Gonadotrophin-releasing hormone antagonists for assisted reproductive technology in women with poor ovarian response. Subgroup analysis of Cochrane systematic review and meta-analysis

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ABSTRACT

BACKGROUND: Treatment of women with poor ovarian response during IVF/ICSI treatment is so complicated. Most of treatment protocols used lead to more cost without improvement in pregnancy rate. The aim of the present subgroup analysis was to evaluate the efficacy of GnRH antagonist in poor responders.

METHODS: We conducted a systematic review and meta-analysis of randomized trials comparing the effect of GnRH agonist versus GnRH antagonist in poor responders undergoing IVF/ICSI cycles.

Outcomes: primary outcome was ongoing pregnancy rate while the Secondary outcomes was clinical pregnancy rate, miscarriage rate, duration of stimulation, amount of FSH, number of retrieved oocytes, number of mature oocytes and cancellation rate. Searches (until Sep. 2010) were conducted in MEDLINE, EMBASE, Science Direct, Cochrane Library and databases of abstracts.

RESULTS: Six randomized trials entailing 919 women were included. There was no evidence of a statistically significant difference in ongoing pregnancy rate (3 RCTs; OR: 1.17, 95% CI 0.53 to 2.58) for GnRH antagonist versus long GnRH agonist protocol.

CONCLUSION: In view of its equivalence, GnRH antagonist protocol for pituitary suppression is an alternative for standard long GnRH agonist protocol in poor responder patients undergoing IVF/ICSI treatment cycles.

INTRODUCTION

Women with poor ovarian response during IVF/ICSI treatment cycles are estimated to comprise approximately 9-24% of IVF/ICSI patients. **(1)** The ideal treatment of those women is a difficult question endlessly without agreeing until now.

Various protocols have been tried to improve pregnancy outcomes in poor responder women **(2)**. These protocols included, long GnRH agonist/high dose of gonadotropin versus short GnRH agonist/high dose of gonadotropin. **(3; 4; 5)** Long GnRH agonist/low dose of gonadotropin versus long GnRH agonist/high dose of gonadotropin **(6; 7)** , short GnRH agonist/high dose of gonadotropin versus natural cycle with no stimulation **(8)** short (flare- up) GnRH agonist protocol plus high fixed dose versus a step-down or step up dose of gonadotropin **(9)**, GnRH antagonist plus high dose of gonadotropins versus high gonadotropin dose alone **(10)**. GnRH antagonist/high gonadotrophin versus short GnRH agonist/high gonadotropin **(11; 12; 13; 14; 15;16)**, stop long GnRH agonist / high gonadotropin protocol versus a non-stop GnRH agonist / high gonadotropin protocol. **(17; 18)**, microdose GnRH agonist versus luteal phase GnRH antagonist protocol.**(19-21)**, clomiphene citrate/ gonadotropin /antagonist (mild protocol) and microdose GnRH agonist flare protocols. **(22)** However, no one is clearly superior to the other**.**

Long GnRH agonist is the standard down regulation protocol used for poor responders, however, there is accumulated evidence that this protocol led to prolonged duration of ovarian stimulation, more injections, and patient's distress and increased the cost without improving IVF outcome. **(23)**

Last decade, GnRH antagonist has been emerged as an alternative to GnRH agonist protocols in IVF/ICSI cycles. The main mechanism of action of GnRH antagonists is competitive occupancy of the GnRH-receptor (**24**). Currently available GnRH antagonists include cetrorelix and ganirelix. Both are available as a 0.25-mg preparation for daily injection **(25)**. There are two regimens for Gonadotropin-releasing hormone antagonists, flexible regimen in which GnRH antagonist commences when the mean diameter of the lead follicle is ≥ 14 mm and fixed regimen on stimulation days 5–6.

GnRH antagonist has many advantages over GnRH agonist such as fewer injections, shorter duration of stimulation, less incidence of OHSS. So it has been promised to be more patient friendly than long GnRH agonist in general, however, there is a great controversy about its impact on pregnancy outcomes in poor responders **(26)** . The aim of this subgroup analysis of Cochrane review is to compare GnRH antagonist suppression protocol with the standard long GnRH agonist in women with poor ovarian response underwent IVF/ICSI treatment cycles.

Methods

Search strategy for identification of studies: The following electronic databases were searched: MEDLINE, EMBASE, Science Direct, Cochrane Central Register of Controlled Trials (CENTRAL), Web of Science, the National Research Register (NRR), and the Medical Research Council's Clinical Trials Register.

A search strategy was carried out based on the following terms: long GnRH agonist protocol, GnRH antagonist protocol, IVF, ICSI, and live birth rate, ongoing pregnancy rate and poor ovarian response, low ovarian response AND "randomized controlled trial(s)" OR " randomised controlled trial(s)".

Furthermore, we examined the reference lists of all known primary studies, review articles, citation lists of relevant publications, abstracts of major scientific meetings (e.g. ESHRE and ASRM) and included studies to identify additional relevant citations.

In addition, references from all identified articles were checked, and a hand search of the abstracts from the annual meetings of the American Society for Reproductive Medicine and the European Society for Human Reproduction and Embryology was performed.

The search was not restricted by language. The searches were conducted independently by H.G AND M.A.Y

Study selection and data extraction

Studies were selected if the target population consisted of subfertile couples with poor ovarian response and the therapeutic interventions were GnRH antagonist protocol versus standard long GnRH agonist protocol in IVF or ICSI treatment. Studies had to be of randomized design. The primary outcome measure was ongoing pregnancy rate per randomized woman. Secondary outcomes were clinical pregnancy rate, early miscarriage rate per randomized woman, number of MII oocytes, cycle cancellation, total duration and amounts of used FSH/HMG.

Studies were selected in a two-stage process. First, the titles and abstracts from the electronic searches were scrutinized by two reviewers independently (H.G and M.Y) 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. Any disagreements about inclusion were resolved by consensus or arbitration by a third reviewer. The selected studies were assessed for methodological quality by using the components of study design that are related to internal validity (Juni et al., 2001) **(27)**. Information on the adequacy of randomization, concealment and blinding was extracted. From each study, outcome data were extracted in 2 X 2 tables. Data extraction was performed in duplicate by. H.G and M. A.M.Y

Statistical analysis

Dichotomous outcomes were expressed as an odds ratio (OR) with 95% CI using a fixed effects model, and a random effects model in case of heterogeneity (Mantel and Haenszel, 1959). Heterogeneity of treatment effects was evaluated graphically using forest plots **(28)** and statistically using the Breslow and Day chi-square test. Continuous outcomes were expressed as a weighted mean difference (WMD) with 95% CI using a fixed effects model and a random effects model. All outcomes were calculated according to intention to treat analysis. All statistical analyses were performed using Rev-Man 5.0 (Cochrane Collaboration, Oxford, UK).

Results

Only six randomized controlled studies fulfilled the inclusion criteria. These trials enrolled 919 randomized women. The quality and the main characteristics of the included trials are presented in table 1.

The studies were generally small and not well powered for the clinically relevant outcomes, with sample sizes varying from 54 to 570 women. Five studies were single centre (Cheung et al., 2005; Inza et al., 2004; Marci et al., 2005,Tazegul et al., 2008; Sbarcia et al., 2009) [4; 12; 23; 29; 30], One study was blind (Cheung et al., 2005) [29] and four studies were not blind (Marci et al., 2005; Inza et al., 2004 ; Tazegul et al., 2008, Kim et al., 2009) [12; 30; 23,31]. One study was three arms (Kim et al., 2009) [31] and the other 5 studies were 2 arms.

All trials were published as full text in peer reviewed journals except (Inza et al., 2004) [30]. Four studies used a GnRH antagonist protocol as 0.25 mg sc. cetrorelix (Cetrotide) for down-regulation. One study used both 0.25 mg sc. cetrorelix (Cetrotide) and Ganirelix, 0.25 mg (Organon, The Netherlands) (Tazegul et al., 2008 [23]). Flexible multiple-dose protocol was used in 4 trials and fixed protocol in one study (Cheung et al., 2005) [29].

Three studies have defined poor responders as women who exhibited a poor ovarian response with <3 mature follicles on a long GnRH agonist protocol in their previous IVF cycles (Cheung et al., 2005, Marci et al., 2005; Tazegul et al., 2008) [29; 12; 23], while the other studies used different definition such as women's age > 40 years (Sbarcia et al., 2009) [4], antral follicles count < 5 (Kim et al., 2009)

Outcomes measurements:

- **Pregnancy rates per women randomized:** There was no evidence of a statistically significant difference in ongoing pregnancy rate (3 RCTs; OR: 1.17, 95% CI 0.53 to 2.58; with less important heterogeneity; I^2 = 28 %, p=0.25) and clinical pregnancy rate (6 RCT; OR: 0.71; 95% CI: 0.49-1.02; with less important heterogeneity; I^2 = 31%, p=0.2) between GnRH antagonist and long GnRH agonist.

  Assuming an ongoing pregnancy rate of 18% after long GnRH agonist this means that the corresponding ongoing pregnancy rate after GnRH antagonist would be 14%.

  There was no evidence of a statistically significant difference in early miscarriage between both groups (5 RCTs; OR: 2.50, 95% CI 0.41 - 2.51; with no heterogeneity; P= 0.55, I^2 =0%)

- **Cancellation rate per woman randomized:** There was no evidence of statistically significant difference between both groups as regards the cancellation rate (5 RCTs; OR: 1.02, 95% CI 0.41 - 2.51; with no heterogeneity; P= 0.55, I^2 =0%)

  There was no evidence of statistically significant difference between both groups as regards the number of oocytes retrieved (5 RCTs; MD: -0.21, 95% CI: -0.52 to 0.10). The trials differed in effect size resulting in moderate heterogeneity; P= 0.00, I^2 =71%)

  There was no evidence of statistically significant difference between both groups as regards the number of days of ovarian stimulation (5 RCTs; MD: -1.76, 95% CI: - 2.00 to - 1.52). The trials differed in effect size resulting in considerable heterogeneity; P < 0.00, I^2 =98%)

  There was no evidence of statistically significant difference between both groups as regards the duration of stimulation (5 RCTs; MD: -1.76, 95% CI: - 2.00 to - 1.52). The trials differed in effect size resulting in considerable heterogeneity; P < 0.00, I^2 =98%)

  There was no evidence of statistically significant difference between both groups as regards the total amount of gonadotropins (5 RCTs; MD: -679, 95% CI: - 820 to - 537). The trials differed in effect size resulting in considerable heterogeneity; P < 0.00, I^2 =98%)

DISCUSSION

To the best of our knowledge, this subgroup analysis of Cochrane systematic review and meta-analysis presents the most recent evidence summarising randomized controlled trials comparing GnRH antagonist with long GnRH agonist in women with poor ovarian response undergoing IVF/ICSI. Gonadotropin-releasing hormone antagonists competitively block pituitary GnRH receptors, and induce immediate, reversible suppression of gonadotropin secretion **(32; 33)**.

Due to these pharmacokinetic characteristics it was anticipated that, GnRH antagonists are an optimal alernative to long GnRH agonist because their use occurs after the commencement of gonadotropin stimulation, thus theoretically minimizing their impact on early follicular recruitment (34; 35) and reduces suppression of endogenous gonadotrophins (36). The present subgroup analysis indeed suggests that GnRH antagonist and long GnRH agonist protocol result in comparable pregnancy rates in assisted reproductive cycles.

Previously published literature included only one study **(12)**, due to the limited published RCTs, it was concluded that there is insufficient evidence to identify the use of any one particular intervention to improve treatment outcomes in poor responders in IVF **(1; 2; 37)**. In contrast, our review which included 6 randomized controlled studies; it is obvious that both protocols are similar as regards pregnancy outcomes. However, patients' preferences studies comparing GnRH antagonist with long GnRH agonist are lacking, GnRH antagonist seems to be more patient friendly protocol because it led to shorter duration of stimulation and less consumption of gonadotropins, which might have less psychological impact on patients and more cost-effectiveness than that associated with long GnRH agonist.

As regards the study limitations, the studies were generally small and not well powered for all the clinical relevant outcomes. In five of six randomized trials, concealment of allocation was not clearly described **(30; 31; 12; 23; 4)**.

In three studies there was no blinding **(4; 12; 23)** and in two studies it was unclearly reported **(30; 31)**.

An intention to treat analysis was stated to have been carried out in only one study **(12)**. Consistencies were found among the studies in outcomes such as ongoing, clinical pregnancy rate and cancellation rate and there were Inconsistencies between studies in outcomes such as number of oocytes, duration of stimulation and amount of FSH used. Although the inconsistency of studies 'results in a meta-analysis reduces the confidence of recommendations about treatment, it is an expected due to clinical and methodological diversity between studies such as inclusion criteria for participation and study quality **(38)**, but it cannot be regarded as a major cause of the differences in the results of the studies included in this review.

In conclusion, in view of its equivalence, GnRH antagonist is an alternative for long GnRH agonist in poor responder patients undergoing ovarian stimulation and IVF/ICSI cycles.

References:

1) Kyrou D, Efstratios M. Kolibianakis,Christos A. Venetis, Evangelos G. Papanikolaou, John Bontis, and Basil C. Tarlatzis, How to improve the probability of pregnancy in poor responders undergoing in vitro fertilization: a systematic review and meta-analysis . Fertil and Steril. 2009; 91(3); 749-766, 49
2) Shanbhag S, Aucott L, Bhattacharya S, Hamilton MA, McTavish AR. Interventions for poor responders' to controlled ovarian hyperstimulation (COH) in in-vitro fertilization (IVF). Cochrane Database of Systematic Reviews, Issue 2, 2007
3) Weissman A, Farhi J, Royburt M, Nahum H, Glezerman M, Levran D. Prospective evaluation of two stimulation protocols for low responders who were undergoing in vitro fertilization-embryo transfer. Fertil Steril 2003; 79:886–92.
4) Sbracia M, Colabianchi J, Giallonardo A, Giannini P, Piscitelli C, Morgia F, Montigiani M, Schimberni M .Cetrorelix protocol versus gonadotropin-releasing hormone analog suppression long protocol for superovulation in intracytoplasmic sperm injection patients older than 40; Fertil. Steril. 2009; 91; 5: 1842-1847.

5) D'Amato G, Caroppo E, Pasquadibisceglie A, Carone D, Vitti A, Vizziello GM. A novel protocol of ovulation induction with delayed gonadotrophin-releasing hormone antagonist administration combined with high-dose recombinant follicle-stimulating hormone and clomiphene citrate for poor responders and women over 35 years. Fertil Steril 2004; 81:1572–7.
6) Klinkert ER, Broekmans FJ, Looman CW, Habbema JD, Te Velde ER. Expected poor responders on the basis of an antral follicle count do not benefit from a higher starting dose of gonadotrophins in IVF treatment: a randomized controlled trial. Human Reproduction 2005; 20(3):611–5
7) Berkkanoglu M, Ozgur K. What is the optimum maximal gonadotropin dosage used in micro dose flare-up cycles in poor responders? Fertil Steril. 2009. [Epub ahead of print]
8) Morgia F, Sbracia M, Schimberni M, Giallonardo A, Piscitelli C, Giannini P, et al. A controlled trial of natural cycle versus microdose gonadotropin-releasing hormone analog flare cycles in poor responders undergoing in vitro fertilization. Fertil Steril 2004; 81:1542–7.
9) Cedrin-Durnerin, I., Bstandig, B., Herve, F., Wolf, J. P., Uzan, M. and Hugues, J.-N. A comparative study of high fixed-dose and detrimental-dose regimens of gonadotropins in a minidose gonadotropin releasing hormone agonist ¯are protocol for poor responders. Fertil. Steril 2000, 73, 1055-1056.]
10) Akman A., Erden F., Tosun S., Bayazit N., Aksoy E., and Bahcec M. Addition of GnRH antagonist in cycles of poor responders undergoing IVF. Hum Reprod 2000, 15(10): 2145-2147.
11) Akman MA, Erden HF, Tosun SB, Bayazit N, Aksoy E, BahceciM. Comparison of agonistic flare-up-protocol and antagonistic multiple dose protocol in ovarian stimulation of poor responders: results of a prospective randomized trial. Human Reproduction 2001; 16(5): 868–70.
12) Marci R, Caserta D, Dolo V, Tatone C, Pavan A, Moscarini M. GnRH antagonist in IVF poor-responder patients: results of a randomized trial. Reprod Biomed Online 2005; 11:189–93.
13) Demirol A, Girgin B, Gurgan T. Comparison of Microdose Flare-up and Antagonist Multiple Dose Protocols in Poor Responder Patients. Fertile steril, abstracts of ASRM 2005 .S325
14) Malmusi S, La Marca A, Giulini S, Xella S, Tagliasacchi D, Marsella T, et al. Comparison of a gonadotropin-releasing hormone (GnRH) antagonist and GnRH agonist flare-up regimen in poor responders undergoing ovarian stimulation. Fertil Steril 2005; 84:402–6
15) Mohamed KA, Davies WA, Allsopp J, Lashen H. Agonist "flare-up" versus antagonist in the management of poor responders undergoing in vitro fertilization treatment. Fertil Steril. 2005 Feb; 83(2):331-5
16) Kahraman K, Berker B, Atabekoglu C.S, Sonmezer M, Cetinkaya E, Aytac R. and Satiroglu K. Microdose gonadotropin-releasing hormone agonist flare-up protocol versus multiple dose gonadotropin-releasing hormone antagonist protocol in poor responders undergoing intracytoplasmic sperm injection–embryo transfer cycle. Fertil.Steril. 2009; 91; 6:2437-2444.
17) Dirnfeld M, Fruchter O, Yshai D, Lissak A, Ahdut A, Abramovici H. Cessation of gonadotropin-releasing hormone analogue (GnRH-a) upon down-regulation versus conventional long GnRH-a protocol in poor responders undergoing in vitro fertilization. Fertil Steril 1999; 72: 406–11
18) Garcia-Velasco JA, Isaza V, Requena A, Martinez-Salazar FJ, Landazabal A, Remohi J, et al. High doses of gonadotrophins combined with stop versus non-stop protocol of GnRH analogue administration in low responder IVF patients: a prospective, randomized, controlled trial. Hum Reprod 2000; 15:2292–6.

19) DiLuigi A J, Lawrence Engmann, David W. Schmidt, Claudio A. Benadiva and John C. Nulsen. A randomized trial of microdose leuprolide acetate protocol versus luteal phase ganirelix protocol in predicted poor responders. Fertility and Sterility, 2011.
20) S.M. Shastri, K.D. Schoyer, E.A. Barbieri, I. Kligman, O. Davis, Z. Rosenwaks Stimulation of the young poor responder: our experience with luteal estradiol/GnRH antagonist suppression versus a standard protocol (OCP microdose lupron). Fertil Steril. 2011 Feb;95(2):592-5.
21) Elassar A, Mann JS, Engmann L, Nulsen J, Benadiva C. Luteal phase estradiol versus luteal phase estradiol and antagonist protocol for controlled ovarian stimulation before in vitro fertilization in poor responders. Fertil Steril. 2011 Jan;95(1):324-6.
22) Karimzadeh MA, Mashayekhy M, Mohammadian F, Moghaddam FM. Comparison of mild and microdose GnRH agonist flare protocols on IVF outcome in poor responders. Arch Gynecol Obstet. 2011 Jan 9.
23) Tazegül A, Görkemli H, Ozdemir S, Aktan TM. Comparison of multiple dose GnRH antagonist and minidose long agonist protocols in poor responders undergoing in vitro fertilization: a randomized controlled trial. Archives of Gynecology and Obstetrics 2008;278(5):467-72.
24) Halmos G, Schally AV. Changes in subcellular distribution of pituitary receptors for luteinizing hormone-releasing hormone (LH-RH) after treatment with the LH-RH antagonist cetrorelix. Proc Natl Acad Sci USA 2002; 99:961–5.
25) Griesinger G, Finas D, Alisch A, Roiha K, Schultze-Mosgau A, Schröder AK, Dafopoulos K, Diedrich K, Felberbaum R. FSH time-concentration profiles before and after administration of 0.25 mg cetrorelix in the gnRH-antagonist multiple-dose protocol for ovarian hyperstimulation. J Assist Reprod Genet. 2004 Jul;21(7):279-82.
26) Tarlatsiz B.C, Zepiridis. L, Grimbizis. G and Bontis J, Clinical management of low ovarian response to stimulation for IVF: a systematic review, Hum Reprod Update 2003, pp. 61–76.
27) Jüni P, Altman DG, Egger M. Systematic reviews in health care: Assessing the quality of controlled clinical trials. BMJ. 2001 Jul 7;323(7303):42-6. Review
28) Lewis S, Clarke M. Forest plots: trying to see the wood and the trees. BMJ. 2001 Jun 16;322(7300):1479-80.
29) Cheung LP, Lam PM, Lok IH, Chiu TT, Yeung1 S Y, Tjer C C and Haines CJ. GnRH antagonist versus long GnRH agonist protocol in poor responders undergoing IVF: a randomized controlled trial. Human Reproduction Vol.20, No.3 pp. 616–621, 2005.
30) Diedrich K, Diedrich C, Santos E, Zoll C, al-Hasani S, Reissmann T, et al. Suppression of the endogenous luteinizing hormone surge by the gonadotrophin-releasing hormone antagonist cetrorelix during ovarian stimulation. Hum Reprod 1994; 9:788 –91.
31) Duijkers IJ, Klipping C, Boerrigter PJ, Machielsen CS, De Bie JJ, Voortman G. Single dose pharmacokinetics and effects on follicular growth and serum hormones of a long-acting recombinant FSH preparation (FSH-CTP) in healthy pituitary-suppressed females. Hum Reprod. 2002 Aug;17(8):1987-93.
32) Mahutte NG, Arici A. Role of gonadotropin-releasing hormone antagonists in poor responders. Fertil Steril. 2007 Feb; 87(2):241-9. Epub 2006 Nov 16.
33) Copperman AB. Antagonists in poor-responder patients. Fertil Steril. 2003, 80 (Suppl 1), S16–S24.
34) Nikolettos N, Al-Hasani S, Felberbaum R, Demirel LC, Kupker W et al. Gonadotropin-releasing hormone antagonist protocol: a novel method of ovarian stimulation in poor responders. Eur J Obstet Gynecol Reprod Biol. 2001 Aug;97(2):202-7.

35) Pandian Z, McTavish AR, Aucott L, Hamilton MP, Bhattacharya S. Interventions for 'poor responders' to controlled ovarian hyper stimulation (COH) in in-vitro fertilization (IVF). Cochrane Database Syst Rev. 2010 Jan 20 ;(1):CD004379.

36) Inza R, Van Thillo G, Lombardi E, Bisioli C, Diradourian M, Kenny A. Reproductive performance in second IVF cycles treated with the use of either GnRH antagonists (-antag) vs GnRH agonists (-ag) after failure with long protocols with GnRH agonists: a prospective randomized trial. Fertility and Sterility 2004;82 Suppl:233-4.

37) Kim CH, Jeon GH, Cheon YP, Jeon I, Kim SH, Chae HD, Kang BM. Comparison of GnRH antagonist protocol with or without oral contraceptive pill pretreatment and GnRH agonist low-dose long protocol in low responders undergoing IVF/intracytoplasmic sperm injection. Fertility and Sterility 2009;92(5):1758-60.

38) Higgins JPT Thompson SG, Deeks JJ: Measuring inconsistency in meta-analyses. BMJ. 2003 September 6; 327(7414): 557–560.

Tables:

Table 1. Characteristics of randomized trials of GnRH antagonist versus long GnRH agonist protocol in poor responders

Cheung et al., 2005	**Patients**: 66 infertile women undergoing IVF/ICSI. **Poor responders** were classified as patients who had exhibited a poor ovarian response with <3 mature follicles on a long GnRH agonist protocol in their previous IVF cycles, or those with repeated high basal levels of FSH >10 IU/l. Patients with polycystic ovaries were excluded from the study.	**GnRH antagonist (n= 33):** OCP (Nordette) 30 µg of ethinyl estradiol and 150 µg of levonorgestrel for 21 days + 300 IU daily rFSH (Gonal-F) + 0.25 mg sc. cetrorelix (Cetrotide) **fixed**, multi-dose GnRH antagonist protocol starting on day 6 of the stimulation **(fixed). Long GnRH agonist (n= 33):** long GnRH agonist protocol, buserelin acetate nasal spray (Suprecur) daily dose of 600 µg starting at the mid-luteal phase of the preceding cycle + 300 IU daily rFSH (Gonal-F). **Oocyte maturation triggering**: 10 000 IU of i.m. HCG (Profasi) when the leading follicles reached 18-20 mm together with at least three mature follicles >16 mm. **Oocyte retrieval**: 36 h later. ICSI was performed only in cases with severe male factor or previous fertilization failure. Depending on the number of embryos available, up to three embryos were transferred on day 3 after oocyte retrieval. **Luteal phase support**: i.m. hCG (Profasi) 2000 IU given every 3 days for four doses starting on the day of oocyte retrieval	Duration of stimulation, consumption of gonadotrophins, cycle cancellation rate, the number of mature follicles recruited and total oocytes retrieved. The hormone levels throughout the cycle, laboratory outcomes and clinical pregnancy rates were also reviewed.	**Randomization**: Random-number table (true), Randomisation: 1:1 (cetrorelix: buserlin acetate) ratio. **Concealed**: Yes. **Sample size**: No. **Blindness**: Yes. **ITT**: No. **Funding**: Financial support by the NV Organon, Oss, The Netherlands.
Marci et al., 2005	60 infertile women **(poor responders)** undergoing IVF/ICSI **Inclusion criteria**: estradiol concentrations <600 pg/ml concentration on the day of HCG administration and a poor response (number of oocyte retrieved <3) after a previous standard long protocol using analogues for down regulation and recombinant gonadotrophin at a dose of 225 IU for stimulation (r-FSH, Gonal-F).	**GnRH antagonist (n= 30)**: 375 IU rFSH (Gonal-F) from cd2 + GnRH antagonist cetrorelix 0.25 mg per day was then administered from when the two lead follicles had reached 14 mm diameter, irrespective of the day of the cycle until the day of HCG injection. **(Flexible). GnRH agonist (n= 30)**: by analogues (GnRHa) from day 23 of the cycle (Enantone 3.75 mg) + 375 IU daily , sc, rFSH, (Gonal-F) from day 3 of the next cycle at a dose of. In group B (n = 30), ovarian stimulation started at day 2 with rFSH at a dose of 375 IU (Gonal-F). **Oocyte maturation triggering**: hCG (Profasi; Serono) 10,000 IU was administered intramuscularly (IM) 24 h after the last r-FSH injection when at least two follicles had reached a diameter of 17 mm. **Oocyte retrieval**: 36 h after HCG administration followed by IVF/ICSI. **Embryo transfers**: were performed 48 h after oocyte retrieval. **Luteal phase**: 2 × 200 mg/day of micronized vaginal progesterone (Prometrium). **Follow up**: Serum HCG concentrations were measured 14 days after embryo transfer. Clinical pregnancies were confirmed 28-35 days after embryo transfer by the presence of a gestational sac under ultrasound.	Cycles with oocyte retrieval, stimulation duration (days), number of ampoules, follicles >15 mm, oocytes retrieved, oocytes fertilized, cycles with transfers, embryos transferred, endometrial thickness (mm), clinical pregnancies	**Randomization**: yes but method of randomization is not reported. **Concealed**: unclear. **Sample size**: yes. **Blindness**: NO. **ITT**: unclear **Funding**: not reported

Table 1. (Cont.)	Characteristics of randomized trials of GnRH antagonist versus long GnRH agonist protocol in poor responders			
Sbarcia et al., 2009	564 **low responders**, undergoing their first IVF cycle were eligible for the study. **Inclusion criteria:** age 40 years or older and no previous IVF cycle, and the exclusion criteria were FSH >10 IU/ mL, a previous IVF cycle, and age 45 years or older. **Exclusion criteria:** PCOS. **Baseline characteristics:** Maternal age, years 42.3 1.4 vs 42.1 1.5, Body mass index 25.1 2.6 vs 24.8 2.4, Basal FSH levels, IU/L 7.0 2.5 vs 6.9 2.4	**Group A (n= 285)**: 300 IU/day rhFSH (Gonal-F) + 0.25 GnRH antagonist (Cetrotide) when the leading follicle ≃ 14 mm or the E2 plasma levels were 600 pg/mL (**flexible multiple-dose protocol**). **Group C (n= 285)**: buserelin 0.4 mg/day long GnRH agonist + 225 IU/day rhFSH (Gonal-F) (**GnRH agonist protocol**). **Oocyte maturation triggering**: 10,000 IU of IM hCG when plasma E2 between 800 and 3500 pg/mL and at least three follicles >16 mm in mean diameter **Oocyte retrieval**: 36 hours later, followed by ICSI. **Maximum of embryo transferred**: 3. **Luteal phase support**: 50 mg daily of P (Prontogest) IM from the day of replacement. **Follow up**: pregnancies were confirmed by a rising titer of serum b-hCG 12 days after ET and ultrasound demonstration of the gestation sac 4 weeks after the transfe	**Primary outcomes**: clinical pregnancy rate per cycle started and per transfer. **Secondary outcomes**: days of stimulation, E2 at the day of hCG, amount of FSH administered, number of oocytes yielded, number of embryos transferred, implantation rate, and abortion rate	**Randomization**: Computer based. **Concealed**: unclear. **Sample size**: yes. **Blindness**: unclear. **ITT**: no. **Funding**: no
Kim et al., 2009	82 **low responders**, aged 28 to 41 years, who were defined as patients with repeated day 3 levels of FSH >8.5 mIU/mL, and/or antral follicle count < 5 and were eligible to undergo IVF/ICSI. **Baseline characteristics**: There were no significant differences in average age, body mass index, proportion of patients with high basal FSH or small number of basal antral follicle, and basal endocrine profile among three groups (data not given)	**Group A (n=27)**: ethinyl estradiol 0.03 mg and levonorgestrel 0.15 (21 days) + 225 IU/day rhFSH (Gonal-F) + 0.25 GnRH antagonist (Cetrotide) when the leading follicle ≃ 14 mm (**flexible multiple-dose protocol**). **Group B (n=27)**: 225 IU/day rhFSH (Gonal-F) + 0.25 GnRH antagonist (Cetrotide) when the leading follicle ≃ 14 mm (**multiple-dose protocol**). **Group C (n= 28)**: Decapeptyl 0.1 mg GnRH agonist luteal low-dose long protocol, The dose of GnRHa was then reduced to 0.05 mg/day + 225 IU/day rhFSH (Gonal-F)(**Low dose GnRH agonist protocol**). **Oocyte maturation triggering**: rhCG (Ovidrel) of 250 mg, when one or more follicles ≥ 18 mm. **Oocyte retrieval**: 35-36 hours later, followed by IVF/ ICSI. **Maximum of embryo transferred**: 3. **Luteal phase support**: intravaginal progesterone gel (Crinone 8%). **Follow up**: up to live birth	Total dose of rhFSH (IU), Days of rhFSH administration, No. of follicles R14 mm on hCG day, Endometrial thickness on hCG day (mm), No. of cycle with premature LH surge No. of cycles with ICSI No. of oocytes retrieved No. of mature oocytes No. of fertilized oocytes No. of grade I, II embryos No. of embryos frozen No. of embryos transferred Clinical pregnancy rate per cycle (%) Implantation rate (%) Miscarriage rate per clinical pregnancy (%) Live birth rate per cycle (%) Twin pregnancy rate per clinical pregnancy (%)	**Randomization**: Computer based. **Concealed**: unclear. **Sample size**: yes. **Blindness**: unclear. **ITT**: no. **Funding**: no
Tazegul et al., 2008	96 poor responders who underwent ICSI-ET cycles. **Inclusion criteria**: baseline follicle stimulating hormone (FSH) < 13 m IU/ml, estradiol level on the day of human chorionic gonadotropin (HCG) injection < 500 pg/ ml and a poor response (failure in obtaining of at least three follicles > 16 mm in diameter and the number of mature oocytes retrieved less than four) after a previous ovarian stimulation cycle. **Exclusion criteria** were: presence of a clinically significant systemic disease; diabetes mellitus; polycystic ovaries or any other endocrine disorder; submucosal polyp, myoma or uterine septum which were detected on hysteroscopy or hysterosalpingography. Intracytoplasmic sperm injection and assisted hatching were performed in all cycles. **Baseline characteristics**: Age (years) 38.3 ± 4.23 vs 37.9 ± 74.87. Baseline FSH (IU/mL) 6.31 ± 2.19 vs 6.27 ± 2.82	**GnRH antagonist (n= 48)**: 300 IU r-FSH and hMG starting on the second day of menstruation for 6 days (adjusted) + 0.25 mg of cetrorelix (Cetrotide) or 0.25 mg ganirelix (Orgalutran) were administered subcutaneously per day when the leading follicle reached 14 mm in diameter until the hCG injection. (Flexible). **GnRH agonist (n= 48)**: 1 mg/ day leuprolide acetate (Lucrin) started on the 21st day prior to menstruation for pituitary desensitization. When exogenous gonadotropins were started on day 2 of menstruation, the dose of leuprolide acetate was decreased to 0.5 mg/day + 300 IU rFSH and hMG starting on the second day of menstruation for 6 days (adjusted). **Oocyte maturation triggering**: When the leading follicle reached 18 mm in diameter or at least two follicles were >17 mm in diameter, a total of 10,000 units of hCG were administered intramuscularly. **Oocyte retrieval**: was performed 35–37 h later. **Embryos transfer**: day 2–3. **luteal phase support**: micronized vaginal progesterone, 600 mg/day, until the tenth week of gestation in cases where a pregnancy was achieved. **Follow up:** Clinical pregnancy was confirmed 28–35 days after embryo transfer by a gestational sac under ultrasound. Ongoing pregnancy was defined as fetal heart beat at 10–12 weeks of gestation. Early pregnancy loss was defined as the proportion of patients with initially positive HCG in whom pregnancy failed to develop before 12 weeks of gestation.	Clinical and ongoing pregnancy per randomized patient, the duration of stimulation, consumption of gonadotrophins, cycle cancellation rate, the number of oocytes retrieved and embryos transferred. The hormone levels throughout the cycle	**Randomization**: Computer based. **Concealed**: no. **Sample size**: yes. **Blindness**: NO. **ITT**: no. **Funding**: not reported
Inza et al., 2004	Patients < 40, with FSH levels on day 3 < 12 IU/ml	GnRH agonist long protocol versus GnRH antagonist protocol (type of antagonist protocol: N/A) (unknown)	number and quality of retrieved oocytes, amount of gonadotropins used days of stimulation, final estradiol levels fertilization rate, number and quality of embryos transferred pregnancy rate implantation rate	**Randomization**: yes but method of randomization is not reported. **Concealed**: unclear. **Sample size**: yes. **Blindness**: NO. **ITT**: no. **Funding**: not reported

Figures:

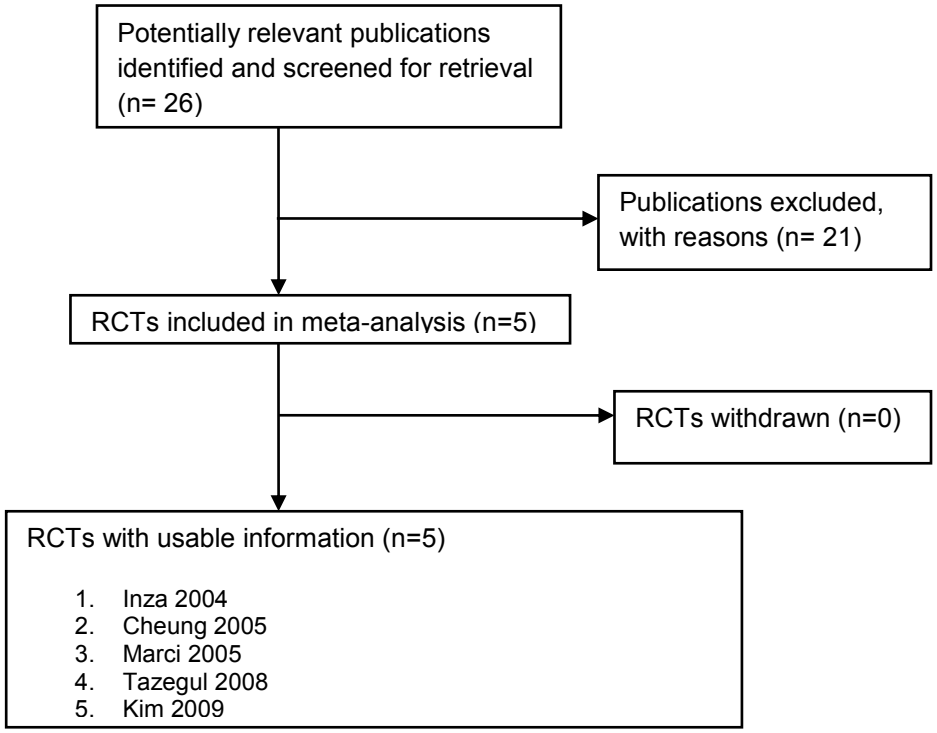

Figure 1. Flow diagram for meta-analysis. Identification and selection of publications

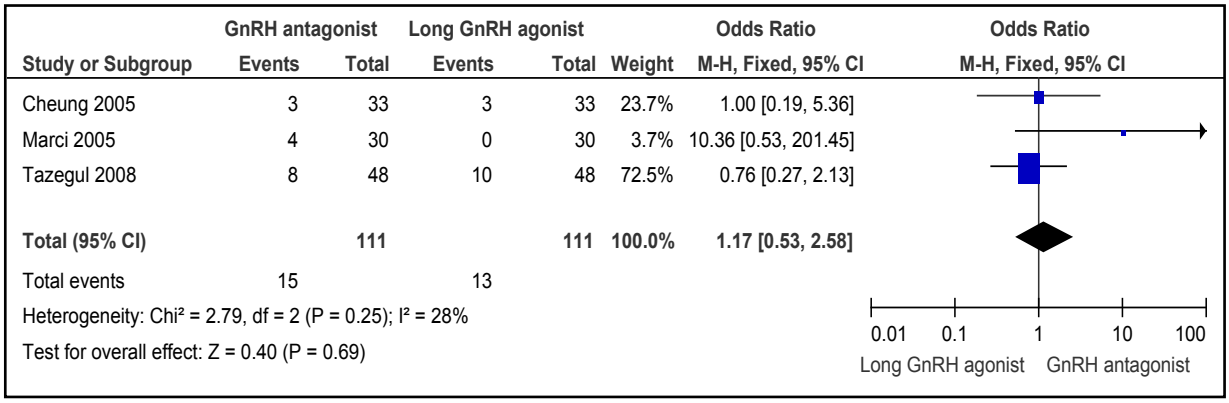

Figure 2. The study specific and pooled OR for ongoing pregnancy outcome per woman randomized

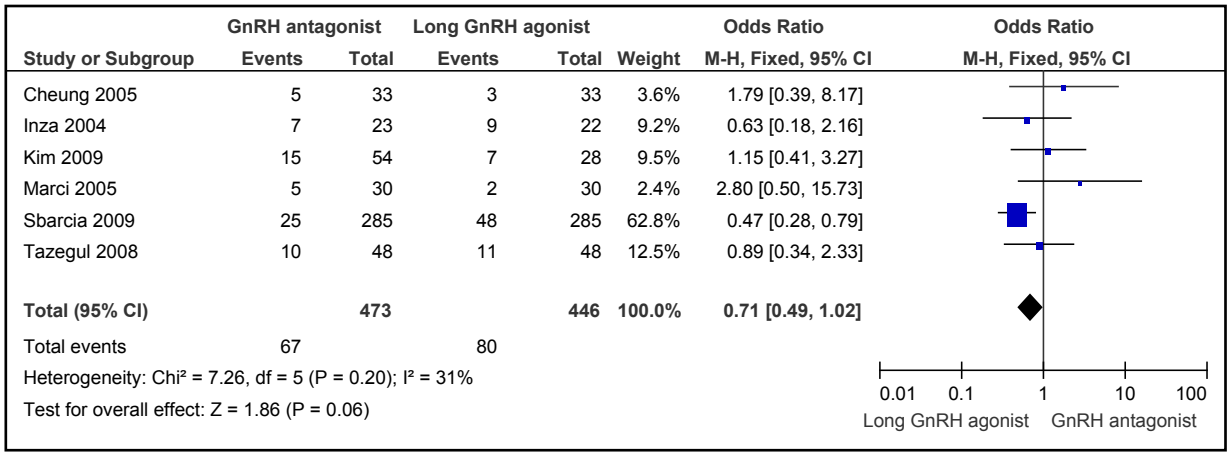

Figure 3. The study specific and pooled OR for clinical pregnancy outcome per woman randomized

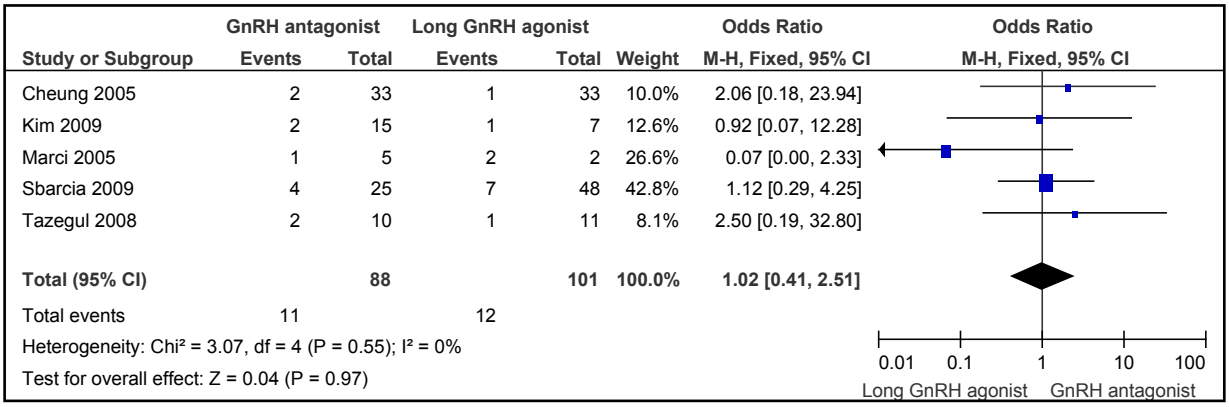

Figure 4. The study specific and pooled OR for cancellation rate outcome per woman randomized

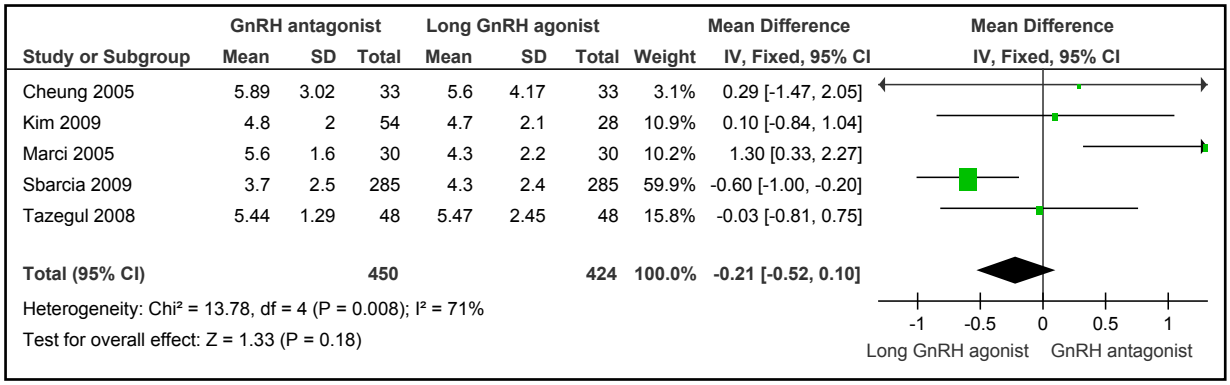

Figure 5. The study specific and pooled MD for number of retrieved oocytes outcome per woman randomized

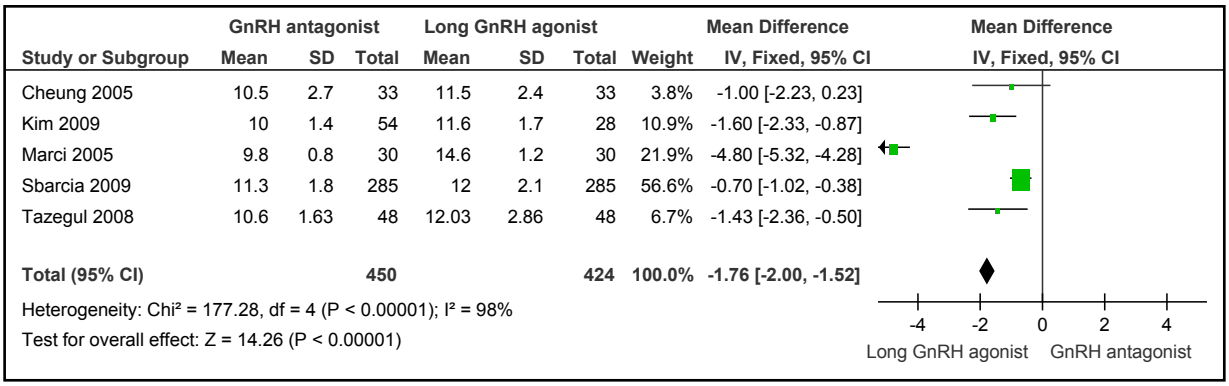

Figure 6. The study specific and pooled MD for duration of ovarian stimulation outcome per woman randomized

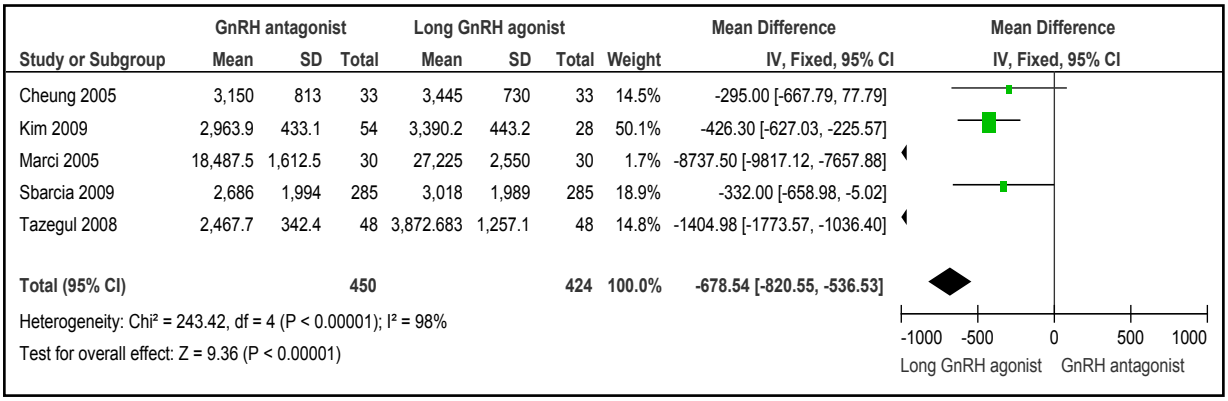

Figure 7. The study specific and pooled MD for amount of FSH outcome per woman randomized