

ORIGINAL ARTICLE

Comparison of follitropin- β administered by a pen device with conventional syringe in an ART programme – a retrospective study

G. A. Rama Raju MBBS DNB, K. Suryanarayana MBBS Dipl (Ortho), G. Jaya Prakash M Pharm PhD and K. Murali Krishna M Pharm PhD
Krishna IVF Clinic, Visakhapatnam, Andhra Pradesh, India

SUMMARY

Objectives: This study compares the efficacy and patient tolerance of follitropin- β (recagon) administered using a pen device with conventional syringe in infertile couples undergoing *in vitro* fertilization/intracytoplasmic sperm injection treatment.

Methods: Data for 481 patients were retrieved retrospectively for the analysis. Conventional syringe group constituted 204 patients with 217 cycles and 265 patients with 294 cycles in the pen-device group. Down-regulation was achieved with GnRH agonist.

Results: Comparison of follitropin- β administered with pen and syringe showed the following data, respectively. A total dose of 1909.38/2100.65 IU ($P < 0.001$), duration of stimulation, 9.70/10.47 days ($P < 0.05$), oestradiol levels on the day of human chorionic gonadotropin, 1488.34/1067.63 pg/ml, number of follicles reaching >16 -mm size, 9.75/7.34 ($P < 0.05$), number of oocytes retrieved, 13.84/9.55 ($P < 0.001$) and number of embryos available for freezing, 4.56/1.30 ($P < 0.05$), the above data were observed in pen/conventional syringe groups, respectively. The live birth rates per cycle were 28.85% and 30.95% in the conventional syringe/pen-device groups, respectively. Patient tolerance with respect to pain at injection site was better with the pen device ($P < 0.025$).

Conclusion: The data show that follitropin- β administered with pen device is well tolerated

and more efficacious with respect to ovarian stimulation outcome compared with the conventional syringe.

Keywords: controlled ovarian stimulation, *in vitro* fertilization, infertility, local tolerance, pen device, recombinant follicle-stimulating hormone

INTRODUCTION

Recombinant DNA technology greatly improved the fertility treatment protocols with the introduction of purified gonadotropins viz. follitropin- α and - β (1–3). The recombinant follicle-stimulating hormone (rFSH) formulations have absolute purity and high reproducibility than gonadotropins derived from urinary source, and moreover these gonadotropins were shown to be suitable for subcutaneous administration (4). Earlier reports concluded that women under WHO-II category (normogonadotrophic anovulatory women) required a lower total dose and shorter duration of treatment with rFSH than when treated with urinary FSH to achieve ovulation in their first cycle (5). In 1999, a meta-analysis by Daya and Gunby revealed that ovarian stimulation with recombinant FSH resulted in increased number of oocytes, embryos and pregnancies in comparison with urinary FSH (6). Recagon (follitropin- β), a recombinant follicle stimulation hormone was available in India only in liquid form until 2003 and was meant for administration using conventional syringe. Later in 2003, a ready-to-use solution in cartridge formulation that can be self-administered by a pen device was introduced. It was the first multiple-use pen device available for subcutaneous administration of rFSH, which could deliver 50–450 IU of rFSH, in dose increments of 25 IU (7).

Received 17 November 2007, Accepted 17 March 2008
Correspondence: G. A. Rama Raju, Krishna IVF Clinic, Visakhapatnam, Andhra Pradesh, India. Tel.: +91 0891 2706164/2706722; fax: +91 0891 2706712; e-mail: krishnaivf@yahoo.com

This study is a retrospective analysis of patients who received the follitropin- β by two formulations, i.e. follitropin- β solution in vial form and follitropin- β solution in cartridge form during the period of 2000–2006. The objective of this study was to compare and evaluate the clinical efficacy and patient tolerance of the follitropin- β administered using pen device and syringe.

MATERIALS AND METHODS

This retrospective analysis included 481 couples who underwent *in vitro* fertilization (IVF)/intracytoplasmic sperm injection treatment (ICSI) at our clinic. The institutional review board gave its approval to review the records of infertile couples who underwent IVF/ICSI treatment at our clinic during the period 2000–2006.

Patient population

Patients were selected according to the inclusion/exclusion criteria as described below. The main inclusion criteria included patients with an age range of 18–36 years at the time of screening and having a body mass index (BMI) of 18–29 kg/m². The inclusion criteria also required the patients to possess both ovaries and have regular menstrual cycles (24–35 days). The exclusion criteria were patients with polycystic ovaries, hyperprolactinaemia, history of poor response to FSH/HMG, previous hospitalization on account of severe ovarian hyperstimulation syndrome (OHSS), contraindications for the use of gonadotropins and gonadotropin-releasing hormone (GnRH) agonists or antagonists.

Study design

A long protocol was followed for down-regulating the patients with GnRH agonist (decapeptyl depot, 3.75 mg, organon) from the mid-luteal phase onwards. When serum oestradiol levels were <50 pg/ml and a transvaginal ultrasound scan ruled out any ovarian activity, ovarian stimulation was started. Serum hCG test was carried out before the first injection of follitropin- β to exclude any ongoing pregnancy. Patients were allowed to administer the follitropin- β (Recagon[®]; Organon, Hyderabad, India) with a pen device equipped with a needle having 29G \times 13 mm diameter (Recagon Pen[™]) or

with a conventional syringe connected with a needle having 25G \times 16 mm diameter (Recagon solution[™]) under medical supervision. Ultrasound was performed every alternate day throughout the stimulation period. Follitropin- β meant to be administered using pen device was available in cartridges containing 300 and 600 IU, and follitropin- β solution was supplied at 50 and 100 IU strengths for administration using conventional syringes. The starting dose of follitropin- β for both the formulations was 200 IU; however, later the dose was adjusted based on the patient response during the stimulation period. Oestradiol levels were measured on day 4 of stimulation and again on the day of hCG administration. If the day 4 oestradiol levels were less than 50 pg/ml or greater than 300 pg/ml, and with a poor ovarian response of less than three follicles or too many follicles with a likelihood development of OHSS, further stimulation was stopped and the cycle was cancelled. Ovulation was induced with 10 000 IU of hCG (pregnyl[®]; Organon) administered as a single i.m. injection, when minimum three follicles with \geq 16-mm size or greater were observed in ultrasound scan. Transvaginal ultrasound-guided oocyte retrieval was carried out under general anaesthesia after 36 h of hCG administration. IVF and ICSI procedures were carried out as per previously described protocols (8–10). After fertilization, embryo transfers were performed on day 3. Luteal support was given by progesterone vaginal suppositories (Uterogestan; Laboratoires Besins International, Paris, France) starting from the day of oocyte retrieval and further continued up to 5 weeks whenever a pregnancy was reported. Two weeks after embryo transfer, hCG was measured for confirmation of pregnancy. A diagnosis of clinical pregnancy was made after visualization of foetal heart pulsation 4 weeks later by transvaginal sonography.

Hormone measurements

Serum oestradiol levels were measured at the initiation of ovarian stimulation, on day 4 of follitropin- β treatment and on the day of hCG administration. Serum hCG was estimated 14 days after embryo transfer for confirmation of pregnancy. Serum oestradiol and hCG levels were measured using enzyme-linked fluourometric assay (ELFA) (Biomerieux, Marcy-l'Étoile, France).

General parameters

Patients' preliminary data include age, BMI, duration of infertility and cause of infertility, which were recorded.

Efficacy parameters

Clinical efficacy was evaluated by estimating the following variables: total dose of follitropin- β , the duration of follitropin- β treatment, the oestradiol levels and number of follicles reaching >16-mm size on the day of hCG administration, the number of oocytes retrieved, the number of oocytes fertilized, the number of embryos transferred, the number of embryos available for freezing, clinical pregnancy, live birth and miscarriage rates.

Local tolerance

The severity of local tolerance symptoms was assessed by the patients themselves after follitropin- β administration either with pen device or conventional syringe throughout the stimulation period and they were asked to report to the staff nurse about any discomfort related to injection of follitropin- β . These were classified as pain, redness, itching, bruising and swelling. The severity of symptoms was scored as none, mild, moderate and severe. The local tolerance data were maintained for individual patients.

Statistical analysis

The descriptive statistics was used for baseline characteristics (age, BMI and duration of infertility) and Student's *t*-test was used to find out the statistical significance for efficacy parameters and chi-squared test was used for local tolerance data. All statistical analyses were done using Sigma stat 3.1 software (Systat Software Asia Pacific Limited, Bangalore, India).

RESULTS

Study population

A total of 481 couples met the eligibility criteria and the data were retrieved for these patients from the patient data base (file maker pro) for the per-

iod between March 2000 and September 2006. In a total of 481 patients, 12 patients failed to start ovarian stimulation on account of spontaneous pregnancy during down-regulation and personal problems. Of the remaining 469 patients, conventional syringe group constituted 204 patients resulting in 217 cycles and 265 patients included in pen-device group resulted in 294 cycles. In the conventional syringe group, about 12 cycles were cancelled and in pen-device group, seven cycles were cancelled on account of poor ovarian response or OHSS. In the end, there were 205 cycles in conventional syringe group and 287 cycles in pen-device group available for final analysis (Table 1). There was no significant difference in the mean maternal age, BMI and duration of infertility between patients of conventional syringe group and pen-device group (Table 2). Male factor infertility was the main reason for the cause of infertility in the patients of both the groups and constituted 44.11% and 42.26% in conventional syringe and pen-device groups, respectively (Table 3).

Local tolerance

The severity of local tolerance symptoms attributed to follitropin- β injections with either pen or conventional syringe are presented in Table 4. To the extent of 20.75% and 36.76% of the patients reported mild discomfort in pen and conventional syringe group, respectively. A moderate level of discomfort was reported by 4.52% of patients in the

Table 1. Subject and treatment allocation patients administered with follitropin- β using pen device and conventional syringe

Parameters	Follitropin- β with pen group	Follitropin- β with conventional syringe group
No. of patients	270	211
Lost for follow up	5	7
No. of cycles	294	217
IVF	59	44
ICSI	208	142
TESE ICSI	20	19
Cancellations	7	12

IVF, *in vitro* fertilization; ICSI, intracytoplasmic sperm injection treatment; TESE, testicular sperm extraction.

Table 2. Subject demographics and baseline parameters

Parameters	Follitropin- β with pen group ($n = 265$)	Follitropin- β with conventional syringe group ($n = 204$)
Age (years)	29.98 \pm 4.99	29.99 \pm 4.82
BMI	26.36 \pm 4.17	26.03 \pm 3.67
Duration of infertility (years)	7.89 \pm 4.64	9.36 \pm 5.38

Values are mean \pm SD.

Table 3. Cause of infertility in the patients of follitropin- β with pen and syringe groups

Parameters	Follitropin- β with pen group ($n = 265$)	Follitropin- β with conventional syringe group ($n = 204$)
Tubal (%)	42 (15.84)	23 (11.27)
Endometriosis (%)	53 (20)	41 (20.09)
Male (%)	112 (42.26)	90 (44.11)
Tubal + male (%)	13 (4.90)	12 (5.88)
Endometriosis + male (%)	23 (8.67)	13 (6.37)
Endometriosis + tubal (%)	2 (0.75)	1 (0.49)
Unexplained (%)	11 (4.15)	14 (6.86)
Other (%)	9 (3.39)	10 (4.90)

pen and 28.43% in the conventional syringe group. There was no severe local discomfort in the pen group; however, about 6.37% of patients in the conventional syringe group reported severe local symptoms in the form of pain and bruising. Though there was no significant difference in most of the local symptoms between patients of pen and conventional syringe groups, the incidence of pain was significantly higher in the syringe-administered group ($P < 0.025$).

Efficacy parameters

A starting dose of 200 IU of follitropin- β was initiated for patients in both the groups and the dose was later adjusted based on the patient response. Patients administered follitropin- β with pen device required significantly ($P < 0.001$) less total dose

Table 4. Local tolerance parameters in the patients of follitropin- β with pen and syringe groups

	Follitropin- β with pen group ($n = 265$)	Follitropin- β with conventional syringe group ($n = 204$)
Itching		
None	259	196
Mild (%)	6 (2.26)	8 (3.92)
Moderate	0	0
Severe	0	0
Pain		
None	240*	133
Mild (%)	20 (7.54)	25 (12.25)
Moderate (%)	5 (1.88)	36 (17.64)
Severe	0	10 (4.90)
Bruising		
None	253	184
Mild (%)	9 (3.39)	12 (5.88)
Moderate (%)	3 (1.13)	5 (2.45)
Severe (%)	0	3 (1.47)
Swelling		
None	263	194
Mild (%)	3 (1.13)	4 (1.96)
Moderate (%)	0	6 (2.94)
Severe	0	0
Redness		
None	244	167
Mild (%)	17 (6.41)	26 (12.74)
Moderate (%)	4 (1.50)	11 (5.39)
Severe	0	0

* $P < 0.05$, compared to follitropin- β with conventional syringe group.

(a difference of 16%) and significantly ($P < 0.05$) shorter duration of stimulation compared with patients administered follitropin- β with conventional syringe. There was no significant difference in basal and day 4 oestradiol levels between the patients of both the groups; however, oestradiol levels on the day of hCG administration were significantly increased in patients of pen-device group compared with patients of conventional syringe group ($P < 0.01$). This data suggest the number of follicles reaching >16-mm size was significantly increased in the patients of the pen-device group compared with the conventional syringe group ($P < 0.05$). There was an increase in number of oocytes retrieved, in patients of pen-device group compared with the conventional

Table 5. Efficacy variables in the patients of follitropin- β with pen and syringe groups

Parameters	Follitropin- β with pen group ($n = 265$)	Follitropin- β with conventional syringe group ($n = 204$)
Total dose (IU)	1909.38 \pm 506.15*	2100.65 \pm 557.71
Duration of treatment (days)	9.70 \pm 1.40**	10.47 \pm 1.51
Basal E ₂ (pg/mL)	26.80 \pm 20.37	27.71 \pm 17.41
Day 4 E ₂ (pg/mL)	159.92 \pm 147.79	139.51 \pm 156.82
E ₂ on the day of hCG administration (pg/mL)	1488.34 \pm 767.82***	1067.63 \pm 837.69
No. of follicles > 16-mm size	9.75 \pm 4.18**	7.34 \pm 5.28
No. of oocytes retrieved	13.84 \pm 6.6*	9.55 \pm 4.12
Fertilization (%)	64.95 \pm 19.92***	60.11 \pm 17.49
No. of Embryos transferred	3.13 \pm 1.45	3.20 \pm 1.09
No. of embryos available for freezing	4.56 \pm 3.72**	1.30 \pm 1.61

Values are mean \pm SD.

* $P < 0.001$, ** $P < 0.05$, *** $P < 0.01$ compared to follitropin- β with conventional syringe group.

Table 6. Clinical outcome in the patients of follitropin- β with pen and syringe groups

Parameters	Follitropin- β with pen group ($n = 265$)	Follitropin- β with conventional syringe group ($n = 204$)
Positive hCG per attempt (%)	43.87	38.70
Positive hCG per transfer (%)	44.94	40.97
Clinical pregnancy rate (%)	41.49	36.40
Implantation rate (%)	38.60	32.15
Live birth rate per cycle	30.95	28.85
Biochemical pregnancies (%)	7 (5.51)	5 (5.95)
Miscarriages (%)	31 (24.40)	17 (20.23)

syringe group ($P < 0.001$). The rate of fertilization was significantly increased in the patients of pen-device group (64.95%) compared with conventional syringe group (60.11%) and number of embryos available for freezing was also increased in pen-device group compared with conventional syringe group ($P < 0.05$) (Table 5).

The pregnancy rate of the patients of the conventional syringe and pen-device group was 40.97% and 44.94% (positive hCG per embryo transfer), respectively. The embryo implantation rate in the patients of conventional syringe groups was 32.15% and 38.60% in case of patients of pen-device group. The live birth rate per cycle was 28.85% and 30.95% in the conventional syringe and pen-device groups, respectively. Although the observed pregnancy rate, rate of implantation and live birth rate in pen-device group are compara-

tively increased with syringe group; however, these findings are not statistically significant. The percentage of biochemical pregnancies and miscarriages was 26.18% in the patients of conventional syringe group and 29.91% in the patients of pen-device group (Table 6).

DISCUSSION

This study is the first retrospective analysis comparing the efficacy of follitropin- β -administered treatment using a pen device with the conventional syringe in the patients of Indian subcontinent undergoing infertility treatment. The introduction of pen-device for administration of recombinant insulin in the treatment of diabetes resulted in a great relief to the patients in terms of local discomfort, accurate dosing and convenience to

the patients (11–13). This is followed by the introduction of pen-device for the delivery of gonadotropins in the year 2001 globally and by 2003 in India. Pharmacokinetic data of follitropin- β indicated that administration of similar amounts of follitropin- β using two different formulations, i.e. either a dissolved cake with a conventional syringe or a ready for use solution with a pen device were bioequivalent with respect to the main pharmacokinetic parameters for rate and extent of absorption (7). The available literature suggests that there were no reported comparative studies to evaluate the efficacy of follitropin- β using pen and syringe. Craenmehr *et al.* (2001) (14) evaluated the local tolerance of follitropin- β pen device with the follitropin- α syringe. Platteau *et al.* (2003) (15), studied the efficacy and local tolerability of follitropin- β pen with the follitropin- α syringe. Kettle *et al.* (2004) evaluated the follitropin- β pen device in clomiphene citrate-resistant women with chronic anovulation (WHO group-II) for its safety and efficacy. In their study, tolerability was assessed on day 2 and day 7 of follitropin- β administration and it was observed that itching, pain, bruising and redness at the site of injection were scored as none or mild in 100% of subjects on treatment day 2 and day 7. Swelling was scored as none or mild on day 2 by 97.7% of the subjects and on day 7 by 100% of the subjects (16).

In this study, pituitary desensitization was achieved with GnRH agonist depot therapy (decapeptyl, 3.75 mg) in contrast to studies by Platteau *et al.* (2003) (15) and Pang *et al.* (2003) (17) wherein buserelin nasal spray and GnRH antagonist protocols were employed. The administration of follitropin- β using pen device resulted in overall decrease in the total dose, number of days of stimulation, recruitment of increased number of follicles, which in turn resulted in higher number of oocytes retrieved and increased availability of embryos for transfer and freezing when compared with conventional syringe and these observations are in accordance with the earlier reports by Platteau *et al.* (2003) (15) and Pang *et al.* (2003) (17). Kettle *et al.* (2004) (16) reported ovulation rate of 95.3% in women with WHO group II anovulatory infertility using follitropin- β pen device, further confirming its efficacy.

The advantage of administration of follitropin- β using pen device in comparison with syringe is

supported by less local problems reported by the patients during the study period. These findings were in accordance with earlier studies, in which volunteers administered with follitropin- β using a pen device experienced less local problems than when receiving follitropin- α using a conventional syringe and this has been attributed on account of the higher gauge needle used and smaller volume of follitropin- β injected by the pen (14). In another study, it was shown that patient compliance in subjects who were administered follitropin- β using a pen was significantly better compared with follitropin- α administered with conventional syringe group (15).

CONCLUSIONS

Many of the infertile couples face obstacles in an attempt to build their family. One such problem is the financial costs involved in the fertility treatment and moreover infertility treatment in countries like India is not covered under insurance policy as is the case with their western counterparts. Hence, the introduction of patient-friendly protocols in terms of ease of administration, efficacy and economics will be of a great advantage to the needy couples. Pang (2005) (18), opined that any device that makes drug delivery easier and simpler for the patient will reduce the anxiety associated with the treatment and makes the treatment less stressful. One such introduction was the development of pen device for the administration rFSH during ovarian stimulation for IVF. Data from this study indicated that the introduction of pen device for the administration of follitropin- β in infertile patients is well tolerated and clinically efficacious in terms of embryological parameters compared with conventional syringe. Economically, introduction of pen device was more beneficial when compared with conventional syringe as total number of days of treatment and dosage required is reduced. Self-administration is possible with proper instructions resulting in fewer visits to the clinic there by minimizing hospital costs.

ACKNOWLEDGEMENT

We would like to thank K. Madan for his help in the preparation of this manuscript.

REFERENCES

- Howles CM, Loumaye E, Giroud D, Luyet G (1994) Multiple follicular development and ovarian steroidogenesis following subcutaneous administration of a highly purified urinary FSH preparation in pituitary desensitized women undergoing IVF: a multicentre European phase III study. *Human Reproduction*, **9**, 424–430.
- Howles CM (1996) Genetic engineering of human FSH (Gonal-F). *Human Reproduction Update*, **2**, 172–191.
- Harlin J, Czemiczky G, Wrambsy H, Fried G (2000) Recombinant follicle-stimulating hormone in in-vitro fertilization treatment—clinical experience with follitropin alpha and follitropin beta. *Human Reproduction*, **15**, 239–244.
- Out HJ, Mannaerts BMJL, Driessen SGAJ, Coelingh Bennink HJT (1997) Recombinant follicle-stimulating hormone (follitropin beta, Puregon) yields higher pregnancy rates in *in vitro* fertilization than urinary gonadotropins. *Fertility and Sterility*, **68**, 138–142.
- Coelingh Bennink HJ, Fauser BC, Out HJ (1998) Recombinant follicle-stimulating hormone (FSH; Puregon) is more efficient than urinary FSH (Metrodin) in women with clomiphene citrate-resistant, normogonadotropic, chronic anovulation: a prospective, multicenter, assessor-blind, randomized, clinical trial. European Puregon Collaborative Anovulation Study Group. *Fertility and Sterility*, **69**, 19–25.
- Daya S, Gunby J (1999) Recombinant versus urinary FSH for ovarian stimulation in assisted reproduction. *Human Reproduction*, **14**, 2207–2215.
- Voortman G, van de Post J, Schoemaker RC, van Gerven JMA (1999) Bioequivalence of subcutaneous injections of recombinant human follicle stimulating hormone (Puregon[®]) by Pen-injector and syringe. *Human Reproduction*, **7**, 1698–1702.
- Devroey P, Tjandraprawira K, Mannaerts B *et al.* (1995) A randomized, assessor-blind, group-comparative efficacy study to compare the effects of Normegon and Metrodin in infertile female patients undergoing in-vitro fertilization. *Human Reproduction*, **10**, 332–337.
- Joris H, Nagy Z, Van de Velde H, De Vos A, Van Steirteghem A (1998) Intracytoplasmic sperm injection: laboratory set-up and injection procedure. *Human Reproduction*, **13**(Suppl. 1), 76–86.
- Van Steirteghem A, Nagy P, Joris H *et al.* (1998) Results of intracytoplasmic sperm injection with ejaculated, fresh and frozen-thawed epididymal and testicular spermatozoa. *Human Reproduction*, **13**(Suppl. 1), 134–142.
- Nancy J, Bohannon MD (1999) Insulin delivery using pen devices – simple to use tools may help young and old alike. *Postgraduate Medicine*, **106**, 57–68.
- Robertson KE, Glazer NB, Campbell RK (2000) The latest development in insulin injection devices. *Diabetes Educator*, **26**, 135–138.
- Bohannon NJ, Ohannesian JP, Burdan AL, Holcombe JH, Zagar A (2000) Patient and physician satisfaction with the Humulin/Humalog pen, a new 3.0 ml pre-filled pen device for insulin delivery. *Clinical Therapeutics*, **22**, 1049–1067.
- Craenmehr E, Bontje PM, Hoomans E, Voortman G, Mannaerts BMJL (2001) Follitropin- β administered by pen device has superior local tolerance compared with follitropin- α administered by conventional syringe. *Reproductive Biomedicine Online*, **3**, 185–189.
- Platteau P, Laurent E, Albano C *et al.* (2003) An open, randomized, single-centre study to compare the efficacy and convenience of follitropin beta administered by a pen device with follitropin alfa administered by a conventional syringe in women undergoing ovarian stimulation for IVF/ICSI. *Human Reproduction*, **18**, 1200–1204.
- Kettel LM, Scholl G, Bonaventura L, Pang S, Sacks P, Chantilis S (2004) Evaluation of pen device for self-administration of recombinant human FSH in clomiphene citrate-resistant anovulatory women undergoing ovulation induction. *Reproductive Biomedicine Online*, **9**, 373–380.
- Pang S, Kaplan B, Karande V, Westphal LM, Scott R, Givens C, Sacks P (2003) Administration of recombinant human FSH (solution in cartridge) with a pen device in women undergoing ovarian stimulation. *Reproductive Biomedicine Online*, **7**, 319–326.
- Pang CS (2005) A pen injection device for self-administration of recombinant follicle-stimulating hormone for fertility treatments. *Expert review of medical devices*, **2**, 27–32.