Efficacy of luteinizing hormone activity in patients undergoing in vitro fertilization and treated only with low-dose recombinant choriogonadotropin alfa (Ovidrel) in the late follicular phase

Ten good prognosis patients underwent IVF with a new ovarian stimulation protocol using only 8 μg of recombinant choriogonadotropin and a GnRH antagonist starting at a 14-mm lead follicle. Six of the 10 subjects delivered and all had supernumerary embryos for cryopreservation. (Fertil Steril® 2006;86:1023–5. ©2006 by American Society for Reproductive Medicine.)

Controlled ovarian stimulation (COS) involves administration of gonadotropins to stimulate multiple follicular growth in assisted reproductive technology (ART). Follicle-stimulating hormone controls early ovarian follicle growth and maturation and stimulates the granulosa cell (GC) aromatase system, which catalyzes the conversion of androgen to estrogen (1). Furthermore, FSH induces the formation of LH receptors on the GCs.

The role of LH in ovulation is well established, but the role of LH in folliculogenesis and late oocyte maturation is not as clearly recognized (2). The concept of an LH threshold has been described, whereby follicular growth is suboptimal in the absence of a minimum LH level. Conversely, when the LH threshold is surpassed, further follicular development is halted (3). The LH threshold concept has been demonstrated in a series of experiments. Sullivan et al. (4) were the first investigators to show that E_2 output is not curtailed by the administration of recombinant LH alone in spite of withdrawing FSH. Filicori et al. (5) demonstrated that once ovarian stimulation has been initiated, it is possible to achieve continued follicular development independently of FSH administration. They also demonstrated LH activity administered as low dose urinary hCG can be used to stimulate the growth of large follicles and hasten the demise of small non-LH responsive follicles when administered in the mid/late follicular phase.

Similarly, Loumaye et al. (6) reported that a daily dose of 225 IU recombinant LH induced follicular arrest and selective follicular growth in anovulatory women who exhibited a hyper-response to COS with recombinant FSH. Although Filicori et al. used hCG and Loumaye et al. used recombinant LH in their studies, the similarity of their findings are not surprising ase hCG and LH interact with the same receptor.

The objective of this study was to assess the efficacy of late follicular phase LH-only activity, administered as recombinant hCG, in good prognosis IVF patients. Because these cycles conclude with pregnancy status and delivery, uterine effects of this novel ovarian stimulation protocol will be indirectly assessed.

The Institutional Review Board, John T. Mather Hospital, Port Jefferson, New York, approved this prospective noncomparative pilot study. All patients provided written consent.

Patients were between 28 and 34 years old, with a mean age of 30.6 years. The body mass index ranged from 20.8 to 33.6 kg/m^2 with a mean of 24.6 kg/m^2. All patients had normal uterine cavities demonstrated by either a saline hysterosonogram or hysteroscopy. Day 3 FSH level was $<$12 IU/L, and E_2 levels were $<$75 pg/mL in all patients at their initial workup. Three couples had coexisting male factor infertility. Two patients each had one previous IVF attempt.

The treatment protocol started with the administration of oral contraceptive (OC) pills from day 1 of normal or P withdrawal–induced menses. Oral contraceptives were given for a period of 14–28 days. On cycle days 2–4, recombinant human FSH (Gonal-f; Serono, Inc., Rockland, MA) was started at a dose of 225 IU/d. All subjects had a transvaginal sonogram and baseline measurement of FSH, E_2, β-hCG, LH, and P levels. The subjects were monitored according to standard IVF protocols by transvaginal sonogram to evaluate the size and growth of the ovarian follicles, and blood samples were collected to assess serum E_2, P, FSH, and LH levels. Once the largest follicle reached 12 mm, 0.25 mg of cetrorelix acetate (Cetrotide; Serono, Inc.) was started and 8 μg of recombinant hCG (choriogonadotropin alfa, r-hCG, Ovidrel; Serono, Inc.) was added to

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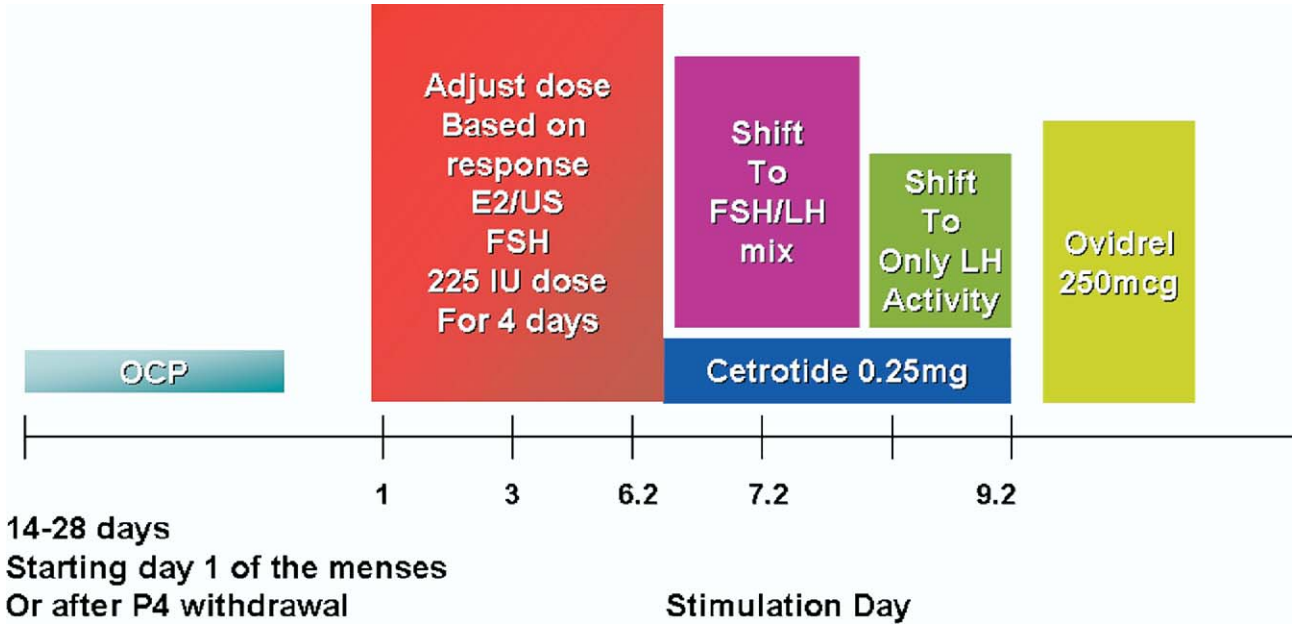
FIGURE 1

Induction protocol. OCP = oral contraceptive pill; P4 = progesterone; US = ultrasound.



Kenigsberg. Late follicular LH-only protocol in IVF. Fertil Steril 2006.

recombinant FSH daily. All the injections were administered SC. The induction protocol is illustrated in Figure 1.

When the largest follicle reached 14 mm in mean diameter, recombinant FSH was discontinued and low-dose recombinant hCG was continued at a dose of 8 μg along with cetrorelix. Final follicular maturation was triggered with 250 μg of recombinant hCG when ≥2 follicles reached 18 mm in mean diameter. Oocytes were retrieved 34.5 hours later; 8 of 10 patients had intracytoplasmic sperm injection (ICSI) on half of their mature oocytes, and all 8 were fertilized both with ICSI and standard IVF. All embryos were graded according to the percentage of fragmentation. Embryos of the highest grade were defined by having <20% fragmentation. Nontransferred embryos with ≥4 cells and <20% fragmentation were cryopreserved on postretrieval day 3.

Two embryos were transferred 3 days later. Progesterone supplementation was administered as 50 mg of P in oil IM from postretrieval days 1–4 and then P vaginal gel (Crinone 8%; Serono, Inc.) was substituted for IM P. Crinone was used once at night until the pregnancy test was performed (10–12 days after the embryo transfer).

At the time of administration of 250 μg of recombinant hCG, the number of follicles ranged from 5 to 24 with a mean of 15. The days of recombinant FSH administration ranged from 5 to 9 days with a mean of 6.3 days, whereas the average days of combination recombinant FSH and recombinant hCG administration were 1.3 with a range of 1–2 days. The administration of recombinant hCG alone ranged from 1 (one patient) to 3 days with a mean of 2.4 days. Cetrorelix administration ranged from 3 to 5 days with a mean of 3.8 days. Serum E_2 levels continued to increase after COS was switched to the recombinant FSH and recombinant hCG combination. Acceleration in the increase of E_2 levels was observed once stimulation consisted of 8 μg of recombinant hCG alone. The E_2 levels continued to increase after the administration of 250 μg of recombinant hCG.

One hundred sixty oocytes were retrieved, and 91% (145) were mature. Normal fertilization, defined as the presence of two pronuclei, was observed in 111 (76.5%) of 145 inseminated oocytes. Seventy-eight percent of all the embryos were of highest grade. A total of 79 embryos met the criteria for cryopreservation. The mean number of cryopreserved embryos was 7.9 for the pregnant patients and 8.75 for the nonpregnant patients. Two embryos of highest quality were transferred in all patients. All patients had trilaminar endometrial patterns on the day of hCG trigger. The endometrial thickness ranged from 7.4 to 13 mm with a mean of 10.23 mm.

Six of 10 patients achieved a delivery and a seventh patient had a chemical pregnancy. Three had twins and three delivered singletons. One patient started as a twin gestation and had a spontaneous loss of one fetus. There was no ovarian hyperstimulation syndrome (OHSS) necessitating additional medical encounters. Historically, our

good prognosis patients typically required a total dose of 2,250 IU of recombinant FSH with approximately 11 oocytes collected at the time of retrieval. In contrast, the novel protocol used in the current study used a lower total dose of recombinant FSH (1,417.5 IU) with a mean of 16 oocytes retrieved per patient. Substitution of LH activity in the form of recombinant hCG successfully completed ovarian stimulation and resulted in oocytes and embryos of excellent quality.

Theoretically, premature follicular luteinization and subsequent P production is a potential side effect of low-dose LH activity in the late follicular phase. We failed to observe unusually elevated P levels in the study subjects throughout the treatment cycle. Fifty percent of the 20 fresh embryos transferred implanted and 9 of the 10 implanted embryos have resulted in deliveries of viable infants. An embryo implantation rate of 50% is greater than would be expected from fresh day-3 embryos from standard IVF protocol. This supports the hypothesis that there are no harmful effects, such as premature decidualization or advancement of the endometrium, from low-dose recombinant hCG stimulation in the late follicular phase.

These results suggest that with an OC/GnRH antagonist stimulation protocol, very low dose recombinant hCG is capable of effectively completing follicular maturation at and beyond a lead follicle of 14 mm. Further studies could determine whether there might be a critical threshold at a smaller follicular size. The particular drug combination and dose is new and not previously described. A dose of 8 μg of recombinant choriogonadotropin corresponds to the 200 IU of hCG mostly used in other reports.

Future well-controlled, prospective, randomized studies using recombinant hCG or recombinant LH in the final days of COS should examine outcomes in patients with poor prognostic factors. Application of preimplantation genetic diagnosis to the embryos generated by this induction protocol will help to identify the aneuploidy rate in this group. The high implantation rates associated with improved embryo quality could permit the more widespread use of single embryo transfer to avoid multifetal gestations and yield future pregnancies from embryo cryopreservation. Other implications and directions for future studies include a decreased overall FSH dose for IVF stimulation and a low or absent incidence of OHSS.

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REFERENCES

1. Adashi EY, Rock JA, Rosenwaks Z. The ovarian follicular apparatus. Reproductive endocrinology, surgery and technology. Philadelphia: Lippincott-Raven, 1996:17–40.
2. Levy DP, Navarro JM, Schattman GL, Davis OK, Rosenwaks Z. The role of LH in ovarian stimulation: exogenous LH. Let's design the future. Hum Reprod 2000;15:2258–65.
3. Shoham Z. The clinical therapeutic window for luteinizing hormone in controlled ovarian stimulation. Fertil Steril 2002;77:1170–7.
4. Sullivan MW, Stewart-Akers A, Krasnow JS, Berga SL, Zeleznik AJ. Ovarian responses in women to recombinant follicle stimulating hormone and luteinizing hormone (LH): a role for LH in the final stages of follicle maturation. J Clin Endocrinol Metab 1999;84:228–32.
5. Filicori M, Cognigni GE, Tabarelli C, Pocognoli P, Taraborrelli S, Spettoli D, et al. Stimulation and growth of antral ovarian follicles by selective LH activity administration in women. J Clin Endocrinol Metab 2002;87:1156–61.
6. Loumaye E, Engrand P, Shoham Z, Hillier SG, Baird DT. Clinical evidence for an LH "ceiling" effect induced by administration of recombinant human LH during the late follicular phase of stimulated cycles in World Health Organization type I and type II anovulation. Hum Reprod 2003;18:314–22.