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Comparison of Two Doses of Recombinant Human Chorionic Gonadotropin (rhCG) During Ovulation Induction in Intrauterine Insemination Cycles: A Prospective Randomized Clinical Trial

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Abstract: The purpose of this study was to compare the effectiveness of two doses of recombinant hCG (500 and 250 μg) on the reproductive outcome through triggering of oocyte maturation. The study was a randomized controlled clinical trial. Healthy women undergoing IUI cycles (n = 66) were randomly assigned to one of two groups at the start of the cycle. Group control (n = 33) received rhCG (250 μg) and group experiment (n = 33) received rhCG (500 μg). Controlled ovarian hyperstimulation was achieved using clomiphene or letrozole and hMG. Semen specimens were washed using the swim up method and IUI using a volume of 0.3 mL was performed 42 h after rhCG injection. No difference was shown in terms of obtained total follicles and pregnancy rate in both groups. However, when all of the cycles who given 250 or 500 μg of rhCG were stratified by the BMI (more and less than 25 kg m^{-2}), Total follicles (more and less than 2 follicles) and infertility duration (more and less than 5 years), the reproductive outcome in the patients with less than 5 years infertility duration and less than 25 kg m^{-2} BMI was more pronounced than the patients with more than 5 years infertility duration and more than 25 kg m^{-2} BMI but the other parameter was not affected the reproductive outcome. No clinical or statistical improvement could be demonstrated, except infertility duration and BMI, for the higher dose of recombinant hCG in women. However, further well-designed studies are essential to offer a final conclusion.

Key words: Recombinant hCG, body mass index, infertility duration, follicles, reproductive outcome

INTRODUCTION

In general, hCG has been used as an alternative to LH for inducing final follicular maturation and ovulation in women undergoing ovarian stimulation for intrauterine insemination (IUI) or intracytoplasmic sperm injection (ICSI)(Melli *et al*., 2007; Moslemizadeh *et al*., 2008).

The success in inducing ovulation of follicles is dependent on the optimum concentration of LH required for the initiating meiosis and triggering the release of the cumulus-oocyte complex into the follicular fluid (Salha *et al*., 2001; Ghasemzad *et al*., 2007; Iheukwumere *et al*., 2008).

Several clinical studies have shown that the 250 and 500 μg of rhCG which represents the lower and upper limits of the dose range, have been inconsistently effective in terms of obtained number of oocytes, clinical and ongoing pregnancies, delivery and miscarriage rates (Kahraman *et al*., 2010; Kashyap *et al*., 2010; Humaidan *et al*., 2010; Bstandig *et al*., 2005). However, the clinical use of rhCG is associated with certain unwanted effects attributed to its biological power, since it is assumed that the biological activity of rhCG is six fold higher than LH, mainly due to its longer half-life and affinity for the common receptor (Gomez *et al*., 2004; Lorzadeh *et al*., 2007). As an example, Ovarian Hyperstimulation Syndrome (OHSS) which is an hCG-dependent phenomenon (Guimera *et al*., 2009; Zargar *et al*., 2011), is mediated through the expression, production and secretion of Vascular Endothelial Growth Factor (VEGF) in human granulosa cells (National Collaborating Centre for Women's and Children's Health, 2004; Rafi *et al*., 2011).

The trouble of high Body Mass Index (BMI), duration of infertility and the poor response in overweight and elderly patients to a standard dosage of rhCG are also discussed (Arora and Samples, 2011).

Therefore, it is not clear, due to controversial literature which dose of recombinant hCG, 250 or 500 μg, is an effective dose to induce final oocyte maturation and also avoid OHSS in patients. The purpose of this study is to conduct a prospective randomized

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study in order to compare two doses of recombinant hCG in women, whether triggering of final oocyte maturation with either 500 μg or the gold standard of 250 μg of rhCG has any effect on the reproductive outcome.

MATERIALS AND METHODS

Drug and media: Recombinant human chorionic gonadotrophin (rhCG) was purchased from Serono Laboratories (Ovidrel; Serono). The sperm wash media was obtained from SAGE (USA).

Study design: The study was a randomized controlled clinical trial. Through the period from June 2009 to April 2010, healthy women between the ages of 22 and 44 years undergoing IUI cycles (n = 66) for the treatment of non-tubal infertility (ovulatory disorders, early-stage endometriosis, mild male factor and idiopathic infertility) were randomly assigned to one of two groups at the start of the cycle. Group 1 (control, n = 33) received rhCG (250 μg) and group 2 (experiment, n = 33) received rhCG (500 μg).

It's worth mentioning that the Infertility is a failure to conceive during a year of unprotected intercourse and as well as for couples who are older or have problems with their reproductive organs, doctors sometimes consider them infertile after six months (Garrido *et al*., 2001).

The Ethics Committee of Jundishapour Ahvaz University of Medical Sciences approved this study.

Patient assessment included demographic information as well as medical and gynaecological histories with physical examination and routine laboratory screening (including BMI, CBC, pap smear, TSH, PRL and viral serology). The partner underwent semen analysis, CBC and viral serology.

Controlled ovarian hyper stimulation was achieved using clomiphene or letrozole and hMG (Pergonal; Serono). Ovarian response was monitored by ultrasound. When two or more follicles were = 16 mm rhCG by dose of 250 or 500 μg was used to induce ovulation.

Semen specimens were washed using the swim up method and a single IUI using a volume of 0.3 mL was performed 42 h after rhCG injection.

Pregnancy was documented by the serum hCG level 2 weeks after the insemination. If pregnant, a vaginal ultrasound was carried out 2 weeks later. The outcome of the pregnancy rate was determined.

End points: The end point of the study was a comparison of side effects and the pregnancy rates between the two groups.

Statistics analysis: All analyses were carried out with the SPSS 16 statistical software. Continuous variables were expressed as mean for numeric variables and also were expressed as number and percent for categorical variables. Categorical variables between groups were compared using Student's t, χ^2 tests or Fisher's exact test where appropriate. Mann-Whitney test was employed to evaluate independent numerical variables because of abnormal distribution. For all other outcomes, a nominal p-value of $p<0.05$ was considered significant

RESULTS

A total of 86 cycles were randomized and available for investigation. Twenty of the women refused to participate to the study. Of the remaining women, 33 patients were injected s.c. with 250 μg of recombinant hCG (rhCG) and 33 women were given 500 μg of rhCG s.c.

The mean ages of all of the patients was 32.3±4.5 years with a range of 22 to 44 years. The median duration of infertility of women was 8.7±8.0 years with a variety of 9 months to 14 years. The mean body mass index (BMI) was 26±6 kg m^{-2} with a range of 20 to 44 kg m^{-2}.

Baseline characteristics were comparable in the two groups. The two groups were found to be identical with respect to age, BMI, kind of infertility, duration of infertility, number of previous trial, duration of stimulation, the type of procedures used, the total dose of gonadotropin injected, number of retrieved follicles and semen analysis(TMC, Motility and morphology of sperm) (Table 1).

When analyzing per cycle, the overall clinical pregnancy rates were 9.9 and 12.1% for group 1 and 2, respectively (Table 1). There was no significant difference in the order of the clinical pregnancy between these two groups.

Table 1: Baseline characteristics women who admitted to the department were comparable in the two groups

	Dosage of rhCG groups		
Parameters	250 μg	500 μg	p-value
Age (years)	28.5±3*	31.9±3	0.963
BMI (kg m^{-2})	27.2±5	28.4±0.69	0.383
Kind of sterility (primary)	19 (57.6)	16 (48.5)	0.970
Kind of sterility (secondary)	14 (42.4)	17 (51.5)	0.241
Duration of infertility	5.03±4.6	6.94±6.8	0.050
Number of follicles	5.5±1	3.8±0.4	0.069
Total sperm count	14500000±505	13200000±710	0.445
Morphology of sperm	8.7±2.9	7.8±3.3	0.470
Motility	38.9%	41.1%	0.231
Dose of gonadotropin	18.0±1.2	17.0±1.9	0.435
Follicle (size = 16 mm)	2.1(25.3)	1.2 (31.6)	0.398
Pregnancy rate	3 (9.9)	4 (12.1)	0.387

*Values are as Mean±SE, Values in brackets are percentage

Table 2: The variety of all of the cycles (n = 66) were categorized by the mean number of follicles into group 1≤2 follicles and group 2>2 follicles

Groups	12≤follicles	2>2 follicles	p-value
250 μg rhCG*	6 (18.2)	27 (81.8)	0.001
500 μg rhCG**	5 (15.2)	28 (84.8)	0.001
Pregnancy rates	0 (0.0)	4.7 (100)	0.001

Values in brackets are percentage, *Patients were given 250 μg rhCG, **Patients were given 500 μg rhCG

Table 3: The variety of all of the cycles (n = 66) were categorized by the mean BMI into group BMI≤25 kg m^{-2} and group BMI>25 kg m^{-2}

Groups	BMI≤25 kg m^{-2}	BMI>25 kg m^{-2}	p-value
250 μg rhCG*	14.0 (24.4)	19.0 (75.6)	0. 05
500 μg rhCG**	11.0 (33.3)	22.0 (66.7)	0.05
Total follicles***	5.2 (52.5)	4.7 (47.5)	0.87
Follicle (Size =16 mm) ****	2.3 (69.7)	1.0 (30.3)	0.05
Pregnancy rates	4.0 (57.2)	3.0 (42.8)	0. 76

Values in brackets are percentage, *Patients were given 250 μg rhCG, **Patients were given 500 μg rhCG, ***Mean total follicles were obtained from each cycles, ****Mean number of follicles obtained that the size of them was more than 16 mm

Table 4: The variety of all of the cycles (n = 66) were categorized by the duration of infertility into group D.I≤5 years and group D.I>5 years

Groups	D.I≤5 years	D.I>5 years	p-value
250 μg rhCG*	20 (60.6)	13 (39.4)	0.001
500 μg rhCG**	16 (48.5)	17 (51.5)	0.387
Mean total follicles***	6.7 (9.6)	3.3 (9.6)	0.001
Follicle (Size =16 mm) ****	2.6 (78.8)	0.6 (11.2)	0.001
Pregnancy rates	3.6 (76.6)	1.1 (23.4)	0.05

Values in brackets are percentage, *Patients were given 250 μg rhCG, **Patients were given 500 μg rhCG, ***Mean total follicles were obtained from each cycles, ****Mean number of follicles obtained that the size of them was more than 16 mm

No significant undesirable reactions at the place of injection were noted in any of the patients. There were no subjects of severe Ovarian Hyper Stimulation Syndrome (OHSS).

Since the mean number of follicles, BMI and also duration of infertility may be have a direct relation with reproductive outcome through triggering of oocyte maturation by the variable dosage of rhCG, the differences between each of them were assessed.

When all of the cycles (n = 66) were categorized by the mean number of follicles into group 1 = 2 follicles and group 2>2 follicles, there was no significant difference in the distribution of rhCG doses between the groups. Also, this difference was not strong statistically and did not affect the pregnancy rates (Table 2). The proportion of follicles with diameters more than 16 mm to the total follicles was not significant in both more and less than two follicles groups whether one or two rhCG preparations were injected (Table 2).

When all of the women were stratified according to their BMI (BMI = 25 kg m^{-2} and BMI>25 kg m^{-2}), there was no significant difference in the distribution of rhCG doses between the groups as well. However, When 250 μg rhCG was used, a significant difference was observed in the mean of the BMI = 25 kg m^{-2} in comparison to the mean BMI>25 kg m^{-2} in terms of total follicles. Nevertheless, this difference was not strong statistically to affect the rate of pregnancy. The proportion of follicles with diameters more than 16 mm to the total follicles was not significant in both more and less than 25 kg m^{-2} BMI groups whether one or two rhCG preparations were injected (Table 3).

When all of the patients were stratified according to their duration of infertility (I.D>5 years and I.D>5 years), there was significant difference in the distribution of rhCG doses between the groups.

Also the significant differences were found in the I.D>5 years group in comparison to the I.D>5 years group in terms of total follicles and follicles with more than 16 mm size ($p<0.05$ and $p<0.05$). Additionally, the pregnancy rates in the patients with less than 5 years infertility duration was more pronounced than the patients with more than 5 years infertility duration($p<0.05$) (Table 4).

DISCUSSION

The results of the present study have shown that both doses were capable of inducing follicular maturation and ovulation. Indeed, the number of oocytes, on the day of intrauterine insemination was not significantly higher with two of recombinant hCG preparations than with the single dose. However, whether the quality of the follicles released in these forms is optimal stays to be clarified. These findings are also in accordance with other three studies done by Chang *et al.* (2001), Sakhel *et al.* (2007) and Tsoumpou *et al.* (2009). Chang *et al.* (2001) were shown that both of doses are effective and well tolerated in the induction of MII oocyte and ovulation in patients undergoing IVF treatment. Sakhel *et al.* (2007) have also found similar clinical outcomes with 250 μg of recombinant hCG or 500 μg urinary hCG during IVF treatment in normal responder patients. Tsoumpou *et al.* (2009) obtained the same number of mature oocytes and similar implantation rates in embryos derived from women treated with single and two dose of hCG.

In contrast, Gomez *et al.* (2004) reported that low dose of rhCG is more efficient to achieve optimal oocyte maturation, with fewer incidences of OHSS, than a high dose of rhCG. Busso *et al.* (2010) observed that the rhCG at the highest dose increase VP and expression of VEGF that may be running the risk of provoking Ovarian Hyper Stimulation Syndrome (OHSS) or Poly Cystic Ovarian Syndrome (PCOS).

The present study also identified that the mean number follicles with more than 16 mm size were identical in single and two doses of rhCG, but comparison of clinical pregnancy and also healthy ongoing rates could be a supplementary step towards understanding the advantages and disadvantages of different gonadotrophin doses.

Although, the events initiated by the mid-phase surge of LH and FSH are presented together, certain amount of LH and FSH may be needed in order for these events to happen (Moeini *et al*., 2009). Bomsel-Helmreich *et al*. (1989) was shown that lower doses of hCG induce nuclear maturation but inducing follicular maturation needed higher doses. This may also be the case in humans where a time-or dose-dependent phenomenon leads to the initial elevation in progesterone 12 h before the LH surge, the final maturation of the oocyte 32 h after the surge and ovulation 36 h after the LH surge (Segers *et al*., 2008; Fauser *et al*., 2002; Kilic *et al*., 2010). Thus, it would be logical to determine the optimal rhCG dose to induce ovulation.

In the present experiment no difference was shown in terms of obtained total follicles and pregnancy rate in patients, if they were not categorized according to their BMI, infertility duration and the range number of follicles, whether 250 or 500 μg of rhCG was injected for the final maturation of follicles. However, when all of the cycles were stratified by the above mentioned parameters (BMI: 25 = vs. >25 kg m^{-2}, group 1 = 2 follicles > group 2 and infertility duration 5 = vs.>5 year), the reproductive outcome in the patients with less than 5 years infertility duration and as well as less than 25 kg m^{-2} BMI was more pronounced than the patients with more than 5 years infertility duration and more than 25 kg m^{-2} BMI but the other parameter was not affected the reproductive outcome.

In patients undergoing IVF treatment, fatness has been associated with require for higher doses of gonadotropins, increased cycle cancellation rates and less oocytes retrieved (Awartani *et al*., 2009; Erel and Senturk, 2009). Moreover, in obese women undergoing IVF treatment, the rate of embryo transfer, pregnancy and live birth was decreased but the miscarriage rates was increased (Devroey *et al*., 2009; Fedorcsak *et al*., 2004). On other hand, other studies have been able to find positive influence of slenderness on ART outcome (Lorusso *et al*., 2008; Anifandis *et al*., 2005; Martinuzzi *et al*., 2008).

In contrast, other studies have reported that cycle parameters, such as clinical pregnancy, implantation and the occurrences of moderate and severe OHSS, were also found to be no significant in both higher and lower BMI, infertility duration and mean number follicles groups (Chen *et al*., 2010; Bastek *et al*., 2008).

Within the considerations of this prospective randomized experiment it can conclude that 250 μg of recombinant hCG, except in women with a BMI higher than 25 kg m^{-2} or infertility duration more than 5 years, is sufficient and safe to trigger ovulation.

No clinical or statistical advantage could be demonstrated, except infertility duration and BMI, for the higher dose of recombinant hCG in women. However, well-designed, further studies are essential to say a final conclusion.

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REFERENCES

Anifandis, G., E. Koutselini, I. Stefanidis, V. Liakopoulos, C. Leivaditis, T. Mantzavinos and N. Vamvakopoulos, 2005. Serum and follicular fluid leptin levels are correlated with human embryo quality. Reproduction, 130: 917-921.

Arora, K.L. and O. Samples, 2011. Role of body weight on reproductive and physiological traits in Japanese quail layers (*Coturnix japonica*). Int. J. Poult. Sci., 10: 640-643.

Awartani, K.A., S. Nahas, S.H. Al-Hassan, M.A. Al-Deery and S. Coskun, 2009. Infertility treatment outcome in sub groups of obese population. Reprod. Biol. Endocrinol., 27: 52-57.

Bastek, J.A., M.D. Sammel, S.K. Srinivas and M.A. Elovitz, 2008. Is routine infectious and toxicologic screening in preterm labor effective in predicting preterm birth?. Am. J. Obstet. Gynecol., 198: 38-42.

Bomsel-Helmreich, O., L.V.N. Huyen and I. Durand-Gasselin, 1989. Effects of varying doses of HCG on the evolution of preovulatory rabbit follicles and oocytes. Hum. Reprod., 4: 636-642.

Bstandig, B., C. Schumaker, V. Isnard, V. Isnard, P. Ferrari and A. Bongain, 2005. Influence of Body Mass Index (BMI) on successful ovulation triggering by urinary hCG (u-hCG) versus recombinant hCG (r-hCG). Fertil. Steril., 84: 422-423.

Busso, C.E., J.A. Garcia-Velasco, C. Simon and A. Pellicer, 2010. Prevention of OHSS: Current strategies and new insights. Middle East Fertility Soc. J., 15: 223-230.

Chang, P., S. Kenley, T. Burns, G. Denton, K. Currie, G. De-Vane and L. O'Dea, 2001. Recombinant human chorionic gonadotropin (rhCG) in assisted reproductive technology: Results of a clinical trial comparing two doses of rhCG (Ovidrel) to urinary hCG (Profasi) for induction of final follicular maturation in *in vitro* fertilization-embryo transfer. Fertil. Steril., 76: 67-74.

Chen, H., W.J. Wang, Y.Z. Chen, M.Q. Mai, N.Y. Ouyang, J.H. Chen and P. Tuo, 2010. Effects of body mass index and age on the treatment of *in vitro* fertilization-embryo transfer among patients with non-polycystic ovarian syndrome. Zhonghua Liu Xing Bing Xue Za Zhi., 31: 567-571.

Devroey, P., M. Aboulghar, J. Garcia-Velasco, G. Griesinger and P. Humaidan *et al*., 2009. Improving the patient's experience of IVF/ICSI: A proposal for an ovarian stimulation protocol with GnRH antagonist co-treatment. Hum. Reprod., 24: 764-774.

Erel, C.T. and L.M. Senturk, 2009. The impact of body mass index on assisted reproduction. Curr. Opin. Obstet. Gynecol., 21: 228-235.

Fauser, B.C., D. De-Jong, F. Olivennes, H. Wramsby, C. Tay, J. Itskovitz-Eldor and H.G. Van-Hooren, 2002. Endocrine profiles after triggering of final oocyte maturation with GnRH agonist after cotreatment with the GnRH antagonist Ganirelix during ovarian hyperstimulation for *in vitro* fertilization. J. Clin. Endocrinol. Metab., 87: 709-715.

Fedorcsak, P., P.O. Dale, R. Storeng, G. Ertzeid and S. Bjercke *et al*., 2004. Impact of overweight and underweight on assisted reproduction treatment. Hum. Reprod., 19: 2523-2528.

Garrido, N., C. Albert, J.S. Krussel, J.E. O'Connor, J. Remohi, C. Simon and A. Pellicer, 2001. Expression, production and secretion of vascular endothelial growth factor and interleukin-6 by granulosa cells is comparable in women with and without endometriosis. Fertil. Steril., 76: 568-575.

Ghasemzad, A., L. Farzadi and O. Ajalli, 2007. Transvaginal ovarian drilling in infertile women with polycystic ovary syndrome resistant to minimal stimulation. J. Medical Sci., 7: 991-996.

Gomez, R., I. Lima, C. Simon and A. Pellicer, 2004. Administration of low-dose LH induces ovulation and prevents vascular hyperpermeability and vascular endothelial growth factor expression in superovulated rats. Reproduction, 127: 483-489.

Guimera, M., M. Morales-Ruiz, W. Jimenez and J. Balasch, 2009. LH/HCG stimulation of VEGF and adrenomedullin production by follicular fluid macrophages and luteinized granulosa cells. Reprod. Biomed. Online, 18: 743-749.

Humaidan, P., J. Quartarolo and E.G. Papamikolaou, 2010. Preventing ovarian hyperstimulation syndrome: Guidance for the clinician. Fertil Steril., 94: 389-400.

Iheukwumere, F.C., A.H. Abu and I.C. Okoli, 2008. Effect of FSH+LH (Pergonal®) treatments on hormonal profile and superovulatory response of West African dwarf does. Asian J. Scientific Res., 1: 281-287.

Kahraman, S., G. Karlikaya, M. Kavrut and H. Karagozoglu, 2010. A prospective, randomized, controlled study to compare two doses of recombinant human chorionic gonadotropin in serum and follicular fluid in woman with high body mass index. Fertility Sterility, 93: 2084-2087.

Kashyap, S., K. Parker, M.I. Cedars and Z. Rosenwaks, 2010. Ovarian hyperstimulation syndrome prevention strategies: Reducing the human chorionic gonadotropin trigger dose. Semin. Reprod. Med., 28: 475-485.

Kilic, S., N. Yilmaz, E. Zulfikaroglu, E. Sarikaya, K. Kose, O. Topcu and S. Batioglu, 2010. Obesity alters retrieved oocyte count and clinical pregnancy rates in high and poor responder women after *in vitro* fertilization. Arch. Gynecol. Obstet, 282: 89-96.

Lorusso, F., M. Palmisano, G. Serrati, E. Bassi, G. Lamanna, M. Vacca and R. Depalo, 2008. Intrauterine insemination with recombinant or urinary human chorionic gonadotropin: A prospective randomized trial. Gynecol. Endocrinol., 24: 644-648.

Lorzadeh, N., S. Kazemirad, M. Lorzadrh and A. Dehnori, 2007. A comparison of human chorionic gonadotropin with magnesium sulphate in inhibition of preterm labor. J. Medical Sci., 7: 640-644.

Martinuzzi, K., S. Ryan, M. Luna and A.B. Copperman, 2008. Elevated body mass index (BMI) does not adversely affect *in vitro* fertilization outcome in young women. J. Assist. Reprod. Genet., 25: 169-175.

Melli, M.S., S. Tagavi, M. Alizadeh, M. Ghojazadeh and M.K. Sheshvan, 2007. Comparison the effect of oxytocin and human chorionic gonadotropin on ovulation. J. Medical Sci., 7: 1126-1134.

Moeini, M.M., F. Alipour and A. Moghadam, 2009. The effect of human chorionic gonadotropin on the reproduction performance in Lory sheep synchronized with different doses of pregnant mare serum gonadotrophin outside the breeding season. Asian J. Anim. Vet. Adv., 4: 9-15.

Moslemizadeh, N., T.G. Moghadam and S. Ehteshami, 2008. Comparison of clomiphene citrate plus estradiol, with tamoxifen citrate effects in induction of ovulation and pregnancy in poly cystic ovarian syndrome patients. J. Med. Sci., 8: 734-738.

National Collaborating Centre for Women's and Children's Health, 2004. Fertility: Assessment and Treatment for People with Fertility Problems. RCOG Press, UK., ISBN-10: 1-900364-97-2.

Rafi, A., D. Ramakrishna, K. Sabitha, S. Mohanty and P. Rao, 2011. Serum copper and Vascular Endothelial Growth Factor (VEGF-A) in dysfunctional uterine bleeding. Am. J. Biochem. Mol. Biol., 1: 284-290.

Sakhel, K., M. Khedr, S. Schwark, M. Ashraf, M.H. Fakih and M. Abuzeid, 2007. Comparison of urinary and recombinant human chorionic gonadotropin during ovulation induction in intrauterine insemination cycles: A prospective randomized clinical trial. Fertil. Steril., 87: 1357-1362.

Salha, O., T. Dada and V. Sharma, 2001. Influence of body mass index and self-administration of hCG on the outcome of IVF cycles: A prospective cohort study. Hum. Fertil., 4: 37-42.

Segers, I., T. Adriaenssens, W. Coucke, R. Cortvrindt and J. Smitz, 2008. Timing of nuclear maturation and postovulatory aging in oocytes of *in vitro*-grown mouse follicles with or without oil overlay. Biol. Reprod., 78: 859-868.

Tsoumpou, I., J. Muglu, T.A. Gelbaya and L.G. Nardo, 2009. Symposium: Update on prediction and management of OHSS. Optimal dose of HCG for final oocyte maturation in IVF cycles: Absence of evidence?. Reprod. Biomed. Online, 19: 52-58.

Zargar, M., N. Nikbakht, E. Pourmatroud, K. Ghasemi and M. Hemadi, 2011. Comparison of the clinical efficacy of two different cabergoline regimens on prevention of Ovarian Hyperstimulation Syndrome (OHSS). Res. J. Obstetrics Gynecol., 4: 51-58.