instance, while some studies report significant improvements in endometrial thickness and pregnancy rates with letrozole, others indicate no substantial differences compared to CC (Chen et al., 2024b, Kar, 2013). Furthermore, the lack of consensus on optimal dosing regimens complicates the interpretation of results across different studies (Yang et al., 2021b). Thus, while letrozole shows promise as a first-line treatment for infertility in women with PCOS, further research with larger sample sizes and standardized protocols is necessary to confirm these findings and address existing inconsistencies (Kivrak et al., 2024). There is a significant difference ($p = 0.001$) in the duration of marriage across age groups. Similarly, while the search results fail to demonstrate a significant difference in the duration of marriage between age groups in couples experiencing infertility due to PCOS, they do indicate that PCOS can significantly affect a couple's satisfaction with their marriage and sexual life, as well as their connections with friends and family (Navid et al., 2018, De Frène et al., 2014). There were non-significant differences in age, BMI, and most hormonal parameters between women with irregular and regular menstrual cycles. However, there is a significant variation in the duration of marriage, with women experiencing irregular cycles having a shorter mean duration of marriage. The duration of marriage has no direct effect on PCOS women's irregular menstrual periods. Nevertheless, irregular periods after marriage can be caused by a variety of circumstances, including stress, changes in practice, weight variations, hormonal birth control, and pregnancy (Khalaf et al., 2015). The condition itself of PCOS can cause irregular menstrual cycles in women, which can impact menstrual regularity and reproductive health (Dason et al., 2024). To address any underlying concerns and receive the proper therapy or management, it is imperative that women who experience irregular periods after

marriage visit healthcare specialists (Navid et al., 2018). The follicular size and endometrial width on Day 12 had significantly enlarged compared to Day 2 of the cycle after administration of oral letrozole. This can be clarified by the fact that letrozole acts as an aromatase inhibitor, which upsurges intra-ovarian androgenic concentrations, particularly in the early antral follicles (6-8mm) at Day 3 or 5 of the menstruation (McGrail et al., 2020). These increased androgen measures in the early ovarian follicles stimulate granulosa cell mitosis, increase expression of FSH receptors, and make the ovarian follicles more resistant to atresia. The latter effect boosts follicular development and eventual ovulation (Rose and Brown, 2020). The increased level of estrogen caused by increased follicle development with early letrozole initiation leads to enhanced endometrial width and maturation (Franik et al., 2022a). Exactly, the published data indicate the endometrial thickness was meaningfully more if letrozole was initiated on Day 5 (9.0mm) compared to Day 3 (8.0mm). The enhanced endometrial response induced by letrozole initiation may contribute to the increased implantation and clinical pregnancy rates detected (Sakar and Oglak, 2020). Hormone replacement therapy led to an increase in proliferative thickening, which is evidence that endometrial growth and responsiveness to hormone stimulation are accepted. These findings might support a new function for letrozole in enhancing endometrial cohesiveness and establishing the perfect environment for the attachment-implantation signal of the embryo and, eventually, conception (15,16).

The search results revealed no studies have compared the impact of oral letrozole initiated on the second day of the cycle for two successive cycles with day 12^{th} of the second cycle regarding follicle size or endometrial thickness. However, data about the effects of letrozole on the endometrial thickness are still controversial. If letrozole was started on the fifth day of the cycle, compared to the third day of the cycle, there were greater rates of ovulation, endometrial thickness (on the day of the HCG intramuscular injection), gestation, and clinical pregnancy in PCOS females documented earlier (Roy et al., 2012, Shi et al., 2022). In contrast, a study on the effect of letrozole on endometrial width in IVF cycles indicated that using letrozole during stimulation reduced the endometrial width by 0.81mm (Ruiter-Ligeti et al., 2021). In comparison to patients who did not get letrozole during their initial IVF cycles, those who received it had a thinner epithelium on the trigger day. The study showed that letrozole thins the endometrium, but it is still unclear how letrozole affects endometrial function. This underscores the need for more research before letrozole is prescribed for new transfers (Ruiter-Ligeti et al., 2021).

The correlational analysis in the current work reveals many substantial correlations among the study parameters, providing insights into the relations between several factors related to reproductive health and fertility. Nevertheless, it's vital to exercise attention when understanding these results and to consider any further confounding issues that could influence the exposed links. The current findings have considerable therapeutic inferences for managing infertility. By clarifying the mechanisms through which letrozole medicine supports ovulation and endometrial receptiveness, healthcare providers can modify treatment schedules to optimize reproductive outcomes for affected individuals. Since letrozole is oral and has a proper safety profile, it is also an appropriate and well-tolerated choice for individuals receiving fertility therapies.

5. Conclusion

According to the research, taking letrozole orally on the second day of the menstrual cycle greatly promotes endometrial and follicular growth. The study considerably enhances our understanding of in what way letrozole aids females with PCOS-related infertility. The conclusions propose that letrozole intervention might be valuable for numerous fertility-related features, including ovulation stimulation, thick endometrium, and lower gestational rates. Prolactin hormone exhibits possible biomarkers to distinguish between hormonal and ultrasonic variations in the context of PCOS medicine. The findings highlight the worth of letrozole as a therapeutic choice that helps females with PCOS-related infertility, and they also stress the necessity for further research to expand treatment tactics and reproductive outcomes.

6. Ethical Consideration

The University of Karbala's College of Pharmacy accepted the study protocol, and the Karbala Health Directorate was also consulted for approval. Furthermore, consent was obtained from every patient following an explanation of the study's nature and goals.

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