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REVIEW

Effect of androgen supplementation or modulation on ovarian stimulation outcome in poor responders: a meta-analysis

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Abstract Many trials have evaluated the use of androgen supplements and androgen-modulating agents to improve outcome of poor responders undergoing IVF treatment. This study conducted a systematic review and meta-analysis of controlled trials of androgen adjuvants (testosterone, dehydroepiandrostereone) and the androgen-modulating agent (letrozole) in poor responders undergoing IVF treatment. Searches were conducted on MEDLINE, EMBASE, Cochrane Library, ISRCTN Register and ISI proceedings. All randomized and non-randomized controlled trials were included. Study selection, quality appraisal and data extraction were performed independently and in duplicate. The main outcome measure was clinical pregnancy rate. The secondary outcome measures were dose and duration of gonadotrophin use, cycles cancelled before oocyte retrieval, oocytes retrieved and ongoing pregnancy rates. A total of 2481 cycles in women considered as poor responders undergoing IVF/intracytoplasmic sperm injection (ICSI) treatment were included in nine controlled trials. Meta-analyses of these studies did not show any significant difference in the number of oocytes retrieved and ongoing pregnancy/live-birth rates with androgen supplementation or modulation compared with the control groups. There is currently insufficient evidence from the few randomized controlled trials to support the use of androgen supplementation or modulation to improve live birth outcome in poor responders undergoing IVF/ICSI treatment.

KEYWORDS: androgens, controlled studies, IVF, poor responders, pregnancy

Introduction

Poor ovarian response is usually defined as failure to achieve at least three oocytes or a certain oestradiol concentration in response to ovarian stimulation, although there is no clear consensus regarding its definition (Sunkara et al., 2007). The incidence of poor response to ovarian stimulation has been reported to vary from 9% to 24% (Keay et al., 1997). Poor ovarian response often results in fewer embryos, reduced implantation rate, decreased pregnancy

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rate (Mahutte and Arici, 2002) and results in high cycle cancellation rate. The burden of poor ovarian response is an increasing problem with women delaying childbirth and presenting for fertility treatment later in their lives. The management of poor responders is still a major challenge in assisted reproduction. Various stimulation regimens and interventions have been proposed for improving the pregnancy outcome in poor responders. These include different regimens of pituitary suppression, ovarian stimulation and adjuvant therapies (Pandian et al., 2010). The proposed adjuvant therapies include growth hormone, growth hormone releasing factor, pyridostigmine, oral L-arginine, recombinant LH, human chorionic gonadotrophin, androgen supplements (testosterone and dehydroepiandrosterone, DHEA) and androgen-modulating agents (letrozole and anastrozole). Most of the interventions proposed to improve pregnancy rates in poor responders showed insufficient evidence to recommend any of them.

The factors that initiate maturation of primordial follicles are poorly understood. Androgens seem to play an important role in ovarian physiology and follicular growth. There are some reports demonstrating that androgens are likely to act on folliculogenesis by increasing the number of FSH receptors expressed in the granulosa cells (Weil et al., 1999). It has been demonstrated in primates that androgens induce a significant increase in the number of primary, preantral and antral follicles, independent of the gonadotrophins (Vendola et al., 1998).

The addition of androgens in ovarian stimulation is thought to have a synergistic effect to FSH in follicular recruitment and granulosa cell proliferation, while the exogenous exposure to FSH protects such recruited follicles from atresia (Feigenberg et al., 2009). With this background, it has been suggested that the addition of androgens in ovarian stimulation, might be synergistic to FSH in follicular recruitment. This has led to the assumption that treating poor responders with androgens might improve the response to ovarian stimulation. Androgens proposed as adjuvants in ovarian stimulation are testosterone and DHEA and the androgen-modulating agents are letrozole and anastrozole. Letrozole is a highly selective, non-steroidal aromatase inhibitor. It blocks the conversion of androgen substrate to oestrogen in the granulosa cells, thereby causing raised intraovarian androgens. Studies that have looked at the addition of androgens and androgen-modulating agents to ovarian stimulation in poor responders have reported inconsistent results. The purpose of this systemic review and meta-analysis is to summarize the existing evidence on the role of androgens and androgen modulators in the management of poor responders to ovarian stimulation.

Materials and methods

This study searched MEDLINE (1950 to August 2010), EMBASE (1980 to August 2010) and Cochrane Library for the relevant studies. The search also included ISI conference proceedings as well as databases for registration of ongoing and archived randomized controlled trials, namely ISRCTN register and meta-register of controlled trials (mRCT). A combination of Medical Subject Headings (MeSH) and text words were used to generate three subsets of citations, for studies of poor ovarian response ('poor response' or 'low response' or 'inadequate response'), studies of IVF ('in vitro fertilization' or 'IVF', or 'intracytoplasmic sperm injection' or 'ICSI'), and studies of androgen ('testosterone', or 'dehydroepiandrostereone' or 'DHEA' or 'letrozole' or 'aromatase inhibitor). These subsets were combined using 'AND' to generate a subset of citations relevant to the research question. The reference lists of all primary and review articles were examined to identify cited articles not captured by electronic searches. The study also made enquiries about unpublished studies from researchers in this field. No language restrictions were placed in any of the searches. The searches were conducted independently by JP and SKS.

Study selection and data extraction

Studies were selected if the target population was poor responders undergoing IVF/intracytoplasmic sperm injection (ICSI) treatment. The study group consisted of cycles with androgen supplementation (testosterone, DHEA) or androgen-modulating drugs (letrozole) as adjuvant, and the control group consisted of cycles with no adjuvant or placebo. The primary outcome was the clinical pregnancy rate and the secondary outcomes were cycle cancellation, number of oocytes retrieved and ongoing pregnancy rates. Data were also obtained on the total dose and duration of gonadotrophin used for ovarian stimulation as a surrogate outcome.

Studies were selected in a two-stage process. Firstly, the titles and abstracts from the electronic searches were scrutinized by two reviewers independently (JP and SKS) and full manuscripts of all citations that were likely to meet the predefined selection criteria were obtained. Secondly, final inclusion or exclusion decisions were made on examination of the full manuscripts. In cases of duplicate publication, the most recent or complete versions were selected. Any disagreements about inclusion were resolved by consensus or arbitration by a third reviewer (YK).

Two reviewers (JP and SKS) completed the data extraction and quality assessment (Berlin and Rennie, 1999). Authors of the primary studies were contacted for any missing or unclear information. From each study, outcome data were extracted in 2×2 tables by the two reviewers JP and SKS.

Statistical analysis

The selected studies were assessed for methodological quality by using the components of study design that are related to internal validity (Centre for Reviews and Dissemination, 2001). For randomized studies, information on the method of randomization, allocation concealment, blinding, intention-to-treat analysis and follow-up rates was sought by examining the full text articles. Study characteristics and participant features (such as the definition of poor ovarian response, androgen protocol) were extracted from each study.

Relative risks (RR) from individual studies were meta-analysed using fixed effects (Mantel and Haenszel, 1959) and random effects models as appropriate

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(DerSimonian and Laird, 1986). Heterogeneity of the exposure effects was evaluated graphically using forest plots (Lewis and Clarke, 2001) and statistically using the I^2 statistic to quantify heterogeneity across studies (Higgins and Thompson, 2002). Exploration of the causes of heterogeneity was planned using variation in features of population, exposure and study quality. Sensitivity analyses were based on the type of the adjuvant (testosterone DHEA and letrozole only) used. Statistical analyses were performed using RevMan 4.2 software (Cochrane Collaboration, Oxford, UK).

Results

The process of literature identification and selection is summarized in **Figure 1**. Of the 235 citations identified, 26 were selected during the initial screening. On examination of manuscripts, 17 studies were excluded: one study was a duplicate study (Garcia-Velasco and Guillen, 2005); six studies were prospective self controled studies (Balasch et al., 2006; Barad and Gleicher, 2006; Casson et al., 2000; Fernandez-Shaw et al., 2008; Mitwally and Casper, 2002; Sonmezer et al., 2009); six studies were clinical reviews (Atay et al., 2007; Feigenberg et al., 2009; Karaer et al., 2004; Mamas and Mamas, 2009; Mitwally and Casper, 2003; Requena et al., 2008) and the remaining four studies did not include poor responders (Lossl et al., 2006, 2008, 2009; Verpoest et al., 2006). Nine studies with suitable information including a total of 2481 cycles were identified that satisfied the selection criteria for the review.

The literature search found five RCTs. Two RCTs compared letrozole versus no letrozole (Goswami et al., 2004; Ozmen et al., 2009), two RCTs compared the testosterone versus no testosterone (Fabregues et al., 2009; Massin et al., 2006) and one RCTs compared DHEA versus no DHEA supplementation (Wiser et al., 2010). Four non-randomized controlled trials were identified, of which two prospective controlled studies (Garcia-Velasco et al., 2005; Schoolcraft et al., 2008) and one retrospective controlled study (Yarali et al., 2009) looked at the addition of letrozole and one retrospective controlled study looked at DHEA supplementation (Barad et al., 2007).

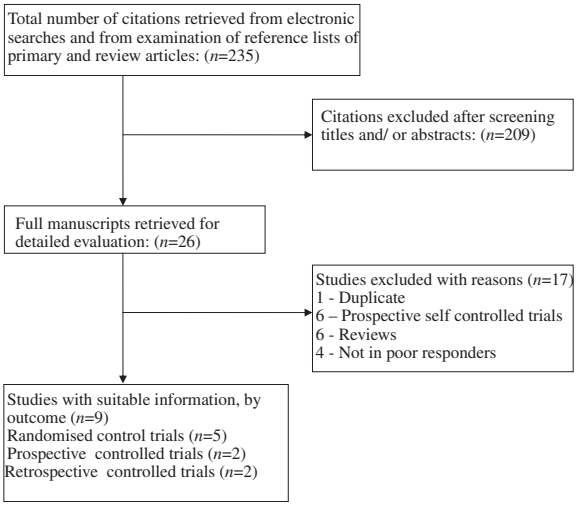

Figure 1 Study selection process for the systematic review of adjuvant androgens in poor responders undergoing IVF treatment.

All five RCTs had undertaken allocation concealment. The method of randomization was not clearly documented in two studies, and two out of the five studies had blinding (**Table 1**). Only one study had placebo in the control group (**Table 2**). These trials were generally small, leading to imprecision in their findings, even when combined in meta-analysis. Furthermore, there was substantial clinical and statistical heterogeneity amongst the trials, weakening any inferences that could be drawn from these studies. The quality and the main characteristics of the included studies are presented in **Tables 1** and **2**.

Primary outcome: clinical pregnancy rate

Meta-analysis of the five RCTs (Fabregues et al., 2009; Goswami et al., 2004; Massin et al., 2006; Ozmen et al., 2009; Wiser et al., 2010) for the outcome of clinical pregnancy rate (CPR) showed no significant difference between those treated with androgen supplementation compared with those without androgen supplementation (RR 1.60, 95% CI 0.91, 2.83; **Figure 2**). Meta-analysis of the four non-randomized controlled studies (Barad et al., 2007; Garcia-Velasco et al., 2005; Schoolcraft et al., 2008; Yarali et al., 2009) for the outcome of CPR showed no significant difference between those treated with androgen supplementation compared with those without androgen supplementation (RR 1.29, 95% CI 0.71, 2.35; **Figure 2**). Pooling of results of all the randomized and non-randomized controlled studies for the outcome of CPR showed no significant difference between those treated with androgen supplementation compared with those without androgen supplementation (RR 1.13, 95% CI 0.82, 1.55; **Figure 2**).

Sensitivity analysis of the five studies using letrozole in the study group (Garcia-Velasco et al., 2005; Goswami et al., 2004; Ozmen et al., 2009; Schoolcraft et al., 2008; Yarali et al., 2009) showed no significant difference in the CPR in the letrozole group compared with the control group (RR 0.91, 95% CI 0.68, 1.22). Sensitivity analysis of the four studies that used either testosterone patches or DHEA in the study group (Barad et al., 2007; Fabregues et al., 2009; Massin et al., 2006; Wiser et al., 2010) showed a significantly higher CPR in the study group compared with the control group (RR 2.86, 95% CI 1.73, 4.73).

Secondary outcomes

Cycle cancellation rate

Meta-analysis of four (Fabregues et al., 2009; Goswami et al., 2004; Massin et al., 2006; Ozmen et al., 2009) out of the five RCTs which reported cycle cancellation as an outcome showed no significant difference between those treated with androgen supplementation compared with those without androgen supplementation (RR 0.70, 95% CI 0.29, 1.69). Meta-analysis of the four non-randomized controlled studies (Barad et al., 2007; Garcia-Velasco et al., 2005; Schoolcraft et al., 2008; Yarali et al., 2009) for the outcome of cycle cancellation showed no significant difference between women treated with androgen

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Table 1 Quality of studies included in the review of adjuvant androgens in poor responders undergoing IVF treatment.

Publication	*Study design*	*Method of randomization*	*Allocation concealment*	*Blinding*	*Intention-to-treat analysis*	*Follow-up rate (%)*
Goswami et al. (2004)	Randomized controlled trial	Random number tables	Yes	Yes	Yes	>95
Massin et al. (2006)	Randomized controlled trial	Allocation sequence was generated by a random permutation table (blocks of four)	Yes	Yes	Yes	>95
Schoolcraft et al. (2008)	Prospective controlled study	1:2 allocation of participants into study and control group	NA	No	Not mentioned	>95
Ozmen et al. (2009)	Randomized controlled trial	NA	Yes	No	Yes	>95
Fabregues et al. (2009)	Randomized controlled trial	Computer generated randomization list	Yes	No	Yes	>95
Garcia-Velasco et al. (2005)	Prospective controlled study	NA	NA	No	Yes	>95
Barad et al. (2007)	Retrospective controlled study	NA	NA	NA	Yes	>95
Yarali et al. (2009)	Retrospective controlled study	NA	NA	NA	Yes	>95
Wiser et al. (2010)	Randomized controlled trial	Computer generated random numbers	Yes	No	Yes	100

NA = not applicable.

supplementation compared with those without (RR 1.10, 95% CI 0.74, 1.63). Pooling of results of the four randomized and four non-randomized controlled studies for the outcome of cycle cancellation showed no significant difference between those treated with androgen supplementation compared with those without (RR 0.91, 95% CI 0.59, 1.41).

Sensitivity analysis of the five controlled studies that used letrozole in the study group (Garcia-Velasco-Velasco et al., 2005; Goswami et al., 2004; Ozmen et al., 2009; Schoolcraft et al., 2008; Yarali et al., 2009) showed no significant difference in the cycle cancellation rate in the letrozole group compared with the control group (RR 1.13, 95% CI 0.75, 1.72). Sensitivity analysis of the study that used DHEA (Barad et al., 2007) and the two studies that used testosterone (Fabregues et al., 2009; Massin et al., 2006) showed no significant difference in the cycle cancellation rate between the study group compared with the control group (RR 0.71, 95% CI 0.35, 1.45).

Total dose of gonadotrophin

Four out of five RCTs (Fabregues et al., 2009; Goswami et al., 2004; Massin et al., 2006; Ozmen et al., 2009), reported on the total dose of gonadotrophin (total units, IU) used as an outcome. One of the studies (Goswami et al., 2004) used a lower daily dose of gonadotrophin in the letrozole group compared with the control group. Therefore this study was excluded from the meta-analysis for the dose and duration of gonadotrophins used. Meta-analysis of the remaining three RCTs (Fabregues et al., 2009; Massin et al., 2006; Ozmen et al., 2009) showed that the total dose of gonadotrophins used was significantly lower in those with androgen supplementation compared with those without androgen supplementation (weighted mean difference (WMD) −811.21, 95% CI −1025.03, −597.39). Meta-analysis of the two (Garcia-Velasco et al., 2005; Yarali et al., 2009) out of four non-randomized controlled studies that reported on the total dose of gonadotrophin used as an outcome (in units) showed that the cycles with androgen supplementation needed a significantly lower total dose of gonadotrophin compared with those without androgen supplementation (WMD −338.69, 95% CI −671.48, −5.89). One non-randomized controlled study (Schoolcraft et al., 2008) could not be included as it reported the total gonadotrophin dose in ampoules. Pooling of results of all the three randomized and two non-randomized controlled studies also showed that the total dose of gonadotrophins used was significantly lower in the study group compared with the control group (WMD −530.45, 95% CI −850.84, −210.06).

Sensitivity analysis of the three (Garcia-Velasco et al., 2005; Ozmen et al., 2009; Yarali et al., 2009) out of five studies using letrozole in the study group which reported on the total dose of gonadotrophin used showed that the study groups needed a significantly lower total dose of gonadotrophins compared with the control group (WMD −506.35, 95% CI −884.21, −128.49). Sensitivity analysis of the two randomized controlled studies that used testosterone patches in the study group (Fabregues et al., 2009; Massin et al., 2006) also showed that the total dose

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Table 2 Characteristics of the studies included in the review of adjuvant androgens in poor responders undergoing IVF treatment.

Publication	*Inclusion criteria (definition of poor responder)*	*No. of cases and controls*	*Androgen protocol*	*Ovarian stimulation protocol*	*Day/mean no. of embryos transferred*	*Outcomes reported*
Goswami et al. (2004), *n* = 38	1–3 failed IVF due to poor ovarian response (no definition) FSH >12 IU/l excluded	13 cases 25 controls	Cases: letrozole Controls: none	Cases: letrozole 2.5 mg orally D3–7, rFSH s.c. 75 IU/day D3–8 Controls: long down-regulation protocol with FSH (300–450 IU)	D2 transfer	Total dose of gonadotrophins Cancellation rate No. of oocytes Transferable embryos
Massin et al. (2006), *n* = 49	One of the following: Oestradiol <1200 pg/ml on HCG day Total oocytes retrieved <5 in previous cycle Decreased ovarian reserve: FSH >12 IU/l, oestradiol >70 pg/ml and inhibin B <45 pg/ml	24 cases 25 controls	Cases: testosterone gel Controls: placebo	Testosterone: 1 g gel/day (10% absorption, 10 mg of testosterone) or placebo for 15–20 days during the period of pituitary desensitization, preceding the stimulation The same GnRH analogue protocol was used for both cases and controls	Up to 3 embryos on D2–3	Total dose of gonadotrophins Duration of gonadotrophin stimulation Cancellation rate No. of oocytes Mature oocytes Implantation rate Clinical pregnancy rate Live birth rate
Schoolcraft et al. (2008), *n* = 534	One of the following: FSH >10 m IU/ml Total AFC <6 Prior cycle cancellation Prior poor response: peak oestradiol <500 pg/ml and/or <6 oocytes Age >41 years	179 cases 355 controls	Cases: Letrozole Controls: none	Cases: Day 3 FSH (300 IU) and HMG (150 IU), letrozole 2.5 mg/day for 5 days. GnRH antagonist when lead follicle >14 mm Controls: 14–21 days of COC pills, 3 days after last COC pill – leuprolide 40 μg s.c. b.d. (until HCG), 2 days after starting leuprolide FSH (300 IU) and HMG (150 IU)	D3–5 transfer	Total dose of gonadotrophins Duration of gonadotrophin stimulation Cancellation rate No. of oocytes Mature oocytes Implantation rate Clinical pregnancy rate
Ozmen et al. (2009), *n* = 70	Cycle cancellation due to oestradiol <130 pg/ml on D6 or <450 on day of HCG <4 retrieved oocytes	35 cases 35 controls	Cases: letrozole Controls: none	Cases: 5 mg letrozole on D3–7, with 450 IU/day FSH started on D5 Controls: 450 IU/day FSH started on D3 Both had GnRH antagonist protocol	D3 transfer	Total dose of gonadotrophin Cancellation rate No. of oocytes Mature oocytes Implantation rate Clinical pregnancy rate

Table 2 (*continued*)

Publication	*Inclusion criteria (definition of poor responder)*	*No. of cases and controls*	*Androgen protocol*	*Ovarian stimulation protocol*	*Day/mean no. of embryos transferred*	*Outcomes reported*
Fabregues et al. (2009), *n* = 62	Women who had first IVF cycle cancelled due to poor follicular response Age 31–39 years BMI normal (19.3–28.9 kg/m^2) Normal ovaries, no pelvic surgery, regular menses, normal ovulatory function	31 cases 31 controls	Cases: testosterone Controls: none	Cases: transdermal testosterone with a daily single patch of 2.5 mg during the 5 days preceding GnRH agonist (standard long down-regulation protocol) Controls: high dose gonadotrophin with a minidose GnRH agonist protocol	D2–3 transfer Up to 3 embryos replaced	Total dose of gonadotrophins Duration of gonadotrophin stimulation Cancellation rate No. of follicles No. of oocytes Implantation rate Clinical pregnancy rate Ongoing pregnancy rate
Garcia-Velasco et al. (2005), *n* = 147	One previous cancelled IVF cycle in which 4 or fewer follicles were obtained and/or serum oestradiol <500 pg/ml and basal FSH <12 IU/ml	71 cases 76 controls	Cases: letrozole Controls: none	Cases: Letrozole 2.5 mg was added for the first 5 days of stimulation. Controls: same protocol without letrozole. Both had GnRH antagonist protocol	D3 transfer	Total dose of gonadotrophins Duration of gonadotrophin stimulation Cancellation rate No. of oocytes Implantation rate Clinical pregnancy rate Miscarriage rate Ongoing pregnancy rate
Barad et al. (2007), *n* = 165	Premature ovarian ageing: baseline FSH <12 mIU/ml but exceeding the 95% CI of the mean value for the patient's age group or Diminished ovarian reserve: baseline FSH >12 mIU/ml and or baseline oestradiol >75 pg/ml	64 cases 101 controls	Cases: DHEA Controls: none	Same protocol for cases and controls: –microdose agonist flare followed by maximum dosage gonadotrophin stimulation Cases: DHEA 25 mg three times a day for up to 4 months, which was continued until a positive pregnancy test	Not mentioned	Cancellation rate No. of oocytes Implantation rate Clinical pregnancy rate Miscarriage rate
Yarali et al. (2009), *n* = 1383	FSH >10 mIU/ml Oestradiol >60 pg/ml, Bilateral AFC <6 Or a history of poor ovarian response: cycle cancellation, peak oestradiol <500 pg/ml, or retrieval of <4 oocytes	357 cases 1026 controls	Cases: letrozole Controls: none	Cases: Letrozole at a dose of 2.5 mg/day from day 2 for 5 days. On D2, 300 IU FSH and 150 IU HMG. GnRH antagonist started once lead follicle >13 mm Controls: microdose leuprolide acetate flare-up regimen. Started on oral contraceptive pills for 21 days. Leuprolide acetate started 3 days after the last pill, 2 days later 300 IU FSH and 150 IU HMG started	D3 embryo transfer	Total dose of gonadotrophins Duration of gonadotrophin stimulation Cancellation rate No. of oocytes Implantation rate Clinical pregnancy rate Miscarriage rate Ongoing pregnancy rate

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Wiser et al. (2010), *n* = 33	Previous IVF cycle (300 IU gonadotrophins) with retrieval of <5 oocytes Cycle cancellation due to poor response Exclusion: age >42 years Previous exposure to DHEA	17 cases 16 controls	Cases: DHEA Controls: none	Cases: DHEA 75 mg orally once a day for atleast 6 weeks before stimulation All patients: standard long protocol with GnRH agonist started in luteal phase. Once down-regulated, 450 IU rFSH with 150 IU rLH commenced	D2–3 embryo transfer Up to 3 embryos	No. of oocytes Clinical pregnancy rate Miscarriage rate Ongoing pregnancy rate

AFC = antral follicle count; COC = combined oral contraceptive; D = day; DHEA = dihydroepiandrostenedione; GnRH = gonadotrophin-releasing hormone; HCG = human chorionic gonadotrophin; HMG = human menopausal gonadotrophin.

of gonadotrophin used in the testosterone group was significantly less compared with the control group that did not have testosterone (WMD −586.69, 95% CI −1056.07, −117.32). Both the studies that used DHEA did not report on the dose of gonadotrophin used.

Total duration of gonadotrophin stimulation

Two (Fabregues et al., 2009; Massin et al., 2006) out of the five RCTs, reported on the total duration of gonadotrophin used as an outcome. Meta-analysis of these studies showed that the total duration of gonadotrophin used was not significantly different in those with androgen supplementation compared with those without androgen supplementation (WMD −0.25, 95% CI −0.93, 0.44). Meta-analysis of the three (Garcia-Velasco et al., 2005; Schoolcraft et al., 2008; Yarali et al., 2009) out of four non-randomized controlled studies for the total duration of gonadotrophin used showed no significant difference between those with androgen supplementation compared with those without (WMD −0.32, 95% CI −1.16, 0.52). Pooling of results of all the randomized and non-randomized controlled studies for the total duration of gonadotrophin used also showed no significant difference between the two groups (WMD −0.31, 95% CI −0.98, 0.37).

Sensitivity analysis of the three (Garcia-Velasco et al., 2005; Schoolcraft et al., 2008; Yarali et al., 2009) out of five studies using letrozole in the study group showed no significant difference in the total duration of gonadotrophin used in the letrozole group compared with the control group (WMD −0.32, 95% CI −1.16, 0.52). Sensitivity analysis of the two RCTs that used testosterone in the study group (Fabregues et al., 2009; Massin et al., 2006) showed no significant difference in the total duration of gonadotrophin used between the two groups (WMD −0.27, 95% CI −1.25, 0.70). Both the studies that used DHEA did not report on the duration of gonadotrophin used.

Oocytes retrieved

Meta-analysis of the five RCTs (Fabregues et al., 2009; Goswami et al., 2004; Massin et al., 2006; Ozmen et al., 2009; Wiser et al., 2010), for the number of oocytes retrieved showed no significant difference between those treated with androgen supplementation compared with those without androgen supplementation (WMD −0.11, 95% CI −0.67, 0.44; **Figure 3**). Meta-analysis of the four non-randomized controlled studies (Barad et al., 2007; Garcia-Velasco et al., 2005; Schoolcraft et al., 2008; Yarali et al., 2009) for the number of oocytes retrieved showed no significant difference between those treated with androgen supplementation compared with those without (WMD −0.92, 95% CI −3.45, 1.61; **Figure 3**). Pooling of results of all the randomized and non-randomized controlled studies for the number of oocytes retrieved showed no significant difference between those treated with androgen supplementation compared with those without the androgen supplementation (WMD −0.45, 95% CI −2.04, 1.14; **Figure 3**).

Sensitivity analysis of the five studies using letrozole in the study group (Garcia-Velasco et al., 2005; Goswami et al., 2004; Ozmen et al., 2009; Schoolcraft et al., 2008; Yarali et al., 2009) showed no significant difference in the number of oocytes retrieved in the letrozole group com-

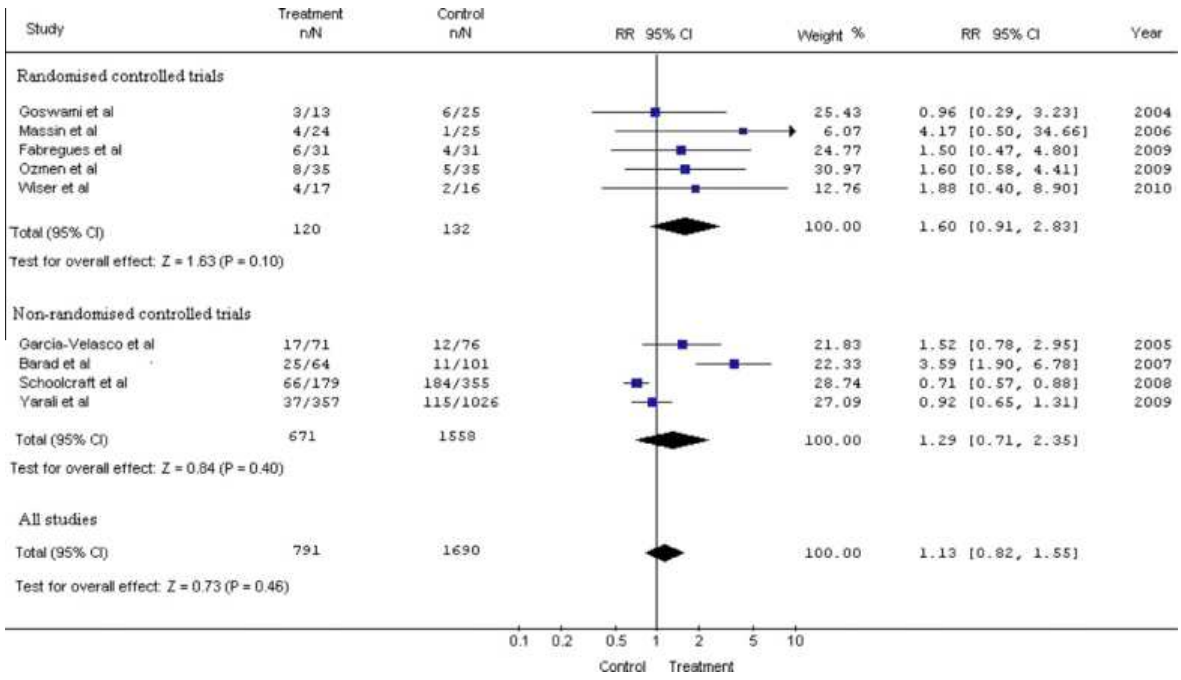

Figure 2 Meta-analysis of studies of adjuvant androgen supplementation versus controls for outcome of clinical pregnancy rates in poor responders undergoing IVF treatment.

pared with the control group (WMD −0.43, 95% CI −2.47, 1.61). Sensitivity analysis of the four studies that used testosterone or DHEA in the study group (Barad et al., 2007; Fabregues et al., 2009; Massin et al., 2006; Wiser et al., 2010) showed no significant difference in the number of oocytes retrieved between the two groups (WMD −0.52, 95% CI −2.04, 1.00).

Ongoing pregnancy rate

Meta-analysis of the three (Fabregues et al., 2009; Massin et al., 2006; Wiser et al., 2010) out of five RCTs, that reported the outcome of ongoing pregnancy rate (OPR) showed no significant difference between the androgen supplementation group compared with those without (RR 1.99, 95% CI 0.72, 5.51). Meta-analysis of the two non-randomized controlled studies (Garcia-Velasco et al., 2005; Yarali et al., 2009) that reported the outcome of OPR showed no significant difference between those with androgen supplementation compared with those without (RR 1.02, 95% CI 0.72, 1.45). Pooling of results of the randomized and non-randomized controlled studies for the outcome of OPR showed no significant difference between those with androgen supplementation compared with those without androgen supplementation (RR 1.10, 95% CI 0.79, 1.53).

Sensitivity analysis of the two (Garcia-Velasco et al., 2005; Yarali et al., 2009) out of the five studies that used letrozole in the study group and reported on OPR showed no significant difference between the letrozole group compared with the control group (RR 1.02, 95% CI 0.72, 1.45). Sensitivity analysis of the two RCTs that used testosterone (Fabregues et al., 2009; Massin et al., 2006) and the RCT that used DHEA (Wiser et al., 2010) in the study group and reported on OPR showed no significant difference in the OPR in the study group compared with the control group (RR 1.99, 95% CI 0.72, 5.51).

Discussion

This review demonstrated that there is insufficient evidence to suggest androgen supplementation improves IVF treatment outcome in poor responders. Systemic androgen administration has a different mode of action on the ovary compared with an androgen-modulating agent (letrozole). This study therefore performed separate subgroup analyses for studies using letrozole and those using testosterone or DHEA as adjuvants. The results showed no significant differences in the cycle cancellation rate, number of oocytes retrieved, CPR and OPR with the letrozole group compared with the control group. There was a significantly higher CPR in the study group with testosterone or DHEA supplementation compared with the control group, but there was no significant difference in the cycle cancellation rate, number of oocytes retrieved and OPR between the study and control groups.

This study explored the possibility of performing a sensitivity analysis based on age, as poor response in young age has better outcome than poor response in old age. However, this was not possible as none of the included studies had women with a mean age of ≤35 years. Eight of the nine included studies had women with a mean age ≥36 years and ≤40 years and only one study (Barad et al., 2007) had women with a mean age ≥40 years.

This review showed that the dose of gonadotrophins used was significantly lower in the androgen supplementation

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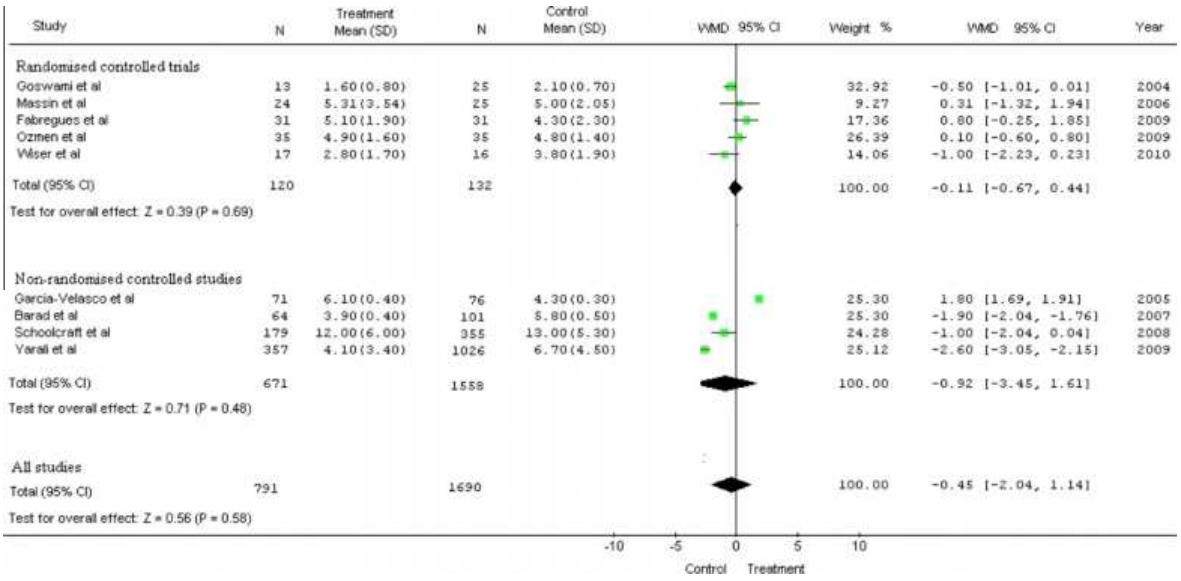

Figure 3 Meta-analysis of studies of adjuvant androgen supplementation versus controls for outcome of number of oocytes retrieved in poor responders undergoing IVF treatment.

group compared with the control group. The selective inhibition of aromatase prevents the production of oestrogen and thereby prevents the negative feedback effect on the hypothalamo-pituitary axis. This is expected to result in more endogenous FSH, which may explain the finding of the need for reduced dose of exogenous FSH required for ovarian stimulation. However, this review showed that there is no significant difference in the total duration of gonadotrophin stimulation used between the study and control groups. In two of the included studies even though the starting dose of gonadotrophins was the same in both the groups, the dose was adjusted on day 3 or 5 based on the ovarian response (Fabregues et al., 2009; Ozmen et al., 2009). Since these two studies and most of the other studies were not blinded a possible bias in the decision making for adjusting the doses of gonadotrophins in patients receiving androgen supplementation or modulation cannot be ruled out.

The first study addressing the supplementation of androgens (Casson et al., 2000) looked at DHEA supplementation and concluded that DHEA supplementation appeared to augment ovulation induction in poor responders. Subsequent to this report various studies tried androgen supplementation (testosterone and DHEA) and androgen-modulating agents (letrozole) in an attempt to improve outcome in poor responders. Two self-controlled studies looked at testosterone and DHEA supplementation in a small sample of poor responders. In one study the patients received transdermal testosterone treatment during the 5 days preceding gonadotrophin treatment. They reported that 80% of patients showed fivefold increase in the number of recruited follicles and clinical pregnancy rate was 30% per oocyte retrieval (Balasch et al., 2006). Another study looking at the effect of DHEA supplementation concluded that DHEA supplementation might enhance ovarian response, reduce cycle cancellation rate and increase embryo quality in such patients (Sonmezer et al., 2009). It is clear that studies such as these have omitted, among other factors, the well-recognized cycle-to-cycle variation by adopting this flawed methodology.

A systematic review on the use of letrozole in normal responders included one randomized controlled study and one cohort study and the pooled pregnancy rate per patient was not found to be significantly different (Requena et al., 2008). A recent systematic review (Kyrou et al., 2009) which evaluated the effectiveness of various interventions and adjuvants in poor responders found only one randomized controlled study on the use of testosterone and one study on the use of letrozole and showed no benefit with either of these interventions (Kyrou et al., 2009).

As far as is known, this is the first study to give a quantitative estimate of the effect of various androgen supplements and modulators used in IVF treatment in poor responders. The validity of this study depends on the methodological rigor of the review and the component primary studies. A prospective protocol was used and a concerted effort made to find all the evidence. Two independent reviewers assessed study quality and extracted data and the agreement between the two reviewers was high.

One major problem usually encountered when synthesizing the evidence in meta-analyses is the clinical and methodological heterogeneity between the studies. Such problems encountered in this review were: the differences between the studies in the populations they evaluated (definition of poor ovarian response); the type, dose and duration of androgen supplement used; and the ovarian stimulation protocol, as well as important differences in the quality features between the studies. In addition, the wide confidence intervals around the relative risks for the outcome measures suggest imprecise results, which could be directly attributed to the relatively small sample sizes of the studies in this review. Furthermore, it should be highlighted that as there were few and small RCT, non-randomized studies were included in the meta-analysis.

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Finally, it is important to highlight that existing studies were not powered to assess differences in pregnancy rates, rendering their use in counselling patients regarding treatment success rates uninformative. This methodological shortcoming is most probably due to the expected low pregnancy rates in poor responders (Surrey and Schoolcraft, 2000), such that sample sizes with enough power to detect differences in pregnancy rate would be prohibitive for a single centre trial.

In conclusion, this systematic review has found that there is currently insufficient evidence from the few and small RCT to support that addition of androgen supplements or androgen-modulating agents to gonadotrophins in poor responders undergoing IVF treatment outcome. As research on poor responders has been hindered by lack of consistent definition, future research should be based on strict evidence based criteria to define poor responders. Furthermore, studies should be powered sufficiently to detect differences in clinically meaningful outcomes in order to make useful conclusions about therapy.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.rbmo.2011.01.015.

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