Effective treatment protocol for poor ovarian response: A systematic review and meta-analysis

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ABSTRACT

Poor ovarian response represents an increasingly common problem. This systematic review was aimed to identify the most effective treatment protocol for poor response. We searched MEDLINE, EMBASE, and The Cochrane Library from 1980 to October 2015. Study quality assessment and meta-analyses were performed according to the Cochrane recommendations. We found 61 trials including 4997 cycles employing 10 management strategies. Most common strategy was the use of gonadotropin-releasing hormone antagonist (GnRHant), and was compared with GnRH agonist protocol (17 trials; $n = 1696$) for pituitary down-regulation which showed no significant difference in the outcome. Luteinizing hormone supplementation (eight trials, $n = 847$) showed no difference in the outcome. Growth hormone supplementation (seven trials; $n = 251$) showed significant improvement in clinical pregnancy rate (CPR) and live birth rate (LBR) with an odds ratio (OR) of 2.13 (95% CI 1.06–4.28) and 2.96 (95% CI 1.17–7.52). Testosterone supplementation (three trials; $n = 225$) significantly improved CPR (OR 2.4; 95% CI 1.16–5.04) and LBR (OR 2.18; 95% CI 1.01–4.68). Aromatase inhibitors (four trials; $n = 223$) and dehydroepiandrosterone supplementation (two trials; $n = 57$) had no effect on outcome.

KEY WORDS: Assisted conception, *in vitro* fertilization, ovarian stimulation, poor ovarian response

INTRODUCTION

Poor ovarian response (POR) is a challenging situation in assisted reproduction. There is a lack of consensus on the definition of POR and a huge variation in treating women with previous POR.[1] However, the most common criterion to diagnose POR is retrieval of low number of oocytes despite adequate ovarian stimulation in an assisted conception cycle. The ESHRE working group on POR definition (the Bologna criteria) reached a consensus on the minimal criteria needed to define POR by the presence of two of the following three features: (i) Advanced maternal age (≥40 years) or any other risk factor for POR; (ii) a previous characterized POR cycle (≤3 oocytes with a conventional stimulation protocol); (iii) an abnormal ovarian reserve test (antral follicle count <5–7 follicles or anti-Mullerian hormone (AMH) <0.5–1.1 ng/ml).[2] It was also proposed by the working group that two episodes of poor ovarian response after maximum stimulation deemed sufficient to define a patient as POR in the absence of other criteria. The suggested incidence of POR ranges from 9% to 25%.[3] Various controlled ovarian hyperstimulation protocols and strategies have been used in this group of women to improve reproductive outcome, but the success rate still remains low.

To date, there are various observational studies, randomized controlled trials (RCTs), and systematic reviews reported on this subject.[4-9] However, either the studies are too specific by trying to address only one treatment strategy,[4,7-10] or they include observational studies and nonrandomized studies in their meta-analysis.[9] The aim of

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our systematic review is to appraise all the existing protocols applied to poor responders by including evidence generated from RCTs.

METHODS

The review was formulated using population, intervention, comparison, outcome, and design structure. Poor responders to ovarian stimulation formed the study population. All types of intervention subjected to RCTs were included in the review. The interventions were analyzed and compared with the control group used in the study. Two or more trials with identical design and interventions were analyzed by meta-analysis. Our outcome measures were number of oocytes retrieved per cycle, live birth rates (LBR), and clinical pregnancy rates (CPR).

We searched the literature on MEDLINE (1980-October 2015), EMBASE (1980-October 2015), and The Cochrane Library (2015) for relevant citations using the keywords, "poor responders, controlled ovarian hyperstimulation, reduced ovarian response, diminished ovarian response, low AMH, assisted conception, and *in vitro* fertilization (IVF)." The reference lists of all known primary and review articles were examined to identify cited articles not captured by the electronic searches. Language restrictions were not applied. A systematic search for all RCTs was carried out. Reference lists from retrieved articles and related articles were checked for relevant studies. All studies addressing the research question and satisfying our inclusion criteria were included in the review. The review protocol was registered with the PROSPERO Registry (CRD42013004190).

Data collection and analysis

The electronic searches were scrutinized, and full manuscripts of all citations that were likely to meet the predefined selection criteria were obtained. Two review authors (Yadava Bapurao Jeve and Harish Malappa Bhandari) independently assessed trial quality and extracted data. Studies which met the predefined and explicit criteria regarding population, interventions, comparison, outcomes, and study design were selected for inclusion in this review. When discrepancies occurred, they were resolved by consensus (Yadava Bapurao Jeve and Harish Malappa Bhandari). We performed meta-analysis when two or more trials were comparable in design and protocol. Data were analyzed using Review Manager (RevMan) [Computer program]. Version 5.1. Copenhagen: The Nordic Cochrane Centre, The Cochrane Collaboration, 2014. For each study, the treatment effect was measured with an odds ratio (OR) for dichotomous outcomes and mean differences for continuous outcomes and random effect models that were presented with their corresponding 95% confidence intervals (CI).

Inclusion criteria

Only RCTs that used suitable definition for POR and used different therapeutic approaches for ovarian stimulation of poor responders in assisted conception were included in the study. The trials reported after publication of the Bologna criteria for poor responders were analyzed as per this criteria.[2]

Exclusion criteria

All observational studies or quasi-randomized studies and studies in which poor responders were not defined were excluded from the study.

Intervention groups

The interventions were grouped as below:

1. Gonadotropin-releasing hormone antagonist (GnRHant) protocols
2. Protocols using luteinizing hormone (LH) as an adjuvant
3. Protocols using growth hormone (GH) as an adjuvant
4. Protocols using transdermal testosterone as an adjuvant
5. Protocols using aromatase inhibitors as an adjuvant
6. Protocols using dehydroepiandrosterone (DHEA) as an adjuvant
7. Protocols using recombinant human chorionic gonadotropin as an adjuvant
8. Natural cycle
9. Protocols using various other adjuvants
10. Various modifications to GnRH agonist (GnRHa) protocol.

Types of outcome measures

To bring uniformity in assessment, we analyzed the most relevant primary outcomes of LBR and CPR per cycle. The secondary outcome measure was the number of oocytes retrieved per cycle.

Quality and risk of bias of included studies

We included only RCTs in this systematic review – some were blinded and/or placebo-controlled, but others were not. Quality analysis was performed using internationally accepted Cochrane tools. GRADEpro. [Computer program on www.gradepro.org]. Version [2014]. McMaster University, 2014, was used to produce a summary of findings, tables for meta-analysis; this shows significant effects with interventions. A risk of bias table was produced using Review Manager (RevMan) [Computer program]. Version 5.1. Copenhagen: The Nordic Cochrane Centre, The Cochrane Collaboration, 2014, and is summarized in Figure 1. Using these tools, we have classified overall quality of evidence as moderate to high grade.

RESULTS

A total of 61 RCTs (4997 assisted conception cycles) were included in this study. The treatment approaches were

categorized into 10 groups (as mentioned above), the most common being the use of GnRHant versus GnRHa for pituitary downregulation in 17 RCTs. The characteristics of the included studies are described in Table 1.

1. GnRHa versus GnRHant for pituitary downregulation: Seventeen RCTs (n = 1696) that met the criteria were subjected to meta-analysis [Figure 2]. The results suggested no significant difference in the number of oocytes retrieved (mean difference 0.09; 95% CI 0.53–0.36) and no difference in CPR with an OR of 1.24 (95% CI 0.88–1.73)
2. LH supplementation: Eight RCTs (n = 847) assessed the role of supplementation to ovarian hyperstimulation but found no difference in CPR (OR 1.32; 95% CI 0.93–1.87)
3. GH supplementation: None of the seven RCTs (n = 251) individually had shown benefit of GH supplementation in improving CPR, but the pooled data from these studies showed a significant improvement in CPR (OR 2.13; 95% CI 1.06–4.28). Of these, only four studies (n = 27) reported LBR and the pooled data showed

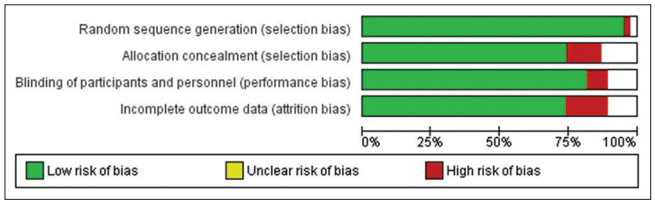

Figure 1: Methodological quality graph

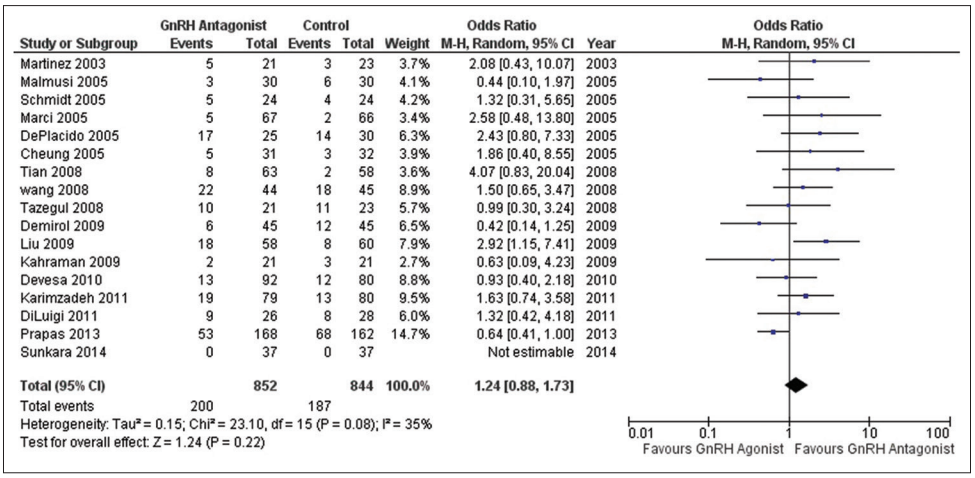

Figure 2: Gonadotropin-releasing hormone agonist (control) versus GnRH antagonist down-regulation protocols

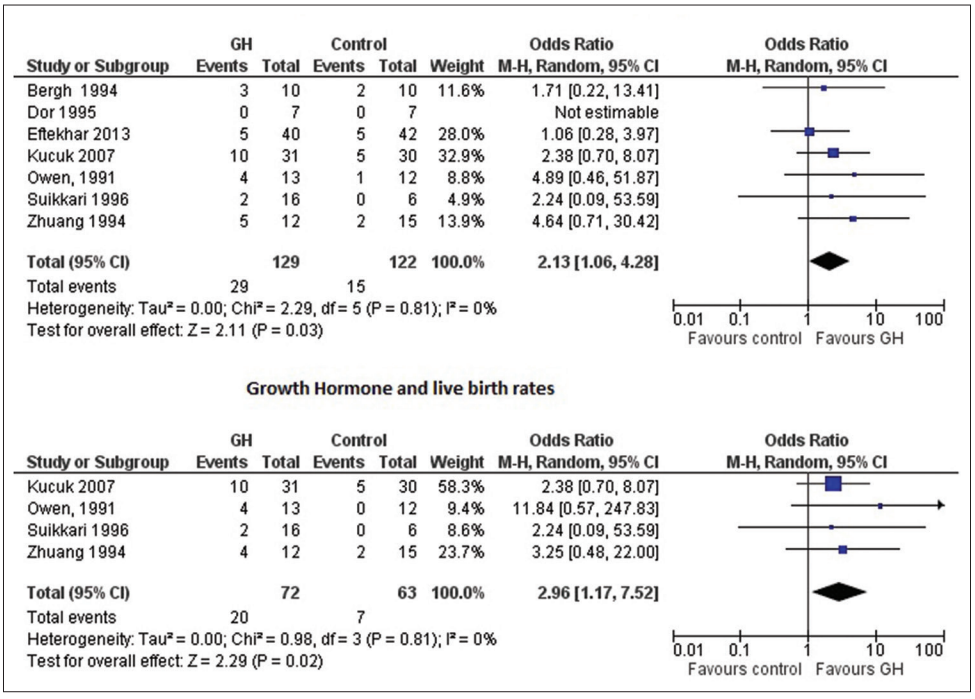

Figure 3: Use of growth hormone supplement

Table 1: Different therapeutic approaches for poor responders

Study	Population	Intervention	Comparison	Outcome	Remark
			GnRHant protocols		
Martinez (2003)	*n*=44 (21 and 23)	Multiple dose (cetrorelix)	Short, multiple dose (triptorelin)	Number of oocytes 3.7 versus 5.6 CPR 5 versus 3	GnRHant and short, multiple GnRHa
Cheung (2005)	*n*=66 (31 and 32)	GnRHant protocol- cetrorelix 0.25 mg daily as a fixed protocol starting on day 6 of the stimulation until the day of hCG	GnRHa - Buserelin acetate nasal spray dose of 600 µg from mid-luteal phase, and co-administered with the final week of OCP pretreatment	Number of oocytes 5.89 versus 5.62 CPR 5 versus 3	Unblinded study: Fixed, multi-dose GnRHant versus - a long GnRHa protocol
Marci (2005)	*n*=60 (30 each)	GnRHa long protocol	GnRHant	Number of oocytes 4.3 versus 5.6 CPR 5 versus 2	Unblinded study: GnRHant versus GnRHa long protocol
Schmidt (2005)	*n*=48 (24 each)	Ganirelix acetate group (received 300 IU of recombinant FSH s.c.+150 IU HMG for 5 days)	21 days of OCP and the microdose leuprolide flare group	Number of oocytes 8.9 versus 9.0 CPR 5 versus 4	Unblinded study: Microdose GnRHa versus GnRHant
Malmusi (2005)	*n*=55 (30 and 25)	rFSH was started on the D1 ganirelix 0.25 mg daily until the hCG injection	Triptorelin 0.1 mg was initiated on D1 followed by rFSH D2 of menstruation	Number of oocytes 3.5 versus 2.5 CPR 3 versus 6	Unblinded study: GnRHa flare-up protocol versus GnRHant
De Placido (2005)	*n*=133 (67 and 66)	Multiple dose (cetrorelix) and flare up	Multiple dose (triptorelin)	Number of oocytes 6.79 versus 6.54 CPR 17 versus 14	Multiple dose GnRHant and flare up, multiple dose
Tazegül (2008)	*n*=89 (44 and 45)	Multiple dose (cetrorelix/ ganirelix)	Long, multiple dose (leuprorelin)	Number of oocytes 5.44 versus 5.44 CPR 10 versus 11	Multiple dose GnRHant versus GnRHa long protocol
Tian (2008)	*n*=44 (21 and 23)	Multiple dose (cetrorelix)	Short, multiple dose (triptorelin)	Number of oocytes 3.25 versus 3.79 CPR 8 versus 2	Multiple dose antagonist versus GnRHa short protocol
Wang (2008)	*n*=121 (63 and 58)	OCP/multiple dose (cetrorelix)	OCP/long, microdose GnRHa (triptorelin)	Number of oocytes 4.40 versus 5.41 CPR 22 versus 18	OCP Multiple dose GnRHant versus microdose GnRHa
Demirol (2009)	*n*=90 (45 each)	Combined OCP for 21 days. On day 3 of menstruation, 40 µg s.c./bid of leuprolide followed by 450 IU/day HMG on day 3	GnRHant multiple dose - 450 IU/day HMG starting on day 3 and 0.25 mg cetrorelix daily	Number of oocytes 4.3 versus 3.1 CPR 6 versus 12	Unblinded study: GnRHa microdose flare-up versus GnRHant multiple dose
Kahraman (2009)	*n*=42 (21 and 21)	Multiple dose (cetrorelix)	Flare-up, microdose (leuprorelin)	Number of oocytes 5.60 versus 5.80 CPR 2 versus 3	Multiple dose GnRHant and GnRHa flare up
Liu (2009)	*n*=118 (58 and 60)	Multiple dose (cetrorelix)	Short, multiple dose (triptorelin)	Number of oocytes 4.12 versus 3 CPR 18 versus 8	Multiple GnRHant and GnRHa short protocol
Devesa (2010)	*n*=172 (92 and 80)	OCP/multiple dose (ganirelix)	OCP/flare-up, (leuprolide)	Number of oocytes 4.89 versus 5.91 CPR 13 versus 12	OCP Multiple GnRHant and GnRHa flare-up
DiLuigi (2011)	*n*=54 (28 and 26)	GnRHant protocol	Microdose leuprolide acetate	Number of oocytes 5.2 versus 5.4 CPR 9 versus 8 LBR 6 versus 7	GnRHant versus GnRHa microdose protocol
Karimzadeh (2011)	*n*=159 (79 and 80)	Clomiphene citrate, gonadotropin and GnRHant	Microdose GnRHa flare	Number of oocytes 6.34 versus 4.1 CPR 19 versus 13	Mild protocol and microdose GnRHa flare protocols

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Table 1: Contd...

Study	Population	Intervention	Comparison	Outcome	Remark
Prapas (2013)	*n*=330 (168 and 162)	Fixed GnRHant protocol	Long GnRHa protocol	Number of oocytes 3.01 versus 2.64 CPR 53 versus 68	GnRHant (fixed) versus GnRHa long protocol
Sunkara (2014)	*n*=74 (37 and 37)	GnRHant	GnRHa long protocol	Number of oocytes 3.3 versus 4.42	GnRHant versus GnRHa long protocol
		Protocols using LH as an adjuvant			
Ferraretti (2004)	*n*=104 (54 and 50)	Recombinant LH in addition to the increased dose of FSH	An increased dosage of FSH	Number of oocytes CPR 22 versus 11	
Demirol (2005)	*n*=106 (53 and 53)	Recombinant LH in addition to the increased dose of FSH	Increased dosage of FSH	Number of oocytes 5.89 versus 5.62 CPR 5 versus 3	
Fernández Ramírez (2006)	*n*=34 (16 and 18)	rLH 75 IU b.i.d. (150 IU/day) starting on the day of GnRHant initiation until hCG trigger	GnRHant without rLH	CPR 2 versus 3	
Polidoropoulos (2007)	*n*=136 (68 and 68)	rLH 75-150 IU/day until hCG criteria were met	GnRHa protocol	CPR 17 versus 15	
Berkkanoglu (2007)	*n*=97 (46 and 51)	600 IU of rFSH plus daily supplementation with 75 IU of rLH	600 IU of rFSH as the control group	Number of oocytes 4.8 versus 5.6 CPR 13 versus 14	Three-arm study with addition of r LH and hCG supplementation
Ruvolo (2007)	*n*=42 (24 and 18)	rLH 75-150 IU/day from 8th day of ovarian stimulation until hCG criteria were met	GnRHa and rFSH without further LH addition	Clinical pregnancy 10 versus 4	
Barrenetxea (2008)	*n*=84 (42 each)	GnRHa and rFSH and rLH	GnRHa and rFSH without further LH addition	Number of oocytes 5.43 versus 5.66 CPR 10 versus 9	Unblinded study
Musters (2012)	*n*=116 and 128	rFSH and rLH (2:1 ratio)	rFSH alone	CPR 16 versus 15	
		Protocols using GH as an adjuvant			
Owen, (1991)	*n*=25 (13 and 12)	GH 24 IU i.m./day on alternate days, starting simultaneously with HMG until the day of hCG administration	Use of placebo	CPR 4 versus 1 LBR 4 versus 0	
Zhuang (1994)	*n*=27 (12 and 15)	GH 12 IU i.m./day on alternate days	No placebo	CPR 5 versus 2 LBR 4 versus 2	
Hughes (1994)	*n*=33 (21 and 12)	In addition to commencing HMG, a second daily injection of GH 12 IU i.m. daily for up to 12 days or until hCG	Placebo	Number of oocytes 5 versus 5	Placebo-controlled double-blind
Bergh (1994)	*n*=40 (20 each)	GH 0.1 IU/kg body weight per day was given s.c. as pretreatment and during stimulation with HMG	Placebo	CPR 3 versus 2	Double-blind, placebo-controlled
Dor (1995)	*n*=14 (7 and 7)	GNRHa/HMG/GH (18 IU on alternate days, total dose 72 IU)	GnRHa/HMG/placebo	CPR 0 both groups	Placebo-controlled double-blind
Suikkari (1996)	*n*=22 (16 and 6)	GnRHa, gonadotropin in a ”boost” flare-up protocol for 4, or 12 IU of human GH	Placebo	CPR 2.4 versus 2	Double-blind placebo-controlled
Howles (1999)	*n*=196 (96 and 100)	Long luteal buserelin 0.5 mg, 1 mg/day GH releasing factor s.c. with HMG 300 IU/day i.m. from the day of down-regulation	No supplement	CPR 8 versus 8 LBR 5 versus 4	Multicenter double-blind, placebo-controlled study

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Table 1: Contd...

Study	Population	Intervention	Comparison	Outcome	Remark
Kucuk (2007)	*n*=61 (31 and 30)	GH co-treatment, daily s.c. injection of 4 mg from day 21 of preceding cycle along with GnRHa	No supplement	Number of oocytes 6.5 versus 3.2	GH co-stimulation to long luteal GnRHa regimen
Eftekhar (2013)	*n*=82 (40 and 42)	GH and GnRHant protocol	GnRHant protocol (no placebo)	CPR 5 versus 5 Number of oocytes 6.1 versus 4.8	
		Protocols using testosterone as an adjuvant			
Massin (2006)	*n*=53 (27 and 26)	Transdermal application of testosterone 15 days before FSH	Placebo	CPR 4 versus 1, LBR 2 versus 1 Number of oocytes 3.17 versus 3.56	Double-blind, placebo-controlled study
Fábregues (2009)	*n*=62 (31 and 31)	Testosterone 20 µg/kg/day for 5 days transdermal application preceding ovarian stimulation	High-dose gonadotropin in association with a minidose GnRHa protocol	CPR 6 versus 4, LBR 6 versus 4 Number of oocytes 4.1 versus 3.6	
Kim (2011)	*n*=110 (55 and 55)	12.5 mg transdermal testosterone daily for 21 days in the cycle preceding ovarian stimulation	No placebo	CPR 17 versus 8, LBR 15 versus 7 Number of oocytes 4.6 versus 3.2	
		Protocols using aromatase inhibitors as an adjuvant			
Goswami (2004)	*n*=38 (13 and 25)	Let 2.5 mg daily from day 3 to 7, and rFSH 75 IU/day on days 3 and 8	Long GnRHa protocol and rFSH (300-450 IU/day)	CPR 3 versus 6 Number of oocytes 1.6 versus 2.1	Unblinded study, Let-FSH group versus GnRHa-FSH group
Kashyap (2005)	*n*=55 (29 and 26)	Let and GnRHant	No placebo, GnRHant protocol	CPR 3 versus 1	
Ozmen (2009)	*n*=70 (35 and 35)	Let 5 mg/day for 5 days and GnRHant 0.25 µg/day	No placebo, GnRH GnRHant 0.25 µg/day	CPR 8 versus 5	
Mohsen (2013)	*n*=60 (30 and 30)	Let supplementation	GnRHa protocol (micro dose flare)	CPR 4 versus 5	
		Protocols using DHEA as an adjuvant			
Artini (2012)	*n*=48 (24 each)	25 mg three times a day of DHEA supplementation for 3 months previous to IVF cycle	No adjuvant	CPR 6 versus 4 Number of oocytes 3.58 versus 2.67	Prospective, randomized, controlled
Wiser (2010)	*n*=51 (26 and 25)	75 mg DHEA orally, once a day, at least 6 weeks before GnRHa long protocol	No adjuvant	CPR 7 versus 3 Number of oocytes 4.6 versus 3.2 3.2 and 3.5	Prospective, randomized, controlled
		Protocols using rhCG as an adjuvant			
Berkkanoglu (2007)	*n*=99 (48 and 51)	600 IU of rFSH plus daily supplementation with 75 IU of rhCG	600 IU of rFSH	CPR 10 versus 14 Number of oocytes 3.8 versus 5.6	Three-arm study recombinant LH, hCG supplementation
		Natural cycle			
Morgia (2004)	*n*=215 (114 and 101)	Natural cycle IVF	Microdose GnRHa flare from day 1 and 600 IU FSH	CPR 17 versus 10 Number of oocytes 0.79 versus 2.1	Unblinded study
		Protocols using other adjutants			
Jinno (1997)	*n*=162 (82 and 80)	Oral 1.25 mg/day bromocriptine from day 4 to 6 in preceding cycle and then 4.5 mg/day, discontinued 7 days before the beginning of HMG administration	GnRHa long protocol	CPR 25 versus 13 Number of oocytes 9.5 versus 8.4	Unblinded, controlled trial: Bromocriptine rebound method

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Table 1: Contd...

Study	Population	Intervention	Comparison	Outcome	Remark
Chung Hoon (1999)	*n*=70 (35 each)	Triptorelin 0.1 mg from day 21; HMG/FSH 300 IU/day i.m. from day of downregulation plus 120 mg/day pyridostigmine until hCG	No adjuvant	CPR 9 versus 4 Number of oocytes 5.9 versus 3.7	Double-blind, placebo-controlled study
Battaglia (1999)	*n*=34 (17 each)	GNRHa + daily supplemented (16 g) with oral L-arginine	No adjuvant	CPR 0 versus 2 number of oocytes 1.6 versus 4.1	Oral L-arginine + GnRHa
Lok (2004)	*n*=60 (30 in each)	Low-dose aspirin (80 mg daily) at the time of commencement of GnRHa in the preceding cycle and continuing until hCG	Placebo	CPR 3 versus 2 Number of oocytes 4 versus 3	Double-blind, placebo-controlled study
		Various modifications to GnRHa protocols			
Dirnfeld (1991)	*n*=54 (28 and 26)	600 µg/day of buserelin nasal spray on day 1 till hCG	1000 µg/day of buserelin nasal spray 15-30 days before ovulation induction, then 600 µg/day till hCG	CPR 3 versus 2 Number of oocytes 7.0 versus 5.6	Unblinded
Von Hoof (1993)	*n*=47 (25 and 22)	225 IU/day i.m. HMG from day 3 for 5 days, increasing to 450 IU/day	Unchanged dose		Unblinded study
Rombauts (1998)	*n*=40 (18 and 20)	GnRHa from day 2. FSH (150 IU) from day 25 of previous cycle	GnRHa from day 2. FSH 150 IU from day 3 of treatment cycle	Number of oocytes 4.5 versus 6	Controlled study
Dirnfeld (1999)	*n*=78 (40 and 38)	GnRHa - started in the midluteal phase and stopped before administration of gonadotropins	GnRHa from the midluteal phase and was continued throughout the follicular phase	CPR 5 versus 9 number of oocytes 6.46 versus 7.73	Unblinded study
Raga (1999)	*n*=30 (15 and 15)	300 IU/day rFSH + 150 IU/day HMG	Purified FSH	CPR 4 versus 1 number of oocytes 7.2 versus 5.6	Prospective randomized study
Garcia-Velasco (2000)	*n*=70 (34 and 36)	Leuprolide acetate 1 mg/d SC till menstruation and then stopped. 3 Amp HMG + 5 Amp FSH on days 1 and 2, 2 Amp HMG + 3 Amp FSH on days 3,4 and 5 from D6	No stop protocol, high dose protocol Long GnRH suppression and gonadotropins	CPR 5 versus 6 number of oocytes 4.1 versus 6.9	Unblinded study
Akman (2000)	*n*=40 (20 each)	Ovarian stimulation with no GnRHa or GnRHant administration	0.25 mg of Cetrorelix daily till hCG	CPR 1 versus 4 number of oocytes 3.46 versus 3.25	Unblinded study
Akman (2001)	*n*=48 (24 in each)	Low-dose oral contraceptive on cycle day 1 for 21 days. On day 2 of menstruation leuprolide acetate (40 µg s.c./day) followed by gonadotropins	Gonadotropins from day 2 and later 0.25 mg of cetrorelix daily	CPR 6 versus 5 Number of oocytes 5.5 versus 4.5	Unblinded study
Marci (2003)	*n*=38 (19 each)	short flare up protocol using GnRHa 0.1 mg/day triptorelin from day 2	Daily dose of 0.25 mg Cetrotide and 375 IU/day rFSH	CPR 3 and 2	Unblinded study
Mohamed (2006)	*n*=30 (15 and 13)	Buserelin 500 µg/day s.c. from day 1 of cycle and ovarian stimulation from day 3	Ovarian stimulation from day 3 and GnRHant 0.25 mg/day from day 8	Number of oocytes 6 each	Unblinded study
Kucuk (2007)	*n*=42 (21 each)	Long protocol triptorelin 0.1 mg, 150 IU rFSH, triptorelin was halved to 0.05 mg and the rFSH was increased to 450 IU	Flare-up regimen of triptorelin. Along with triptorelin, 450 IU rFSH	Number of oocytes 6.8 versus 3.8	Long protocol flare up regimen

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Table 1: Contd...

Study	Population	Intervention	Comparison	Outcome	Remark
Lainas (2008)	*n*=270 (180 and 90)	Ganirelix 0.25 mg and including the day of hCG administration	0.05 mg/day triptorelin from day 2 of the cycle including the day of hCG administration	CPR 29 versus 7 number of oocytes 3 and 3	

CPR=Clinical pregnancy rate, LBR=Live-birth rate, GnRH=Gonadotropin-releasing hormone, GnRHa=Gonadotropin-releasing hormone agonist, GnRHant=GnRH antagonist, FSH=Follicle-stimulating hormone, rFSH=Recombinant FSH, HCG=Human chorionic gonadotropin, rhCG=Recombinant hCG, OCP=Oral contraceptive pill, HMG=Human menopausal gonadotropin, LH=Luteinizing hormone, rLH=Recombinant LH, GH=Growth hormone, Let=Letrozole, DHEA=Dehydroepiandrosterone, s.c=subcutaneous, IVF=*In vitro* fertilization

Table 2: Summary of findings for use of growth hormone supplementation

Growth hormone supplementation to gonadotropins compared to placebo or no supplementation for poor responders

Patient or population: Patients with poor responders
Settings: Randomized controlled trials
Intervention: Growth hormone supplementation to gonadotropins
Comparison: Placebo or no supplementation

Outcomes	Illustrative comparative risks* (95% CI)		Relative effect (95% CI)	Number of participants (studies)	Quality of the evidence (grade)	Comments
	Assumed risk Placebo or no supplementation	Corresponding risk Growth hormone supplementation to gonadotropins				
Study population						
Clinical pregnancy rates	123 per 1000	230 per 1000 (129-375)	OR 2.13 (1.06-4.28)	251 (7 studies)	⊕⊕⊕⊕ High	
	Moderate					
Live birth rates	111 per 1000	270 per 1000 (128-485)	OR 2.96 (1.17-7.52)	135 (4 studies)	⊕⊕⊕⊕ High	
	Moderate					

*The basis for the assumed risk (e.g., the median control group risk across studies) is provided in footnotes. The corresponding risk (and its 95% CI) is based on the assumed risk in the comparison group and the relative effect of the intervention (and its 95% CI). CI=Confidence interval, OR=Odds ratio. Grade working group grades of evidence - High quality=Further research is very unlikely to change our confidence in the estimate of effect, Moderate quality=Further research is likely to have an important impact on our confidence in the estimate of effect and may change the estimate, Low quality=Further research is very likely to have an important impact on our confidence in the estimate of effect and is likely to change the estimate, Very low quality=We are very uncertain about the estimate

Table 3: Summary of findings for the use of transdermal testosterone supplementation

Transdermal testosterone supplementation to gonadotropins compared to placebo or no supplementation for poor responders

Patient or population: Patients with poor responders
Settings: Randomized controlled trials
Intervention: Transdermal testosterone supplementation to gonadotropins
Comparison: Placebo or no supplementation

Outcomes	Illustrative comparative risks* (95% CI)		Relative effect (95% CI)	Number of participants (studies)	Quality of the evidence (grade)	Comments
	Assumed risk Placebo or no supplementation	Corresponding risk Transdermal testosterone supplementation to gonadotropins				
Study population						
Clinical pregnancy rates	116 per 1000	240 per 1000 (132-398)	OR 2.41 (1.16-5.04)	225 (3 studies)	⊕⊕⊕⊕ High	
Low						
	Moderate					
Live birth rates	107 per 1000	207 per 1000 (108-360)	OR 2.18 (1.01-4.68)	225 (3 studies)	⊕⊕⊕⊕ High	
	Moderate					

*The basis for the assumed risk (e.g., the median control group risk across studies) is provided in footnotes. The corresponding risk (and its 95% CI) is based on the assumed risk in the comparison group and the relative effect of the intervention (and its 95% CI). CI=Confidence interval, OR=Odds ratio. Grade working group grades of evidence - High quality=Further research is very unlikely to change our confidence in the estimate of effect, Moderate quality=Further research is likely to have an important impact on our confidence in the estimate of effect and may change the estimate, Low quality=Further research is very likely to have an important impact on our confidence in the estimate of effect and is likely to change the estimate, Very low quality=We are very uncertain about the estimate

significantly improved LBR (OR 2.96; 95% CI 1.17–7.52) with GH supplementation [Figure 3 and Table 2]

4. Testosterone supplementation: A relatively smaller number of trials tested transdermal testosterone

supplementation in assisted conception cycles (three RCTs; n = 225). The meta-analysis showed significantly improved CPR (OR 2.41; 95% CI 1.16–5.04) and LBR (OR 2.18; 95% CI 1.01–4.68), but the number of oocytes retrieved was not statistically significant (mean difference 0.94; 95% CI 0.24–1.64), [Figure 4 and Table 3]

5. DHEA supplementation: Two RCTs (n = 99). DHEA supplementation was found to have no significant effect on the number of oocytes (mean difference 0; 95% CI − 1.07–1.07) and CPR (OR 2.10; 95% CI 0.75–5.85)
6. Use of aromatase inhibitors: Letrozole supplementation was used in four trials (n = 223) and the pooled data failed to find any statistically significant CPR (OR 1.28; 95% CI 0.60–2.73)
7. Natural cycle: The natural cycle IVF was tested by only one trial (n = 215).[11] The CPR and number of oocytes retrieved were statistically similar in both groups
8. Other interventions: Various authors modified the GnRHa protocols or used various supplementations such as bromocriptine, pyridostigmine, L-arginine, and low-dose aspirin which are described in Table 1. None of these interventions showed any significant improvement in outcomes.

DISCUSSION

Our systematic review updates on the evidence on various strategies to improve reproductive outcome for POR. We analyzed 61 RCTs and 4997 assisted conception cycles which were divided into 10 categories based on the interventions used.

The use of GnRHant protocol for pituitary downregulation is a commonly used approach for poor responders. GnRHant protocol offers several advantages. They cause immediate, rapid gonadotropin suppression by competitively blocking GnRH receptors in the anterior pituitary gland, thereby preventing endogenous premature release of LH and FSH. Our meta-analysis of 17 RCTs did not show any significant difference in CPR or number of oocytes retrieved with the use of GnRHant.[12-28]

LH aids maintain adequate concentrations of intraovarian androgens and promote steroidogenesis and follicular growth. It has been proposed that addition of LH to ovarian stimulation protocol may benefit poor responders. Meta-analysis of eight trials[13,29-33] did not show significant improvement in CPR with use of recombinant LH.

GH, insulin-like growth factor-1, and GH-releasing hormone increase the sensitivity of ovaries to gonadotropin stimulation and enhance follicular development. GH enhances oocyte quality by accelerating and coordinating cytoplasmic and nuclear maturation. There are some suggestions that GH-releasing factor supplementation may improve pregnancy rates in poor responders. The pooled data from eight RCTs in this review show significantly improved CPR and LBR with GH supplementation.[13,29-36] There was no significant heterogeneity in the included studies (τ^2 = 0.00, χ^2 = 0.98, df = 3 [P = 0.81]; I^2 = 0%). However, none of the studies had independently found any significant benefit with GH supplementation. The

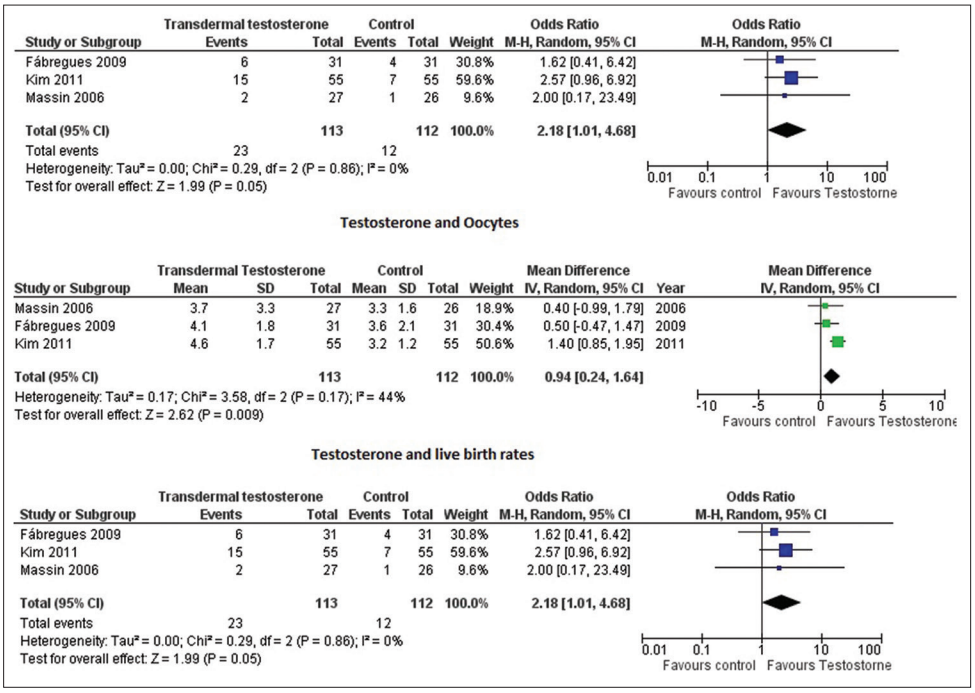

Figure 4: Use of testosterone supplement

total numbers in the meta-analysis are small to draw any definitive conclusions.

Androgen stimulates early stages of follicular growth and increases the number of preantral and antral follicles by the proliferation of granulosa and thecal cells and reduction in granulosa cell apoptosis. It is hypothesized that positive change in microenvironment in the ovaries may lead to an increase in the number and the maturity of oocytes in poor responder group.[37] Three randomized trials[38-40] have tested this approach and the meta-analysis shows significant improvement in LBR and CPR.

Aromatase inhibition was proposed to improve ovarian response to FSH in poor responders. Our meta-analysis included four RCTs and failed to show any improvement in outcome with the use of aromatase inhibitors.

It is proposed that DHEA changes the follicular microenvironment by reducing hypoxic inducible factor-1, thus improving the quality of oocytes. Pooled data from 2 RCTs showed no significant difference in CPR with DHEA supplementation.[41]

Natural cycle IVF offers several advantages such as low cost and low risk of multiple pregnancies and most importantly eliminates the risk of ovarian hyperstimulation syndrome. Morgia *et al*.[11] randomized natural cycle IVF and microdose GnRHa flare along with FSH. It was found that natural cycle IVF may be as effective as IVF using controlled ovarian hyperstimulation. No further trials with this approach were found for meta-analysis.

Strengths and limitations

Our study provides most comprehensive and up-to-date review on the topic of assessing most effective treatment for poor responders and included only RCTs. We divided different approaches into 10 categories and performed meta-analysis as appropriate. Previous reviews were very specific in addressing one treatment strategy, and they failed to provide any conclusive answer. Some reviews were methodologically limited as they included observational studies and nonrandomized studies in their meta-analysis.[4,7,9]

The major limitation of this review is related to its small population size. Although some adjuvant supplementations may appear to improve ovarian response and reproductive outcome, we recognize that the numbers are small to recommend their routine use in poor responders. There was significant heterogeneity in the definition of poor responders in these trials conducted before Bologna consensus criteria were recommended.

Interpretation

Our meta-analysis showed no difference in the number of oocytes retrieved or the CPRs with use of GnRHant. The pooled data from seven studies show significantly improved CPR and LBR with GH supplementation in the previous review.[4] Our meta-analysis adds a further RCT[36] (n = 82) which results in a 48% increase in sample size. GH supplementation showed some promising results; however, the numbers are small to draw any convincing conclusion. Our results for testosterone supplementation are consistent with the results of previous meta-analyses as there were no new RCTs.[5,7] Letrozole supplementation may result in improved FSH sensitivity and concentration, but this beneficial effect was not reflected in the results. A systematic review by Bosdou *et al*.[7] previously showed no difference in outcome with the use of letrozole. Two more RCTs have been undertaken[37,42] since the previous review, and we added a total of 68 cycles (43%) to the sample size in our review. However, the pooled data showed no significant difference in outcome with use of letrozole. The anti-aging effect of the adrenal androgen DHEA is thought to be the mechanism to improve ovarian response. Recent meta-analysis did not show significant improvement with the use of DHEA.[9] Only two RCTs were eligible for our meta-analysis, which failed to demonstrate any benefit.

CONCLUSION

Evidence from this review suggests that GH supplementation or transdermal testosterone supplementation to assisted conception treatment cycles is associated with an improved CPR and LBR in poor responders. However, it is essential to recognize that this evidence is derived from a small number of studies; hence, we feel that the current evidence is insufficient to recommend the routine use of either of these approaches. Other treatment strategies are not found to be useful in improving clinical outcome in poor responders. We recommend that the empirical use of adjuvants should be avoided pending good quality evidence from well-designed studies.

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Conflicts of interest

There are no conflicts of interest.

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