Adding luteinizing hormone to follicle stimulating hormone from day 3-5 improves pregnancy outcome in normal but not poor responders using gonadotropin releasing hormone antagonists

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Summary

Purpose: To determine if the addition of luteinizing hormone (LH) to follicle stimulating hormone (FSH) stimulation for controlled ovarian hyperstimulation (COH) protocols using gonadotropin releasing hormone (GnRH) antagonists improves pregnancy rates following in vitro fertilization-embryo transfer (IVF-ET). *Materials and Methods:* All IVF-ET cycles using a GnRH antagonist were evaluated according to whether FSH was used exclusively or if LH was added. The cycles were further stratified according to age (≤ 39 and 40-42 years) and according to good responders (≥ five oocytes retrieved) or poor responders (≤ four oocytes). *Results:* Combining all data, a significantly higher clinical and live delivered pregnancy rates were found in those adding LH (34.7% and 32.3%) vs those taking all FSH (33.4% and 25.8%). The only subgroup not showing this effect was the women aged 40-42 years with diminished oocyte reserve. *Conclusions:* LH should be added not only to COH protocols using GnRH agonists but also those using GnRH antagonists.

Key words: Luteinizing hormone; Follicle stimulating hormone; Gonadotropin releasing hormone antagonist; In vitro fertilization-embryo transfer.

Introduction

The introduction of highly purified follicle stimulating hormone (FSH) products where the presence of luteinizing hormone (LH) was minimized, and the later introduction of recombinant DNA technology where a completely pure FSH product could be manufactured, made it clear that except in circumstances of hypogonadotropic hypogonadism ovulation induction can be achieved in anovulatory women with FSH alone. This led to a large number of subsequent studies to determine if pregnancy rates are similar, higher, or lower with exogenous FSH stimulation alone vs LH and FSH together both in in vitro fertilization-embryo transfer (IVF-ET) cycles and non-IVF cycles.

To ascertain small significant differences, a study with sufficient power would be needed and it is unlikely that any given IVF center's prospective or retrospective data would be sufficiently large enough to demonstrate significant differences. Thus one method to gain sufficient power is to combine studies and perform a meta-analysis. One of the first meta-analyses performed evaluating this question was a Cochrane meta-analysis [1]. It found a clinical pregnancy rate of borderline significance in favor of LH/FSH combination in ovulation induction cycles and IVF-ET and thus recommended the use of the less expensive human menopausal gonadotropins (hMG) preparation [1].

Subsequently a larger Cochrane meta-analysis was published [2]. This study found a significantly higher ongoing pregnancy rate with highly purified hMG compared to recombinant FSH in women undergoing IVF-ET [2].

This meta-analysis led to the largest randomized controlled trial to date comparing menotrophin versus recombinant FSH in-vitro fertilization trial (MERIT). A trend was found for higher ongoing pregnancy rates with highly purified hMG vs recombinant FSH (rFSH) (27% vs 22%) [3].

A subsequent meta-analysis evaluating only gonadotropin releasing hormone (GnRH) agonist protocols involving 2,519 IVF cycles found a significantly higher live birth rate with hMG (25.5%) vs rFSH (21.6%) [4].

A meta-analysis by Al-Inany et al included both long GnRH agonists and short GnRH antagonist protocols and still found higher pregnancy rates with hMG [5]. There are less data concerning the benefits of having LH in the controlled ovarian hyperstimulation (COH) protocol when one exclusively uses a GnRH antagonist protocol. Although meta-analyses always favor prospective studies, and there is always the possibility of potential selection biases when performing a retrospective study, there are still meaningful conclusions that can be reached from a large retrospective study. This is especially true if one is evaluating a group not likely to ever be studied in a prospective study.

The objective of this study was to evaluate whether adding LH to FSH stimulation increases the pregnancy

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outcome following IVF-ET in women with diminished oocyte reserve using a GnRH antagonist protocol. Furthermore, similar comparisons of women using all FSH vs LH added would be retrospectively compared in a large group of women with normal oocyte reserve.

Materials and Methods

A retrospective review of all IVF cycles using GnRH antagonists were used from January 2, 2003 through April 30, 2010. To increase the statistical power, all cycles were included so that a woman not conceiving previously could have been used multiple times. Though some would claim that using the same couple more than once could introduce a bias, on the other hand, studies only using a couple's first IVF cycle may select a population with a better prognosis. By including all cycles, this study would include women with a worse prognosis since they could have failed to conceive despite several previous cycles.

The data were stratified not only according to poor responders (poor responder arbitrarily assigned to a group having four or less oocytes retrieved) vs normal responders, but according to two age groups (≤ 39 and 40-42 years). There were no restrictions for day 3 serum FSH and estradiol (E2) levels. Cycles having intracytoplasmic sperm injection were included.

Comparisons were made using Chi-square analysis and included clinical pregnancy rate/transfer (ultrasound evidence of pregnancy at eight weeks) viable pregnancy rate (live fetus at 12 weeks), and live delivered pregnancy rates and implantation rate.

The relative amount of LH to FSH could have been as little as 25% and as much as 50%. The antagonists were either cetrorelix or ganirelix and were used at a dosage of 250 mcg, once at least one follicle reached an average diameter of 14 mm.

If day 3 serum FSH was > 12 mIU/ml or serum E2 > 50 pg/ml, or a diminished antral follicle count was found on day 3, there was a marked reduction in the dosage of gonadotropins [6, 7].

Results

The comparison of pregnancy outcome according to taking all FSH vs FSH plus LH in normal responders using a GnRH antagonist is shown in Table 1.

The comparison of pregnancy outcome according to taking all FSH vs FSH plus LH in poor responders using a GnRH antagonist is shown in Table 2.

Combining all data (all ages and good and poor responders), there was a significantly higher clinical pregnancy rate in those taking FSH plus LH (423/1,066, 39.7%) vs those taking all FSH (248/743, 33.4%) with p = 0.007, chi-square analysis.

Significantly higher viable and live delivered pregnancy rates were also seen in the FSH plus LH group (35.5% and 32.3% vs 28.3% and 25.8%) with p = 0.007 and p = 0.0038, respectively.

If one evaluates separately the poor responder group (Table 2), no significant differences were found or even a trend for superiority of COH regimens adding LH to the FSH. For poor responders, the pregnancy rates for those taking all FSH vs LH added to FSH were: clinical pregnancy rate/transfer – 25.5% (103/404) vs 26.4% (60/227), viable pregnancy rate/transfer – 20.5% vs 22.5%, live delivered pregnancy rate/transfer – 17.8% vs 20.3% (Chi-square analysis = no significance). Evaluating the percentage of first IVF cycles in each group, there were 350/814 (43.0%) first cycles in normal responders taking FSH and LH vs 150/337 (44.5%) of those women using FSH exclusively.

Table 1. — *The comparison of pregnancy outcomes following IVF-ET according to taking all FSH vs FSH plus LH in normal responders using a GnRH antagonist protocol.*

	Antagonist cycles w/FSH only		Antagonist cycles w/FSH and LH started at the same time	
Age at time of retrieval (years)	≤ 39.9	40-42	≤ 39.9	40-42
# retrievals	337	68	814	205
# transfers	281	58	678	161
Average age (years)	34.0	41.2	34.1	41.2
Avg. E2 day of hCG (pg/ml)	1,913.7	1,366.8	2,184.4	1,916.7
Avg. P day of hCG (ng/ml)	1.5	1.3	1.4	1.4
% fertilized	64.4	67.5	67.3	65.4
% clinical preg./transfer	44.8	32.8	45.9	31.1
% viable preg./transfer	40.6	22.4	41.6	28.0
% miscarriage/clinical preg.	13.5	42.1	17.0	20.0
% delivered/ongoing	38.8	19.0	38.1	24.8
Avg. number embryos transferred	2.8	3.5	2.7	3.5
Implantation rate (%)	25.0	10.8	26.5	12.6

Table 2. — *The comparison of pregnancy outcomes following IVF-ET according to taking all FSH vs FSH plus LH in poor responders using a GnRH antagonist protocol.*

	Antagonist cycles w/FSH only		Antagonist cycles w/FSH and LH started at the same time	
Age at time of retrieval (years)	≤ 39.9	40-42	≤ 39.9	40-42
# retrievals	363	244	201	130
# transfers	245	159	143	84
Average age (years)	36.6	41.7	36.4	41.3
Avg. E2 day of hCG (pg/ml)	551.1	473.3	681.4	699.1
Avg. P day of hCG (ng/ml)	0.9	0.8	1.0	1.0
% fertilized	71.1	70.0	69.2	72.4
% clinical preg./transfer	28.6	20.8	30.1	20.2
% viable preg./transfer	24.5	14.5	28.0	13.1
% miscarriage/clinical preg.	24.3	42.4	11.6	52.9
% delivered/ongoing	21.6	11.9	26.6	9.5
Avg. number embryos transferred	1.7	1.6	1.9	2.0
Implantation rate (%)	20.6	13.8	19.4	11.6

Discussion

A recent Cochrane meta-analysis evaluated the effect of recombinant LH for COH in 14 prospective trials involving 2,612 women. However 11 of the 14 trials involved 2,396 women using a GnRH agonist and thus only three trials involving 216 women used a GnRH antagonist [8].

Only seven of the 13 trials provided ongoing pregnancy rates (only three provided live births) and this Cochrane analysis did not find that the addition of LH improved the outcome. A priori, because of prolonged pituitary suppression, one might suspect that adding LH would be even more important for GnRH agonist cycles which rep-

resented the majority of patients in this study [8]. Interestingly three trials in this Cochrane meta-analysis used only poor responders and this sub-group did show that adding LH improved pregnancy outcome [8-11]. Though retrospective, the present study included 1,810 women which thus had five times the power of the three prospective studies evaluating GnRH antagonist protocols. Only two of the 14 prospective studies in the recent Cochrane meta-analysis evaluated the live birth rates and these two studies used GnRH agonists not antagonists. The data in this study provided the live birth rates.

Theoretically women taking GnRH agonists should have a greater need for addition of LH to FSH stimulation, compared to GnRH antagonists because of prolonged pituitary suppression and longer recovery to producing LH when using the former. Yet this retrospective data showed a significant 20% increase in live delivered pregnancy rates when adding LH to FSH from the early follicular phase using a GnRH antagonist protocol for COH.

Retrospective studies have several inherit flaws, so one has to favor conclusions made from well-designed prospective studies. However, frequently prospective studies are flawed by type I statistical errors, i.e., insufficient power. Theoretically, combining several prospective studies and evaluating these combined data as in a meta-analysis can obviate type I errors; however until there are sufficient numbers of prospective studies evaluating addition of LH with GnRH antagonist protocols, the data from larger retrospective studies in deciding COH protocols may be considered.

Using more than one treatment cycle per patient in a retrospective study adds a potential bias that if one group has more first cycles, that group could be favored. However, there was no favoring of first cycles for FSH only vs LH plus FSH and the data from the largest of the groups was provided under results. The authors purposely included all cycles not only to increase the power but to include more difficult patients, i.e., those failing to conceive in previous IVF-ET cycles. One of the problems in prospective studies is that they tend to exclude more difficult cases and favor the ones with the best prognosis.

It is interesting that with the recent meta-analysis by Mochtar *et al*., the only subgroup that improved with LH and FSH were the poor responders [8]. In contrast in this retrospective study, this group did not show an advantage of adding LH to FSH. There was the same 20% increase in live delivery pregnancy rates in poor responder women ≤ 39.9 years adding LH, so the lack of significance could be from a type I error. However it should be noted that in women 40-42 years of age, there was a 20% decreased live delivered pregnancy rate in those taking LH and FSH.

References

[1] Van Wely M., Westergaard L.G., Bossuyt P.M., Van der Veen F.: "Human menopausal gonadotropins versus recombinant follicle stimulation hormone for ovarian stimulation in assisted reproductive cycles". Cochrane Database of Systematic Reviews, 2003, CD003973.

[2] Platteau P., Smitz J., Albano C. *et al*.: "Exogeneous luteinizing hormone activity may influence the treatment outcome in in-vitro fertilization but not in intracytoplasmic sperm injection cycles". *Fertil. Steril*., 2004, *81*, 1401.

[3] Andersen A.N., Devroey P., Arce J.C.: "Clinical outcome following stimulation with highly purified hMG or recombinant FSH in patients undergoing IVF: a randomized assessor-blind controlled trial". *Hum. Reprod*., 2006, *21*, 3217.

[4] Coomarasamy A., Afnan M., Cheema D. *et al*.: "Urinary hMG versus recombinant FSH for controlled ovarian hyperstimulation following an agonist long down-regulation protocol in IVF or ICSI treatment: a systematic review and meta-analysis". *Hum. Reprod.,* 2008, *23,* 310.

[5] Al-Inany H.G., Abou-Setta A.M., Aboulghar M.A., Serour G.I.: "Efficacy and safety of human menopausal gonadotrophins versus recombinant FSH: a meta-analysis". *Reprod. BioMed. Online*, 2008, *16,* 81.

[6] Check J.H., Summers-Chase D., Yuan W., Horwath D., Wilson C.: "Effect of embryo quality on pregnancy outcome following single embryo transfer in women with a diminished egg reserve". *Fertil. Steril.,* 2007, *87,* 749.

[7] Check J.H.: "Mild ovarian stimulation". *J. Assist. Reprod. Genet*., 2007*, 24,* 621.

[8] Mochtar M.H., Van der Veen F., Ziech M., van Wely M., Musters A.: "Recombinant luteinizing hormone (rLH for controlled ovarian hyperstimulation in assisted reproductive cycles (Review)". *Cochrane Database Syst. Rev*., 2007, *18,* CD005070.

[9] Ferraretti A.P., Gianaroli L., Magli M.C., D'Angelo A., Farfalli V., Montanaro N.: "Exogeneous luteinizing hormone in controlled ovarian hyperstimulation for assisted reproductive techniques". *Fertil. Steril*., 2004, *82,* 1521.

[10] De Placido G., Alviggi C., Perino A., Strina I., Lisi F., Fasolino A. *et al*.: "Recombinant human LH supplementation versus recombinant human FSH (rFSH) step-up protocol during controlled ovarian stimulation in normogonadotrophic women with initial inadequate ovarian response to rFSH. A multicentre, prospective, randomized controlled trial". *Hum. Reprod*., 2005, *20,* 390.

[11] Barrenetxea G., Agirregoika J.A., Lopez de Larruzea A., Ganzabal T., Jimenez M.R., Carbonero K. *et al*.: "LH supplementation in ovarian stimulation in poor responder women. A randomized controlled trial". Abstracts of the 22nd Annual Meeting of the ESHRE, i55, 2006.

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