"Recombinant human follicle-stimulating hormone (r-hFSH) plus recombinant luteinizing hormone versus r-hFSH alone for ovarian stimulation during assisted reproductive technology: Systematic review and meta-analysis"

Lehert, Philippe ; Kolibianakis, Efstratios M. ; Venetis, Christos A. ; Schertz, Joan ; Saunders, Helen ; Arriagada, Pablo ; Copt, Samuel ; Tarlatzis, Basil

Abstract

Background: The potential benefit of adding recombinant human luteinizing hormone (r-hLH) to recombinant human follicle-stimulating hormone (r-hFSH) during ovarian stimulation is a subject of debate, although there is evidence that it may benefit certain subpopulations, e.g. poor responders.Methods: A systematic review and a meta-analysis were performed. Three databases (MEDLINE, Embase and CENTRAL) were searched (from 1990 to 2011). Prospective, parallel-, comparative-group randomized controlled trials (RCTs) in women aged 18-45 years undergoing in vitro fertilization, intracytoplasmic sperm injection or both, treated with gonadotrophin-releasing hormone analogues and r-hFSH plus r-hLH or r-hFSH alone were included. The co-primary endpoints were number of oocytes retrieved and clinical pregnancy rate. Analyses were conducted for the overall population and for prospectively identified patient subgroups, including patients with poor ovarian response (POR).Results: In total, 40 RCTs (6...

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Among patients treated with FSH and GnRH analogues for *in vitro* fertilization, is the addition of recombinant LH associated with the probability of live birth? A systematic review and meta-analysis

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The aim of this systematic review and meta-analysis was to assess whether the addition of recombinant luteinizing hormone (LH) increases live birth rate, among patients treated with follicle stimulating hormone (FSH) and gonadotrophin-releasing hormone (GnRH) analogues for *in vitro* fertilization (IVF). Eligible studies were randomized controlled trials (RCTs) answering the research question that contained sufficient information to allow ascertainment of whether randomization was true and whether equality was present between the groups compared, regarding baseline demographic characteristics, gonadotrophin stimulation protocol, number of embryos transferred and luteal phase support administered. A literature search identified seven RCTs (701 patients) that provided the information of interest, among which five reported agonist and two antagonist cycles. The reported outcome measure, clinical pregnancy, was converted to live birth using published data in one study. No significant difference in the probability of live birth was present with or without rLH addition to FSH (odds ratio [OR]: 0.92, 95% confidence interval (CI): 0.65–1.31; $P = 0.65$). This finding remained stable in subgroup analyses that ordered the studies by dose of rLH added, the type of analogue used to inhibit premature LH surge, the time rLH was added during the follicular phase, the age of patients analysed, the presence of allocation concealment and by the way the information on live birth was retrieved. In conclusion, the available evidence does not support the hypothesis that the addition of recombinant LH increases the live birth rate in patients treated with FSH and GnRH analogues for IVF.

Keywords: luteinizing hormone; GnRH agonists; GnRH antagonists; live birth rate

Introduction

The role of endogenous LH levels during ovarian stimulation has attracted a lot of attention since the early in-vitro fertilization (IVF) years (Stanger and Yovich, 1985; Howles *et al.*, 1987; Thomas *et al.*, 1989). At present, available evidence suggests that among women with normal ovulation or World Health Organization (WHO) II oligo-anovulation, low endogenous LH levels during ovarian stimulation for IVF using gonadotrophin-releasing hormone (GnRH) analogues are not associated with a decreased probability of ongoing pregnancy beyond 12 weeks (Kolibianakis *et al.*, 2006).

On the basis of these data, an adverse role of low endogenous LH levels on the probability of pregnancy cannot serve as a rationale for LH supplementation in ovarian stimulation for IVF. However, it cannot be excluded that LH supplementation during the follicular phase might be beneficial for pregnancy achievement, independently of any effect of endogenous LH levels. Several studies have so far evaluated the addition of recombinant LH (rLH) to FSH in ovarian stimulation for IVF (Table 1). Due to sample size restrictions, however, these individual studies are usually not conclusive as regards the effect of LH supplementation on pregnancy likelihood.

The purpose of the current systematic review and meta-analysis was to summarize the available published evidence regarding the role of rLH addition in ovarian stimulation for IVF by answering the following clinical question: among patients treated with FSH and GnRH analogues for IVF, does the addition of rLH increase live birth rate?

Materials and Methods

Identification of studies

In April 2006, a computerized literature search was performed on the bibliographic databases EMBASE, MEDLINE and CENTRAL without time limitations. Additionally, references of retrieved articles were hand-searched. The search strategy aimed at identifying randomized controlled trials (RCTs) on the basis of the following clinical question: among patients treated with FSH and GnRH analogues is the addition of rLH associated with the probability of live birth? Search terms used were 'luveris', 'lutropin alfa', 'recombinant LH' and 'recombinant luteinizing hormone'. The search was limited to RCTs in humans. Meeting proceedings were not considered, since unpublished studies cannot be adequately evaluated for their design and quality. Moreover, it has been shown that although there is a considerable publication deficit in reproductive medicine for RCTs, there is no concomitant publication bias (Evers, 2000).

Selection of studies

Criteria for inclusion/exclusion of studies were established prior to the literature search. Eligible studies were RCTs answering the research question that contained sufficient information to allow ascertainment of whether randomization was true and whether equality was present between the groups compared, regarding baseline demographic characteristics, gonadotrophin stimulation protocol, number of embryos transferred and luteal phase support (LPS) administered. An effort was made to contact all authors or sponsors of studies to retrieve missing or additional information.

Studies were excluded if no down-regulation was used for ovarian stimulation for IVF or if the gonadotrophin used for ovarian stimulation contained LH. Language of publication or number of patients analysed was not amongst the exclusion criteria.

Studies identified

Literature search yielded 74 studies that were potentially able to answer the research question. Further evaluation based on study titles and abstracts and/or assessment of full manuscripts resulted in 15 studies that evaluated the role of LH addition on the probability of live birth. Thirty-seven studies were excluded after screening the titles and a further 22 were excluded after screening the abstracts. The question of interest was answered in seven of these studies (Tables 1 and 2). Eight studies were excluded because a quasi-randomization was used for patient allocation ($n = 2$), because randomization method was unclear ($n = 1$), because, besides rLH addition, the dose of FSH was also modified at the same time ($n = 3$) or because problems occurred during the study period ($n = 2$) (Table 3).

Data extraction

The following data were recorded from each of the studies in parallel by two of the authors (L.K., E.M.K.): demographic (type of study, country of origin and period of enrolment), methodological (randomization method, allocation concealment, randomization ratio, whether sample size calculation was performed), procedural (whether financial support was declared, number of patients included,

Table 1: Characteristics of the RCTs included in the meta-analysis

Study, country of origin (number of centres)	Journal	Study period	Randomization method; allocation concealment	Sample size calculation	Number of participants	Financial support by pharmaceutical company
Sills *et al.* (1999), USA (single centre)	Hum Reprod	February 1996–November 1996	Randomization table; no	No	ITT population: 30; hpFSH: 17; hpFSH + rLH: 13	Yes
Balasch *et al.* (2001), Spain (single centre)	J Assist Reprod Genet	Not stated	Computer-generated randomization table; yes (sealed envelopes)	No	ITT population: 30; rFSH: 14; rFSH + rLH: 16	Yes
Humaidan *et al.* (2004), Denmark (single centre)	Reprod Biomed Online	November 2001–October 2002	Computer-generated randomization table; yes (sealed envelopes)	Yes, for pregnancy rate	ITT population: 231; rFSH: 115; rFSH + rLH: 116	Not stated
Sauer *et al.* (2004), USA (mutlicentre)	Reprod Biomed Online	Not stated	Computer-generated randomization table; no	No	ITT population: 73; rFSH: 24; rFSH + rLH: 25	Yes
Griesinger *et al.* (2005), Germany (single centre)	Hum Reprod	June 2003–May 2004	Computer-generated randomization table; yes (sealed envelopes)	Yes, for the duration of gonadotrophin treatment (days)	ITT population: 127; rFSH: 65; rFSH + rLH: 62	Not stated
Tarlatzis *et al.* (2006), multinational (multicentre)	Hum Reprod	Not stated	Computer-generated randomization table; yes (sealed envelopes)	Yes, for metaphase II oocytes retrieved	ITT population: 114; rFSH: 59; rFSH + rLH: 55	Yes
Fabregues *et al.* (2006), Spain (single centre)	Fertil Steril	November 2003–September 2004	Computer-generated randomization table; yes (sealed envelopes)	Yes, for pregnancy rate	ITT population: 120; rFSH: 60; rFSH + rLH: 60	Not stated

hpFSH: highly purified follicle stimulating hormone, rFSH: recombinant follicle stimulating hormone, rLH: recombinant luteinizing hormone, ITT: intention to treat.

Table 2: Characteristics of the RCTs included in the meta-analysis

Study	GnRH analogue protocol; analogue (dose)	FSH type in control group; starting dose; dose adjustments	Gonadotrophin treatment regimen in the study group	hCG	Criteria for hCG administration	Fertilization	ET day	LPS
Sills *et al.* (1999)	Long luteal; leuprorelin (1 or 0.5 mg/day, s.c.)	hpFSH; 150–450 IU; yes: step down protocol	hpFSH protocol as in control group, plus 75 IU rhLH daily from initiation of stimulation (no rLH dose adjustments)	Up to 10 000 IU hCG	At least two follicles $\geq$17 mm	IVF or ICSI	Day 3	Progesterone
Balasch *et al.* (2001)	Long luteal; leuprorelin (1 or 0.5 mg/day, s.c.)	rFSH; 450 IU; yes: step-down protocol	rFSH protocol as in control group, plus 75 IU rhLH daily from initiation of stimulation (no rLH dose adjustments)	5000 IU hCG	Consistent E_2 rise observed in the presence of $\geq$2 follicles $>$18 mm	IVF or ICSI	Not stated	hCG
Humaidan *et al.* (2004)	Long luteal; buserelin (0.5 or 0.2 mg/day, s.c.)	rFSH; 150–300 IU; yes, from day 8 onwards if necessary	rFSH protocol as in control group, plus rLH from day 8 onwards (rFSH and rLH given in 2 : 1 ratio)	10 000 IU hCG	At least three follicles $\geq$17 mm	IVF or ICSI	Day 2, 3 or 5	Progesterone
Sauer *et al.* (2004)	(Pretreatment with OCP-single dose antagonist; cetrorelix (3 mg s.c.)	rFSH; 225 IU; yes, from day 6 onwards if necessary	rFSH protocol as in control group, plus 150 IU rLH from day of cetrorelix initiation onwards (no rLH dose adjustments)	250 μg rhCG	At least one follicle $\geq$18 mm, at least two other follicles $\geq$16 mm and serum E_2 $\sim$150 pg/ml per mature follicle	ICSI	Not stated	Progesterone
Griesinger *et al.* (2005)	Cetrorelix starting on stimulation day 6 (0.25 mg/day)	rFSH; 150 IU; yes, from day 6 onwards if necessary	rFSH protocol as in control group, plus 75 IU rLH. rLH dose adjustments from day 6 according to rFSH adjustment	250 μg rhCG	Three follicles $\geq$18 mm	IVF or ICSI	Day 2	Progesterone and hCG
Tarlatzis *et al.* (2006)	Long follicular; buserelin (0.2 mg/day, s.c.)	rFSH; 150 IU; yes, from day 6 onwards if necessary	rFSH protocol as in control group, plus 75 IU rLH when leading follicle reached 14 mm (no rLH dose adjustments)	10 000 IU hCG	At least two follicles $>$17 mm	IVF or ICSI	Day 2	Progesterone
Fabregues *et al.* (2006)	Long luteal; triptorelin (0.1 or 0.05 mg/day, s.c.)	rFSH; 450 IU; yes, step down protocol	rFSH protocol as in control group, plus 150 IU rhLH from day 6 onwards (no rLH dose adjustments)	250 μg hCG	$\geq$2 follicles $\geq$18 mm, with $\geq$4 follicles $\geq$14 mm in association with consistent rise in E_2	IVF or ICSI	Day 2 or 3	Progesterone

OCP: oral contraceptive pill, rFSH: recombinant follicle stimulating hormone, rLH: recombinant luteinizing hormone, hCG: human chorionic gonadotrophin, IVF: in vitro fertilization, ICSI: intra cytoplesmic sperm injection, rhCG: recombinant hCG, s.c.: subcutaneous, hpFSH: highly purified follicle stimulating hormone, LPS: luteal phase support, GnRH: gonodotrophin releasing hormone, E_2: estradiol, ET: embryo transfer.

type and protocol of ovarian stimulation, type of gonadotrophin administered, criteria for human chorionic gonadotrophin (hCG) administration and dose of hCG, type of fertilization, day of embryo transfer and type of luteal support administered) and outcome data (live birth rate, duration of stimulation, dose of FSH required, fertilization rate, number of cumulus–oocyte complexes (COCs) retrieved, estradiol (E_2) and progesterone level on the day of hCG administration). Where standard deviation (SD) was not reported by the authors, it was calculated from the standard error of the mean (SEM). The values reported as SDs in the study by Fabregues *et al.* (2006) were considered as SEMs following communication with the authors. Any disagreement between the persons responsible for data extraction was solved by discussion.

Where live birth was not reported in a study that fulfilled the inclusion criteria, an effort was made to contact the corresponding authors to retrieve the missing information. If this was not possible, the reported outcome measure, clinical pregnancy, was converted to live birth using published data (84% probability of live birth after

Table 3: Studies excluded from the meta-analysis

Study, country	Main reason for exclusion
	Quasi-randomization
Lisi *et al.* (2002), Italy	Allocation of treatment to every third patient
Cedrin-Durnerin *et al.* (2004), France	In centers 2 and 3, patients randomization according to the even or uneven year of the woman's birth.
Toporcerova *et al.* (2005), Slovenia	Unclear method of randomization
	Simultaneous systematic change of FSH at LH initiation
Ferraretti *et al.* (2004), Italy	Increase of FSH dose in addition to rLH initiation in both study arms
Levi-Setti *et al.* (2005), Italy	Decrease of FSH dose in the patients that received rLH
De Placido *et al.* (2005), Italy	Increase of FSH dose at LH initiation in patients that did not receive rLH
	Problems during the study period
Lisi *et al.* (2005), Italy	Treatment was not performed according to random allocation in 34 patients
Marrs *et al.* (2004), USA	Significantly different number of embryos were transferred in the two arms of the study

FSH: follicle stimulating hormone, rLH: recombinant luteinizing hormone.

confirmation of clinical pregnancy with a living fetus at the 7th week of gestation) (Arce *et al.*, 2005). This live birth equivalent was calculated in one study: Sauer *et al.* (2004). In addition, information was sought from the corresponding authors regarding live birth in patients who were randomized but did not start treatment within the study period. Where this information was not available (Griesinger *et al.*, 2005), these patients were considered as not pregnant ($n = 1$).

Outcome variables

The primary outcome variable was live birth rate per randomized patient (described as delivery rate in the study by Sills *et al.*, 1999). Secondary outcome variables were clinical pregnancy rate, gonadotrophin consumption (per cycle); duration of stimulation (per cycle), E_2 level on the day of hCG, progesterone level on the day of hCG, number of COCs retrieved (per cycle), fertilization rate (per cycle) and number of 2PN oocytes (per cycle).

Quantitative data synthesis

Study features and results were assembled in tabular form, and a formal meta-analysis was performed. The dichotomous data results for each study were expressed as an odds ratio (OR) with 95% confidence intervals (CI). These results were combined for meta-analysis with Comprehensive Meta-analysis software (Biostat, 14 North Dean St, Englewood NJ 07631, USA), using the Mantel/Haenszel method. Live birth rate was calculated for each study per patient randomized.

When the outcome of interest was of a continuous nature, the differences were pooled across the studies that provided information on this outcome variable, resulting in a weighted mean difference (WMD) with 95% CI.

Study-to-study variation was assessed by using χ^2 statistic (the hypothesis tested was that the studies are all drawn from the same population, i.e. from a population with the same effect size). A fixed effects model was used where no heterogeneity was present, whereas in presence of significant heterogeneity a random effects model was applied.

Subgroup analyses were carried out to check the stability of the main finding. These analyses ordered the studies according to the dose of rLH added (75 IU, 150 IU), the type of analogue used to inhibit premature LH surge (agonist, antagonist), the time rLH was added during the follicular phase (early follicular phase, mid-follicular phase), the age of patients analysed (>35 years of age, all ages), the type of allocation concealment (allocation concealed, concealment unknown) and according to the way the information on live birth was retrieved (reported by authors or calculated).

Power analysis

It was calculated that the optimal information size required to reject the null hypothesis (no difference in live birth rates between the two treatment groups) was 2504 subjects, assuming a clinically important difference of 5%, a baseline live birth rate of 25% and using beta 0.2, alpha 0.05 and a two-tailed hypothesis test.

Results

Seven RCTs fulfilled the inclusion criteria and were included in the analysis with no disagreement noted between the authors responsible for study selection. Characteristics of the included studies are listed in Tables 1 and 2. All analysed studies were published between 1999 and 2006. The size of the studies ranged from 29 to 231 patients (median 114), whereas a total of 701 patients were analysed (FSH only: $n = 354$, FSH + rLH: $n = 347$). Characteristics of the patients included in these trials are shown in supplementary Table 1. Two studies were multicentre trials and all others were single-centre studies. In five studies, patient allocation was concealed, whereas in the remaining studies concealment of allocation had not been performed or was not reported.

All the studies were double arm with the exception of the study by Sauer *et al.* (2004). In this three-armed study, data were extracted for analysis from the study arms that provided the information with respect to the study research question.

Financial support by a pharmaceutical company was declared in four out of the seven analysed studies. Two out of the seven studies reported a power analysis aiming to detect differences in the probability of pregnancy achievement (Table 1).

To inhibit premature LH surges, agonists were used in five studies (leuprorelin: $n = 2$, daily triptorelin: $n = 1$, buserelin $n = 2$) and antagonists in two studies (cetrorelix daily: $n = 1$, single dose cetrorelix: $n = 1$).

For ovarian stimulation, one study used highly purified gonadotrophins, whereas the remaining studies used recombinant gonadotrophins. Criteria for triggering final oocyte maturation varied

across studies and were based on follicular data ($n = 4$) or on a combination of follicular data and hormonal levels ($n = 3$). In three studies, recombinant hCG was used for triggering final oocyte maturation, whereas the remaining studies used urinary hCG (10 000 IU in three studies; 5000 IU in one study).

Fertilization methods included both IVF and intracytoplasmic sperm injection (ICSI). Embryo transfers were performed 2–5 days after oocyte retrieval. Luteal support varied between studies. The majority of the studies analysed used micronized progesterone for luteal support.

Live birth rate

The OR for live birth was 0.92 (95% CI: 0.65 to 1.31, $P = 0.65$; heterogeneity: $P = 0.39$, fixed effects model), suggesting that the probability of live birth was not associated with the addition of rLH to FSH stimulation (Fig. 1). The rate difference was 1.5% in favour of the FSH only group (95% CI: −7.7 to +4.85, $P = 0.65$; heterogeneity $P = 0.32$, fixed effects model).

Subgroup analyses of the likelihood of live birth ordered the studies by dose of rLH added (75 IU, 150 IU), the time rLH was added during the follicular phase (early follicular, mid-cycle), the category of analogue used for inhibition of premature LH surge (agonist, antagonist), the age of the patients analysed (all ages, >35 years of age), the type of allocation concealment (allocation concealed, concealment unknown) and the way the information on live birth was retrieved (reported by authors or calculated). As illustrated in Figures 2–3 and supplementary Figures 1–4 the difference in live birth rate between patients treated with FSH only and those treated with FSH + rLH remained not significant in all subgroup analyses. In none of these analyses was heterogeneity present, and thus a fixed effects model was used.

In five studies, information was reported both for live birth and for clinical pregnancy. rLH addition was not associated with either the probability of clinical pregnancy (OR: 0.85, 95% CI: 0.57–1.26) or live birth (OR: 0.94, 95% CI: 0.62–1.43) in these studies.

Repeating the analysis with the three excluded quasi-randomized studies (Lisi *et al.*, 2002; Cedrin-Durnerin *et al.*, 2005; Toporcerova *et al.*, 2005) sums 1414 patients and does not materially change the OR for live birth: 1.05 (95% CI: 0.82–1.36, $P = 0.69$; heterogeneity: $P = 0.50$, fixed effects model). The rate difference was 0.9% in favour of the FSH + LH group (95% CI: −3.7 to +5.5, $P = 0.89$; heterogeneity $P = 0.39$, fixed effects model).

The OR for clinical pregnancy in the included studies was 0.86 (95% CI: 0.61–1.20, $P = 0.37$; heterogeneity: $P = 0.35$, fixed

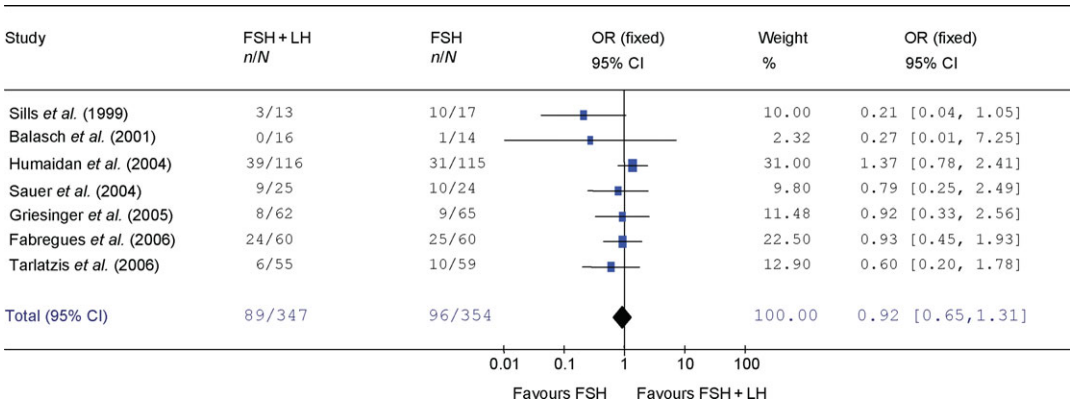

Figure 1: OR of live birth rate per randomized patient (heterogeneity: $P = 0.39$)

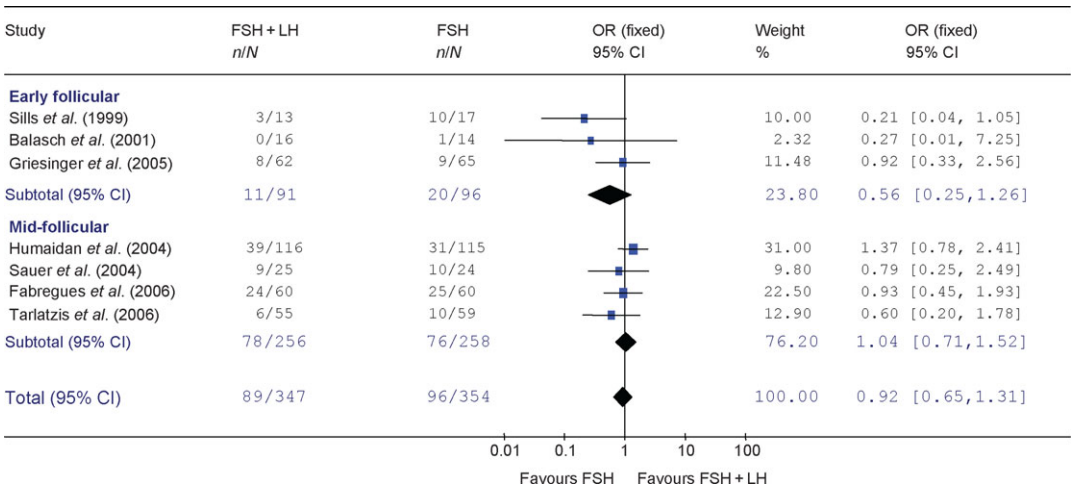

Figure 2: OR of live birth rate per randomized patient according to time during the follicular phase that rLH supplementation was initiated (heterogeneity in different groups: early follicular: $P = 0.28$; mid-follicular: $P = 0.53$)

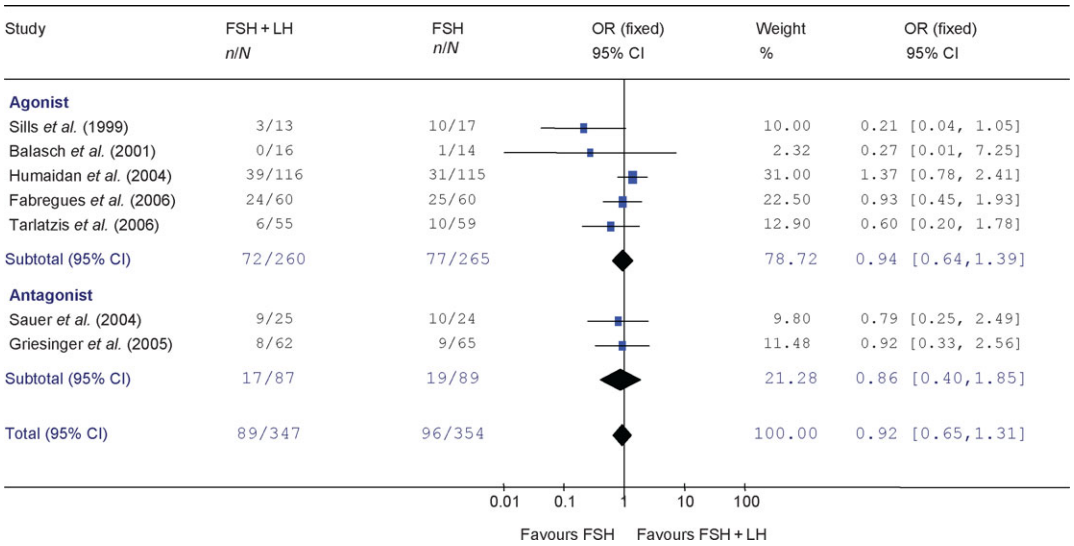

Figure 3: OR of live birth rate per randomized patient according to type of analogue used for ovarian stimulation (heterogeneity in different groups: agonist: $P = 0.18$; antagonist: $P = 0.84$)

effects model), suggesting that the probability of clinical pregnancy was not associated with the addition of rLH to FSH stimulation. The rate difference was 3.0% in favour of the FSH only group (95% CI: −9.5 to +3.5, $P = 0.36$; heterogeneity $P = 0.34$, fixed effects model).

Secondary outcomes

FSH requirement

No significant difference was detected regarding the number of units of FSH required for ovarian stimulation in patients treated with FSH + rLH as compared with those treated with FSH only (WMD: +64.44 IU, 95% CI: −51.15 to +180.02; $P = 0.27$; heterogeneity: $P = 0.69$, fixed effects model). Six studies offered data for this outcome measure.

Duration of FSH stimulation

The duration of stimulation was not significantly different between patients treated with FSH + rLH as compared with those treated with FSH only (WMD: −0.04 days, 95% CI: −0.31 to +0.23; $P = 0.77$; heterogeneity: $P = 0.19$; fixed effects model). Seven studies offered data for this outcome measure.

E_2 on the day of hCG administration

Serum E_2 was not significantly higher in patients treated with FSH and rLH as compared with those treated with FSH only (WMD: +214.42 pg/ml, 95% CI: −107.93 to +536.78 $P = 0.019$; heterogeneity: $P = 0.04$; random effects model). Six studies offered data for this outcome measure.

Progesterone on the day of hCG administration

Only two studies offered data on the level of serum progesterone on the day of hCG, which was not significantly different between patients treated with FSH + rLH as compared with those treated with FSH only (WMD: +0.06 ng/ml, 95% CI: −0.18 to +0.29 $P = 0.63$; heterogeneity: $P = 0.38$; fixed effects model).

COCs retrieved

The number of COCs retrieved was not significantly different between patients treated with FSH + rLH as compared with those treated with FSH only (WMD: −0.57 COCs, 95% CI: −1.46 to +0.31 $P = 0.21$; heterogeneity: $P = 0.42$; fixed effects model). Seven studies offered data for this outcome measure.

Fertilization rate

Fertilization rate was not significantly different between patients treated with FSH + rLH as compared with those treated with FSH only (WMD: −4.15%, 95% CI: −9.24 to +0.93 $P = 0.11$; heterogeneity: $P = 0.24$; fixed effects model). This result is based on the analysis of four studies.

2PN oocytes

The mean number of 2PN oocytes was not significantly different between patients treated with FSH only and those treated with FSH and rLH (WMD: +0.03 2PN oocytes, 95% CI: −0.51 to +0.58; $P = 0.90$; heterogeneity: $P = 0.38$, fixed effects model). This result is based on the analysis of five studies.

Discussion

The current systematic review suggests that, among patients treated with FSH and GnRH analogues for IVF, the addition of rLH does not increase live birth rate.

It has to be noted that this result should be viewed with caution since the optimal sample size, required to exclude a clinically significant difference, has not yet been reached. Thus the addition of further RCTs to examine the effect of adding rLH to FSH in ovarian stimulation for IVF is necessary. Nevertheless, the main finding of the current meta-analysis is supported by the fact that it remained stable in all subgroup analyses performed, whereas no significant heterogeneity was present between the RCTs analysed.

Interestingly, no trend for a beneficial effect of rLH addition was present depending on the dose of rLH added to FSH stimulation. As yet, an optimal dose of rLH has not been systematically assessed in normogonadotropic women (De Placido *et al.* 2004).

The current evidence suggests that an improvement of the probability of live birth does not depend on the addition of either 75 or 150 IU of rLH.

In addition, the time that rLH addition was initiated during the follicular phase (early or mid-follicular), did not affect the probability of live birth. It should be noted that the three studies in which rLH was started during the early follicular phase did not favour such an intervention (Fig. 2). Finally, the type of down-regulation (agonist or antagonist) did not seem to modify the effect of rLH addition to FSH. The two studies performed using GnRH antagonists for down-regulation did not favour the addition of rLH to FSH.

In the current meta-analysis, one study out of the seven analysed reported only clinical pregnancy rates for the population analysed, whereas information on live birth was not available. In this study, live birth was calculated using published data (Arce *et al.* 2005). This choice was made in order to include this eligible trial in the current meta-analysis, since otherwise it would have to be ignored. The assumption for this calculation is that, following confirmation of clinical pregnancy, the probability of live birth is not different between patients that received rLH and those that did not receive rLH in addition to FSH. No published data exist to object to this assumption, which is further supported by the fact that no significant effect of rLH addition was detected either for clinical pregnancy or for live birth in the studies that reported both outcomes.

Regarding secondary outcomes no significant differences were observed between patients who received rLH and those who did not, which is in line with the non-significant difference observed in the probability of live birth.

In conclusion, the current review suggests that, among patients treated with FSH and GnRH analogues for IVF, the addition of rLH does not increase live birth rate. Moreover, the available evidence does not indicate a beneficial effect of rLH addition on secondary outcome variables.

Conflict of interest

Dr G.G. and Dr B.C.T. are co-authors of two of the randomized controlled studies included in the current meta-analysis (Griesinger *et al.*, 2005; Tarlatzis *et al.*, 2006).

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