

Endocrine changes and follicular development in patients during ovulation induction using Goserelin and different gonadotropin treatments

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Summary: The aim of this study was to compare the endocrine changes and the follicular development in patients receiving pure FSH alone or in association with LH after desensitization with an LH-RH agonist depot. Thirty four cycles were selected for this prospective randomized study. Desensitization was obtained using Goserelin the cycle before the stimulation. Induction of ovulation for IUI was carried out with 225 IU/day of pure FSH or with 225 IU/day of hMG.

The number of days and ampules required for follicular maturation were equivalent in the two groups. The same number of follicles were developed, while different, but not significant, pregnancy rates were obtained. Estradiol values at the end of stimulation were significantly lower for FSH group. In conclusion the contemporary administration of LH with FSH does not exert any effect on follicular development, but it seems to facilitate E2 synthesis, probably providing more substrate for the aromatization process.

Key words: Gonadotropins; Induction of ovulation; Steroidogenesis.

INTRODUCTION

Ovulation induction in assisted conception has long been carried out with the use of exogenous gonadotropins in addition to the endogenous secretion of Luteinizing Hormone (LH) and Follicle Stimulating Hormone (FSH).

Several studies were initiated to determine the relative importance of FSH and

LH in follicular development and Estradiol (E2) synthesis (^{1,2}).

The introduction of the use of Luteinizing Hormone Releasing Hormone (LH-RH) agonists to reduce or exclude the endogenous release of gonadotropins during induction of ovulation (³) provided the possibility of studying more precisely the role of the single pituitary hormones during follicular development (⁴).

The cooperation of both gonadotropins in ovarian steroidogenesis has already been described with the formulation of the "two-cell theory" (⁵). Recent reports have confirmed this hypothesis, suggesting that very low levels of LH may be required and are probably sufficient for E2 syn-

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thesis, thus supporting a primary role of FSH in ovarian steroidogenesis with an enhancing, critical role of LH (^{6, 7}).

A reevaluation of this theory has been proposed and diminished importance or none was attributed to LH in E2 production (^{1, 8}).

Moreover the relative importance of both gonadotropins in follicular development has still to be determined (⁹), although several studies suggest that preovulatory secretion of LH is not required for follicular growth (^{10, 11}).

The aim of our study was to compare the endocrine changes and the follicular development in patients receiving pure FSH alone or in association with LH after desensitization with an LH-RH agonist.

MATERIAL AND METHODS

Thirtytwo patients, for a total of 34 cycles, were selected for this study. No ovulatory dysfunction, tubal or uterine factor and/ or male factor were observed. Patients were enrolled in a program of artificial insemination, and cycles were randomly subdivided into two groups. Both groups received an LH-RH agonist depot Goserelin (Zoladex, ICI Pharma, Milan, Italy) as a single subcutaneous injection of 3.6 mg in the midluteal phase of the cycle preceeding the stimulation.

Induction of ovulation was started approximately two weeks later, when desensitization was completed. At that time E2 levels were <50 pg/ml, ultrasound monitoring did not reveal any follicle >5 mm in diameter and an evident endometrial growth.

Patients in the first (FSH) group were daily administered 225 IU i.m. of pure FSH (Metrodin, Serono, Rome, Italy) for five days. Patients in the second group (hMG group) were daily given 225 IU i.m. of human Menopausal Gonadotropins (hMG) (Pergonal, Serono, Rome, Italy) for five days. Dosages of FSH or hMG were then adjusted according to ultrasound monitoring.

The first day of gonadotropin administration was designated day 0. Human Chorionic Gonadotropin (hCG) (Profasi, Serono, Rome, Italy) at a dosage of 5,000 IU i.m. was administered when leading follicles reached 18 mm size in diameter.

Two intrauterine inseminations (IUI) were performed 12 and 36 hours later, using the "swim-up" technique of sperm preparation. Patients started the luteal phase support with daily i.m. injections of 50 mg of progesterone in oil on the day of the second IUI. Blood was taken every 2-3 days starting on the day of LH-RH agonist injection, during the desensitization, immediately before the gonadotropin treatment and during the ovarian stimulation until the day of hCG administration. All samples were stored at -20° C and assayed for FSH, LH, E2, Progesterone (P) and Prolactin (PRL).

Ultrasound and hormonal monitoring evidenced a relevant number of growing follicles in some cases. In these patients, considered at risk of hyperstimulation, hCG was withheld and the cycle was cancelled.

RESULTS

Mean age \pm standard deviation (SD) with duration of infertility in both groups are indicated in Table 1.

Two cycles were cancelled because of the risk of hyperstimulation, one in each group. One cycle was cancelled in FSH group because of a poor response. No hyperstimulation syndrome was observed in patients who completed the IUI cycle.

Characteristics of desensitization and ovarian stimulation are represented in Table 1. Patients of hMG group evidenced a higher pregnancy rate per completed cycle. Assays of PRL and P did not vary

Table 1.

	FSH group	hMG group
Age	30.9 \pm 2.7	31.4 \pm 3.6
Duration of infertility	2.3 \pm 0.6	2.6 \pm 0.8
Patients	17	15
Cycles	17	17
Cancelled cycles	2	1
Days of suppression	13.0 \pm 3.2	12.7 \pm 2.4
Days of stimulation	12.4 \pm 2.0	10.4 \pm 1.6
Ampules	40.2 \pm 7.5	35.0 \pm 8.0
Follicles	5.1 \pm 3.0	4.9 \pm 3.4
Pregnancies	1	5
Pregnancy/ completed cycle	6.6	31.2

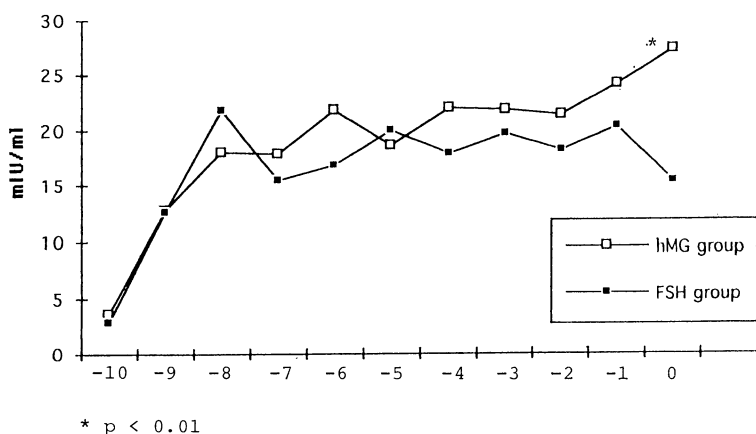


Fig. 1. — Mean values of FSH during ovarian stimulation.

during the desensitization or the stimulation periods.

Levels of FSH, LH and E2 during the stimulation period in both groups are represented in Figs. 1, 2 and 3 respectively. At the end of the stimulation E2 values were significantly lower in FSH group. Levels of FSH and LH were not statistically different.

DISCUSSION

The role of FSH and LH in follicular growth and steroidogenesis has recently been reevaluated and debated.

Initial studies demonstrated the importance of estradiol in folliculogenesis⁽⁹⁾. The "two-cell hypothesis" originally described by Short considered both gonadotropins essential for steroidogenesis, LH stimulating the androgen production of theca cells and FSH inducing granulosa cells to aromatize androgen precursors in estrogens⁽⁵⁾.

Thus LH seemed to play an essential role for both, steroidogenesis and follicular growth⁽⁹⁾.

On the contrary in a more recent report infertile patients stimulated first with

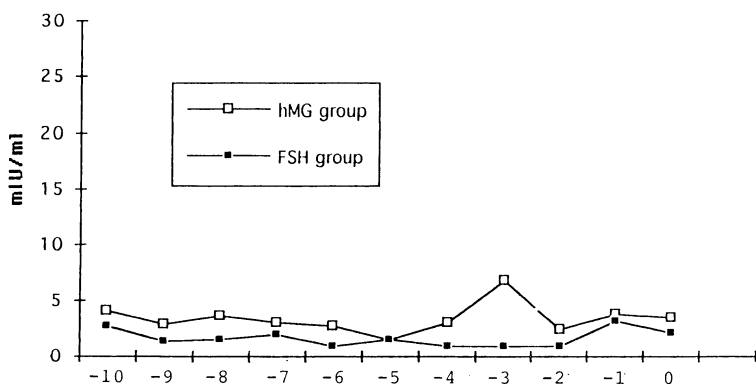


Fig. 2. — Mean values of LH during ovarian stimulation.

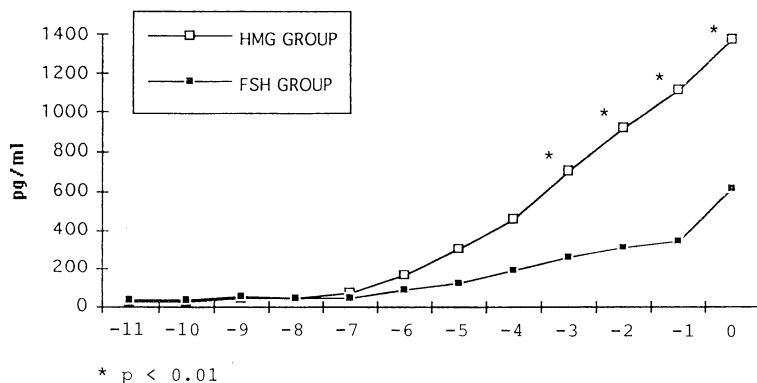


Fig. 3. — Mean values of E2 during ovarian stimulation.

hMG and second with FSH achieved similar E2 levels and follicular development⁽¹⁾. The authors concluded that the presence of LH was not important for steroidogenesis and folliculogenesis.

Both studies^(1,9) considered follicular maturation and steroidogenesis to be closely dependent, with estradiol playing a relevant role in granulosa cells proliferation.

The clinical introduction of LH-RH agonist^(3,12) and the experimental use of LH-RH antagonist⁽⁸⁾ provided the possibility of studying more precisely the influence on folliculogenesis and steroidogenesis of each gonadotropin exogenously administered in absence of an endogenous release. A study on a monkey model using LH-RH antagonist demonstrated that FSH treatment produced E2 levels not dissimilar from those obtained with hMG. Luteinizing hormone was supposed to play a minor role or none in steroidogenesis, leading to a possible reexamination of the "two-cell theory"⁽⁸⁾.

At the same time several researchers showed the relative importance or unimportance of LH in ovarian events leading to follicular maturation^(11,13). An interesting study of Galway using recombinant

FSH in hypophysectomized rats demonstrated a regular follicular growth in absence of LH⁽¹⁴⁾. Therefore the importance of LH was reexamined and E2 synthesis and folliculogenesis started to be considered as two distinct phenomena of ovarian physiology.

In our study desensitized patients developed the same number of follicles, of the same size, with FSH or hMG stimulation. Our results are in agreement with recent studies: LH does not seem to play any role in folliculogenesis. Follicular growth is an expression of the influence exerted by FSH only on its own receptors.

Furthermore we found that estradiol levels produced by FSH alone were lower than those obtained by the contemporary administration of LH, thus indicating some importance of LH in steroidogenesis.

In our study E2 was produced either by FSH or hMG, but the different amount of E2 detected at the end of induction of ovulation led to the hypothesis of an enhancing, stimulating role of LH in E2 biosynthesis. During the gonadotropin treatment in medically hypophysectomized patients, LH was regularly administered in the hMG group, while in the FSH group very low levels of LH were revealed, firstly because a minimal amount of LH

was given with pure FSH and secondly because the LH-RH agonist did not completely suppress pituitary function⁽¹⁵⁾. It might be possible that in the hMG group more androgenic substrate was available for aromatization into estrogens, and in the FSH group even minimal levels of LH could stimulate the theca cells to produce sufficient androgens, which would be converted by FSH into estrogens. This theory, supporting the "two-cell hypothesis", is in strict correlation with previous studies carried out in infertile patients with similar protocols^(10, 16), or in hypogonadotropic patients^(6, 7).

Different results have recently been published demonstrating no difference between desensitized cycles stimulated with FSH or hMG in regard to the E2 levels reached, indicating an equivalence of the two study groups^(4, 11, 17). In one of these studies Edelstein started the induction of ovulation with levels of LH three-four fold higher than levels found in our study at the beginning of stimulation⁽¹⁷⁾. It is possible to argue that during FSH stimulation more androgenic substrates synthesized by higher levels of LH and more estradiol is obtained at the end of stimulation.

Although the aim of this study was not the analysis of pregnancy rates, we obtained different results for the two protocols: we achieved more pregnancies, though not significantly, with hMG than using FSH. The small population could explain the absence of significance of the result, but similar data need more investigations to clarify this possible interesting aspect.

In summary, this study supports the hypothesis that LH does not play any role in folliculogenesis and, following the "two-cell theory", LH is essential, even at low levels, for steroidogenesis, augmenting the amount of E2 synthesized.

Now it is important to demonstrate whether there is a threshold level of LH

necessary to guarantee androgen and estrogen production.

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