

MOTHERISK ROUNDS

Fetal Safety of Letrozole and Clomiphene Citrate for Ovulation Induction

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J Obstet Gynaecol Can 2007;29(8):668–671

INTRODUCTION

Letrozole is a third-generation selective aromatase inhibitor that blocks the rate-limiting step in the production of estrogen from androstenedione and testosterone substrates. Letrozole is approved in Canada for use in the treatment of postmenopausal women with breast cancer.¹ Letrozole has no significant active metabolites. It is completely absorbed after oral administration and has a mean terminal half-life of approximately 45 hours (range 30–60 hours). It is cleared from the systemic circulation mainly by the liver.

In the late 1990s, aromatase inhibitors, including letrozole, began to be used to induce ovulation by being administered in the early part of the menstrual cycle.^{2–4} Estrogen production from all sources is blocked by inhibiting aromatization, releasing the hypothalamic-pituitary axis from estrogenic negative feedback and resulting in increased gonadotropin secretion and ovarian follicular stimulation.^{5,6} In the ovary, aromatase inhibitors increase follicular sensitivity to FSH, as there is an accumulation of intraovarian androgens.⁷

At the level of the endometrium, estrogen receptors may be upregulated, resulting in rapid endometrial growth once

estrogen secretion is restored (after clearance of letrozole).⁸ In its ability to induce ovulation, letrozole compares favourably to clomiphene citrate, which has been the first line treatment for ovulatory disorders for more than 40 years, and it has emerged as an alternative to CC for ovulation induction. The clinical outcome of early pregnancies achieved through the use of letrozole for ovulation induction or controlled ovarian hyperstimulation for intrauterine insemination was reported in 2005 in a cohort study.⁹ The outcomes of pregnancies achieved through letrozole and other ovarian stimulation regimens (CC) were compared with a control group composed of women who had conceived spontaneously. Pregnancies conceived after use of letrozole were associated with rates of miscarriage and ectopic pregnancy that were comparable to rates associated with all other pregnancies, including spontaneous conceptions. Letrozole use was associated with a significantly lower multiple gestation rate than use of CC.⁹ It seems unlikely that there would be significant exposure of the embryo to letrozole, as the short half-life of letrozole and the timing of administration in the early follicular phase should result in clearance of the drug before implantation takes place.

Concern was raised at the 2005 Annual Meeting of the American Society for Reproductive Medicine about the safety of the fetus in mothers who used letrozole.¹⁰ One hundred fifty babies from 130 pregnancies conceived after the use of letrozole were compared with 36 000 babies conceived spontaneously and born to women at low risk in a community hospital. Although there was no difference in the overall rate of congenital anomalies between the two

Key Words: Letrozole, clomiphene citrate, pregnancy, teratogenicity, fetal anomalies

Table 1. Comparing the effects on pregnancy outcomes of letrozole and clomiphene citrate with controls

Group (n all offspring, n singletons)	Median maternal age at birth (years) [25%, 75%]	Median birthweight [25%, 75%] (kg)*	Median gestational age at birth (weeks) [25%, 75%]*	Number of offspring with malformations n (%)
Motherisk (112, 78)	33 [30, 37]	3.320 [3.097, 3.749] 3.391 [3.099, 3.808]†	39 [36, 40] 39 [37.6, 40]†	3 (3.2)
Letrozole (94, 78)	33 [30, 37]	3.220 [2.540, 3.720] 3.538 [3.039, 3.820]†	38.5 [36.7, 40] 39 [38, 40]†	0 (0)
CC (271, 211)	33 [30, 36]	3.240 [2.815, 3.526]‡ 3.325 [2.950, 3.606]†	38.4 [37.1, 39.6] 38.6 [37.4, 39.6]†	7 (2.6)

*data from multiple births (i.e., twins, triplets) are excluded from each group (Motherisk controls, letrozole and CC) in this analysis.

†Singleton only data

‡ $P < 0.05$ compared with Motherisk controls

groups, the authors reported that the incidence of cardiac and bone anomalies was higher in the letrozole group than in the control group. There were numerous concerns regarding the methodology of this study: the small size of the letrozole group, the choice of a control group that would have a lower risk of pregnancy complications and congenital malformations than an infertile population, and the under-representation of congenital anomalies in the control group (noting that any babies identified as abnormal on prenatal ultrasound would be delivered at a tertiary care hospital rather than a community hospital).

The present study was designed to compare the risks to the fetus of letrozole and CC and to compare them with the risks to the fetus in the general obstetrical population. The primary objective was to compare the malformation rates in the offspring of women who conceived using letrozole, women who conceived spontaneously (age-matched controls), and women who conceived using CC (disease-matched controls). The secondary objective was to compare other pregnancy outcomes (birth weight and gestational age at birth) among the three groups.

METHODS

We reviewed the records of women who had delivered after using either letrozole or CC for ovulation induction during treatment at the McGill Reproductive Centre in Montreal or the Toronto Centre for Advanced Reproductive

Technology. The data recorded were maternal age at birth, gender of offspring, gestational age at birth, birth weight, and congenital malformations. Each woman in the letrozole group was matched by age with a control from the Motherisk database. All Motherisk controls conceived spontaneously. In each group, data were analyzed with and without exclusion of multiples, and centiles for birthweight adjusted for GA were calculated for all available data using centile charts.¹¹

Statistical Analysis

Data were analyzed using GraphPad InStat 3.05 (GraphPad Software Inc, San Diego, CA). The Kolmogorov and Smirnov method was used to test the normality of data distribution, specifically for maternal age, gestational age, and for birth weight including and excluding multiple births. If the data passed the normality test, then a one-way ANOVA was conducted; if not, then the Kruskal-Wallis one way ANOVA on Ranks test (a nonparametric ANOVA) was used for analysis. For any statistically significant differences found in these data, Dunn's Multiple Comparison test was used. The chi-square test was used to compare time of delivery and incidence of malformations between groups. Unpaired *t* tests were used to compare the mean centiles between each group.

RESULTS

In this retrospective multicentre study, we analyzed data from 94 women who conceived using letrozole, 242 women who conceived using CC, and 94 women who conceived spontaneously. In the letrozole group, 112 babies were born (including 14 sets of twins and 2 sets of triplets). In the CC group, 271 babies were born (including 27 sets of twins and 2 sets of triplets). There were no multiple births in the group who conceived spontaneously (Motherisk control group). When the letrozole group was compared with the CC and

ABBREVIATIONS

ANOVA	analysis of variance
CC	clomiphene citrate
CI	confidence interval
FSH	follicle stimulating hormone
GA	gestational age

the Motherisk control group, there were no statistically significant differences in the median maternal age at time of delivery (33 years), GA at birth (38.5, 39, and 38.4 weeks, respectively), or the rate of malformations (0%, 3.2%, and 2.6%, respectively) (Table 1). The median birth weight of babies in the letrozole group was not significantly different from that of babies in the Motherisk control group, either when multiples were included in the letrozole group (3220 vs. 3320 g) or when multiples were excluded (3538 vs. 3391 g). In contrast, the median birth weight of all babies born to women who conceived using CC was significantly reduced compared to the controls (3240 vs. 3320 g, $P < 0.01$). Further analysis was performed to determine if there were differences between groups in median birth weight adjusted for GA and with multiples excluded. Mean centiles for babies in the letrozole group did not differ significantly from the controls (54.8 vs. 61.7, $P > 0.05$), and the mean centiles for babies in the CC group were lower (37.2, $P < 0.0001$) (Table 2).

DISCUSSION

The present matched control study provides evidence that letrozole is not a human teratogen. Instead, it suggests that use of CC may result in small for gestational age infants. A recent Canadian retrospective multicentre study¹² addressed some of the methodological issues in the 2005 American Society for Reproductive Medicine abstract.¹⁰ This new study included a much larger number of women who had used letrozole to conceive. In addition, the populations being compared were both composed of patients with infertility. The study did not find any increase in the rates of major malformations in babies conceived after letrozole treatment.¹² The present study provides support for these findings. We identified no major congenital malformations in the letrozole group, and the number of major malformations in the CC group was not statistically significantly different from those in the Motherisk control group.

Aside from the retrospective Canadian study described above,¹² there are no published reports of congenital anomalies in human offspring delivered after the use of letrozole. When letrozole was given to pregnant rats at 1% of the dose used in humans for ovulation induction, fetal anomalies involving the kidney and ureter, and incomplete skeletal ossification were seen. Treatment of pregnant rats with letrozole 1 mg/kg/day on gestation days 21 and 22 resulted in altered sexual function in male offspring.¹³ Although these male offspring underwent puberty at the normal age and had normal body and testicular weights, there was a 24% decrease in rates of pregnancy when they were mated with normal females. Sexual activity was reduced and there was a mild decrease in testicular spermatid number. The conclusions of this study are limited, however, by failure to

Table 2. comparing the birthweight adjusted for gestational age between letrozole, disease-matched (CC), and age-matched (Motherisk) controls*

Group (n)†	Mean centile ± SD [lower 95% CI, upper 95% CI]
Motherisk (73)	61.7 ± 33.5 [53.8, 69.5]
Letrozole (41)	54.8 ± 30.0 [45.3, 64.2]
CC (54)	37.2 ± 30.6‡ [28.9, 45.6]

*Data from multiple births (i.e., twins, triplets) are excluded from each group (Motherisk controls, letrozole, and CC) in this analysis
 †Data for gestational age and/or birthweight were not available for all women
 ‡ $P < 0.05$ compared with Motherisk controls

control for possible effects of litter of origin on male sexual function.

Because CC has been used for ovulation induction for more than 40 years, there is significantly more published information on congenital anomaly rates in human and animal offspring following use of CC than use of letrozole. In human pregnancies conceived after use of CC for induction of ovulation, the reported overall rates of major or minor malformations have not been significantly different from those observed in spontaneously conceived pregnancies.^{14,15} An association between maternal use of CC and coarctation of the aorta (odds ratio 4.5; 99% CI 1.0–19.9) was observed in a case–control study that included 126 children. This association was not seen in a subsequent case–control study of 83 infants with conotruncal cardiac defects.¹⁶ A possible association between the use of CC for ovulation induction and an increased incidence of neural tube defects in the offspring has been debated in the literature. Proponents of the association do agree, however, that any increased risk associated with CC is not large.¹⁷ A small number of reports have described an association between use of CC or other agents that may alter hormone levels in early pregnancy and an increased incidence of neuroendocrine tumours, including neuroblastoma.^{18–20} In other reports acardic twins have been reported twice in pregnancies when CC was used for ovulation induction.¹² The risk of craniosynostosis was increased in a small population of infants whose mothers had used CC for ovulation induction,^{21,22} although the authors of these reports indicated that the results were not adjusted for multifetal gestations.

Our present comparison of the outcome of babies conceived after the use of letrozole with matched controls conceived after use of CC and a control group of babies whose mothers conceived spontaneously found that the birth

weight of babies in the CC group was significantly lower than the birth weight of babies in both the letrozole group and the control group, even after controlling for maternal age and GA at birth, and excluding multiple gestations. Although this observation has not been previously reported in human studies, there is support from a small amount of animal data. In some reports, CC treatment during pregnancy was associated with decreased fetal growth. Decreased implantation rates and increased rates of fetal growth retardation and exencephaly were observed among offspring of mice treated with CC just before ovulation in doses similar to those used in humans.²³ Blastocyst transfer experiments in mice indicate that the preovulatory administration of CC impairs uterine function, which subsequently reduces embryonic growth and development.²³ A possible mechanism for the observed intrauterine growth restriction in animals and humans when CC is used for ovulation induction is the long half-life of the CC isomers (elimination time 5–7 days) and the possible presence of anti-estrogenic effects in the endometrium and uterus causing reduced blood flow or affecting embryogenesis.

Despite the use of matched controls, it remains a possibility that the women in the two treatment groups were given CC or letrozole based on different characteristics and therefore were not directly comparable. After reviewing the prescribing practices of the clinics involved in this study, we do not believe this to be the case. Some clinics have a preference for one medication and treat all patients with this medication first unless a drug allergy or adverse reaction has been reported. In some instances, patients are treated with CC first and are treated with letrozole if CC is unsuccessful. Occasionally, a patient who conceived after use of letrozole or CC will wish to use this medication when trying to become pregnant again. Letrozole and CC are comparable in price, so the groups would not likely differ on the basis of on socioeconomic factors.

CONCLUSION

The use of letrozole for ovulation induction does not appear to increase the risk of congenital malformations and does not affect birth weight. The results of the present study suggest use of CC increases the incidence of small for gestational age infants.

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