Original Article
Efficacy of growth hormone supplementation with gonadotrophins in vitro fertilization for poor ovarian responders: an updated meta-analysis

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Abstract: Growth hormone (GH) is involved in the regulation of male and female infertility. Several clinical studies reveal that adjuvant GH treatment has a possible role in vitro fertilization (IVF) or intracytoplasmic sperm injection (ICSI), especially in poor ovarian responders (POR) undergoing IVF/ICSI. Recent studies suggest that GH addition in POR patients significantly improves the rate of clinical pregnancy and live birth. Databases including PubMed, Embase, the Cochrane Central China National Knowledge Infrastructure (CNKI) and Google Scholar were searched for randomized controlled trials (RCTs) or controlled clinical trials (CCTs) on the effectiveness of GH supplementation with gonadotrophins in IVF/ICSI for POR. Two reviewers independently screened literature according to the inclusion and exclusion criteria, extracted data, and assessed methodological quality. Meta Analyst Beta 3.13 software was used to meta-analysis. Eleven studies (six RCTs and five CCTs) and 3788 subjects (613 subjects in cases group and 3175 subjects in controls group) were included in our study. The results of meta-analysis showed that GH addition significantly increased serum E2 level on the day of HCG (OR = 0.55; 95% CI = 0.127-0.973) and MII oocyte number (OR = 0.827; 95% CI = 0.470-1.184). Furthermore, GH addition significantly improved the number of 2PN (OR = 0.934; 95% CI = 0.206-1.661) and obtained embryos (OR = 0.934; 95% CI = 0.206-1.661). However, no significant difference was found for the overall implantation rate was 8.8% (95% CI = -0.062-0.237) and clinical pregnancy rate was 5.1% (95% CI = -0.033-0.134). The present result revel that GH supplementation for IVF/ICSI in POR increases the probability of serum E2 level on the day of HCG, the number of MII oocyte, 2PN and obtained embryos. However, GH addition does not increase implantation rate and clinical pregnancy rates. Due to the limited quantity and quality of the included studies as well as the difference in methodology, we suggest this above could be taken as a reference for clinical analysis which needs to be further evaluated in its effects.
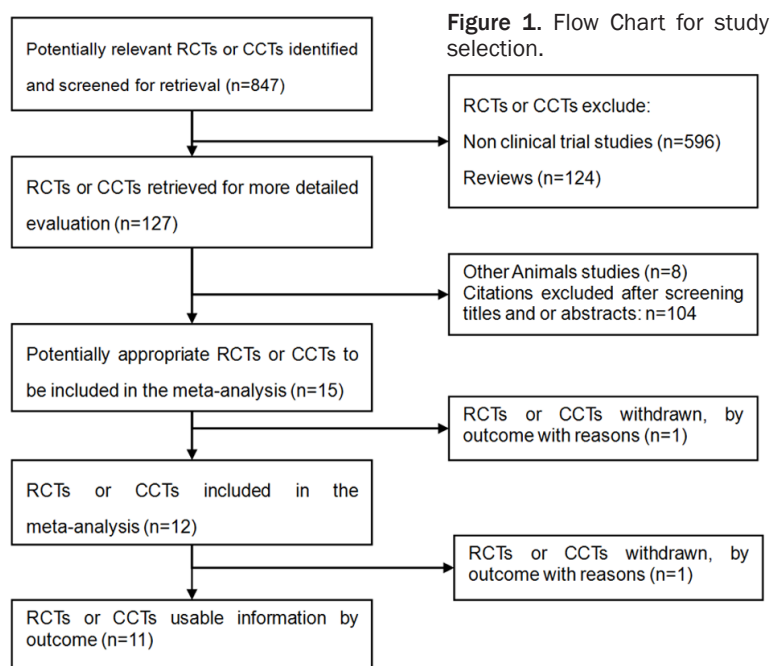
Keywords: Growth hormone, poor ovarian response, gonadotrophin releasing hormone, gonadotrophins, in vitro fertilization, intracytoplasmic sperm injection

Introduction

Poor ovarian response (POR), defined as failure of the development of sufficient number of mature follicles to proceed to oocyte retrieval or yielding only a few oocytes following gonadotrophin stimulation in women undergoing in vitro fertilization (IVF) or intracytoplasmic sperm injection (ICSI) treatment. In 1988, GH was first used as an adjuvant to gonadotrophins for ovarian stimulation [1]. However, a randomized double blind placebo controlled trial conducted by Owen et al [2]. showed that

there are no significant improvement in ovarian response after co-treatment with GH.

Previous study reported that the incidence of POR is increasing and vary from 9% to 24% [3], also, the prevalence of POR was 11.9% in Chinese [4]. Epidemiology study showed that ovarian ageing [5], previous ovarian surgery [4] and high body mass index [6] are associated with POR. However, pathogenesis of POR is still unclear now. Women with POR often respond poorly to controlled ovarian stimulation (COS), which resulting in retrieval of fewer oocytes,

Figure 1. Flow Chart for study selection.

Potentially relevant RCTs or CCTs identified and screened for retrieval (n=847)

RCTs or CCTs exclude:
Non clinical trial studies (n=596)
Reviews (n=124)

RCTs or CCTs retrieved for more detailed evaluation (n=127)

Other Animals studies (n=8)
Citations excluded after screening titles and or abstracts: n=104

Potentially appropriate RCTs or CCTs to be included in the meta-analysis (n=15)

RCTs or CCTs withdrawn, by outcome with reasons (n=1)

RCTs or CCTs included in the meta-analysis (n=12)

RCTs or CCTs withdrawn, by outcome with reasons (n=1)

RCTs or CCTs usable information by outcome (n=11)

controlled trials (RCTs) and controlled clinical trials (CCTs), to further confirm whether receiving regarding have effect on the probability of serum E2 levels on the HCG day, the number of MII oocytes, 2PN, obtained embryos and implantation rate, clinical pregnancy rate.

Materials and methods

Literature searches

We searched PubMed, Embase, the Cochrane Central database, China National Knowledge Infrastructure (CNKI) and Google Scholar databases for relevant article published between 1988 and 2013. Using the following Medical Subject Headings (MeSH) terms "Growth hormone" or "GH", "Low response" or "Poor ovarian response", "Premature ovarian aging" or "Diminished ovarian reserve", "Gonadotrophin releasing hormone (GnRH)", "in vitro fertilization (IVF)" or "intracytoplasmic sperm injection (ICSI)". We only searched the article written in in English and Chinese. The electronically databases were searched up to May 2013. In addition, we also reviewed the references included studies.

Study selection

Only RCTs and CCTs were included in the current review. Include criteria: (a) the target population was either poor-responders or those with diminished ovarian reserve, as described above ,who were undergoing ovarian stimulation plus IVF/ICSI; (b) GH was supplemented before ovarian stimulation in the study group while neither was used in the control; (c) The primary outcomes were E2 levels of HCG day, No. of oocytes collected or MII oocyte number, 2PN rate or No. of 2PN, No. of obtained embryos, implantation rate and Clinical pregnancy rate.

Excluded studies included studies not published as full manuscripts in peer-reviewed journals, reviews, editorials and metaanalyses.

producing poorer quality embryos, reduced implantation rates and pregnancy rates [3]. Several stimulation protocols have been used for the management of poor responders [7, 8]. However, the pregnancy rate of POR after IVF remain disappointingly low especially for women aged > 40 years become accelerated [4].

Growth hormone (GH) supplements have been used as a part of the adjunctive therapy to POR to COS protocols [9]. Three meta-analysis studies reviewed the value of providing GH supplements in IVF treatment [10-12]. GH addition increases the probability of clinical pregnancy and live birth in poor responders undergoing ovarian stimulation with gonadotrophin releasing hormone (GnRH) analogues and gonadotrophins for IVF [10]. Some evidence suggests that addition of GH, as well as performing embryo transfer on day 2 versus day 3, appear to improve the probability of pregnancy [11]. Although using growth hormone in poor responders can significant improve live birth rates, lack of information that in poor responders who will be more suitable for adjuvant growth hormone. Therefore, further research is necessary to fully define its role before recommending growth hormone adjuvant in IVF [12].

Thus, the purpose of this meta-analysis was to update the existing evidence from randomized

Table 1. Characteristics of the studies included

Articles (years)	Study period	Study design	Inclusion criteria	Cases/Controls
Dor J, *et al*. (1995)	Not reported		(i) 17-β oestradiol concentrations on the day of human chorionic gonado-trophin (HCG) administration were <501pg/ml; (ii) number of follicles (> 14 mm) observed in ultrasonography was less than four; (iii) number of re-trieved oocytes following ovum retrieval was three or less.	7/7
Liu DE, *et al*. (2006)	January 2004-December 2005	Randomized controlled trial (RCT)	(i) Basal FSH levels(bFSH)>15 u/L; (ii) number of follicles (> 14 mm) observed was ≤3; (iii) E2 concentrations on the day of human chorionic administration were <1830 pg/ml; (iv) The doses of gonadotropin was > 300 IU/d.	32/56
Huang XH, *et al*. (2007)	006	Case-control	(i) Women age > 40 years; (ii) bFSH> 10 u/L or E2 > 80 pg/ml; (iii) bAFC (Basal antral follicle count) < 5 or no follicles was observed in COH or Women who have failed to obtain fol-licles in previous cycles.	25/25/
Guang Q, *et al*. (2007)	December 2003-December 2004	Randomized controlled trial (RCT)	(i) ≥ 14 mm follicles < 3 follicles was observed in COH on HCG day in previous cycles; (ii) Number of oocytes collected ≤ 3; (iii) Number of embryos transferred per transfer ≤ 2.	20/20
Kucuk T, *et al*. (2008)	January 2005-June 2007	Prospective, randomized trial	patients who responded poorly to high dose gonadotropin treatment in their first cycles in the same center.	31/30
Hazout A, *et al*. (2009)	J7	An open, non-compar-ative and non-random-ized study	(i) at least three previous assisted cycle failures; (ii) regular spontane-ous menstrual cycles of 25-30 days; (iii) FSH, LH, oestradiol, inhibin B and anti-Müllerian hormone concentra-tions in the normal range during the early follicular phase; (iv) unexplained infertility with normal spermatozoa before IVF, or subnormal spermatozoa justifying ICSI; (v) less than 50% of dysmorphic oocytes in their previous assisted cycles; (vi) no treatment with gonadotrophins within 1 month of the treated cycle for the study; (vii) normal uterine cavity; (viii) negative pregnancy test; (ix) willingness to participate and to comply with the protocol.	245/2780
Yang XL, *et al*. (2012)	ber 2010	Case-control	< 5 follicles was observed in COH	54/54

Song HL, et al. (2012)	January 2010-February 2011	Case-control	(i) bFSH≥10 mIU/mL; (ii) Women age >25 years and < 35 years; (iii) Normal menstruation and Duration of infertility ≥ 3 years; (iv) the most female subfertility causes were tubal factors, and exclusion criteria were endocrine or metabolic disorders, such as thyroid disorder, diabetes, hyperpro-lactinemia (HPRL), polycystic ovary syndrome (PCOS) and severe oligoasthenosper-mia			
Eftekhar M, et al. (2013)	January 2009-June 2011	Randomized prospec-tive study	Women who had one or more previous failed IVF-ET cycles with three or fewer retrieved oocytes and with subsequent three or less obtained embryos using GnRH agonist long protocol, and/or E2 levels B 500 pg/mL on the day of human chorionic gonadotropin (hCG) injection enrolled the study. Exclusion criteria were BMI ≥ 30 mg/m², FSH >15 IU/L, endocrine or metabolic disorders, such as diabetes, thyroid disorder, and polycystic ovary syndrome (PCOS), severe endometriosis and azospermia. There was no age limitation for select-ing the subjects.	40/42		
Dor J, et al. (1995)	GnRHa/HMG/GH	18 IU on alternate days, total dose 72 IU	10 000 IU uhCG	IVF	Number of follicles > 14 mm; Number of oocytes collected Fertilization rate (%); Number of embryos replaced	
Liu DE, et al. (2006)	GnRHa/rFSH/GH	4.5 U on every days, total dose 27 U	5000-10000IU uhCG	IVF	Doses of gonadotropin; Days of use gonadotropin E2 on HCG day (pg/ml); Number of oocytes collected 2PN (%); Cleavage rate (%); Im-plantation rate (%); Clinical pregnancy rate (%)	
Huang XH, et al. (2007)	GnRHa/rFSH/HMG/GH	2.0 U on every days, total dose 10 U	10 000 IU uhCG	IVF/ICSI	Doses of gonadotropin; Days of use gonadotropin E2 on HCG day (pg/ml); Number of oocytes collected 2PN (%); Cleavage rate (%); Im-plantation rate (%) Clinical pregnancy	
Guang Q, et al. (2007)	GnRHa/rFSH/GH/aspirin	4.0 IU on alternate days, un-til the criteria for triggeringfi-nal follicular maturation and aspirin 75mg/d on day 1~20 of the cycle	10 000 IU uhCG	IVF	Number of follicles; Number of oocytes collected Number of oocytes collected (MII); Fertilization rate (%) Num-berof embryos transferred per transfer Clinical preg-nancy rate (%)	

Kucuk T, et al. (2008)	GnRHa/rFSH/HMG/GH	2.0 U on every days, total dose 10 U	10 000 IU uhCG	IVF/ICSI	Doses of gonadotropin; Days of use gonadotropin E2 on HCG day (pg/ml); Number of oocytes collected 2PN (%); Cleavage rate (%); Implantation rate (%) Clinical pregnancy	
Hazout A, et al. (2009)	GnRHa/rFSH/HMG/GH	8.0 U (1.33 mg) on every days, until the criteria for triggering final follicular maturation (at least three follicles > 16 mm in diameter and an oestradiol concentration of 140 pg/ml per follicle)	10 000 IU uhCG	IVF	Total gonadotrophins (IU); Stimulation length (days) Oestradiol on HCG day (pg/ml); Number of oocytes collected; Numberof dysmorphic oocytes; Numberof inseminated/injected oocytes; Number of day-2 embryos; Cleavage rate (%) Numberof embryos transferred per transfer Total number of pregnancies (%)	
Niu F and Li LM. (2011)	GnRHa/Gn /GH	4.0 IU on every days, until the criteria for triggering final follicular maturation (at least two follicles > 18 mm)	10 000 IU uhCG	IVF/ICSI	Number of oocytes collected; 2PN (%) Fertilization rate (%) Cleavage rate (%); Number of embryos obtained Number of embryos transferred per transfer Clinical pregnancy rate (%)	
Song HL, et al. (2012)	GnRHa/rFSH/GH	4.5 IU on every days, total dose 22.5 IU	10 000 IU uhCG	IVF	Days of use gonadotropin; Doses of gonadotropin E2 on HCG day (pg/ml); Number of oocytes collected 2PN (%); Number of embryos transferred per transfer Implantation rate (%); Clinical pregnancy (%)	
Yang XL, et al. (2012)	GnRHa/rFSH/GH	4.0 IU on every days, until the criteria for triggering final follicular maturation.	10 000 IU uhCG	IVF/ICSI	Days of use gonadotropin; Doses of gonadotropin Number of oocytes collected (MII); Fertilization rate (%) Cleavage rate (%); Clinical pregnancy rate (%)	
Song HL,et al. (2012)	GnRHa/rFSH/GH	4.5 IU on every days, total dose 22.5I U	10 000 IU uhCG		IVF	Days of use gonadotropin; Doses of gonadotropin E2 on HCG day (pg/ml); Number of oocytes collected 2PN (%); Number of embryos transferred per transfer Implantation rate (%); Clinical pregnancy rate (%)

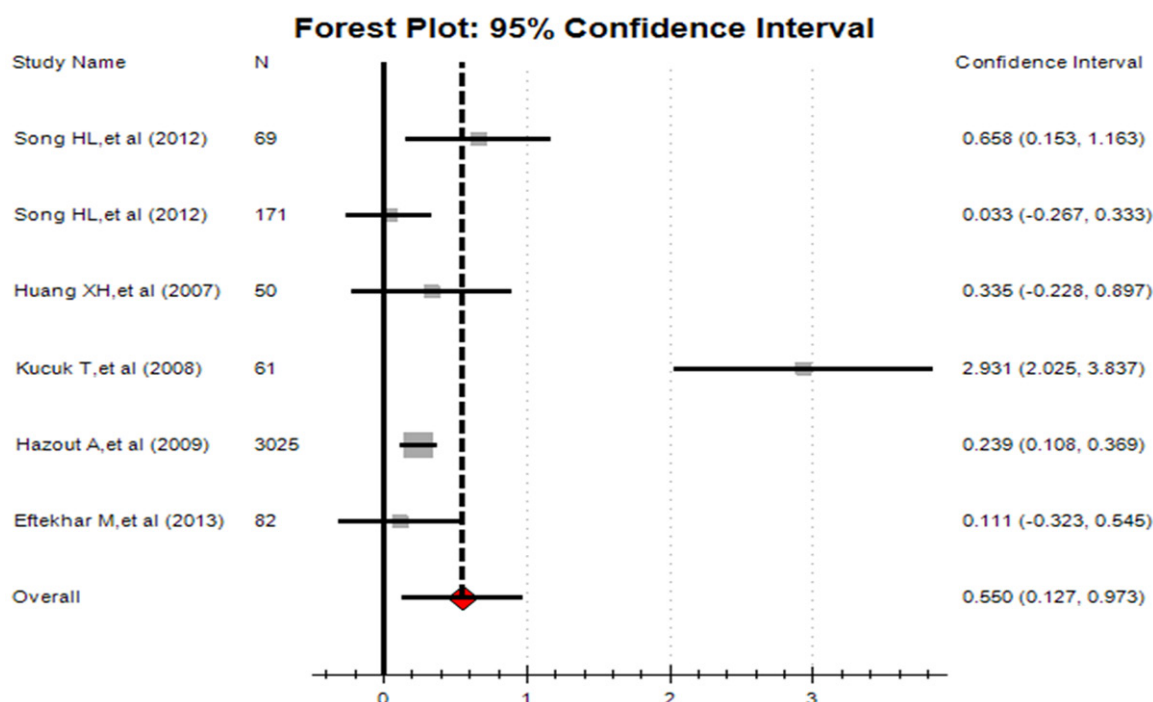| Eftekhar M,*et al.* (2013) | GnRHant/HMG/GH | 4.0 IU on every day, from day 21 of previous cycle until the day of hCG injection. | 10 000 IU uhCG | IVF/ICSI | Stimulation days (days); Total doses of gonadotropin (IU) Endometrial thickness(mm); E2 levels of hCG day (pg/mL); Failed oocytes retrieval, (%); Failed fertilization, (%); Total cycle cancelation, n/n (%); Number of MII oocytes No. of retrieved oocytes; No. of two pronucleate No. of obtained embryos; No. of transferred embryos ICSI cycles (%) IVF cycles (%); Fertilization rate (%) Implantation rate (%); Chemical pregnancy rate/cycle (%) Clinical pregnancy rate/cycle (%); Chemical pregnancy rate/transfer, (%); Clinical pregnancy rate/transfer (%) Abortion rate (%) |

Figure 2. Forest plot of E2 level of HCG day between two groups.

Data extraction

Demographic, methodology, procedural and outcome data (Stimulation days, Total doses of gonadotropin, No. of MII oocytes, Endometrial thickness, E2 levels of HCG day, No. of retrieved oocytes, Cleavage rate, Number of two pronucleate or 2PN rate, Number of obtained embryos or Number of transferred embryos, Fertilization rate, Implantation rate, Clinical pregnancy rate and Abortion rate) were extracted in article included. Data extraction was performed by two independently reviewers (Xiaoying Yu and Jian Ruan).

Outcomes

The main outcome measures included E2 levels of HCG day (pg/ml), Number of MII oocytes, Number of 2PN and Number of obtained embryos. Secondary outcome measures were implantation and clinical pregnancy rates.

Quality assessment

The same two reviewers (Xiaoying Yu and Lianping He) will also independently evaluate risk of bias among the included studies. The quality of observational studies will be assessed using the framework described by Hayden et al., which evaluates study participation, study attrition, outcome measurement, and statistical analyses among studies included in systematic reviews using a simple 'yes', 'partly', 'no', 'unsure' scale [13]. We will also describe the following characteristics for each study included in our analysis:

Quantitative data synthesis

Meta Analyst for Windows [14] was used for performing meta-analysis. Heterogeneity across studies was measured using the Q-test based on the x^2 statistic, considering significant statistical heterogeneity as $P < 0.1$. By heterogeneity test, the random-effect or fixed-effect models were used to merging sets of data and data analysis. The final data subdivided into several groups for statistical analysis and chart description.

Results

Search results and characteristics of the studies included

847 relevant papers were found after searching electronic database (**Figure 1**). Non-clinical
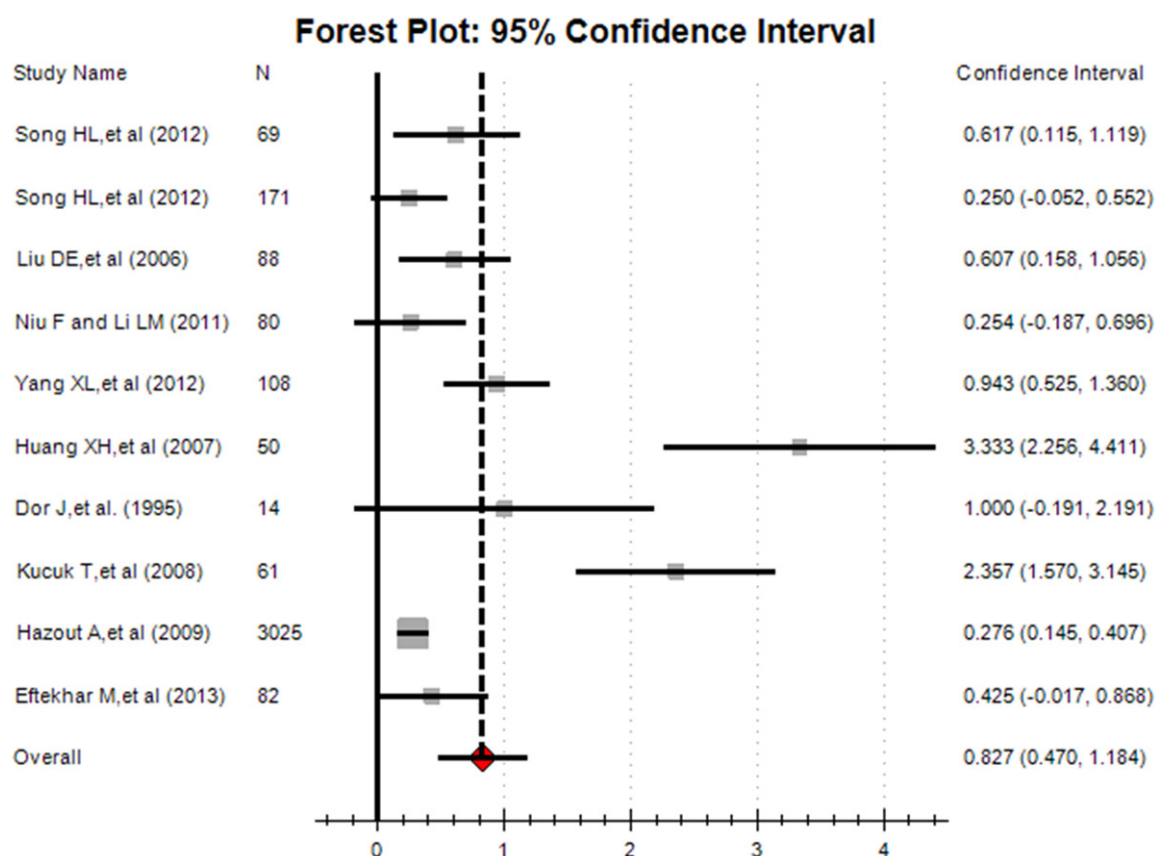
Figure 3. Meta-analysis of No. of MIIoocytes.

trial studies (n = 596), reviews (n = 124) and other Animals studies (n = 8). After screening titles and or abstracts (n = 104). We excluded three meta-analyses and one study in our study, because they cannot offer data regarding the primary outcome measure. Finally, Eleven articles (See **Table 1**) were included in our meta-analysis (six RCTs and five CCTs). Characteristics of the including studies is showed in **Table 1**.

Meta-analysis

Primary outcome measure

E2 levels of HCG day (pg/ml): Total six studies, two RCTs [15, 16] and four CCTs [17-20] were selected for meta-analysis for the E2 levels of HCG day outcome. Heterogeneity between studies was significantly high ($Q = 38.671$, $I^2 = 0.845$, $P < 0.05$). Therefore, random-effect models was used to estimate the deference of E2 levels of HCG day and significantly difference (OR: 0.55; 95% CI: 0.127-0.973; $P = $

0.000) was found between groups with adjuvant growth hormone and without adjuvant growth hormone therapy for E2 levels of HCG day (see **Figure 2**).

No. of MIIoocytes (n): Five RCTs [15, 16, 21-23] and five CCTs [17-20, 24] were included for meta-analysis of MII oocyte number. The controls has less number of oocytes than women treated with adjuvant GH by random-effect models (OR, 0.827; 95% CI, 0.470-1.184; $P = $ 0.000) (**Figure 3**). Heterogeneity between studies was significantly high ($Q = 65.92$, $I^2 = 0.863$, $P < 0.05$).

No. of 2PN (n): Only three studies, two RCTs [15, 16] and one CCT[20] were selected for meta-analysis for the No. of 2PN outcome. There was significant difference in the No. of 2PN between women adjuvant GH compared to those without adjuvant GH (OR, 1.531; 95% CI, 0.701-2.360; $P = 0.000$) (**Figure 4**). Heterogeneity between studies was significantly high ($Q = 20.340$, $I^2 = 0.092$, $P < 0.05$).
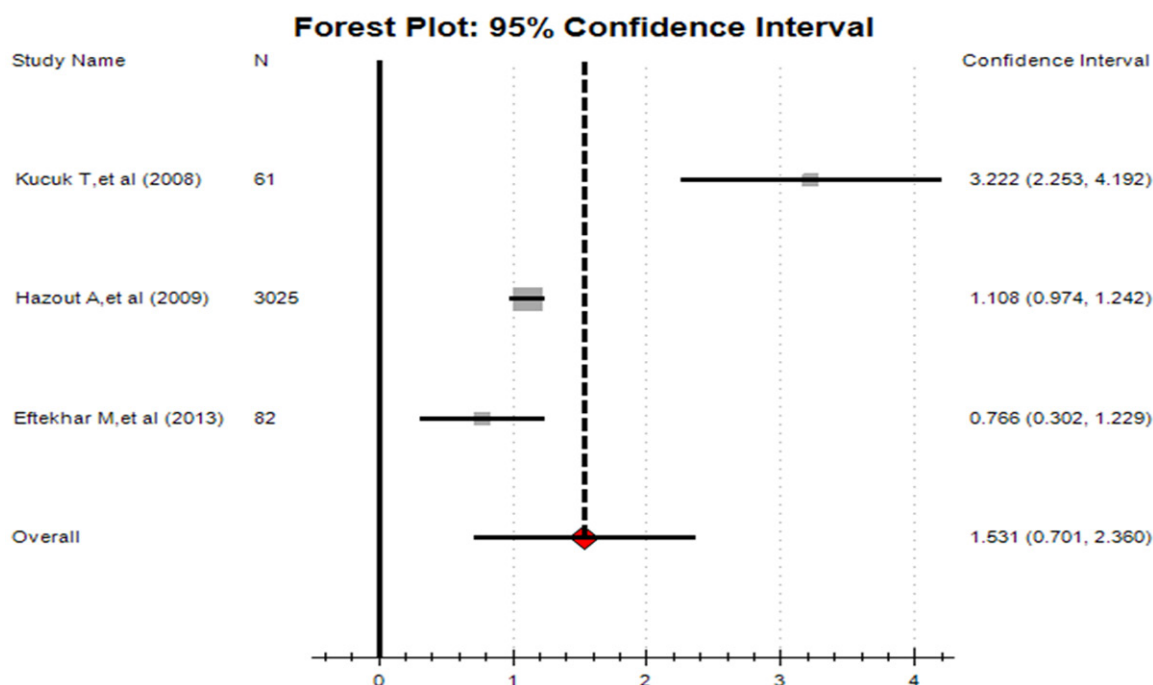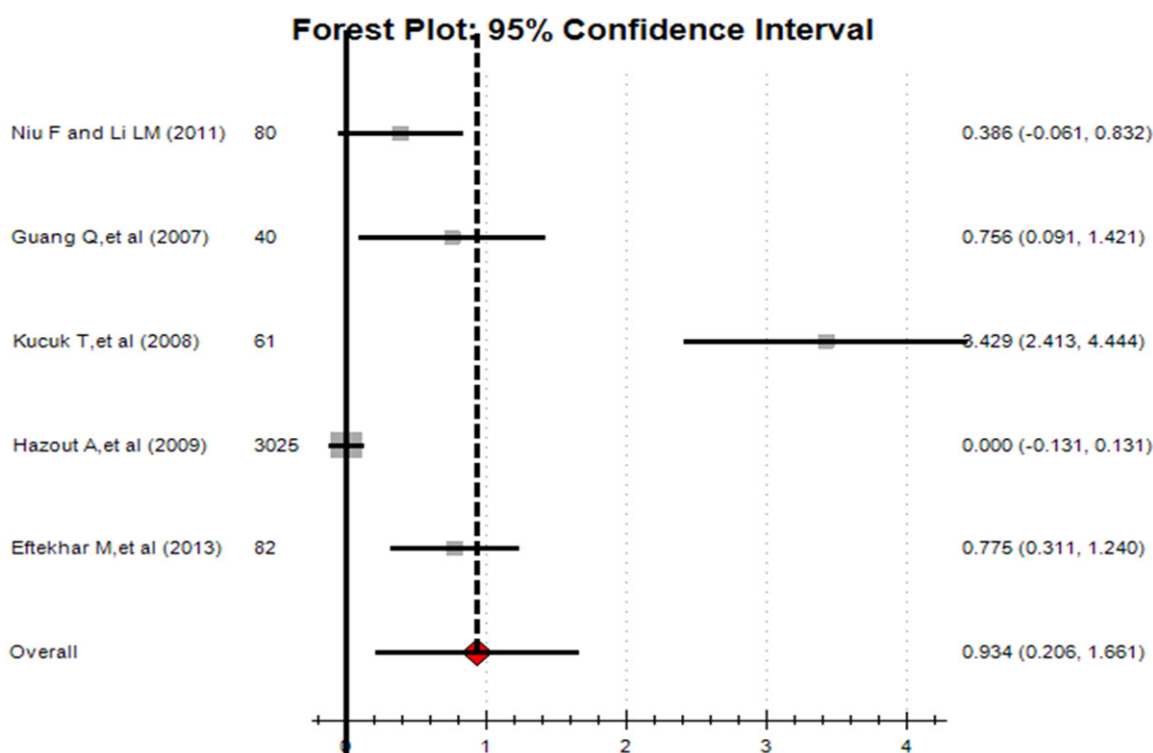
Figure 4. Meta-analysis of No. of 2PN.

Figure 5. Meta-analysis of No. of obtained embryos.

No. of obtained embryos (n): Five studies [15, 16, 20, 23, 25] offered data for this outcome measure. The results from these studies in the number of obtained embryos showed that there was significant difference between the GH and control groups (OR, 0.934; 95% CI, 0.206-
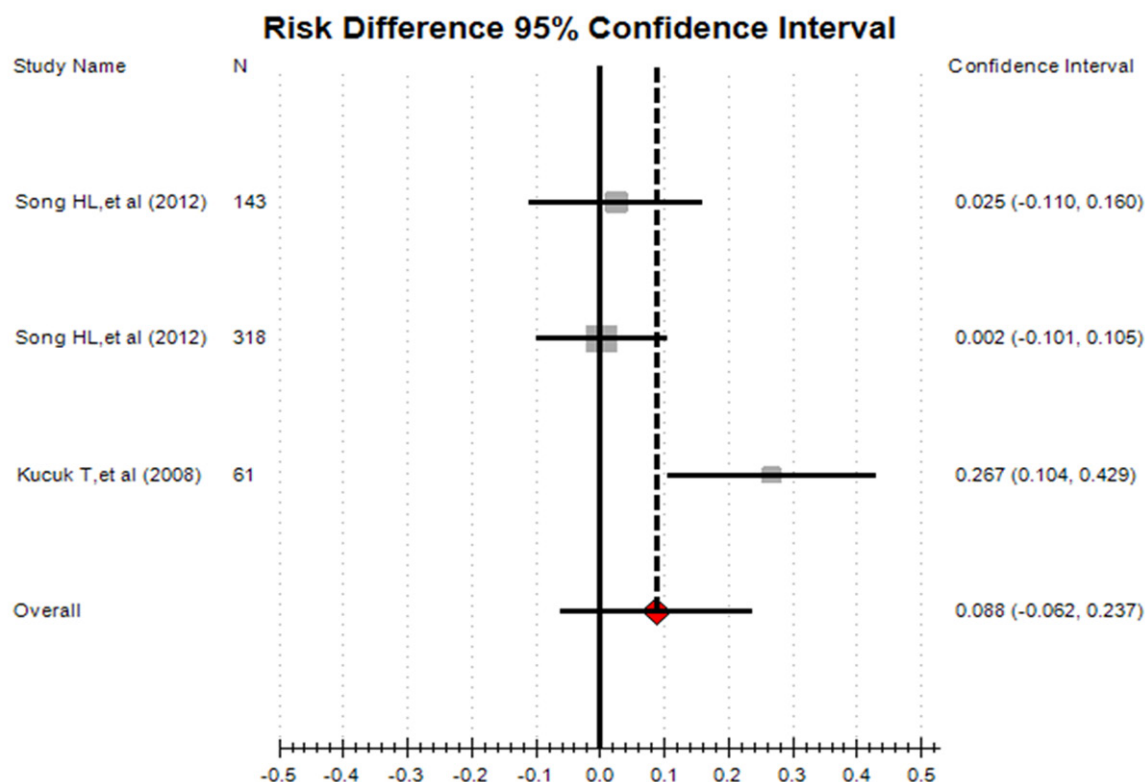
Risk Difference 95% Confidence Interval

Study Name	N		Confidence Interval
Song HL,et al (2012)	143		0.025 (-0.110, 0.160)
Song HL,et al (2012)	318		0.002 (-0.101, 0.105)
Kucuk T,et al (2008)	61		0.267 (0.104, 0.429)
Overall			0.088 (-0.062, 0.237)

-0.5 -0.4 -0.3 -0.2 -0.1 0.0 0.1 0.2 0.3 0.4 0.5

Figure 6. Meta-analysis of implantation rate.

1.661; P = 0.000) (**Figure 5**). Heterogeneity between studies was significantly high (Q = 56.361, I^2 = 0.929, P < 0.05).

Secondary outcome measures

Implantation rate (%): In three studies [15, 18, 19], information was reported implantation rate. The pooled implantation rate for GH addition was 6.8% (95% CI, -0.062-0.237; P = 0.206) in these studies (**Figure 6**). Heterogeneity between studies was significantly high (Q = 7.661, I^2 = 0.739, P < 0.05).

Clinical pregnancy rate (%): Heterogeneity between studies was not significantly (Q = 1.986, I^2 = 0.000, P = 0.738). The OR for clinical pregnancy rate was 0.051 (95% CI: -0.033 to 0.134, P = 0.197), suggesting that the probability of clinical pregnancy rate was not associated with the addition of GH addition [15, 16, 18, 19, 25] (**Figure 7**).

Discussion

The present meta-analysis demonstrated that the number of MII oocytes and obtained embry-

os were significantly higher in co-treatment with GH. Receiving GH could improve the serum E2 levels of HCG day and the number of 2PN but the implantation rates and clinical pregnancy rates, which are inconsistent with previous meta-analyses [10-12].

GH is a pleiotropic, multifunctional hormone with effects ranging far beyond those on linear growth, which binds to GH receptors on granulosa, the theca, and luteal cells, thus promoting steroidogenesis and gametogenesis. GH, IGF-1, and GHRH, all increase the sensitivity of ovaries to gonadotropin stimulation and enhance follicular development [26, 27]. GH also enhances aromatase and 3-β-hydrogenase activity, thus increasing the conversion of androgen into estrogens in females. This effect on the ovarian steroidogenic enzymes occurs through a direct and/or IGF-I mediated mechanism. Growth hormones, IGF-1 and IGF-2, all affect the maturation of the follicle and gamete as well.GH directly inhibits follicle apoptosis in conjunction with gonadotropins and may enhance follicular survival and cell proliferation by strengthening LH action. Both GH and IGF-I
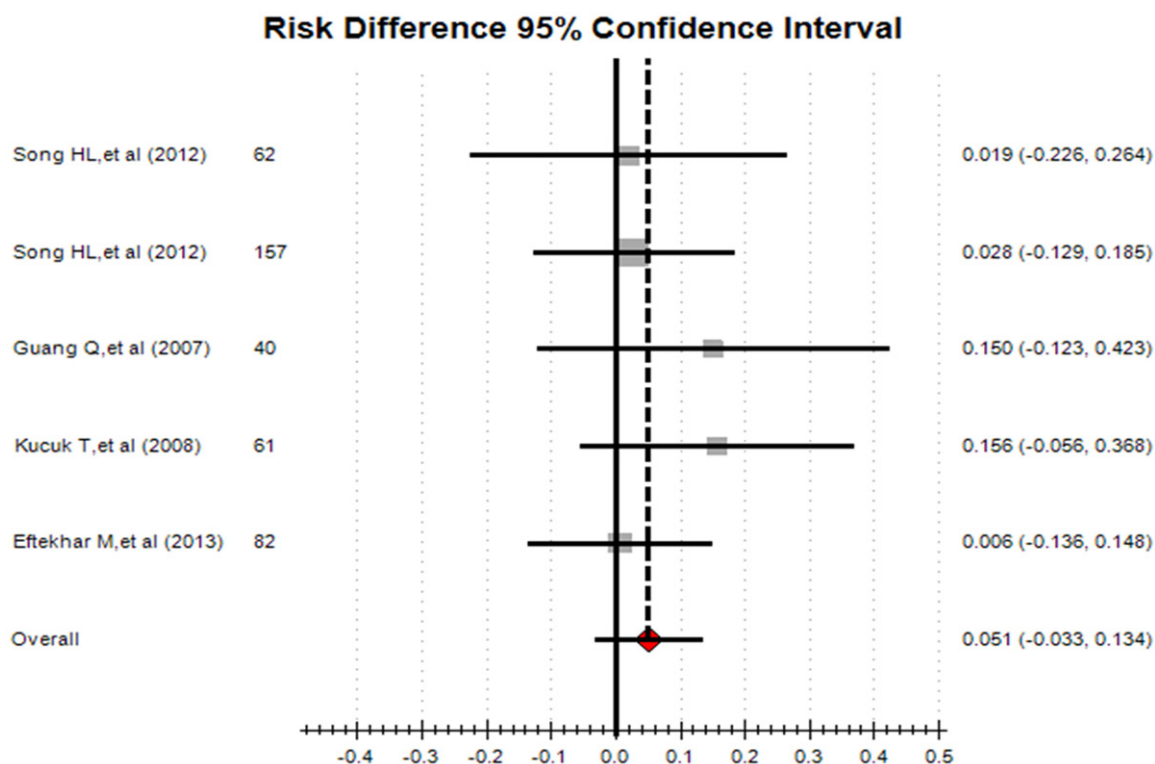
Risk Difference 95% Confidence Interval

Study	N	Risk Difference (95% CI)
Song HL,et al (2012)	62	0.019 (-0.226, 0.264)
Song HL,et al (2012)	157	0.028 (-0.129, 0.185)
Guang Q,et al (2007)	40	0.150 (-0.123, 0.423)
Kucuk T,et al (2008)	61	0.156 (-0.056, 0.368)
Eftekhar M,et al (2013)	82	0.006 (-0.136, 0.148)
Overall		0.051 (-0.033, 0.134)

Figure 7. Meta-analysis of clinical pregnancy rate.

play a role in the recruitment of the dominant follicle from its cohort, leading to monofollicular growth in women [28].

GH can regulates the effect of follicle-stimulating hormone (FSH) on granulose cells via increasing the synthesis of insulin-like growth factor-I, augments the effect of gonadotropin on granulose and theca cells, and plays an essential role in function of ovarian, including follicular development, estrogen synthesis and oocyte maturation [15, 29].

A prospective randomized trial performed by Kucuk T et al [15] found a shorter mean duration of stimulation in the GH group of 2 days. The total dose of FSH was 21% lower in the GH group but the number of 2PN embryos was significantly higher than in the control group. Their conclusions showed that poor responder women undergoing repeated assisted reproduction treatment and co-stimulated with GH achieve more oocytes, higher fertilization rate if growth hormone started in the luteal phase of previous cycle, as compared with women of the same status treated with GnRHa long protocol, which is inconsisit with our study.

Slot KA et al [30]. Reported that the amount of primordial follicles was significantly elevated in GH receptor knock-out (GHR/GHBP-KO) mice, while the numbers of primary, preantral and antral follicles were lower compared with wild-type values. IGF-I treatment of GHR/GHBP-KO mice for 14 days resulted in a reduced number of primordial follicles, an increased number of healthy antral follicles, and a decreased percentage of atretic follicles. Their results suggest that GH may play a role, either directly or indirectly, via for instance IGF-I, in the recruitment of primordial follicles into the growing pool. Furthermore, GH seems to protect antral follicles, directly or indirectly from undergoing atresia. The IGF stimulate ovarian function by acting synergistically with gonadotropins to promote growth and steroidogenesis of ovarian cells [31]. Hassan HA et al [32]. Study showed that in vivo administration of GH enhanced in vitro maturation and fertilization of human GV oocytes retrieved from small antral follicles.

Results of Kiapekou E et al [33] suggest that GH and IGF-1, alone or in combination, affect mouse oocyte maturation significantly. The lack of a synergistic effect on oocyte cultures

when supply both hormones indicates that both hormones act through the same signal pathway.

Bachelot A et al [29] used a mouse model, in which the GH receptor (GHR) and GH-binding protein (GHR/GHBP) gene was disrupted by homologous recombination. The major effect on reproductive function seen in GHR/GHBP knockout (KO) compared with wild type animals is a dramatic decrease in litter size; this defect is due to a reduction of the ovulation rate. Their study indicated that the reduction of litter size in GHR/GHBP KO mice is the consequence of an alteration of the growth of follicles and suggest that the effects of GH effects on follicular growth are independent of IGF-I. Some studies demonstrated that growth hormone is associated with good quality embryo, and high concentration of growth hormone in follicular fluid is related to good embryo morphology and increases embryo implantation [34, 35].

Our study demonstrated that co-treatment with GH in POR undergoing ovarian stimulation with GnRH analogues and gonadotrophins for IVF, increased the number of MII oocyte (OR, 0.827; 95% CI, 0.470 to 1.184; P = 0.000) (**Figure 3**), and obtained embryos (OR, 0.934; 95% CI, 0.206 to 1.661; P = 0.000) (**Figure 5**).

Gregoraszczuk et al [36]. Demonstrate that the influence of exogenous GH on steroid secretion by granulosa and theca interna cells recovered from small (1-3 mm), medium (4-6 mm) and large (8-12 mm) follicles. GH added to the culture media had no effect on estradiol and progesterone secretion by granulosa cells isolated from small and medium follicles while it stimulated both estradiol and progesterone secretion by Gc isolated from large preovulatory follicles. GH did not stimulate progesterone secretion by Tc isolated from small follicles but stimulated progesterone secretion by Tc isolated from medium and large preovulatory follicles. Both co-culture systems exhibited synergistic effect on estradiol secretion. The stimulatory effect on progesterone secretion under the influence of GH was observed in Gc cultured alone and Tc cultured alone. In contrast, the secretion of progesterone decreased in both co-culture systems and the addition of GH further augmented this attenuation. Our result also shows that addition of GH increase the serum E2 on the day of HCG in poor

responders undergoing COH with GnRH agonist and gonadotropins for ART.

Meta-analysis conducted by Kolibianakis et al [10], showed that the addition of GH in POR undergoing COH with GnRH agonist and gonadotropins for ART, increases the clinical Pregnancy and live birth rates. However, our meta-analysis showed that the use of GH as an adjuvant treatment in POR patients in IVF-ET cycles does not increase pregnancy rate and implantation rate.There are also limitations of the meta-analysis itself. Firstly, the current analysis based on data derived from only six RCTs and five CCTs studies. However, this overcomes the common criticism of meta-analysis about the clinical heterogeneity arising from different centres and countries. Additionally, data of individual studies all pointed in the same direction and the observations regarding the conventional protocol are in accordance with the existing literature, which confirms the authenticity of the observed findings.

In conclusion, the current meta-analysis suggests that the addition of GH does increase the serum E2 level of HCG day and the number of MII oocyte, 2PN and obtained embryos. In POR undergoing ovarian stimulation with GnRH analogues and gonadotrophins for IVF/ICSI, However, the available evidence does not indicate a beneficial effect of GH addition on implantation rate and clinical pregnancy rate.

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Disclosure of conflict of interest

None.

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