Reproductive BioMedicine Online (2010) **21**, 66–76

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ARTICLE

Corifollitropin alfa for ovarian stimulation in IVF: a randomized trial in lower-body-weight women

The corifollitropin alfa Ensure study group [1]

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Abstract In this double-blind, double-dummy, randomized, equivalence trial (Ensure), 396 women weighing 60 kg or less who underwent controlled ovarian stimulation prior to IVF or intracytoplasmic sperm injection were randomized in a 2:1 ratio to a single dose of 100 μg corifollitropin alfa or daily 150 IU recombinant FSH (rFSH) for the first 7 days of stimulation in a gonadotrophin-releasing hormone antagonist protocol. The mean ± SD number of oocytes retrieved per started cycle was 13.3 ± 7.3 for corifollitropin alfa versus 10.6 ± 5.9 for rFSH. The estimated treatment difference of +2.5 oocytes (95% CI 1.2—3.9) in favour of corifollitropin alfa ($P < 0.001$) was well within the predefined equivalence margin. The median (range) duration of stimulation was 9 (6—15) days in both groups. In 32.8% of the patients, one injection of corifollitropin alfa was sufficient to reach the human chorionic gonadotrophin criterion. The incidence of moderate and severe ovarian hyperstimulation syndrome was 3.4% for corifollitropin alfa and 1.6% for rFSH. A dose of 100 μg corifollitropin alfa offers a simplified treatment option for potential normal responder patients with a lower body weight.

KEYWORDS: body weight, corifollitropin alfa, follicle stimulant, IVF, ovarian stimulation, sustained

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1472-6483/$ - see front matter
doi:10.1016/j.rbmo.2010.03.019

Introduction

Corifollitropin alfa is a novel recombinant hormone designed as a sustained follicle stimulant. A single dose administered in the early follicular phase of the menstrual cycle initiates and sustains multiple follicular development for 7 days (Corifollitropin Alfa Dose-Finding Study Group, 2008). Corifollitropin alfa contains the alfa-subunit of human FSH coupled to a hybrid subunit composed of the sequence of the β-subunit of human FSH and the carboxy-terminal peptide of the β-subunit of human chorionic gonadotrophin (HCG) (Fares et al., 1992; Fauser et al., 2009). The available preclinical and clinical data on corifollitropin alfa show that the compound has a prolonged half-life and a slower absorption to serum peak concentrations. Therefore, a single dose of corifollitropin alfa remains effective for a whole week in contrast to recombinant human FSH (rFSH), which is to be injected daily (Bouloux et al., 2001; Duijkers et al., 2002; Fares et al., 1992). The efficacy of corifollitropin alfa has initially been investigated in a small feasibility trial (Devroey et al., 2004), followed by a larger multicentre dose-finding trial in women undergoing ovarian stimulation for IVF or intracytoplasmic sperm injection (ICSI) (Corifollitropin Alfa Dose-Finding Study Group, 2008). The results of the dose-finding trial indicated a significant dose—response relationship with respect to the number of cumulus—oocyte—complexes retrieved.

The phase II data were combined with historical data in a modelling and simulation project that was initiated to predict the effects of a range of single doses of corifollitropin alfa followed by daily rFSH treatment for ovarian stimulation. By taking various variables (including age and body weight) into account, the modelling revealed that 100 μg is the most optimal corifollitropin alfa dose in the desired 1-week regimen for women with a body weight up to and including 60 kg and provides an exposure similar to the exposure provided by 150 μg in women weighing more than 60 kg. Equal exposure to those two dosages would also imply equal ovarian response (De Greef et al., 2007).

The anticipated therapeutic indication for corifollitropin alfa is ovarian stimulation for the development of multiple follicles and pregnancy in women participating in an assisted reproductive technology programme. Corifollitropin alfa has been developed in a gonadotrophin-releasing hormone (GnRH) antagonist protocol (Al-Inany et al., 2007; Hohmann et al., 2003; Tarlatzis et al., 2006). The application of a simplified GnRH antagonist protocol has been suggested as the preferred option for predicted normal responders (Devroey et al., 2009a). This treatment regimen may reduce the psychological distress and the drop-out rates of IVF patients (Heijnen et al., 2007; Verberg et al., 2008). It is anticipated that the replacement of the first seven injections of daily FSH by a single injection of corifollitropin alfa may further lower the injection burden of patients undergoing ovarian stimulation (Fauser et al., 2009).

In the Engage trial, more than 1500 patients were treated either with 150 μg corifollitropin alfa or 200 IU rFSH in a standardized GnRH antagonist protocol. The pregnancy rates (38.9% versus 38.1%) confirmed equal efficacy in terms of ongoing pregnancy rates (Devroey et al., 2009b).

The main objective of this comparative, double-blind trial (Ensure) in patients undergoing ovarian stimulation prior to IVF or ICSI was to assess the efficacy and safety of a lower dose of 100 μg corifollitropin alfa in patients weighing up to 60 kg, using a fixed daily dose of 150 IU rFSH as a reference.

Materials and methods

The Ensure trial was a multicentre, multinational, randomized, double-blind, double-dummy equivalence trial involving 14 centres in Europe (three in Austria, two each in Czech Republic, France, Spain, Poland and Sweden and one in Denmark) and five centres in Asia (three in Korea and two in Taiwan). The study was approved by the local health authorities and the independent medical ethics committee for each centre and conducted in accordance with the Declaration of Helsinki, the International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use (ICH) Guidelines and Good Clinical Practice. Written informed consent was provided by all patients.

Patients

Women aged 18—36 years, weighing 60 kg or less, a body mass index of 18—32 kg/m^2, a normal menstrual cycle length (24—35 days), access to ejaculatory spermatozoa and an indication for ovarian stimulation for IVF or ICSI were eligible to enrol in the study. The exclusion criteria were the same as those reported in the Engage trial (Devroey et al., 2009b); thus, patients with a history of ovarian hyperresponse to ovarian stimulation (more than 30 follicles ≥11 mm) or ovarian hyperstimulation syndrome (OHSS), polycystic ovary syndrome or more than 20 basal antral follicles on ultrasound (<11 mm, both ovaries combined) were excluded. Similarly, patients with a history of no or low ovarian response (i.e. cycle cancelled due to insufficient response or less than four oocytes obtained) or more than three unsuccessful ovarian stimulation cycles since the last established ongoing pregnancy were excluded.

Study design

The Ensure trial was designed as a multicentre, multinational, randomized, double-blind, double-dummy equivalence trial. After evaluation of screening data to confirm eligibility, patients were randomized just before the start of stimulation. Randomization to one of the two treatment groups in a 2:1 ratio (investigational group: reference group) was performed at each centre and stratified by age (<32 or ≥32 years) and planned fertilization procedure (IVF or ICSI) by central remote allocation using randomly permutated blocks with an undisclosed fixed block size of three.

The treatment regimen is depicted in **Figure 1**. All patients were to start ovarian stimulation on day 2 or 3 of their menstrual cycle with a single subcutaneous (s.c.) injection of 100 μg (0.5 ml) corifollitropin alfa (N.V. Organon, The Netherlands) or placebo injection. Injections could be given

by the patient herself, her partner or the medical staff. To conceal treatment allocation, all patients also started daily s.c. injection of rFSH (150 IU/day) or placebo on the same day (stimulation day 1) using the Puregon/Follistim Pen (N.V. Organon). Daily active or placebo rFSH injections were continued through the first 7 days of stimulation. The chosen reference dose of 150 IU rFSH daily was fixed for the first 5 days of stimulation, but could be reduced or increased up to a maximum of 200 IU from day 6 onwards. If no follicle ≥11 mm was visible on ultrasound scan (USS) before injection on day 8, the cycle was to be cancelled due to insufficient ovarian response. From day 8 onwards, treatment with (open-label) rFSH was continued in both the investigational and reference group and the dose could be reduced or increased from 150 IU based on the observed follicular response (maximum rFSH dose 200 IU/day) up to the day of HCG administration. The investigator was allowed to withhold rFSH administration for a maximum of 3 days (coasting) up to and including the day of HCG administration. If, in the opinion of the investigator, the ovarian response was too high, the investigator was allowed to cancel the cycle at any time. However, in case of a risk for OHSS, i.e. more than 30 follicles ≥11 mm on USS, HCG was to be withheld and the treatment cycle was to be cancelled per protocol. Starting on stimulation day 5, all patients were scheduled to receive 0.25 μg ganirelix acetate injection (Orgalutran; N.V. Organon) up to and including the day of HCG to prevent premature LH surges. To induce final oocyte maturation 10,000 IU HCG (Pregnyl; N.V. Organon), or 5000 IU HCG in case of a high ovarian response, was to be given when three follicles ≥17 mm in mean diameter were observed by transvaginal USS. Investigators were allowed to delay HCG administration for 1 day when preferred for practical reasons. All IVF or ICSI procedures after HCG administration were equal to those described by Devroey et al. (2009b) for the Engage trial. At embryo transfer, which took place 3 or 5 days after oocyte retrieval, a maximum of two embryos could be transferred. Daily progesterone (at least 600 mg/day vaginally or at least 50 mg/day intramuscularly, to be prescribed locally) for luteal phase support was to be administered for at least 6 weeks if pregnant or until menses.

Assessments

USS assessments were performed on stimulation days 1, 3, 5 and 8 and then daily up to and including the day of HCG. Serum FSH, LH, oestradiol, progesterone and inhibin B were analysed prior to injection on days 1, 3, 5 and 8, day of HCG, day of embryo transfer and at the visit 2 weeks after embryo transfer or at cycle discontinuation and 2–3 weeks after cycle discontinuation.

Local injection site tolerance was assessed by each centre's medical staff at 30 min after drug administration on stimulation day 1. Injection site reactions were scored none, mild, moderate or severe for four parameters (pain, itching, swelling and redness). Assessment of serum anti-corifollitropin alfa antibodies was performed by radioimmunoassay on day 1 prior to injection and 2 weeks after embryo transfer or at cycle discontinuation and 2–3 weeks after cycle discontinuation.

Validated immunoassays (Devroey et al., 2004) were performed at a central laboratory (MSD, Oss, The Netherlands and Waltrop, Germany) to measure serum concentrations of corifollitropin alfa, FSH, LH, oestradiol, progesterone, inhibin B and anti-corifollitropin alfa antibodies.

A pregnancy test (serum or urinary HCG) was performed at least 2 weeks after embryo transfer. In case of a pregnancy, USS was performed at 5–6 weeks and at ≥10 weeks after embryo transfer to establish the presence of an ongoing pregnancy.

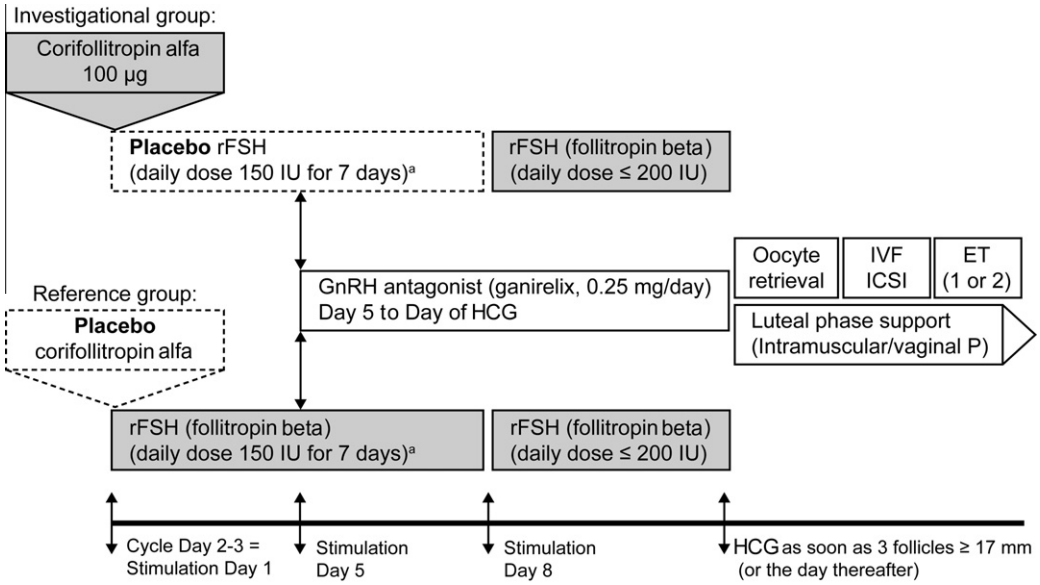

Figure 1 Graphical illustration of the treatment regimens applied in this double-blind, double-dummy trial. ET, embryo transfer; GnRH, gonadotrophin-releasing hormone; HCG, human chorionic gonadotrophin; ICSI, intracytoplasmic sperm injection; P, progesterone; rFSH, recombinant FSH. [a]Only when required, in the opinion of the investigator, the rFSH dose could be adjusted from day 6 onwards.

Study endpoints and statistics

The primary objective was to show that the corifollitropin alfa regimen, in terms of the number of cumulus–oocyte–complexes retrieved, was equivalent to the reference treatment (predefined equivalence range: −3 to +5 oocytes). The number of oocytes was chosen as the primary endpoint of the trial, as it reflects best the pharmacological effect of the new corifollitropin alfa regimen, which in comparison to daily FSH should not provide less than three oocytes (which usually result in one good-quality embryo) and not more than five oocytes as that could increase the risk of OHSS significantly. Randomizing patients in a 2:1 ratio (twice as many patients in the investigational group as in the reference group), a total of at least 330 patients (220 patients in the investigational group, 110 patients in the reference group) ensured 90% power of the trial, assuming a standard deviation of almost 8 for the number of oocytes retrieved. The treatment groups were formally compared with analysis of variance for the number of cumulus–oocyte–complexes, including covariates treatment group, age ($<$32 or $\geq$32 years), planned fertilization procedure (IVF or ICSI) and centre.

Descriptive statistics, including mean and standard deviation, were calculated for other endpoints, including dose of rFSH required from day 8 to the day of HCG administration, serum FSH, LH, oestradiol, inhibin B and progesterone concentrations, number and size distribution of follicles ($\geq$11, $\geq$15 and $\geq$17 mm) during stimulation and on the day of HCG administration, number and quality of oocytes, fertilization rate (defined as 100 times the ratio of the number of fertilized two pronuclei (2PN) oocytes obtained and the number of oocytes used for fertilization), number and quality of embryos, implantation rate (defined as 100 times the maximum number of gestational sacs as assessed by any USS after embryo transfer divided by the number of embryos transferred (per subject), maximized to 100%), miscarriage rate and pregnancy rate. Patients who received corifollitropin alfa or rFSH but did not have embryo transfer were considered to be cancelled. The percentage of cancelled patients was compared between the treatment groups using Fisher's exact test.

Occurrence of (serious) adverse events, including moderate and severe OHSS as per the World Health Organization criteria (WHO, 1973), outcome of local tolerance and immune response assessments were evaluated as safety endpoints. The percentage of patients with moderate or severe OHSS was compared between the treatment groups using Fisher's exact test.

All efficacy analyses were based on the intent-to-treat (ITT) population, which included all randomized patients who received corifollitropin alfa or at least one dose of rFSH. Patients were grouped based on the treatment to which they had been randomized. One patient treated with rFSH was not randomized via the interactive voice response system and is therefore not part of the ITT population. Safety analyses were performed on the all-subjects-treated group, which comprised all the patients who received either corifollitropin alfa or rFSH, with patients grouped according to the active treatment that they actually received.

Results

Patient characteristics and disposition

A total of 396 patients were randomized (2:1 ratio) and treated: 268 patients with corifollitropin alfa and 128 patients with rFSH (**Table 1**). The two treatment groups were

Table 1 Patient disposition.

	100 μg corifollitropin alfa	*150 IU rFSH*
Randomized	268 (100)	128 (100)[a]
Started stimulation	268 (100)	128 (100)
Cancelled	2	1
	Insufficient ovarian response (investigator opinion) (n = 1)	Too high ovarian response (n = 1)
	Patient's decision (n = 1)	
Treated with HCG	266 (99.3)	127 (99.2)
Oocyte retrieval	266 (99.3)	127 (99.2)
Cancelled	20	7
	Risk of OHSS (n = 1)	Too high ovarian response (n = 1)
	Too high ovarian response (n = 5)	No/too few/bad quality oocytes retrieved (n = 2)
	No/too few/bad quality oocytes retrieved (n = 2)	No or abnormal fertilization (n = 2)
	No or abnormal fertilization (n = 4)	No/too few/bad quality embryos for transfer (n = 1)
	No/too few/bad quality embryos for transfer (n = 7)	No fertilization possible (n = 1)
	Suspicious pulmonary tuberculosis (n = 1)	
Embryo transfer	246 (91.8)	120 (94.5)

Values in brackets are percentages. HCG, human chorionic gonadotrophin; OHSS, ovarian hyperstimulation syndrome; rFSH, recombinant FSH.
[a]Excluding one patient who was not randomized via the interactive voice response system, but was treated.

comparable with respect to demographics, fertility characteristics, lifestyle characteristics, ultrasound findings and hormone profiles (Table 2). The mean age ± SD of the patients included in this trial was 31.0 ± 3.1 years and their mean body weight was 54.2 ± 4.2 kg. In total, 44.4% of all patients were Asian. Overall, the average duration of infertility was 3.2 ± 2.2 years, 61.9% of the patients presented with primary infertility and 56.8% of the patients had no previous IVF cycle. The most frequently reported cause of infertility was male factor (49.5%). On stimulation day 1, the number of basal antral follicles was comparable between the treatment groups and serum LH and FSH concentrations were normal for the early follicular phase.

Three patients who started ovarian stimulation did not receive HCG: two in the corifollitropin alfa group (0.7%) and one in the rFSH group (0.8%). Overall, in 22 (8.2%) patients in the corifollitropin alfa group and eight (6.3%) in the rFSH group, the cycle was cancelled (i.e. did not have embryo transfer). The cancellation rates in both treatment groups were low and not statistically significantly different between the groups (Fisher's exact test). The main reason for cycle cancellation was 'No/too few/bad quality embryos for transfer', occurring in 2.6% and 0.8% of patients of the corifollitropin alfa group and the rFSH group, respectively (Table 1).

Primary endpoint

The mean ± SD number of cumulus–oocyte–complexes retrieved per started cycle in the ITT group was 13.3 ± 7.3 in the corifollitropin alfa group versus 10.6 ± 5.9 in the reference group. The estimated treatment difference was +2.5 oocytes in favour of corifollitropin alfa ($P < 0.001$). With a 95% confidence interval of 1.2–3.9, the two treatment groups are considered equivalent based on the predefined equivalence range of (−3, +5) oocytes, despite any statistical significance of the difference.

Table 2 Demographics, fertility and lifestyle characteristics and baseline (stimulation day 1) ultrasound scan and serum hormone concentrations per treatment group (intent-to-treat population).

	100 μg corifollitropin alfa (n = 268)	*150 IU rFSH (n = 128)*
Demographics		
Age (years)	30.9 ± 3.2	31.1 ± 3.0
Body weight (kg)	54.1 ± 4.2	54.4 ± 4.2
Body mass index (kg/m^2)	20.5 ± 1.5	20.6 ± 1.6
Race		
Asian	120 (44.8)	56 (43.8)
Black	1 (0.4)	0 (0.0)
Caucasian	147 (54.9)	72 (56.3)
Fertility characteristics		
Primary infertility	60.8	64.1
Duration of infertility (years)	3.2 ± 2.2	3.3 ± 2.1
Cause of infertility[a]		
Male factor	127 (47.4)	69 (53.9)
Tubal factor	70 (26.1)	31 (24.2)
Endometriosis	32 (11.9)	11 (8.6)
Cervical mucus problems	3 (1.1)	1 (0.8)
Unexplained fertility	74 (27.6)	33 (25.8)
Other	7 (2.6)	2 (1.6)
No previous IVF cycles	55.2	60.2
Lifestyle characteristics		
Smoking (max. five/day)	20 (7.5)	10 (7.8)
Alcohol consumption	65 (24.3)	31 (24.2)
Stimulation day 1		
Total ovarian volume (ml)	10.7 ± 6.2	10.4 ± 5.4
Number of basal antral follicles (<11 mm)	11.1 ± 4.4	11.4 ± 4.3
FSH (IU/l)	6.5 (1–16)	6.6 (2–15)
LH (IU/l)	4.5 (<0.6–13)	4.1 (<0.6–15)

Values are mean ± SD, number (%), % or median (range). rFSH, recombinant FSH.
[a]A patient can have multiple causes of infertility.

Other endpoints

Pharmacokinetic evaluation of the serum corifollitropin alfa concentrations revealed an average elimination half-life ($t_{1/2}$) of 73.1 h. The time to reach the maximum serum concentration (t_{max}) was 46.2 h.

Figure 2 presents serum rFSH immunoreactivity during ovarian stimulation for corifollitropin alfa and daily rFSH. Upon starting stimulation with a single subcutaneous 100 μg corifollitropin alfa injection, median serum rFSH immunoreactivity showed a rapid increase at day 3 (median value: 26.3 IU/l) and a rapid decline thereafter to reach a median value of 10.1 IU/l at day 8. In the daily rFSH group, median serum FSH immunoreactivity increased until day 5 (10.2 IU/l) and then reached a plateau to a median value of 10.3 IU/l at day 8. From day 8 onwards, there were no relevant differences in median serum FSH immunoreactivity between the two treatment groups.

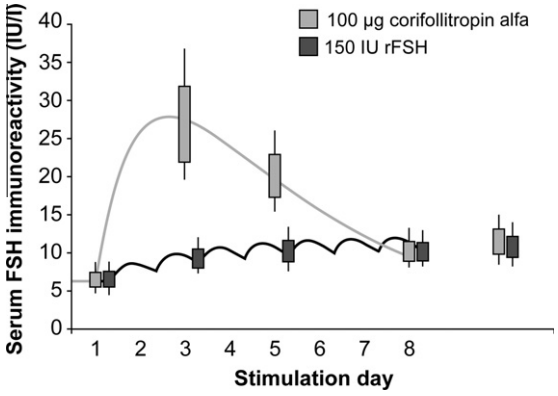

Figure 2 Serum recombinant FSH (rFSH) immunoreactivity during ovarian stimulation for the corifollitropin alfa regimen and the reference treatment with daily rFSH. The boxes represent interquartile ranges and the whiskers represent 10–90th centiles. The lines represent typical time profiles of FSH immunoreactivity for a subject with a body weight of 55 kg (simulated from available data).

The median duration of stimulation was 9 days in both treatment groups (Table 3); thus, after corifollitropin alfa injection on average only 2 days of rFSH were required until HCG administration. The median total amount of rFSH required from day 8 to HCG administration was 300 IU for the corifollitropin alfa group and 275 IU for the rFSH treatment group (Table 3). Figure 3 presents the frequency distribution of the day when HCG criteria were met in each treatment group. One-third (32.8%) of the patients in the corifollitropin alfa group met the criterion for HCG injection before or on stimulation day 8. From stimulation day 8 onwards, a fixed dose of 150 rFSH per day was used in 56.1% of the corifollitropin alfa group and in 68.5% of the rFSH group. Per protocol, the dose of (placebo) rFSH could be reduced or increased from stimulation day 6 onwards. Dose decreases on stimulation days 6 and/or 7 were recorded more often in the corifollitropin alfa group (6.4%) than in the rFSH group (0.8%), while this was vice versa for dose increases (3.4% versus 6.4%, respectively). Most patients (all-subjects-treated group) received 10,000 IU HCG (80.2% and 89.1% of the patients in the corifollitropin alfa and rFSH group, respectively) and a dose of 5000 IU HCG was used for 19.0% and 10.1% of the respective groups.

In the corifollitropin alfa group, slightly more follicles (≥11 mm) were recruited during the first week of stimulation compared with the rFSH group (Table 3). On stimulation day 8, the mean ± SD number of follicles ≥11 mm was 11.8 ± 6.1 in the corifollitropin alfa group and 10.6 ± 5.3 in the rFSH group. On the day of HCG, the total number of follicles ≥11 mm had increased to 14.9 ± 6.6 follicles in

Table 3 Stimulation characteristics and follicle growth (intent-to-treat population).

	100 μg corifollitropin alfa (n = 268)	*150 IU rFSH (n = 128)*
Stimulation characteristics[a]		
Total duration of stimulation (days)	9 (6–15)	9 (6–15)
Total dose of rFSH (IU)	300 (0–1550)	1350 (825–2650)
Total dose of rFSH from day 8 onwards (IU)	300 (0–1550)	275 (0–1600)
Patients reaching HCG criterion on or before stimulation day 8 (%)	32.8	39.8
Follicles, stimulation day 8		
≥11 mm	11.8 ± 6.1	10.6 ± 5.3
≥15 mm	5.0 ± 4.6	5.1 ± 4.5
≥17 mm	2.0 ± 3.0	2.4 ± 3.1
Follicles on day of HCG[a]		
≥11 mm	14.9 ± 6.6	12.9 ± 5.8
≥15 mm	9.4 ± 4.9	8.5 ± 4.4
≥17 mm	5.3 ± 3.0	5.1 ± 3.0

Values are median (range), % or mean ± SD unless otherwise stated. HCG, human chorionic gonadotrophin; rFSH, recombinant FSH.
[a]Restricted to patients with human chorionic gonadotrophin injection.

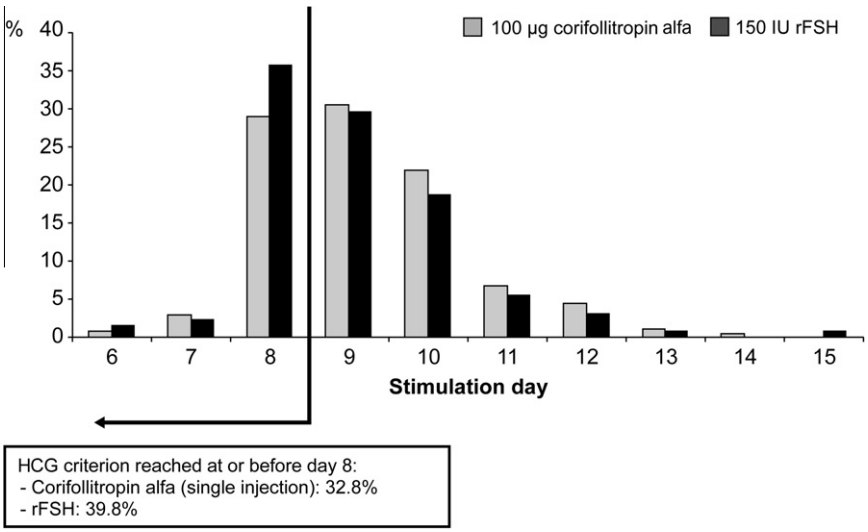

Figure 3 Graphical presentation of the percentage of patients per stimulation day reaching the human chorionic gonadotrophin (HCG) criterion (as soon as three follicles ≥17 mm) (intent-to-treat group). rFSH, recombinant FSH.

the corifollitropin alfa group and to 12.9 ± 5.8 follicles in the rFSH group.

Serum concentrations of oestradiol, inhibin B, LH and progesterone for all patients who received HCG are shown in **Figure 4A—D**. During the first 5 days of stimulation, serum oestradiol and inhibin B concentrations tended to increase more rapidly in the corifollitropin alfa group than in the rFSH group. From stimulation day 5 to stimulation day 8, serum oestradiol and inhibin B concentrations continued to rise in both treatment groups, but the increase was

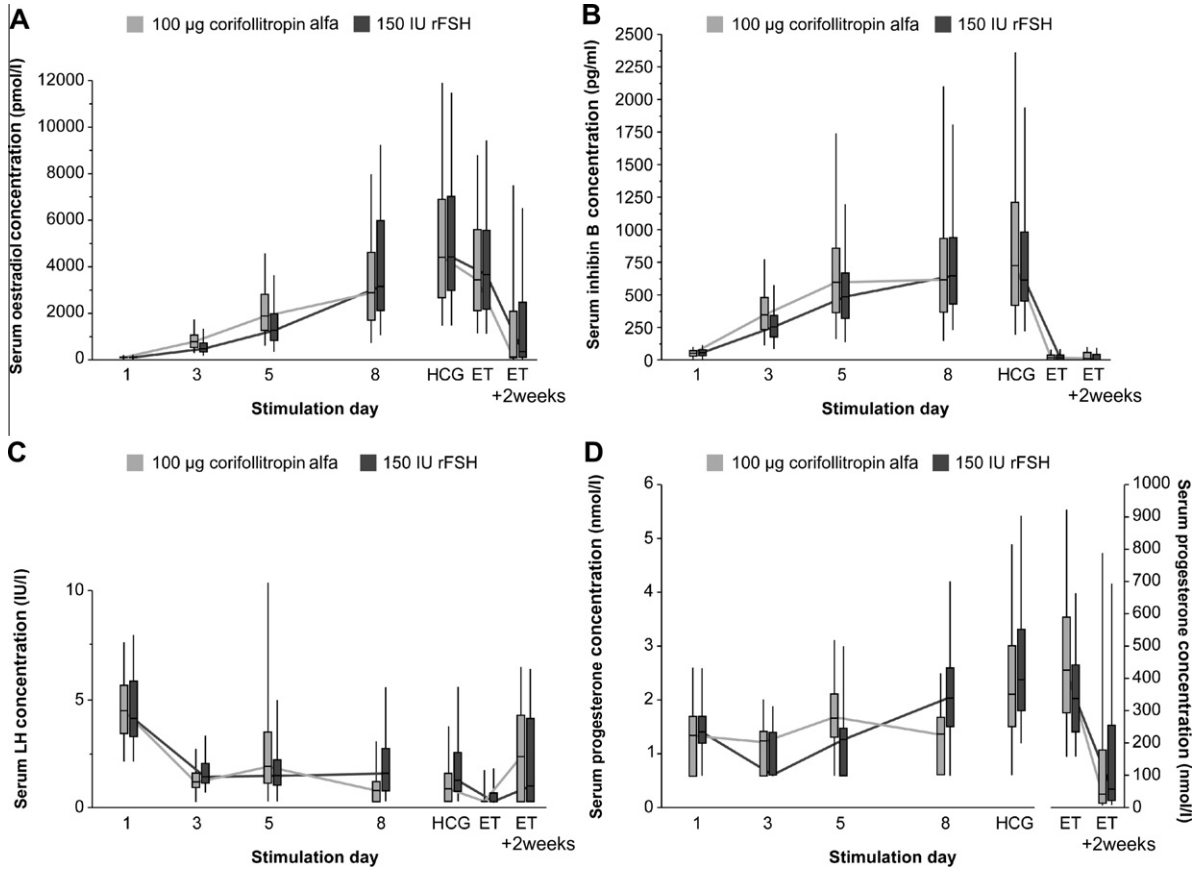

Figure 4 Serum concentrations of (**A**) oestradiol, (**B**) inhibin B, (**C**) LH and (**D**) progesterone per treatment group during and after stimulation restricted to patients with human chorionic gonadotrophin (HCG) injection (intent-to-treat group). The boxes indicate the 25—75% centiles, the whiskers indicate the 5—95% centiles and median values are connected. ET, embryo transfer; rFSH, recombinant FSH.

less pronounced in the corifollitropin alfa group than in the rFSH group.

Serum LH concentrations rapidly declined from stimulation day 1 to 3 in both treatment groups. On stimulation day 5 (prior to starting GnRH antagonist co-treatment) the incidence of premature LH rises (value $\geq$10 IU/l) was 5.2% (14 out of 268) of the patients in the corifollitropin alfa group and 3.9% (five out of 128) of the patients in the rFSH group. Overall, on stimulation day 5, a larger variation in LH concentrations was observed in the corifollitropin alfa group with a somewhat higher median LH value as compared with rFSH. Upon initiation of GnRH antagonist administration at day 5, LH concentrations decreased again in the corifollitropin alfa group, whereas the median LH values in the rFSH group did not change from day 3 until day 8. During GnRH antagonist treatment, none of the patients in the corifollitropin alfa group and two patients (1.6%) in the rFSH group experienced an LH rise.

Table 4 Clinical outcome per started cycle (intent-to-treat population).

	100 μg corifollitropin alfa (n = 268)	*150 IU rFSH (n = 128)*
Cumulus—oocyte—complexes retrieved (primary endpoint)	13.3 ± 7.3	10.6 ± 5.9
Fertilization procedure[a]		
IVF only	83 (31.4)	37 (29.8)
ICSI only	137 (51.9)	72 (58.1)
IVF + ICSI	44 (16.7)	15 (12.1)
Number and quality of oocytes (ICSI only)		
Number of oocytes	12.7 ± 6.8	9.9 ± 5.4
Metaphase II oocytes	10.7 ± 6.4	7.8 ± 4.8
Percentage of total	82.5	79.1
Fertilization outcome		
Fertilization rate[a,b]	67.6 ± 22.5	67.7 ± 25.4
2PN fertilized oocytes obtained[a]	7.8 ± 4.7	6.2 ± 3.9
2PN fertilized oocytes used for embryo development[a]	6.8 ± 3.6	5.8 ± 3.8
Number and quality of embryos		
Total number of embryos obtained (day 3)[a]	7.1 ± 4.2	6.1 ± 4.1
Good-quality embryos (grade 1 and 2)[a]	3.4 ± 3.0	3.0 ± 3.0
Percentage of total[a]	50.1	49.1
Single embryo transfer[c]	46 (18.7)	26 (21.7)
Embryos transferred[c]	1.8 ± 0.4	1.8 ± 0.4
Good-quality embryos transferred[c]	1.3 ± 0.8	1.3 ± 0.8
Embryos cryopreserved	2.0 ± 3.0	1.7 ± 2.6
Implantation rate[c]	23.4 ± 37.1	28.5 ± 38.6
Clinical outcome		
Biochemical pregnancy[d]	101 (37.7)	58 (45.3)
Clinical pregnancy[e]	78 (29.1)	48 (37.5)
Vital pregnancy[f]	69 (25.7)	45 (35.2)
Ongoing pregnancy[g]	68 (25.4)	44 (34.4)
Twin pregnancy[h]	19 (27.9)	10 (22.7)
Miscarriage[i]	10 (12.8)	4 (8.3)

Values are mean ± SD or number (%) unless otherwise stated.
[a] Restricted to patients with IVF and/or intracytoplasmic sperm injection (ICSI).
[b] Defined as 100 times the number of mature oocytes (with two pronuclei (2PN)) obtained divided by the number of oocytes used for fertilization.
[c] Restricted to patients with embryo transfer.
[d] Positive human chorionic gonadotrophin (HCG) test performed 2 weeks after embryo transfer.
[e] Clinical pregnancy: gestational sac on ultrasound scan.
[f] Vital pregnancy: gestational sac + fetal heartbeat.
[g] Ongoing pregnancy: vital fetus at least 10 weeks after embryo transfer or live birth.
[h] Per ongoing pregnancy.
[i] Per clinical pregnancy.

At the start of stimulation, serum progesterone concentrations were similar between the two treatment groups and remained low throughout ovarian stimulation in both groups (**Figure 4D**).

ICSI was the most frequently used fertilization procedure (51.9% in the corifollitropin alfa group and 58.1% in the rFSH group). In patients with ICSI only, the mean number of metaphase II oocytes as a percentage of the total number of oocytes was 82.5% in the corifollitropin alfa group and this was comparable to the mean percentage of 79.1% in the rFSH group (**Table 4**).

For patients with IVF and/or ICSI, the fertilization rate was similar between the two treatment groups (67.6% versus 67.7%). The mean number of fertilized 2PN oocytes obtained and used for embryo development was 7.8 and 6.8, respectively, for the corifollitropin alfa group and 6.2 and 5.8, respectively, for the rFSH group (**Table 4**). This difference between obtained and used is explained by the fact that for a total of 49 patients a subset of fertilized 2PN oocytes were reported to be lost or used for other purposes, of which more than 90% was cryopreserved.

The mean ± SD number of good-quality (Grade 1 and 2) embryos obtained at day 3 in patients with IVF and/or ICSI was 3.4 ± 3.0 and 3.0 ± 3.0 in the corifollitropin alfa and rFSH groups, respectively (**Table 4**). Although the majority of patients had two embryos transferred, the mean ± SD number of good-quality embryos transferred was 1.3 ± 0.8 in both groups. The mean ± SD number of embryos cryopreserved was 2.0 ± 3.0 and 1.7 ± 2.6, in the corifollitropin alfa and rFSH groups, respectively. The mean ± SD implantation rates for patients with embryo transfer were 23.4 ± 37.1% in the corifollitropin alfa group and 28.5 ± 38.6% in the rFSH group.

Per started cycle, the biochemical and clinical pregnancy rates were 37.7% and 29.1%, respectively, for the corifollitropin alfa group and 45.3% and 37.5%, respectively, for the rFSH group. The vital and ongoing pregnancy rates per started cycle were 25.7% and 25.4%, respectively, for the corifollitropin alfa group and 35.2% and 34.4%, respectively, for the rFSH group. Analysis of the ongoing pregnancy rate per started cycle showed that the *P*-value for the estimated difference in ongoing pregnancy rate between the corifollitropin alfa and rFSH treatment groups was not statistically significant at a 5% level. For both treatment groups, the majority of the ongoing pregnancies were singletons: 72.1% ($n = 49$) in the corifollitropin alfa group and 77.3% ($n = 34$) in the rFSH group. In total, 29 twin pregnancies were reported: 19 (27.9%) in the corifollitropin alfa group and 10 (22.7%) in the rFSH group. Eight patients (7.9%) in the corifollitropin alfa group and four patients (6.9%) in the rFSH group had an ectopic pregnancy per biochemical pregnancy. In total, 14 patients with a clinical pregnancy had a miscarriage: 10 (12.8%) in the corifollitropin alfa group and four (8.3%) in the rFSH group.

Safety

In total, 20 patients (7.5%) in the corifollitropin alfa group reported 22 serious adverse events and eight patients (6.3%) in the rFSH group reported nine serious adverse events. The most frequently reported serious adverse event was ectopic pregnancy, which occurred with the similar incidence of 3.0% and 3.1%, in the corifollitropin alfa and rFSH groups, respectively. The percentage of patients reporting adverse events was comparable between the two treatment groups: 55.2% in the corifollitropin alfa group and 53.5% in the rFSH group. None of the patients had discontinued the trial due to an adverse event or serious adverse event. The most frequently reported adverse events in the corifollitropin alfa and rFSH groups, respectively, were pelvic discomfort (10.1% and 14.7%), pelvic pain (10.4% and 10.9%), antepartum haemorrhage (6.0% and 12.4%) and headache (8.2% and 8.5%). A total of 18 patients in the corifollitropin alfa-treated group (6.7%) and six patients in the rFSH-treated group (4.7%) developed OHSS in this trial. The incidences of (moderate/severe) OHSS were 3.4% and 1.6% for the corifollitropin alfa and rFSH groups, respectively; the difference was not statistically significant at a 5% level (Fisher's exact test).

No drug-related hypersensitivity reactions were reported following corifollitropin alfa injection. With respect to local tolerance, none of the patients had any moderate or severe local reaction at the site of injection. In total, 267 patients treated with corifollitropin alfa and screened in the anti-corifollitropin alfa antibody assay were found negative, indicating that none of the patients had developed anti-corifollitropin alfa antibodies.

Discussion

The current trial was undertaken to confirm the efficacy and safety of a single subcutaneous injection of 100 μg corifollitropin alfa in patients weighing up to 60 kg using daily 150 IU rFSH as a reference. With respect to the primary endpoint, corifollitropin alfa provided significantly more oocytes (+2.5 oocytes, 95% confidence interval 1.2−3.9), but the estimated difference was well within the pre-set equivalence margin. Twice as many patients were randomized to the corifollitropin alfa group as compared with the rFSH reference group. This 2:1 randomization ratio was used to collect more (safety) information on the investigational product and did not introduce bias due to the double-blind, randomized design of the trial. rFSH (reference group) has already been on the market for several years and the safety and efficacy profile of this reference compound is well established (Kolibianakis et al. 2007).

Previously, the dose-finding trial of corifollitropin alfa indicated that body weight is a major determinant of exposure to corifollitropin alfa and treatment outcome (Corifollitropin Alfa Dose-Finding Study Group, 2008). In the current trial, a single dose of 100 μg corifollitropin alfa was sufficient to maintain multiple follicular development during the first week of stimulation in patients weighing ≤60 kg given the low cancellation rate in this group prior to the day of HCG.

The median duration of stimulation in the 100 μg corifollitropin alfa group was 9 days and was equal to the reference group using daily 150 IU rFSH in the same GnRH antagonist protocol. After a single injection of 100 μg corifollitropin alfa, patients required on average 2 days of additional stimulation with rFSH to reach the criterion for triggering final oocyte maturation. In 32.8% of the patients,

a single injection of corifollitropin alfa was sufficient to reach the HCG criterion, without the need for any additional rFSH.

The stimulation characteristics of 100 μg corifollitropin alfa in patients weighing at most 60 kg are identical to those observed after a single injection of 150 μg corifollitropin alfa in patients weighing more than 60 kg, as investigated in the large Engage trial (Devroey et al. 2009b). In the current double-blind trial, treatment with corifollitropin alfa resulted in an equal number of cumulus—oocyte—complexes retrieved per started cycle as in the Engage trial, i.e. 13.3 in the Ensure trial versus 13.7 in the Engage trial. These data confirm that the two dosages in the recommended body weight groups provide similar exposure and, therefore, induce the same degree of ovarian response. In comparison to the reference groups, the difference in number of oocytes retrieved is determined by the daily dose of rFSH, which was 150 IU in the current trial and 200 IU in the Engage trial. Accordingly, the estimated treatment difference in terms of the number of oocytes retrieved was 2.5 oocytes in the current trial versus 1.2 oocytes in the Engage trial. In terms of other clinical outcome parameters such as the maturity of oocytes retrieved, the fertilization rate and the number of good-quality embryos obtained, comparable results were obtained in both the Ensure and Engage trials.

In the Engage trial involving more than 1500 patients, results demonstrated an equal and high ongoing pregnancy rate in patients treated with corifollitropin alfa versus daily rFSH. In the current trial, ongoing pregnancy rate was a secondary endpoint and, in contrast to the Engage trial, this trial was not powered to assess non-inferiority in the ongoing pregnancy rates between the treatment groups. Observed ongoing pregnancy rates were 25.4% and 34.4% for the 100 μg corifollitropin alfa group and 150 IU rFSH group, respectively, and this difference was not statistically significant. Given the fact that other outcome parameters such as oocyte maturity, fertilization rate and number of good-quality embryos transferred were comparable to the reference group, the apparent difference in ongoing pregnancy rates in the current trial is considered a chance finding.

In this trial, the incidence of ectopic pregnancy in both treatment groups was twice as high as the reported incidence in IVF practice (Fernandez and Gervaise, 2004). Known risk factors for ectopic pregnancy include the method of embryo transfer, previous ectopic pregnancy, previous tubal surgery or pathology, previous spontaneous abortion and previous genital infections. In the current study, one patient had a history of three previous ectopic pregnancies whereas seven additional patients had a history of previous spontaneous abortion, endometriosis, tubal or unexplained infertility, which may have contributed to the relatively high incidences in this trial.

A single injection of 100 μg corifollitropin alfa had a safety profile comparable to daily doses of 150 IU rFSH in terms of the incidence and type of reported serious adverse events and adverse events. In line with the higher ovarian response, the incidence of moderate/severe OHSS tended to be higher after treatment with corifollitropin alfa than after treatment with rFSH, although the difference was not statistically significant.

Corifollitropin alfa was well tolerated at the site of injection. No drug-related hypersensitivity reactions or anti-corifollitropin alfa antibodies were reported following injection of 100 μg corifollitropin alfa. These findings are consistent with the outcome of previous phase I to III trials (Balen et al. 2004; Beckers et al. 2003; Bouloux et al. 2001; Corifollitropin Alfa Dose-Finding Study Group, 2008; Devroey et al. 2004, 2009b; Duijkers et al. 2002).

In conclusion, a lower dose of corifollitropin alfa (100 μg) offers a simplified treatment option for potential normal-responder patients with a lower body weight (at most 60 kg) undergoing controlled ovarian stimulation prior to IVF or ICSI. Compared with the reference group, treated with a fixed starting dose of 150 IU rFSH, the ovarian response is higher following corifollitropin alfa but well within the predefined equivalence margin whereas the duration of stimulation is equally short. One-third of the patients studied had complete multiple follicular development up to three follicles ≥17 mm based on a single injection of 100 μg corifollitropin alfa and did not need additional rFSH injections.

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Declaration: Financial support for this trial was provided by Schering-Plough, part of Merck and Co., Inc.

Received 21 August 2009; refereed 1 January 2010; accepted 24 February 2010.