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ARTICLE

A multi-centre phase 3 study comparing efficacy and safety of Bemfola® versus Gonal-f® in women undergoing ovarian stimulation for IVF

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Manfred Rettenbacher obtained his medical degree in 2000 from the University of Bern in Switzerland. He completed his doctoral thesis 'Localization and characterization of neuropeptide receptors in human colon', published in *Naunyn-Schmiedeberg Archives of Pharmacology* in 2001. Manfred Rettenbacher has held roles as Advisor, Clinical Research Physician and Medical Director in the pharmaceutical and biotechnology industry. He currently works at Finox Biotech AG, with responsibility for the entire drug development process and joint determination of the company's strategic direction. He holds an executive MBA with major 'Innovation Management' from Bern University of Applied Sciences, Engineering and Information Technology.

Abstract Bemfola (follitropin alfa) (Finox AG, Switzerland), a new recombinant FSH, has a comparable pharmacological profile to that of Gonal-f (Merck Serono, Germany), the current standard for ovarian stimulation. A randomized, multi-centre, Phase 3 study in women undergoing IVF or intracytoplasmic sperm injection ($n = 372$) showed Bemfola yielding similar efficacy and safety profiles to Gonal-f. Women aged 20–38 years of age were randomized 2:1 to receive a single, daily, subcutaneous 150 IU dose of either Bemfola or Gonal-f. This study tested equivalence in the number of retrieved oocytes using a pre-determined clinical equivalence margin of ±2.9 oocytes. Compared with Gonal-f, Bemfola treatment resulted in a statistically equivalent number of retrieved oocytes (Bemfola

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10.8 ± 5.11 versus Gonal-f 10.6 ± 6.06, mean difference: 0.27 oocytes, 95% confidence interval: −1.34, 1.32) as well as a similar clinical pregnancy rate per embryo transfer in first and second cycles (Bemfola: 40.2% and 38.5%, respectively; Gonal-f: 48.2% and 27.8%, respectively). No difference in severe ovarian hyperstimulation syndrome was observed between treatment groups (Bemfola: 0.8%; Gonal-f: 0.8%). This study demonstrates similar clinical efficacy and safety profiles between Bemfola and Gonal-f, and suggests that Bemfola can be an appropriate alternative in ovarian stimulation protocols.

KEYWORDS: IVF-ICSI, oocytes retrieved, ovarian stimulation, r-FSH

Introduction

The recombinant FSH (r-FSH) preparations Gonal-f (Merck Serono, Darmstadt, Germany) and Puregon have been commercially available since 1995 and 1996, respectively (Lunenfeld, 2004). Recently, biosimilar versions of r-FSH have been developed to provide high quality but economically attractive alternatives to the present FSH products.

For treating infertility, Gonal-f (Merck Serono, Germany) is used in the following indications: ovulation induction with human FSH in normogonadotrophic (World Health Organization [WHO] type II) anovulatory women; ovulation induction in WHO type I anovulatory infertility in association with a LH preparation; and ovarian stimulation in patients undergoing assisted reproduction technique treatment or in milder forms of intrauterine insemination. It has been recognized that the high treatment costs restrict access to high-quality biological medicines (Engelberg et al., 2009; Schellekens and Moors, 2010). 'Biosimilar' medicines, which are medical products whose active drug substance is made by or derived from a living organism by recombinant DNA or controlled gene expression, meet high standards for comparability to the originator medicine and are approved for use in the same indications (Weise et al., 2012).

Bemfola (Finox AG, Switzerland) is the first r-FSH biosimilar on the market that has been demonstrated to have similar physicochemical properties in pre-clinical studies and is intended for use in the same therapeutic indications as Gonal-f (Merck Serono, Germany). Bemfola (Finox AG, Switzerland) has similar non-clinical pharmacological, pharmacokinetic and toxicological profiles to those of Gonal-f (Merck Serono, Germany) (European Public Assessment Report, Bemfola, (European Medicines Agency, 2014)). An important difference between the two products is that Bemfola (Finox AG, Switzerland) does not contain m-cresol as an excipient because Bemfola (Finox AG, Switzerland) is presented as a single-use injection system with no required preservative. In contrast, the Gonal-f injection device (Merck Serono, Germany) is for multiple use and includes preservatives in the formulation. The Bemfola pen (Finox AG, Switzerland) offers a fine-tuned dosing scheme of a minimum of 12.5-IU increments. Other characteristics of the single-use pen are volume and injection-control mechanisms and visual aids that may improve treatment compliance. Additionally, an in-built lock prevents re-use of the pen device by the patient.

The objective of this study was to demonstrate the equivalence of Bemfola (Finox AG, Switzerland) and the comparator product Gonal-f (Merck Serono, Germany) in ovarian stimulation for IVF or intracytoplasmic sperm injection (ICSI). Because of the similarity in the active substance as well as the formulation, Bemfola treatment (Finox AG, Switzerland) was expected to result in efficacy and safety profiles similar to those of Gonal-f (Merck Serono, Germany). This expectation was based on results from a Phase 1 clinical trial, FIN1001, which demonstrated equivalent pharmacokinetic profiles between Bemfola (Finox AG, Switzerland) and Gonal-f (Merck Serono, Germany). Bemfola (Finox AG, Switzerland) was designed to be a biosimilar of Gonal-f (Merck Serono, Germany), thereby showing familiar efficacy and safety profiles compared with Gonal-f (Merck Serono, Germany).

Materials and methods

Study FIN3001 was an assessor-blinded, randomized, parallel-group, multi-centre, Phase 3 trial conducted in women undergoing assisted reproduction techniques ($n = 372$), who were treated with a fixed-dose regimen of a daily, single subcutaneous dose of 150 IU Bemfola (Finox AG, Switzerland) or 150 IU Gonal-f (Merck Serono, Germany). A total of 15 centres in six European countries received study medication and recruited eligible patients into the study: the participating countries (number of centres) were Austria ($n = 5$), Denmark ($n = 3$), Germany ($n = 2$), Spain ($n = 2$), Switzerland ($n = 1$) and UK ($n = 2$). The study was conducted between July 2010 and April 2012, with a consecutive pregnancy follow-up period. The study was registered in clinicaltrials.gov (NCT01121666).

To test immunogenicity, patients who did not become pregnant after a completed cycle of recombinant human FSH (r-hFSH) treatment and IVF or ICSI had the option of undergoing a second cycle of r-hFSH treatment at least 4 weeks after termination of the first treatment cycle; patients remained allocated to the study group arm Bemfola (Finox AG, Switzerland) or Gonal-f (Merck Serono, Germany), respectively. The schedule of assessment and follow-up procedures were according to the main study. The study was primarily powered for the first treatment cycle but not for the second treatment cycle, which was carried out to evaluate anti-FSH antibodies based on repeated administration of FSH as agreed upon by the European Medicinal Agency (EMA); as a consequence, only descriptive analysis was conducted for the second treatment cycle. Numbers presented in the present study reflect the full data of the first treatment cycle, including immunogenicity data and the pregnancy rates of the second treatment cycle.

The study was approved by the Independent Medical Ethics Committee or Institutional Review Board of respective countries and was conducted in accordance with Good Clinical Practice, Declaration of Helsinki, the International Conference on Harmonisation guidelines for Good Clinical Practice CHMP/

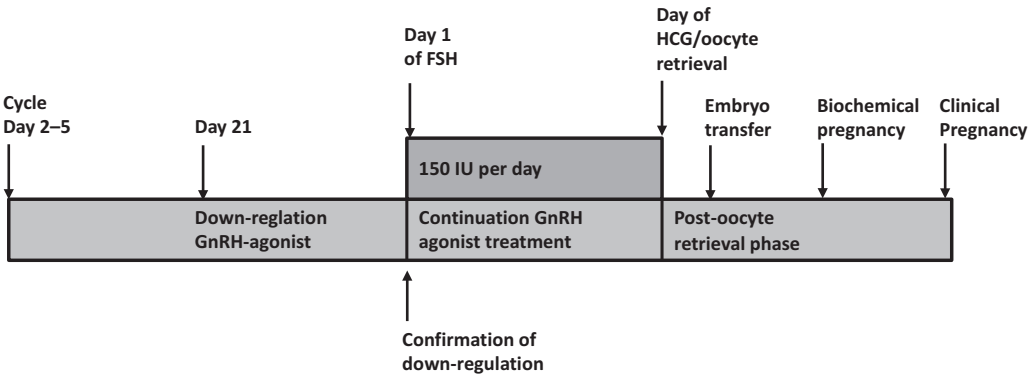

Figure 1 Study design.

ICH/135/95, and local regulatory requirements/laws. The aims, methods, anticipated benefits and potential hazards of the study were expained to all participants, and written informed consent was obtained from each.

Study population

Women who fulfilled the following inclusion criteria were eligible: age between 20 and 38 years; body mass index (BMI) between 18 and 30 kg/m^2; regular menstrual cycles of 25–35 days; and first or second cycle in the present series of assisted reproduction techiques. Other key inclusion criteria were basal FSH less than 10 IU/L (cycle day 2–5); oestradiol levels less than 50 pg/mL on the first day of FSH administration; a total antral follicle count of 10–25 follicles; infertility resulting from tubal factors; mild endometriosis (American Society of Reproductive Medicine [ASRM] stage 1–2 (American Society for Reproductive Medicine (ASRM), 1997); male factor; unexplained infertility; and presence of both ovaries and normal uterine cavity as confirmed by transvaginal ultrasound within 6 months before randomization. Key exclusion criteria were a history of more than two earlier assisted reproduction technique retrieval cycles; the presence of endocrine disorder; known tumours of the hypothalamus and pituitary gland, or both; a history of severe ovarian hyperstimulation syndrome (OHSS); severe endometriosis (ASRM stage 3 or 4) (American Society for Reproductive Medicine (ASRM), 1997); the presence of a hydrosalpinx; polycystic ovaries (Rotterdam ESHRE/ASRM-Sponsored PCOS consensus workshop group, 2004); history of poor response to gonadotrophin treatment (defined as fewer than five oocytes retrieved in a previous attempt); and any hormonal treatment within 1 month before starting FSH treatment (with the exception of levothyroxin).

Study design

The goal of this assessor-blinded, randomized, parallel-group, multi-centre study was to compare the efficacy and safety profiles of Bemfola (Finox AG, Switzerland) with those of Gonal-f (Merck Serono, Germany) after daily administration of 150 IU. This fixed daily dose (150 IU FSH/day) design was based on the Scientific Advice provided by the EMA in 2008 and used to reduce inter-group variability that might occur if different investigator-initiated dosing schemes had been applied. Eligible participants were randomized in a 2:1 ratio to receive either Bemfola (Finox AG, Switzerland) or Gonal-f (Merck Serono, Germany), respectively. Patient randomization into the two treatment groups was also stratified by age (<35 and ≥35 years). Centralized treatment allocation with an interactive web response system was used and filed in the study centre file. A minimization algorithm was used to achieve an optimally balanced patient allocation, overall and across the strata. The outcome of pregnancies was followed up within the context of safety follow-up information.

Treatment administration

All eligible participants were down-regulated with a gonadotrophin-releasing hormone (GnRH) agonist initiated in the luteal phase. The GnRH agonists triptorelin (Ipsen, France), buserelin (Sanofi-Aventis, Germany) and leuprorelin (Sandoz, Switzerland), were subcutaneously administered according to local practice. After successful down-regulation with an oestradiol level of less than 50 pg/mL, a shedded endometrium thickness of less than 5 mm and no ovarian cysts, participants were randomized 2:1 to receive either Bemfola (Finox AG, Switzerland) or Gonal-f (Merck Serono, Germany) at a dose of 150 IU/day (**Figure 1**); FSH in both groups was administered subcutaneously. After 6 days of FSH administration and only in cases of risk of OHSS or other safety concerns, could the fixed daily 150 IU FSH dose be decreased, coasting applied or treatment terminated. Once at least one follicle reached a diameter of 18 mm or over and two additional follicles reached a diameter of 16 mm or over, a dose of 250 μg r-hCG (Ovitrelle; Merck Serono, Germany) was given to trigger ovulation and 34–36 h later, oocytes were retrieved. If the criteria for ovulation triggering could not be reached by FSH stimulation on day 16, treatment was to be stopped. Both ICSI and IVF were carried out according to the centre's standard procedures, and a maximum of two embryos or blastocysts were transferred 2–5 days after oocyte retrieval.

Assessments

Ovarian response was assessed by vaginal ultrasound at baseline, at days 6 and 8, and on the day of human chorionic

gonadotrophin (HCG) administration for a maximum of 16 days after the start of treatment with r-hFSH. For the purpose of subject safety, serum oestradiol concentration was measured as well. After day 8, follicular development was monitored by vaginal ultrasound at 2- to 3-day intervals. The vaginal ultrasound assessor and the investigator were blinded regarding the r-hFSH preparation of the patient. Surplus pronuclear stages or embryos were cryopreserved according to local regulations. Further safety follow-up visits were scheduled. The pregnancy rate was determined biochemically (i.e. about 2 weeks after oocyte retrieval), and, clinically, (i.e. 5–6 weeks after oocyte retrieval), respectively. Clinical pregnancy was defined as the presence of at least one intrauterine gestational sac. Utrogestan (Besins Healthcare, Belgium) was vaginally administered and used for luteal support at a concentration of 3×200 mg per day beginning at the day of embryo or blastocyst transfer until confirmation of clinical pregnancy. A blood sample was taken for anti-Müllerian Hormone (AMH) determination at the baseline visit (day of the start of FSH treatment). Processed blood samples were sent to a central laboratory where they were measured, after the study was completed, with the AMH Gen II enzyme linked immunosorbent assay test kit of Beckman-Coulter.

Immune response was investigated using a validated surface plasmon resonance method, an immunoassay method using adsorption properties of possible antibodies to FSH bound to specific platelets (Szolar et al., 2007). At baseline, 3 weeks and 8 weeks after completing the FSH treatment in both treatment cycles, blood samples were taken to detect possible anti-FSH antibodies. In case anti-FSH antibodies were detected, further confirmation assays were run to characterize the cross-reactivity to other proteins sharing the common alpha chain with FSH and the neutralization potential of such antibodies. Patients in both treatment arms who did not become pregnant in the first treatment cycle, and who underwent an additional assisted reproduction technique cycle, were investigated for potential immunogenicity after repeated treatment cycles. Surface plasmon resonance method, an immunoassay detecting and characterizing possible antibodies by binding to FSH bound to specific platelets was used to detect anti-FSH antibodies expressed in relative units (relU). The cut-off point for a positive response, which is a certain signal above the serum background of untreated serum samples, was set to 104 relU; above this value, a serum sample was regarded as a 'positive hit', and a subsequent confirmation assay was therefore required.

End-points

The primary end-point was the number of oocytes retrieved. Secondary end-points included the quality of oocytes retrieved; fertilization rate of oocytes; embryo quality; number of cryopreserved pronuclear stages, embryos or blastocysts; total dose of r-hFSH required; number of days of r-hFSH stimulation; number of patients with cycle cancellation owing to excessive response and low response. Adverse events were recorded as secondary safety end-points. Pharmacodynamically, the number and size of follicles 12 mm or over in diamter at day 8 of stimulation as well as on the day of HCG administration were evaluated as secondary end-points. Other secondary end-points included implantation and clinical pregnancy rates, ongoing pregnancy and live birth rate.

Statistical methods

This study was powered to test equivalence using a two one-sided test (TOST) of the primary end-point (i.e. the number of oocytes retrieved) with a power of 90%, an alpha error of 2.5% and a pre-determined clinical equivalence margin of ±2.9 oocytes for the relevant population. Because of the non-normality of the distribution of the primary end-point, which was proven by the Shapiro-Wilk test, a Mann-Whitney U test was carried out in both populations. A poor responder rate of 5% was included. A sample size of 351 patients with a 2:1 ratio (234/117) was calculated under the hypothesis (±2.9 is the considered clinical equivalence margin for the mean difference): H_0: $\Delta < -2.9$ or $\Delta > 2.9$ null hypothesis; H1: $-2.9 \leq \Delta \leq 2.9$ alternative hypothesis. The corresponding 95% confidence interval was approximated by a bootstrap-t approach. Schuirmann's TOST test was carried out to confirm the robustness of the results. Analyses of covariance stratified by demographic and baseline characteristics were used to investigate the treatment effects of (Bemfola Finox AG, Switzerland) and Gonal-f (Merck Serono, Germany) on number of oocytes retrieved. The primary efficacy analysis was based on the per-protocol populations. The analysis of adverse events, laboratory data, vital signs, physical examination findings and other safety evaluations was carried out on the intention-to-treat (ITT) population, which included all patients who received any amount of study drug. $P < 0.05$ was considered statistically significant.

Results

A total of 460 patients were enrolled in this trial; 88 patients were reported as screening failures (**Figure 2**). A total of 372 patients were randomized whereas 39 randomized patients were excluded from the per-protocol population because of protocol deviations. Therefore, a total of 333 patients were analysed in the per-protocol population (i.e. 220 patients in the Bemfola group and 113 patients in the Gonal-f group).

Most patients (>75%) were younger than 35 years of age, and over 90% of patients were white (**Table 1**). No appreciable differences were observed in baseline FSH concentration, antral follicle count, and GnRH agonist duration. Subsequent evaluation of the AMH level, however, revealed that, although a higher percentage of patients treated with Bemfola (Finox AG, Switzerland) had an AMH level of ≥24 pmol/L compared with those on Gonal-f treatment (Merck Serono, Germany) (42.6% versus 38.2%, respectively), the difference between treatment groups was not statistically significant (**Table 1**). The cut-off level of ≥24 pmol/L was defined on the basis of the evaluated level by Lee et al. (2008), and was applied as the suggested cut-off level for this study to

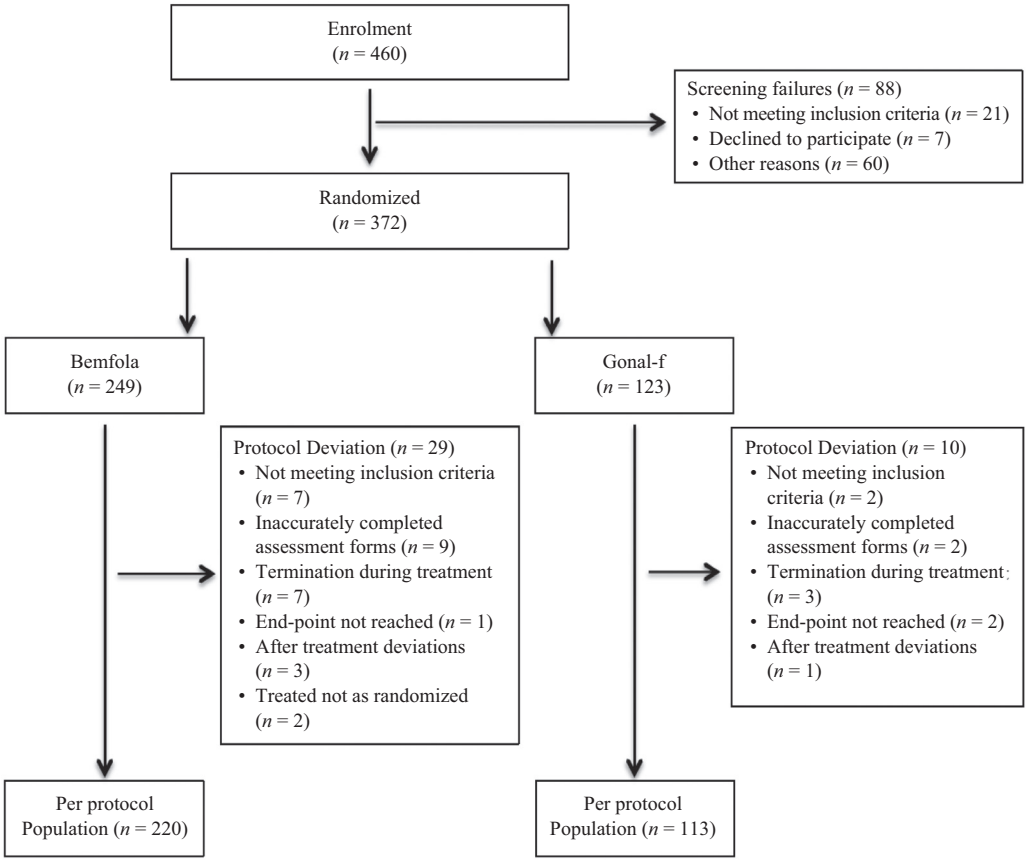

Figure 2 Patient disposition scheme.

evaluate potential high-responder patients in both treatment groups.

Primary end-point: number of retrieved oocytes from patients undergoing assisted reproduction techniques

Bemfola (Finox AG, Switzerland) treatment resulted in a number of oocytes statistically equivalent to that of patients treated with Gonal-f (Merck Serono, Germany) (Table 2). The treatment difference was 0.27 oocytes (95% confidence interval: −1.34, 1.32) yielding a *P*-value for equivalence of $P = 0.0003$, demonstrating equivalence as pre-defined in the statistical analysis plan (less than three oocytes). The results from ITT analysis was similar to the data showed from per protocol analysis, thereby confirming the observed equivalence between Bemfola (Finox AG, Switzerland) and Gonal-f (Merck Serono, Germany).

Evaluation of FSH stimulation phase

No significant difference was observed in the total dose of r-hFSH administered between participants treated with Bemfola compared with those in the Gonal-f treatment group ($P = 0.9638$), and in mean duration of stimulation between both treatment arms ($P = 0.8926$) (Table 1). Participants treated with Bemfola (Finox AG, Switzerland) were reported with comparable numbers of follicles 12 mm or over, 15 mm or over, and 17 mm or over compared with those in the Gonal-f (Merck Serono, Germany) group ($P = 0.2357$, 0.1395, and 0.3992, respectively). The concentration of oestradiol was also comparable in patients treated with Bemfola (Finox AG, Switzerland) and Gonal-f (Merck Serono, Germany) on day of HCG administration (Table 1).

Assessment of response and oocyte quality

Most patients (>90%) in both treatment arms were reported with an oocyte retrieval of four or more oocytes (Table 2). The percentage of oocytes with a high degree of cumulus oophorus maturity (i.e. 'very mature') was similar in both groups, but was lower than that reported for mature oocytes; this was also the case in the percentage of immature oocytes. Most oocytes in both treatment groups were in the metaphase-II stage of nuclear development, and no significant difference in the fertilization rate was observed between treatment arms.

Bemfola (Finox AG, Switzerland) treatment resulted in a similar number of patients with embryo transfer and cryopreservation compared with those receiving Gonal-f (Merck Serono, Germany) (Table 2). The main embryo quality parameters observed (i.e. mean number of blastomeres and the absence of multinucleation) were similar in both groups.

Table 1 Demographic characteristics and assessment of stimulation phase.

	Bemfola n = *249*	*Gonal-f* n = *123*
Demographic characteristics		
Age (±SD), years	31.8 (4.03)	32.1 (3.76)
BMI (±SD), kg/m^2	22.7 (2.88)	22.4 (2.56)
Race, *n* (%)		
White	226 (91.9)	117 (95.1)
Asian	12 (4.9)	3 (2.4)
African	2 (0.8)	1 (0.8)
Other	6 (2.4)	2 (1.6)
FSH baseline concentration (IU/L)	6.9 (1.50)	6.9 (1.56)
Antral follicle count (mean SD)	15.1 (3.77)	15.3 (3.83)
GnRH-agonist duration (days; mean SD)	23.5 (7.92)	22.7 (7.46)
AMH ≥ 24 pmol/L, *n* (%)	106 (42.6)	47 (38.2)
Assessment of stimulation phase		
Mean total dose of r-hFSH (SD) IU	1555.7 (293.00)	1569.2 (259.20)
Duration of FSH medication (SD) days	10.6 (1.91)	10.7 (1.72)
Number of follicles (SD)		
≥12 mm	11.8 (4.73)	11.1 (4.23)
≥15 mm	8.3 (3.81)	7.7 (3.60)
≥17 mm	4.9 (3.29)	4.5 (2.71)
Oestradiol serum concentrations (SD), pmol/L		
Day of HCG	8982.3 (6535.3)	7704.2 (5345.8)
Day 8	3958.9 (3699.4)	3234.0 (2428.1)
Safety assessment		
OHSS, *n* (%)	14 (5.6)	4 (3.3)
Patients with OHSS and AMH ≥ 24 pmol/L	8 (57.1)	1 (25.0)
Patients with dose reduction due to risk of hyperstimulation, *n* (%)	38 (15.3)	16 (13.0)
Patients with coasting due to risk of hyperstimulation, *n* (%)	3 (1.2)	3 (2.4)
Patients with hCG withdrawal due to risk of hyperstimulation, (*n*, %)	5 (2.0)	1 (0.8)

Numbers are mean (SD) values unless otherwise indicated.
Differences in each of the parameters were not statistically significant.
AMH = anti-Müllerian hormone; BMI = body mass index; GnRH = gonadotrophin-releasing hormone; OHSS = ovarian hyperstimulation syndrome; r-hFSH = recombinant FSH; SD = standard deviation.

Implantation rate and pregnancy outcome per embryo transferred

Treatment cycle 1 resulted in an implantation rate for Bemfola (Finox AG, Switzerland) that was similar to that reported for Gonal-f (Merck Serono, Germany) (Bemfola: 31.8%; Gonal-f: 36.7%) (Table 2). Biochemical and clinical pregnancy rates were also similar between treatment groups. The percentages of patients with an ongoing pregnancy and live born children did not reveal any significant differences between both treatments groups. The mean number of transferred embryos and blastocysts was slightly higher in the Gonal-f (Merck Serono, Germany) group compared with the Bemfola (Finox AG, Switzerland) group, with a trend towards a higher number of blastocysts transferred in the Gonal-f (Merck Serono, Germany) group (Table 2).

Only results of treatment cycle 2 on the clinical and ongoing pregnancy rate (Table 2) and the repeated tests for immunogenicity are shown (Table 3). In contrast to data from treatment cycle 1, the clinical pregnancy rate for Bemfola (Finox AG, Switzerland) and Gonal-f (Merck Serono, Germany) evaluated from treatment cycle 2 was, although not significant, higher in the Bemfola (Finox AG, Switzerland) group compared with the Gonal-f (Merck Serono, Germany) group. With the ongoing pregnancy rate, the same non-significant difference in favour of Bemfola (Finox AG, Switzerland) could be reported (Table 2).

Adverse event profile

The adverse effects in patients treated with Bemfola (Finox AG, Switzerland) were relatively similar to that observed in Gonal-f (Merck Serono, Germany) patients. The most commonly reported adverse reactions reported in both study arms

Table 2 Number and quality of retrieved oocytes, secondary efficacy parameters, and clinical efficacy outcome.

	Bemfola	*EU-Gonal-f*	*Treatment difference*	P-*value*
Per protocol population (patients with oocyte retrieval after completed treatment cycle)	*n* = 220	*n* = 113		
Number of oocytes retrieved ± SD	10.8 ± 5.11	10.6 ± 6.06	0.27 (−1.34, 1.32)	0.0003[a]
Intention-to-treat population	*n* = 249	*n* = 123		
Number of oocytes retrieved ± SD	10.7 ± 5.62	10.4 ± 6.14	0.29 (−1.29, 1.34)	0.0003[a]
Number of embryos/blastocysts transferred (mean ± SD)	1.5 ± 0.52	1.6 ± 0.53		
Number of patients with transferred embryos, *n* (%)				
Day 2	82 (32.9)	36 (29.2)		
Day 3	53 (21.3)	28 (22.8)		
Number of patients with transferred blastocysts, *n* (%)				
Day 4	10 (4.0)	2 (1.6)		
Day 5	76 (30.5)	46 (37.4)		
Patients with good response[b]	225 (90.4)	112 (91.0)		
Cycle cancellations	13 (5.2)	5 (4.1)		
Cumulus oophorus Maturity, *n* (%)				
Very mature	214 (9.1)	106 (9.4)		
Mature	1773 (75.7)	849 (75.3)		
Immature	339 (14.5)	160 (14.2)		
Nuclear maturity, *n* (%)				
Germinal vesicle	203 (9.5)	92 (9.1)		
Metaphase I	154 (7.2)	78 (7.7)		
Metaphase II	1788 (83.4)	845 (83.3)		
Number of blastomeres at day 3 (mean ± SD)	6.6 ± 2.41	6.4 ± 2.49		
Absence of multinucleation at day 3, *n* (%)	927 of 990 (93.6)	512 of 545 (93.9)		
Fertilization rate (mean ± SD)	66.1 ± 24.84	64.0 ± 24.76		
Patients with embryo transfer, *n* (%)	224 (89.9)	114 (92.7)		
Patients with cryopreservation, *n* (%)	103 (41.4)	55 (44.7)		
Implantation rate, *n* (%)[c]	110 of 346 (31.8)	66 of 180 (36.7)		
Biochemical pregnancy rate per embryo transfer, *n* (%)	116 (51.8)	60 (52.6)		
Clinical pregnancy rate per embryo transfer, *n* (%)[d]	90 (40.2)	55 (48.2)		
Ongoing pregnancy per embryo transfer, *n* (%)[e]	84 (37.5)	51 (44.7)		
Patients with liveborn children, *n* (%)	80 (35.7)	50 (43.9)		
Second treatment cycle	*n* = 72	*n* = 38		
Clinical pregnancy rate per embryo transfer, *n* (%)[d]	25 (38.5)	10 (27.8)		
Ongoing pregnancy per embryo transfer, *n* (%)[e]	22 (33.8)	9 (25.0)		

[a]*P*-value indicating a high significance for clinical equivalence; differences in remaining parameters are not statistically significant.
[b]Patients with an oocyte retrieval of four or more oocytes.
[c]Defined as fetal sac per embryo transferred.
[d]Presence of at least one intrauterine gestational sac.
[e]Presence of at least one viable fetus 10 weeks after embryo transfer.

were headache, ovarian cysts and local injection site reactions (e.g. pain, erythema, haematoma, swelling, irritation, or both, at the site of injection) (data not shown). All grades of OHSS were reported in 5.6% of the patients in the Bemfola (Finox AG, Switzerland) group and in 3.3% of the patients in the Gonal-f (Merck Serono, Germany) group ($P = 0.4428$), whereas severe OHSS (The Practice Committee of the American Society for Reproductive Medicine, 2004), occurred in less than 1% of patients in either study arm (0.8% in both treatment groups). Eight out of 14 patients with reported OHSS in the Bemfola (Finox AG, Switzerland) group had an AMH level of 24 pmol/L or over compared with one out of four patients in the Gonal-f (Merck Serono, Germany) group (**Table 1**).

Immunogenicity profile

Analysis of antibodies specific for Bemfola (Finox AG, Switzerland) or Gonal-f (Merck Serono, Germany) indicated no evidence of immunogenicity (i.e. antibody reactivity to Bemfola or Gonal-f) in either treatment group during both treatment cycles. All patients had anti-follitropin antibody levels below the serum background of untreated serum samples at baseline in both treatment cycles (**Table 3**). No patient showed any increase in response units, as measured 3 and 8 weeks after completion of FSH treatment. This was also the case for patients being re-treated with the same FSH preparation in the second treatment cycle.

Table 3 Mean anti-follitropin antibody levels (full analysis set).

		Actual values (relU)		
Treatment group	*Visit*	n	*Mean*	*SD*
Treatment cycle 1				
Bemfola	Baseline	247	42.2	43.87
	3 weeks after FSH treatment	219	40.4	43.80
	8 weeks after FSH treatment	192	42.2	42.07
Gonal-f	Baseline	121	39.6	30.36
	3 weeks after FSH treatment	106	39.6	30.47
	8 weeks after FSH treatment	114	38.8	29.40
Treatment cycle 2				
Bemfola	Baseline	72	37.8	30.52
	3 weeks after FSH treatment	67	38.6	30.74
	8 weeks after FSH treatment	58	41.8	32.17
Gonal-f	Baseline	37	35.5	21.55
	3 weeks after FSH treatment	33	35.2	22.32
	8 weeks after FSH treatment	35	37.9	25.37

Discussion

This study has shown that Bemfola (Finox AG, Switzerland) and Gonal-f (Merck Serono, Germany) are statistically equivalent in the primary physiological function of FSH; that is, in stimulation of follicular development. In addition, Bemfola (Finox AG, Switzerland) and Gonal-f (Merck Serono, Germany) seem to have similar efficacy and safety profiles. The evaluation of potential biosimilarity between Bemfola (Finox AG, Switzerland) and Gonal-f (Merck Serono, Germany) was based on data obtained from an assessor-blinded, randomized, parallel-group, multi-centre, Phase 3 trial conducted in women undergoing assisted reproduction techniques. The study design was chosen to examine the degree of biosimilarity between both treatments, whereas a 2:1 randomization ratio was intended to allow a more comprehensive assessment of the safety profile of Bemfola (Finox AG, Switzerland) in this patient population.

Patients enrolled in this study were representative of the patient population usually treated in this clinical setting. The mean age of the patients (abouty 32 years of age), the proportions of patients above and below 35 years of age, and the mean BMI (about 23) correspond well to the characteristic general assisted reproduction technique population documented in the medical literature (Andersen et al., 2006; Bergh et al., 1997; Frydman et al., 2000; Schats et al., 2000). Overall, the population for this Phase 3 study met the expected criteria and was well balanced between the treatment arms. No baseline characteristics could be identified that showed variance between the two groups.

To investigate potential immunogenicity, a second treatment cycle for patients who did not become pregnant during the first treatment cycle and who still met the eligibility criteria was offered and performed (if accepted) to evaluate the possible emergence of anti-FSH antibodies following repeated exposure to the same recombinant FSH (either Bemfola or Gonal-f). Pooling the results from both treatment cycles was not intended in the statistical analysis, because no re-randomization was carried out in the second treatment cycle. Safety and efficacy results for a non-randomized sub-population would be potentially biased. Therefore, the data from each treatment cycle must be analysed independently. The results presented reflect the outcome of the first treatment cycle as statistically relevant; the study was not powered for the detection of statistically relevant differences of any variable in the second treatment cycle.

The primary end-point for the FIN3001 study was the number of oocytes retrieved in the relevant per protocol patient population. The data showed that Bemfola (Finox AG, Switzerland) administered subcutaneously at a dose of 150 IU daily was statistically equivalent (within the protocol-defined equivalence margin) to Gonal-f (Merck Serono, Germany) in terms of number of retrieved oocytes. The secondary end-points used in this Phase-3 study are standard end-points in assisted reproduction technology clinical studies. Similar results were also observed in secondary efficacy end-points such as oestradiol levels, number and size of follicles and pregnancy outcome. The numbers of oocytes retrieved from patients in this study are similar to those of other studies of r-FSH in assisted reproduction technologies (Bergh et al., 1997; Bühler and Naether, 2011; Frydman et al., 2000; Jayaprakasan et al., 2010; Schats et al., 2000). Although this study was not powered to detect a difference between treatments for secondary efficacy end-points, the results were consistent with those found in other studies.

The clinical pregnancy outcome as a secondary end-point of the study was not significantly different between treatment groups in both treatment cycles. Although the study was not powered to evaluate significant differences in clinical pregnancy rate, however, an 8% difference in clinical pregnancy rate in favour of Gonal-f (Merck Serono, Germany) in the first treatment cycle and a 10.7% difference in clinical pregnancy rate in favour of Bemfola (Finox AG, Switzerland) in the second treatment cycle was found. In view of there being slightly more day-5 blastocyst transfers in treatment cycle 1 in the Gonal-f (Merck Serono, Germany) group compared with the Bemfola (Finox AG, Switzerland) group (37.4% versus 30.5%), this may have contributed to the non-significant differences in clinical pregnancy and live birth rate in this study

(Table 2). Additional clinical trials are warranted to further document these end-points.

The biosimilarity between Bemfola (Finox AG, Switzerland) and Gonal-f (Merck Serono, Germany) for various efficacy parameters was coupled with a safety profile that is consistent with other studies in this clinical setting (Recombinant Human FSH Study Group, 1995; Bergh et al., 1997; Frydman et al., 2000; Schats et al., 2000; Balen et al., 2007). In addition, Bemfola (Finox AG, Switzerland) treatment did not result in increased immunogenicity compared with the reference comparator Gonal-f (Merck Serono, Germany). Findings were consistent between per protocol and ITT analysis sets in the first treatment cycle, thereby confirming the robustness of the data.

The overall OHSS rate (including mild, moderate and severe forms) was 5.6% for Bemfola (Finox AG, Switzerland) and 3.3% for Gonal-f (Merck Serono, Germany). Severe OHSS occurred in less than 1% of patients (0.8% in both treatment groups), which is in line with that reported in a similar patient population and using a long GnRH agonist design with Gonal-f (Merck Serono, Germany) as a comparator (Andersen et al., 2006; Olivennes et al., 2009; Schats et al., 2000) Sensitivity analysis revealed a high AMH level (cut-off level $\geq$ 24 pmol/L) to be related to the OHSS rate. Taking into consideration AMH levels in both treatment groups, an ancillary analysis revealed the same overall (mild, moderate, severe) OHSS rate of 2.4% (with 0.8% of the clinical relevant severe form) in both groups.

At the time the study was designed, AMH was still not fully acknowledged to be an important, response-predictive clinical parameter. Therefore, it was not included as a stratification factor in the study design. As the blood sample was measured after the study was finished, the investigators did not know the AMH level at the time the patient was randomized and consequently the start of the FSH treatment. These findings, however, support the current view that stratification for AMH as an important indicator of ovarian response in clinical trials is necessary (La Marca and Sunkary, 2013; Nelson et al., 2012).

The EMA approved Bemfola (Finox AG, Switzerland) on the basis of these study results, showing that Bemfola (Finox AG, Switzerland) meets the efficacy and safety requirements as a FSH product used in IVF treatments. As biologics, including biosimilars, are biological products recovered by recombinant methods from human or animal cell-lines, the evaluation of the glycosylation pattern, which has been suggested as the main reason for adverse events, is of particular interest to health authorities, healthcare providers and patients. The similar safety results of Study FIN3001, coupled with the comparable physico-chemical profiles of Bemfola (Finox AG, Switzerland) and Gonal-f (Merck Serono, Germany), indicate that possible differences in glycosylation pattern between the two molecules have no clinical consequences. The cohort size of the study was defined on the basis of the primary end-point of oocytes retrieved, and thus too small to detect a significant difference in any secondary end-points, such as pregnancy, and hence further studies are warranted to assess these as primary end-points.

The fixed FSH-dose protocol used has strengths but also has limitations. The objective of this study was to show a true comparison of primary end-points. The fixed-dose protocol supports this approach, as FSH stimulation is less prone to centre practices, as they might be if dose adaptations had been permitted. The limitation of such a design, however, is that optimization of the FSH dose in the case of unexpected low response was not possible. In this respect, the objective of the clinical study does not correspond completely to the usual objectives of clinical practice.

In conclusion, the demonstrated clinical equivalence of Bemfola (Finox AG, Switzerland) to the reference comparator Gonal-f (Merck Serono, Germany) in this clinical trial suggests that Bemfola (Finox AG, Switzerland) as the first FSH-biosimilar on the market may be a viable and familiar alternative to Gonal-f (Merck Serono, Germany).

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