Letrozole in poor responders undergoing ICSI: an egyptian experience

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Abstract

Objective: to compare gonadotropine /antagonist protocol with and without Letrozole for controlled ovarian hyperstimulation for poor responder cases undergoing ICSI.
Materials & Methods: A prospective randomized case control study performed in Mansoura Fertility Care unit, Mansoura University hospital. Controlled ovarian hyperstimulation using gonadotropins/antagonist protocol with (group A) and without Letrozole (group B). The primary outcome is to measure duration of stimulation, total dose of gonadotropins, serum E2 level and endometrial thickness on day of HCG administration. The number and degree of maturation of retrieved oocytes, fertilization rate and number and grade of the developed embryos were evaluated. Secondary outcome includes ongoing pregnancy and take home baby rates.

Results: Duration of stimulation was shorter in group A (10.2± 1.5 days) compared to (11.5± 1.9days) in group B, total dose of gonadotropins was lower in group A (2602±433) in contrast to (2752±515IU) in group B, endometrial thickness was thicker in group B (9±2.2mm) compared to (7.8±2.2 mm) in group A, but no significant difference was detected between the two groups apart from mean duration of stimulation.

Conclusions: Letrozole as an adjuvant to gonadotropins seems to reduce stimulation period and total gonadotropins units used.
Key wards: Letrozole, poor responders, controlled ovarian hyperstimulation, ICSI cycles.

Introduction

The response to controlled ovarian stimulation (COS) regimens differs from woman to another, it could be normal leading to growth of reasonable number of follicles, poor leading to growth of reduced number of follicles or over response referred to as ovarian hyper stimulation syndrome(OHSS) (1).The success of intracytoplasmic sperm injection (ICSI) depends on a properly selected protocol of controlled ovarian stimulation and adequate oocyte recruitment. (2).

It is important to predict poor ovarian response in order to determine the most suitable stimulation protocol. Several ovarian reserve tests have been studied to predict outcome of (ICSI) in terms of response to ovulation induction and likelihood of pregnancy. Some of these tests have become part of the routine diagnostic procedure for infertility patients that will undergo (ICSI). Female age, for instance, is the basic factor that is related to both quantity and quality of oocytes. Basal Follicle stimulating hormone (FSH) gives a more thorough indication of quality aspects. By choosing the right thresholds these tests may eventually correctly predict oocyte quality (3).

The incidence of poor responders varies in the literature between 9 and 24%. Till now, there is no agreement upon its definition; failure to respond adequately to standard protocols and to recruit adequate follicles could be used as a definition (4). At ICSI, a patient is considered a poor responder when three or fewer follicles are recruited and serum estradiol concentrations are lower than 300 pg/ml (if one follicle) or 500 pg/ml (if two or three follicles) at the time of human chorionic gonadotropin (hCG) administration (5). Reduced ovarian reserve represents the most frequent etiological factor (4).which could be due to advanced patient age. Furthermore, a young patient with advanced endometriosis, previous ovarian surgery or pelvic adhesions may present with diminished ovarian reserve. Two other situations that can lead to poor response are the elevated basal serum FSH levels and the reduced inhibin production by granulosa cells (6).

Aromatase is an enzyme that catalyzes the rate-limiting step in the production of estrogens. It is a good target for selective inhibition because estrogen production is a terminal step in the biosynthetic sequence. (7) An aromatase inhibitor (e.g. Letrozole) could be

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used for ovulation induction, its mechanisms of action could be explained by blocking estrogen-negative feedback, without depletion of estrogen receptors (ERs), A second hypothesis involves an increased follicular sensitivity to FSH, resulting from temporary accumulation of intraovarian androgens because conversion of androgen substrate to estrogen is blocked. Data support a stimulatory role for androgens in early follicular growth in primates (8). Peripherally, it is possible that Letrozole, with suppression of estrogen concentrations in the circulation and peripheral target tissues, results in up-regulation of ERs in the endometrium, leading to rapid endometrial growth once estrogen secretion is restored. (9). In this study, the role of Letrozole as an adjuvant to gonadotropins was evaluated in a subgroup of poor responders undergoing ICSI cycles.

Materials and Methods

This study was conducted at the Fertility Care Unit, Mansoura University Hospital, Mansoura , Egypt. A group of 60 women were recruited during the period from January, 2010 till December, 2011. Women were diagnosed as poor responders for ICSI cycles. They were recruited if they have one or more of the following criteria:

1. Poor ovarian response with <3 mature follicles on a long GnRH agonist protocol in their previous IVF/ICSI cycles.
2. Repeated high basal levels of FSH >10 IU/l.
3. Older than 40 years.

Participants were asked to sign a written consent to study and were randomized into two groups:

- ***Group A:*** 30 participants received Letrozole 2.5mg/12h (Femara® , Novartis, Egypt) from day 2 of the cycle for 5 days plus gonadotropines 375 IU/day (Merional®, IBSA, Switzerland) followed by gonadotropines given according to response to stimulation detected by follow-up until day of hCG.
- ***Group B:*** 30 participants received gonadotropins 375 IU / day (Merional®, IBSA, Switzerland) from day 2 of the cycle for 5 days then gonadotropins given according to response to stimulation detected by follow-up until day of hCG.

Participants were followed up by the same observer using 7 MHz vaginal probe mounted to an ultrasound machine (Sonoace 3200, Medison, South Korea) for folliculometry after the first 5 days of stimulation then every other day thereafter. Doses of gondotropins were modulated according to the response. Cancellation of cycles were based on low E2 level during follow up (<300 pg/ml) correlated with less than three ovarian follicles detected at the follow up visits after counseling the couple regarding success rate.
Both groups had pituitary suppression by flexible multidose GnRH antagonist protocol (Citrotide®,EMD Serono, Switzerland) 0.25 mg per day SC when the largest follicle reaches 14mm. Citrotide was continued by daily injection until day of hCG administration. All cases received 10000 IU hCG (Choriomon®, IBSA, Switzerland) when the mature follicle reached 18 mm. Serum E2 concentration and endometrial thickness were measured on the day of hCG administration.

Oocytes were collected ultrasound-guided under general anesthesia 34-36 h after hCG administration and categorized as regards number and degree of maturation into metaphase I (MI) and metaphase II (MII). Oocytes were manipulated in the laboratory by ICSI and incubated for 48-72 hours until dividing embryos were formed which were also categorized into grades A, B and C.

Endometrial preparation for embryo transfer was started the day of oocyte collection by giving cases progesterone (Prontogest®, IBSA, AMSA, Italy) 100 mg ampoules IM once per day. Normally cleaving embryos were transferred into the uterine cavity 48-72h after oocyte retrieval using soft embryo transfer catheter under ultrasound guidance. Luteal phase support was continued by the same regimen started on the day of oocytes collection until two weeks after embryo transfer when serum pregnancy test was done. Cases with positive pregnancy tests were examined by ultrasound two weeks later to document intrauterine clinical pregnancy. Pregnant participant were followed up and ongoing pregnancy rate and take home baby rate were determined.

Results

A total of 60 cycles were recruited to the study and the participants were randomized into two groups (A and B). The demographic criteria of the participants regarding age, basal FSH and previous poor response in ICSI cycle were plotted in table (1).
The results of the studied groups were shown in Table (2). Fertilization rate (developed embryos per injected oocytes) was slightly lower in group A (52%) compared with (53%) in group B with no significant difference (P = 0.9). The mean number of embryos transferred was (1.4± 0.9) in group A compared to (1.3±0.9) in group B. Yet, no statistically significant difference could be detected (P = 0.8).

As regards implantation rate, it was equal in both groups (22.2%). Clinical pregnancy rate was equal in both groups (4 cases in each group) which represent (13.3%) of initiated cycle.Also, pregnancy rate per embryos transferred the rate was equal in both groups (14.81%).

There were three cases (two in group A and one in group B) reached full term and were delivered by cesarean section while, there were five cases(two in group A and three in group B)were miscarriaged in the first trimester. The delivered babies in both groups revealed no congenital anomalies.

Table (1): Demographic criteria of the studied groups

Variables	Group A*	Group B **	P
	Mean ±SD	Mean ±SD	
Age	36.4 ± 4.9	35.6 ± 4.8	0.523
Basal FSH	10.1 ± 2.8	10.3 ± 2	0.730
Previous ICSI	11	10	0.061

Table (2): Results of the studied groups

Variables		Group A* (Mean ±SD)	Group B** (Mean ±SD)	P
Stimulation period(days)		10.2 ± 1.5	11.5 ± 1.9	0.008
HMG units used		2602 ± 433	2752 ± 515	0.227
Oocyte quality	MI	1.6± 1.2	0.6± 0.5	0.044
	MII	1.6 ± 0.8	1.8 ± 1	0.445
Embryos Transferred		1.4 ± 0.9	1.3 ± 0.9	0.773
Serum. E2		475.5± 182.7	517.3 ± 276.9	0.533
endometrial thickness		7.8 ± 2.2	9± 2.2	0.068
Cancellation rate		7	8	0.338

Discussion

Letrozole is an aromatase inhibitor. It was introduced as an ovulation induction agent either alone or as an adjuvant with gonadotropins. It acts through transient inhibition of aromatase activity in early follicular phase with subsequent induction of ovulation. Its role in poor responder was evaluated in many studies with varying results regarding its advantages (10- 15).

In the current study the mean duration of stimulation was shorter in group A compared to group B (10.2±1.5 vs. 11.5±1.9). These results support the results of Yarali et al. (10) who found shorter duration of stimulation in cases received Letrozole. However another study (11) showed no difference regarding days of stimulation on addition of letrozole but this study compared different stimulation protocols, namely, microdose GnRH agonist flare with GnRH antagonist /Letrozole protocol.

Regarding the total gonadotropins units used, in the current study were lower in group A in contrast to group B (2602±433 vs. 2752±515) this could support the results obtained by some authors (10,12). Letrozole used in combination with gonadotropins reduced the cost of infertility treatment by decreasing the gonadotropins dose required for optimum ovarian stimulation. This could make assisted reproductive technology available to a larger group of infertile couples. If this was the case, this could have a reflection on the total cost of the cycle which is an important factor in low income societies like Egypt.

Serum estradiol at day of hCG administration was found lower in group A compared to group B (475.5±182.7vs517.3±276.9) which is comparable to results of Goswami et al (12), Who found significant reduction in serum estradiol level. Other authors (10, 13) found similar results. The current study evaluated endometrial thickness on day of hCG administration and found that endometrium was thinner in group A this is in contrary to the results of Verpoest et al. (13) which showed increased endometrial thickness with Letrozole co-treatment. Although the addition of Letrozole to gonadotropins in the current study reduced the mean serum E2 level and endometrial thickness, there was no negative impact on implantation and pregnancy rates. Although Cancellation rate was slightly lower in group A (23.3% vs. 26.7%), it was not statistically significant difference could be detected. This could reflect the impact of low income of the population studied, as some cases refused cancellation of their cycles despite cancellation criteria. They could not afford another cycle, so they decided to continue. Regarding the oocytes number and quality, no statistically significant difference was found between the studied groups. These findings support the results of some authors (11, 12, 14). In contrast, other authors (13, 15) found Increase in the number of oocytes retrieved this could be explained by the effect of Letrozole which improves ovarian response to gonadotropins.

Fertilization rate in the current study was lower in group A compared to group B(52% vs. 53%) which is comparable to the results of some authors(10). Furthermore the current present study could not demonstrate a statistically significant difference in number and quality of resulting embryos which supports the findings of some authors (11, 12). Ozmen et al (14) showed increased number of embryos but no difference in quality. No difference was found in the current study regarding implantation and pregnancy rates in contrary to the results found by some authors (10, 13, 15).

This study has some limitations and some points remain to be evaluated. First, it has low statistical power regarding the number of the studied ICSI cycles of poor-responders. Second, concerning the issue of Letrozole and congenital anomalies the limited number of delivered babies limited the ability to comment on possible congenital malformation developed in Letrozole group. There were no congenital anomalies detected in the current study which could add to the finding of Tulandi T. et al, who stated that the use of letrozole for ovulation induction was not associated with congenital malformations (16). The current study could be used as a base for planning a larger multicentric study for better evaluation of the role of Letrozole in the management of poor responders undergoing ICSI cycles.

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