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ORIGINAL ARTICLE *Reproductive endocrinology*

The non-ergot derived dopamine agonist quinagolide in prevention of early ovarian hyperstimulation syndrome in IVF patients: a randomized, double-blind, placebo-controlled trial†

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BACKGROUND: Ovarian hyperstimulation syndrome (OHSS) seems to be induced by the ovarian release of vascular endothelial growth factor (VEGF), which increases vascular permeability. Dopamine agonists inhibit VEGF receptor phosphorylation and thereby decrease vascular permeability.

METHODS: A randomized, double-blind, placebo-controlled, multicentre study assessing three oral doses (50, 100, 200 μg/day) of the non-ergot derived dopamine agonist quinagolide started on the day of human chorionic gonadotrophin (hCG) and continued for 17–21 days without dose-titration in comparison to placebo in preventing moderate/severe early OHSS (onset ≤9 days after hCG administration) in 182 IVF patients with ≥20 but less than 30 follicles ≥10 mm.

RESULTS: The incidence of moderate/severe early OHSS was 23% (12/53) in the placebo group and 12% (6/51), 13% (7/52) and 4% (1/26) in the quinagolide 50, 100 and 200 μg/day groups, respectively. The moderate/severe early OHSS rate was significantly lower with all quinagolide groups combined compared with placebo [$P = 0.019$; OR = 0.28 (0.09–0.81)]. The incidence of ultrasound evidence of ascites among patients with no clinical pregnancy was significantly reduced from 31% (8/26) with placebo to 11% (8/70) with all quinagolide groups combined [$P = 0.033$; OR = 0.29 (0.10–0.88)], although there was no difference for those with clinical pregnancy. Quinagolide did not have a detrimental effect on pregnancy or live birth rates. The incidence of gastrointestinal and central nervous system adverse events increased with increasing doses of quinagolide.

CONCLUSIONS: Quinagolide appears to prevent moderate/severe early OHSS while not affecting treatment outcome. The effect is more marked in patients who did not achieve a clinical pregnancy. Quinagolide administered in high doses without dose-titration is associated with poor tolerability.

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Key words: randomized controlled trial / OHSS prevention / dopamine agonists / quinagolide

† These data were presented in part at the Annual ESHRE Meeting 2008 (Barcelona, Spain).

Introduction

Ovarian hyperstimulation syndrome (OHSS) is the most concerning complication of controlled ovarian hyperstimulation (COH) for assisted reproduction techniques (ART). It is a broad spectrum of signs and symptoms that include abdominal distention and discomfort, enlarged ovaries, ascites and other complications of enhanced vascular permeability (Golan *et al*., 1989; Whelan and Vlahos, 2000). The incidence of OHSS reported in the literature varies depending on the data collection method (registries, clinical trials), the population studied and the interpretation of the definitions/classifications used; however, it is estimated to be around 6–12%, with the presentation of severe cases being about 2–4% (Out *et al*., 1995; Enskog *et al*., 1999; Devroey *et al*., 2009). There are two forms of OHSS, the early with onset within 9 days after human chorionic gonadotrophin (hCG) administration, and the late onset which is believed to be the consequence of the hCG released by the trophoblast starting ~2 days after hCG administration (Mathur *et al*., 2000; Papanikolaou *et al*., 2005). The latter form is clinically more difficult to manage (Lyons *et al*., 1994).

The pathophysiology of early OHSS remains unknown but it is clear that its appearance is mediated by the administration of hCG in COH cycles. hCG administration leads to vascular permeability enhancement resulting in loss of fluid to the third space and the full blown syndrome as a consequence. Vascular endothelial growth factor (VEGF) has emerged as the main angiogenic factor responsible for increased vascular permeability (Rizk *et al*., 1997; Busso *et al*., 2009). The relevance of VEGF in increased vascular permeability has been demonstrated using an OHSS animal model (Gómez *et al*., 2002, 2003, 2006). In these assays, it was shown that: (i) vascular permeability is increased after hCG administration in superovulated animals; (ii) there is a strong correlation between ovarian mRNA VEGF expression and vascular permeability (also proving the ovarian origin of VEGF); (iii) there is an increase of VEGF receptor-2 (VEGFR-2) expression in the ovaries coincidental in time with maximal vascular permeability, demonstrating the involvement of VEGF–VEGFR-2 system in OHSS; (iv) SU5416, a VEGFR-2 inhibitor, can reverse hCG action on vascular permeability, providing new insights into the development of strategies to prevent and treat the syndrome based on its pathophysiological mechanism; (v) the dopamine agonist cabergoline can reverse VEGFR-2 dependent increased vascular permeability (Gómez *et al*., 2002, 2003, 2006).

The concept has been tested in humans and it was observed that cabergoline can significantly reduce the incidence of moderate OHSS as well as pelvic fluid accumulation and hemoconcentration when compared with placebo in oocyte donors at risk of OHSS (Alvarez *et al*., 2007b). Implantation and ongoing pregnancy/live birth rates of IVF patients at risk of OHSS appear to not be affected by cabergoline administration (Alvarez *et al*., 2007a; Carizza *et al*., 2008), although a randomized, double-blind, placebo-controlled study in which a dopamine agonist has been used to prevent OHSS in women undergoing ART has not been performed yet. Cabergoline, however, has been associated with valvular heart disease when administered chronically in patients with Parkinson's disease (Antonini and Poewe, 2007). Although the prevalence of clinically relevant valvular heart disease did not appear increased when cabergoline is given at lower doses to patients with prolactinomas, such long-term use has also been associated with valvular fibrotic changes (Kars *et al*., 2008; Martin *et al*., 2009).

Quinagolide is a non-ergot-derived dopamine D2 receptor agonist, with lack of effect on the serotonin (5-hydroxytryptamine [5-HT]) receptor subtype 5-HT_{2b} at relevant concentrations. This may be clinically important as stimulation of 5-HT_{2b} receptors in cardiac valve tissue may lead to proliferation of fibroblasts (Roth, 2007). The use of non-ergot-derived dopamine agonist in patients with Parkinson's disease does not seem to be associated with an increased prevalence of heart complications (Zanettini *et al*., 2007). Dopamine agonists are also differentiated by their pharmacokinetic profile, as indicated by a much shorter half-life of quinagolide (~17 h) compared with cabergoline (~63–69 h), thus minimizing exposure during organogenesis when used in an IVF setting. These two features make quinagolide an interesting dopamine agonist for use in prevention of OHSS. The present investigation is the first randomized, double-blind study to evaluate the efficacy of ~3 weeks treatment with three different doses of quinagolide versus placebo in preventing the development of OHSS in patients undergoing COH for ART.

Materials and Methods

This was a randomized, double-blind, parallel groups, dose-finding, placebo-controlled, multicentre study. It was a proof of concept study assessing the effect of quinagolide in preventing moderate/severe early OHSS. The primary objective of the study was to identify the effective doses of quinagolide, including the lowest effective dose, compared with placebo in preventing moderate/severe early OHSS.

The trial was conducted at seven IVI clinics in Spain (Barcelona, Bilbao, Madrid, Murcia, Sevilla, Valencia and Vigo) during 2006 and 2007, with the follow-up period covering collection of pregnancy outcome extending into 2008. The trial was conducted in accordance with the Declaration of Helsinki and Good Clinical Practice, and the required approvals from the health authorities and ethics committees were obtained. All participants gave written, signed, informed consent prior to entering the trial.

Women included in this study were 21–37 years old, had a BMI of 18–29 kg/m^2 and early follicular phase serum levels of FSH within normal limits (1–12 IU/l) as well as a uterus consistent with expected normal function and presence of both ovaries. They had been infertile for at least 1 year, and were currently undergoing a COH cycle for IVF/ICSI in which they presented with ovarian response suggestive of being at risk of developing OHSS because of the presence of at least 20 follicles of 10 mm or greater on the day of hCG administration. Patients at very high risk of OHSS, presenting with more than 30 follicles and/or serum E2 of ≥6000 pg/ml (local laboratory) were not included in this study as cycle cancellation in such patients would be appropriate. If coasting or other preventive measures for managing OHSS had been applied, the patient could not enter the trial. Women with any clinically significant systemic disease, endocrine or metabolic abnormalities or history of recurrent miscarriage were excluded from participation. Also, women with hypertension, hypotension, orthostatic hypotension, recurrent syncope or any contra-indication to the use of gonadotrophins or dopamine agonists were prohibited from inclusion. Screening could be initiated at any time point during the COH cycle, but the last checks for eligibility could only be made on the day of hCG administration (day of randomization).

Subjects were randomized in a 1:1:1:1 fashion to one of the following four treatment groups: quinagolide 50 μg/day, quinagolide 100 μg/day, quinagolide 200 μg/day or placebo. A computer-generated randomization list was prepared for each center by a statistician not involved in the trial, and based on this the clinics were provided with individual code envelopes that were sealed in order to conceal the treatment group allocation. The block size was not disclosed during the study. At the time of

randomization, the investigator assigned the subject to the lowest available number and she was thereby allocated to the corresponding treatment group. The study was double-blind; subjects as well as staff at clinics and trial sponsor were blinded to treatment allocation.

Treatment consisted of four tablets (combination of indistinguishable quinagolide 50 μg and placebo tablets) to be taken orally once daily. On the first day of dosing, the tablets were taken at least 2 h before the hCG injection; while on subsequent days administration occurred at bedtime. The dose-titration approach used for quinagolide in patients with hyperprolactinemia was not applied given the acute onset of OHSS after hCG administration. Treatment was continued until the day before the serum hCG test which took place 17 ± 2 days after oocyte retrieval (OR). All subjects received a single dose of hCG (Ovitrelle 250 μg SC, Merck-Serono, Geneva, Switzerland) to trigger final maturation when ≥2 follicles of ≥18 mm were observed. OR took place 36 ± 2 h after hCG administration and transfer of 1–2 embryos was done on Day 3 or Day 5–6 after OR. Luteal support was provided with vaginal progesterone (Utrogestan, SEID Laboratories, Barcelona, Spain), 200 mg twice daily from the day after OR to negative βhCG test or to the day of clinical pregnancy assessment. Clinical pregnancy was defined as at least one intrauterine gestational sac with fetal heart beat documented by transvaginal ultrasound ~5 weeks after embryo transfer (Fig. 1). As post-study follow-up, information on ongoing pregnancy (at least one intrauterine viable fetus documented by transvaginal ultrasound 10–11 weeks after embryo transfer) and pregnancy outcome (live birth and neonatal safety) was collected for all subjects with clinical pregnancy at the end of the study. Furthermore, the post-study follow-up also covered patients who discontinued from the trial after exposure to trial medication and for whom pregnancy data were available (hereafter 'outside of trial').

Evaluation of early OHSS signs/symptoms was performed on the day of OR, Day OR +3 and Day OR +6, corresponding to the first 9 days after hCG administration. Evaluation of late OHSS signs/symptoms, i.e. onset more than 9 days after hCG administration, was performed on Day OR +10 (± 2) as well as on the day of serum βhCG test and the day of clinical pregnancy assessment. Transvaginal ultrasound to measure pelvic fluid pockets, body measurements and blood samples were taken at each visit. All individual signs/symptoms of OHSS were assessed and OHSS was classified as moderate or severe using Golan's classification system (Golan *et al.*, 1989) with certain modifications and specifications. Moderate OHSS was defined as abdominal distension and discomfort, enlarged ovaries and ultrasound evidence of ascites. Nausea, vomiting and diarrhoea were not included in the interpretation of moderate OHSS, as these symptoms are also among the undesirable effects of dopamine agonists and therefore could not with certainty be attributed to OHSS; thus being less appropriate parameters for evaluation of OHSS incidence. Ultrasound evidence of ascites was quantified by measuring the size of the fluid pockets in the pelvis. This parameter was first analyzed using peritoneal fluid volume ≥9 cm^2 based on data from a study in oocyte donors (Alvarez *et al.*, 2007b), but this arbitrarily selected cut-off did not discriminate among groups in IVF/ICSI patients with subsequent embryo transfer and potential pregnancy and a cut-off of ≥24 cm^2 was used for the final analysis. Severe OHSS was defined as features of moderate OHSS plus clinical evidence of ascites and/or hydrothorax or breathing difficulties; and/or by the presence of change in blood volume, increased blood viscosity due to haemoconcentration, coagulation abnormalities and diminished renal perfusion and function. Finally, it was specified that the symptoms of OHSS did not have to be present at the same visit, however, all symptoms were to be present in the period of early OHSS (≤9 days after hCG administration) or the period of late OHSS (>9 days after hCG administration).

The primary end-point was the percentage of subjects with a moderate/severe early OHSS (onset ≤9 days after hCG administration). The sample size calculation assumed comparison of two binomial proportions using a two-sided significance level (α) of 0.05 and a power of 80%. On the basis of interim data from a study in oocyte donors at risk of OHSS (Alvarez *et al.*, 2007b), in which moderate/severe OHSS was reported for 24% (7/29) and 52% (13/25) in the dopamine agonist and placebo groups, respectively, 50 subjects per group were required for the present trial to show a significant difference between the highest dose of quinagolide and placebo. Test for overall treatment effect and comparison between each dose of quinagolide versus placebo were based on the likelihood ratio test (in the full model) with a significance level of 0.05. Prespecified analyses were also conducted for all quinagolide dose levels combined versus placebo. In addition to the primary end-point, treatment comparisons were also made for secondary trial end-points which included the proportion of patients with ultrasound evidence of ascites, peritoneal fluid volume, the proportion of patients with moderate/severe late OHSS, clinical pregnancy rate and adverse event frequency. The main analysis of treatment effect was adjusted, estimated using a logistic regression analysis including the factors 'treatment group' and 'centre' and moderate/severe early OHSS as the response. Treatment differences were presented with odds ratios and two-sided 95% confidence interval and corresponding *P*-values. Furthermore, to assess dose-linearity a Cochrane–Armitage linear trend test was performed. The statistical

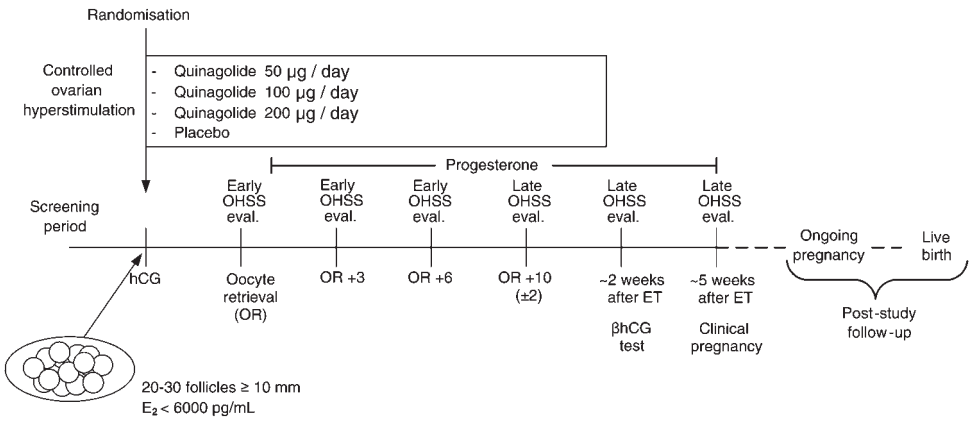

Figure 1 Trial schedule and overview.

package from SAS version 9.1 was used. No adjustment for multiplicity to correct for repeated testing was applied to the primary end-point due to the hierarchical testing strategy or to the secondary variables. The analyses are based on the intention-to-treat (ITT) population, i.e. all randomized, unless otherwise indicated. On the basis of the trial findings, subgroup analyses of efficacy parameters were conducted according to pregnancy status, i.e. whether the patient had a clinical pregnancy in the trial cycle or not. Pregnancy and live birth data were analyzed for the population consisting of patients who had undergone embryo transfer.

During the trial, a safety committee monitored the overall frequency of adverse events while remaining blinded to treatment allocation. When noticing a potential safety signal in the form of a high total frequency of adverse events, an independent statistician was requested to perform an unplanned interim analysis. The analysis and the dissemination of results were handled in a manner that maintained blinding to the individual treatment allocation of subjects, investigators and other personnel at the clinic as well as Sponsor staff. Thus, the double-blind design of the trial remained intact.

Results

A total of 182 IVF/ICSI patients were randomized to the trial, with the following distribution to the four treatment groups: 53 (29%) were randomized to placebo, 51 (28%) to quinagolide 50 μg/day, 52 (29%) to quinagolide 100 μg/day and 26 (14%) to quinagolide 200 μg/day. Randomization to the quinagolide 200 μg/day group was stopped after inclusion of 97 subjects into the trial due to poor tolerability within the first 1–3 days of treatment with quinagolide 200 μg/day when administered without dose-titration. The distribution of patients to the four treatment groups was balanced within each center. Table I summarizes the main demographics and baseline characteristics of the trial population. There were no statistically significant differences between treatment groups with respect to age, body weight or infertility reason. The treatment groups were also comparable with respect to serum E2 and number of follicles on the day of hCG administration as well as number of oocytes retrieved. The mean number of follicles ≥10 mm ranged from 23.2 to 25.0 in the four treatment groups, and an average of 21.0 oocytes were retrieved (Table I). The flow of participants is illustrated in Fig. 2, which depicts the number of patients attending each trial visit.

The serum prolactin concentrations over time are illustrated in Fig. 3. A rapid and dose-dependent reduction in serum prolactin levels was observed with quinagolide. On the day of OR (before administration of the third dose of quinagolide), the mean prolactin concentration was 30.7 ng/ml in the placebo group, compared with 10.8, 5.4 and 2.9 ng/ml in the quinagolide 50, 100 and 200 μg/day groups, respectively (Fig. 3).

Moderate/severe early OHSS occurred at the following frequencies: 23% (12/53) for placebo, 12% (6/51) for quinagolide 50 μg/day, 13% (7/52) for quinagolide 100 μg/day and 4% (1/26) for quinagolide 200 μg/day (Table II). When combining the three dose levels, quinagolide was associated with a significant [$P = 0.019$; OR = 0.28 (0.09–0.81)] decrease in the frequency of moderate/severe early OHSS. Concerning the individual dose levels of quinagolide, the 200 μg/day group had a significantly [$P = 0.046$; OR = 0.11 (0.01–0.96)] lower proportion of patients with moderate/severe early OHSS compared with placebo. The 12 and 13% rates of moderate/severe early OHSS with the lower doses of 50 and 100 μg/day were not significantly different from the 23% rate observed with placebo [$P = 0.142$; OR = 0.43 (0.14–1.32)

Table I Demographics and baseline characteristics.

	Placebo	Quinagolide			Total
		50 μg/day	100 μg/day	200 μg/day	
Subjects randomized (ITT)	$n = 53$	$n = 51$	$n = 52$	$n = 26$	$n = 182$
Age (years)	30.9 ± 2.7 [30.2; 31.6]	31.7 ± 2.6 [31.0; 32.5]	32.0 ± 3.3 [31.1; 32.9]	31.8 ± 3.8 [30.3; 33.3]	31.6 ± 3.0 [31.1; 32.0]
Weight (kg)	60.6 ± 7.7 [58.8; 62.6]	63.8 ± 9.9 [61.1; 66.5]	60.5 ± 8.8 [58.1; 62.9]	61.0 ± 7.4 [58.2; 63.9]	61.5 ± 8.7 [60.3; 62.8]
BMI (kg/m^2)	22.8 ± 2.6 [22.1; 23.5]	23.7 ± 3.2 [22.9; 24.6]	23.0 ± 2.9 [22.2; 23.8]	23.2 ± 2.5 [22.2; 24.1]	23.2 ± 2.9 [22.8; 23.6]
Follicles ≥10 mm, day of hCG	23.7 ± 3.2 [22.8; 24.5]	23.8 ± 2.8 [23.0; 24.6]	25.0 ± 4.0 [23.9; 26.1]	23.2 ± 2.7 [22.1; 24.2]	24.0 ± 3.3 [23.5; 24.5]
Estradiol (pg/ml), day of hCG	2975 ± 1221 [2647; 3304]	3068 ± 1140 [2755; 3381]	3262 ± 1079 [2969; 3556]	2741 ± 967 [2369; 3112]	3050 ± 1128 [2889; 3214]
Oocytes retrieved	21.4 ± 8.7 [19.1; 23.8]	19.5 ± 7.0 [17.6; 21.5]	22.4 ± 7.8 [20.1; 24.7]	20.1 ± 6.0 [17.4; 22.7]	21.0 ± 7.7 [19.8; 22.1]

Data are mean ± standard deviation [95% confidence interval].

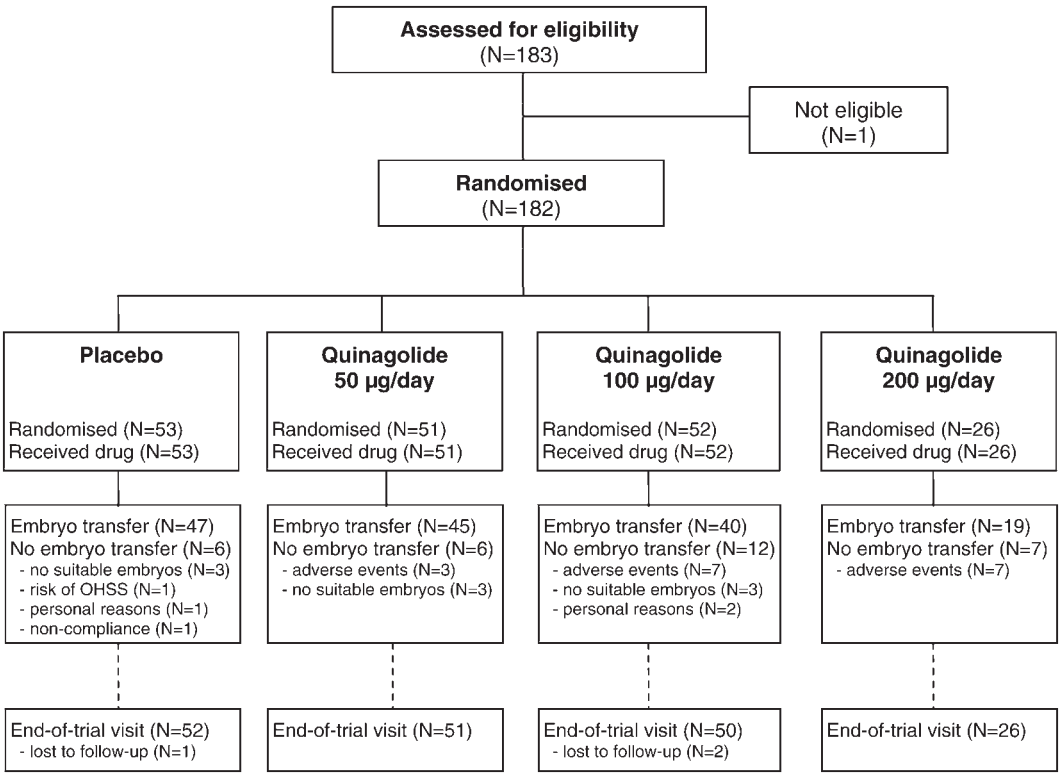

Figure 2 Disposition of patients by trial visit.
N indicates the number of patients attending the trial visits/undergoing the trial procedures. Patients who stopped trial medication prematurely were discontinued from the trial, but may have undergone procedures related to the controlled ovarian hyperstimulation cycle (e.g. OR and embryo transfer) outside the trial; these patients are not included in Fig 2. End-of-trial visits were scheduled both for patients who completed the trial and patients who discontinued early (three patients were lost to follow-up after negative βhCG test).

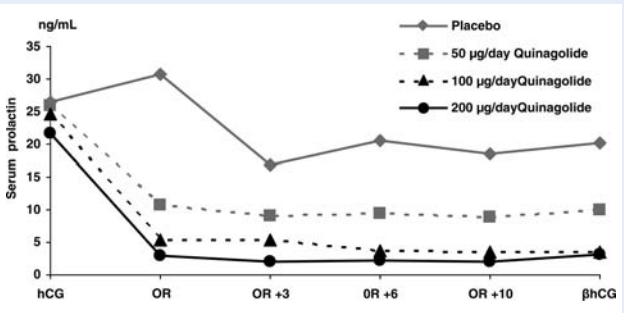

Figure 3 Mean serum prolactin concentrations (ng/ml) during the trial.

and $P = 0.161$; OR = 0.45 (0.15–1.37)], respectively. The combination of the quinagolide 100 and 200 μg/day groups yielded a frequency of moderate/severe early OHSS of 10% (8/78) [$P = 0.026$; OR = 0.22 (0.06–0.83)]; the corresponding rate for the combination of the 50 and 100 μg/day dose levels was 13% (13/103) [$P = 0.086$; OR = 0.44 (0.17–1.12)].

The effect of quinagolide appeared to be consistent among patients following a GnRH agonist ($n = 148$) or GnRH antagonist ($n = 34$) down-regulation protocol with reductions in moderate/severe early OHSS rate from 20–33% in the placebo group to 0–6% in the quinagolide 200 μg/day group.

Early OHSS Grade 4 or 5 occurred for 6% (3/53) of the subjects in the placebo group and 2% (1/51) in the quinagolide 50 μg/day group, and none of the subjects in the 100 and 200 μg/day groups experienced severe early OHSS (Table II). The incidence of patients with ultrasound evidence of ascites within the initial 9 days after hCG was significantly [$P = 0.027$; OR = 0.09 (0.01–0.77)] reduced from 28% (15/53) in the placebo group to 4% (1/26) in the quinagolide 200 μg/day group.

Clinical pregnancy was found to be a statistically significant factor ($P = 0.044$) in the logistic regression model analyzing moderate/severe early OHSS, and the results are therefore also presented separately for subjects who achieved a clinical pregnancy in the study cycle and those who did not. The incidence of moderate/severe early OHSS among subjects who did not obtain a clinical pregnancy was significantly reduced from 23% (6/26) with placebo to 4% (3/70) with all groups of quinagolide combined [$P = 0.011$; OR = 0.15 (0.03–0.65)] (Fig. 4a). In patients who had a documented clinical pregnancy in the study cycle the incidence of moderate/severe early OHSS was not significantly different among any of the study groups (individually or combined; Fig. 4b). With respect to ultrasound evidence of ascites within the initial 9 days after hCG administration, quinagolide was able to reduce the incidence of this OHSS sign among the patients who did

Table II Overview of main end-points.

	Placebo	Quinagolide			P^a odds ratio [95% confidence interval]
		50 μg/day	100 μg/day	200 μg/day	
Subjects randomised (ITT)	$n = 53$	$n = 51$	$n = 52$	$n = 26$	
OHSS and symptoms					
Moderate/severe early OHSS	23% (12)	12% (6)	13% (7)	4% (1)	$P = 0.019$/OR = 0.28 [0.09; 0.81]
Severe early OHSS, Grade 4/5	6% (3)	2% (1)	0% (0)	0% (0)	$P = 0.075$/OR = 0.13 [0.00; 1.67]
Ultrasound evidence of ascites	28% (15)	22% (11)	19% (10)	4% (1)	$P = 0.028$/OR = 0.33 [0.13; 0.89]
Treatment outcome parameters (within the trial)					
Embryo transfer during the trial	$n = 47$	$n = 45$	$n = 40$	$n = 19$	
Positive βhCG	70% (33)	56% (25)	75% (30)	63% (12)	$P = 0.578$/OR = 0.77 [0.34; 1.70]
Clinical pregnancy	57% (27)	49% (22)	65% (26)	58% (11)	$P = 0.470$/OR = 1.34 [0.62; 2.87]
Implantation rate	44%	32%	49%	39%	$P = 0.538$
Post-trial follow-up					
Embryo transfer outside the trial[b]	$n = 1$	$n = 1$	$n = 4$	$n = 3$	
Embryo transfer, total	$n = 48$	$n = 46$	$n = 44$	$n = 22$	
Ongoing pregnancy	56% (27)	50% (23)	68% (30)	64% (14)	$P = 0.862$/OR = 1.11 [0.53; 2.31]
Live birth	56% (27)	50% (23)	64% (29)	64% (14)	$P = 0.863$/OR = 1.07 [0.51; 2.22]

ITT, intention-to-treat; OHSS, ovarian hyperstimulation syndrome.
[a]*P*-values are for the analysis of all quinagolide groups combined versus placebo.
[b]Activities that took place after the patient had discontinued/withdrawn from the trial.

not obtain a clinical pregnancy from 31% (8/26) in the placebo group to 11% (8/70) with all groups of quinagolide combined [$P = 0.033$; OR = 0.29 (0.10–0.88)] (Fig. 4c). Among the patients with a clinical pregnancy, there was no significant difference between quinagolide and placebo with respect to the presence of ultrasound evidence of ascites (Fig. 4d). Peritoneal fluid over time followed a dose–response pattern in patients with no clinical pregnancy although there was no relationship between peritoneal fluid accumulation and the use of quinagolide in patients with a clinical pregnancy (Fig. 5a and b).

The average number of embryos transferred ranged from 1.9 to 2.0 in the four treatment groups. Transfer of three embryos occurred in one subject in each treatment group. The clinical pregnancy rates were comparable across treatment groups. Among subjects with embryo transfer, the clinical pregnancy rate was 57% in the placebo group and in the range 49–65% in the quinagolide groups with no dose–response pattern (Table II). The frequency of moderate/severe late OHSS, i.e onset >9 days after hCG administration, was 4% (2/53) in the placebo group and 4% (2/51), 6% (3/52) and 4% (1/26) in the quinagolide 50, 100 and 200 μg/day groups, respectively. All subjects with moderate/severe late OHSS had a clinical pregnancy, of which 50% were multiple pregnancies.

No subjects in the placebo group discontinued because of adverse events, however this was the case for 6% (3/51), 13% (7/52) and 27% (7/26) in the quinagolide 50, 100 and 200 μg/day groups, respectively; mainly due to gastrointestinal and central nervous system adverse events. The most common adverse events in the trial were vomiting, nausea, dizziness and somnolence, which all were reported at a higher frequency with quinagolide compared with placebo (Table III). The frequency of nausea, vomiting and somnolence increased with increasing quinagolide dose, although dizziness occurred at a similar frequency in all three quinagolide dose groups. The majority of gastrointestinal adverse events occurred after the first and second administration of investigational medicinal product, with a dose-dependent increase in the quinagolide groups. By the end of the first week of treatment, the onset of new gastrointestinal adverse events was similar across treatment groups (Fig. 6a). Similarly, most of the central nervous system adverse events were reported on the day of the first administration, and with the frequency increasing with the dose of quinagolide. The frequency of nervous system adverse events with onset after the initial 1–3 days of treatment was similar for placebo and quinagolide (Fig. 6b).

Post-study follow-up information was collected for all 86 subjects with a clinical pregnancy documented at the trial visit and their 119 fetuses. In addition, nine subjects who discontinued from the trial after exposure to quinagolide/placebo but who had embryo transfer outside the trial contribute with safety post-study follow-up data. The ongoing pregnancy and live birth rates were similar among treatment groups (Table II). There was one case of miscarriage between the clinical and the ongoing pregnancy assessments in the placebo group and one patient in the quinagolide 100 μg/day group was lost to follow-up after confirmed ongoing pregnancy. The majority of neonates were born at term and there were no very premature deliveries. The birthweight and length were comparable across treatment groups. Congenital malformations were recorded for none of the 38 neonates in the placebo group, 3 of the 30 neonates in the quinagolide 50 μg/day group (syndactyly, hydrocephalus, congenital cystic kidney disease), 3 of the 38 neonates in the quinagolide 100 μg/day group (two cases of double ureter, trisomy 21) and none of the 17 neonates in the quinagolide 200 μg/day group. On the basis of a teratological review by an independent expert in embryology development, the double ureter cases

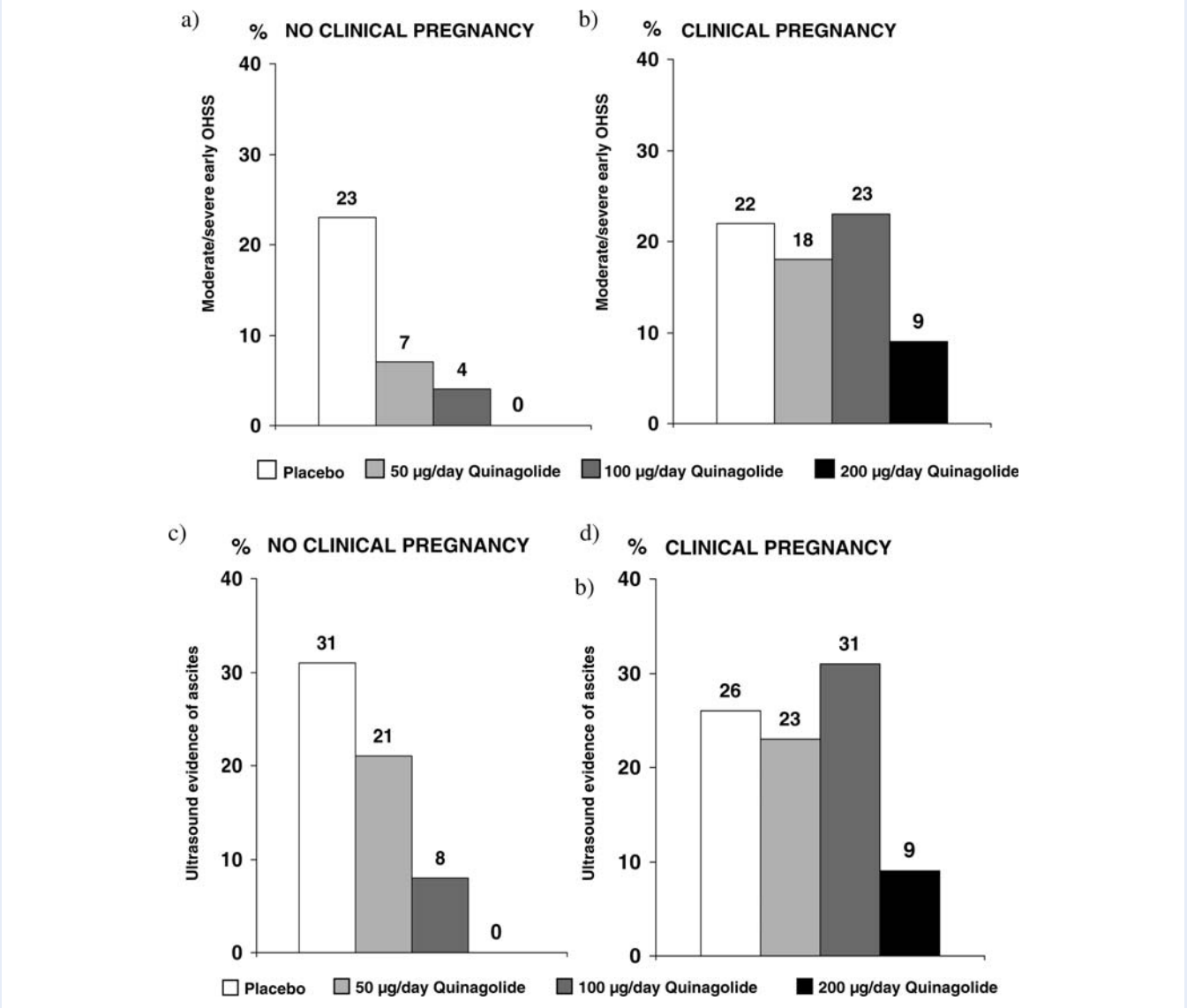

Figure 4 Moderate/severe early OHSS rate according to the patient's pregnancy status in the trial cycle; no clinical pregnancy (**a**) and clinical pregnancy (**b**); and percentage of patients with ultrasound evidence of ascites within the initial 9 days after hCG administration according to the patient's pregnancy status in the trial cycle; no clinical pregnancy (**c**) and clinical pregnancy (**d**).

were considered to be random occurrences. The other reports were all judged by the investigator to be unrelated to treatment.

Discussion

The oral administration of a dopamine agonist to oocyte donors and IVF/ICSI patients provides a potential preventive therapeutic tool in current ART practice, as early OHSS is an undesirable complication that should be avoided. This is the first randomized, double-blind, placebo-controlled trial testing several doses of a dopamine agonist in ART patients at risk of OHSS. The study demonstrated that quinagolide at a dose of 200 µg/day can prevent moderate/severe early OHSS in IVF patients and suggested that also lower doses of quinagolide may be efficacious.

The overall incidence of moderate/severe early OHSS observed in this trial was lower than expected, with an incidence of 23% in the placebo group compared with 52% reported in the trial used for the sample size calculations (Alvarez *et al*., 2007b). Possible explanations for this include differences in trial populations and methodology. Although the follicular response required for participation was the same in the two trials, a further restriction was made in the single-centre study, which only reported data for the subset of subjects who had at least 20 oocytes retrieved rather than for all subjects treated (Alvarez *et al*., 2007b). Furthermore, the cut-off of peritoneal fluid volume used to define ultrasound evidence of ascites plays a role in the frequency of moderate/severe OHSS, and in this respect the 9 cm^2 cut-off used in the previous trial (Alvarez *et al*., 2007b) will have contributed to a higher rate than when applying 24 cm^2 as done in this trial. Despite that the OHSS rates in the placebo

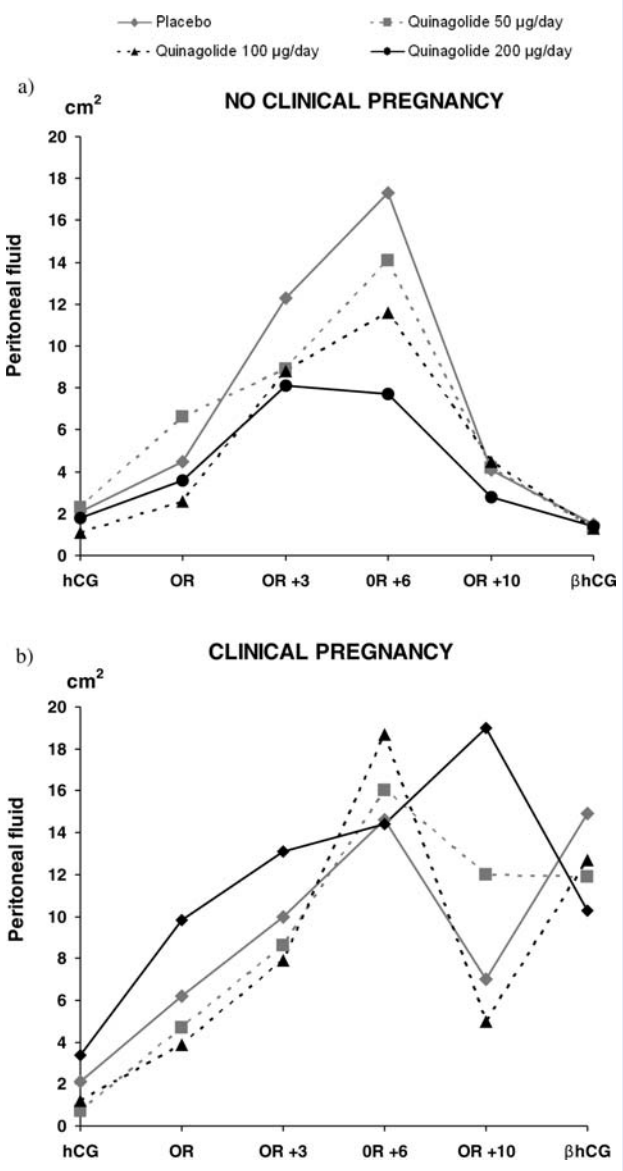

Figure 5 Mean peritoneal fluid (cm^2) according to the patient's pregnancy status in the trial cycle; no clinical pregnancy (**a**) and clinical pregnancy (**b**).

groups are not comparable between the two studies, it is important to note that the relative reduction in OHSS rate in the dopamine agonist group versus the placebo group was similar in the two studies (around 50%).

Although other studies have indicated a reduction of the incidence of OHSS or ultrasound evidence of ascites with cabergoline (Alvarez *et al*., 2007b; Carizza *et al*., 2008) the present study provides for the first time dose–response information and the identification of clinical pregnancy as a major confounding factor in the interpretation of the efficacy data. In patients who did not become pregnant, quinagolide prevented moderate/severe OHSS by decreasing the incidence of ascites in a dose–response fashion. This finding leads to the hypothesis that in women who get pregnant, either higher doses of the dopamine agonist is required to prevent OHSS and/or other vascular

Table III Frequent adverse events.

	Placebo	Quinagolide		
		50 µg/day	100 µg/day	200 µg/day
Subjects randomized (ITT)	*n* = 53	*n* = 51	*n* = 52	*n* = 26
Nausea	25% (13)	35% (18)	50% (26)	69% (18)
Dizziness	15% (8)	41% (21)	52% (27)	42% (11)
Somnolence	13% (7)	25% (13)	33% (17)	42% (11)
Diarrhoea	13% (7)	12% (6)	13% (7)	12% (3)
Vomiting	8% (4)	24% (12)	71% (37)	69% (18)
Abdominal pain lower	8% (4)	8% (4)	8% (4)	4% (1)
Headache	6% (3)	10% (5)	10% (5)	12% (3)
Abdominal distension	6% (3)	0%	2% (1)	0%
Flatulence	4% (2)	8% (4)	2% (1)	4% (1)
Abdominal pain upper	4% (2)	6% (3)	0%	0%
Syncope	0%	2% (1)	8% (4)	12% (3)

ITT, intention-to-treat.

permeability parameters and angiogenesis mediators, less influenced by dopamine agonists, may be present. In patients who did not get pregnant, quinagolide treatment had a significant effect on reducing the incidence of ultrasound evidence of ascites. Abdominal fluid accumulation over time shows two completely different behaviors in pregnant and non-pregnant patients. Among the subjects who did not obtain a clinical pregnancy in the trial cycle, the highest volume of peritoneal fluid observed within the first 9 days of treatment was reduced by ~50% in the quinagolide 200 µg/day group compared with placebo.

The population included in the trial were women developing at least 20 follicles during COH, but not more than 30, and with serum estradiol less than 6000 pg/ml after end of stimulation. Thus, they were at risk of moderate OHSS, but only to a lesser extent at risk of severe OHSS. Nevertheless, the findings on severe early OHSS are also supportive as none of the subjects in the quinagolide 100 and 200 µg/day groups experienced severe early OHSS, although this was the case for three subjects in the placebo group and one in the quinagolide 50 µg/day group.

Late OHSS is associated with the most severe forms of OHSS that lead to severe complications and hospitalization (Mathur *et al*., 2000; Papanikolaou *et al*., 2005). The present study included monitoring of late OHSS, although it was not designed for evaluation of late OHSS, as the participants were selected based on being at risk of early OHSS. Consistent with the data on cabergoline in COH patients (Carizza *et al*., 2008), quinagolide did not have an effect on the frequency of moderate/severe late OHSS. This is in line with the current thinking that late OHSS has a different pathophysiology than early OHSS, and further supported by the observation that all late OHSS cases occurred in patients with a clinical pregnancy. It seems that either the high levels of hCG released by the trophoblast, and/

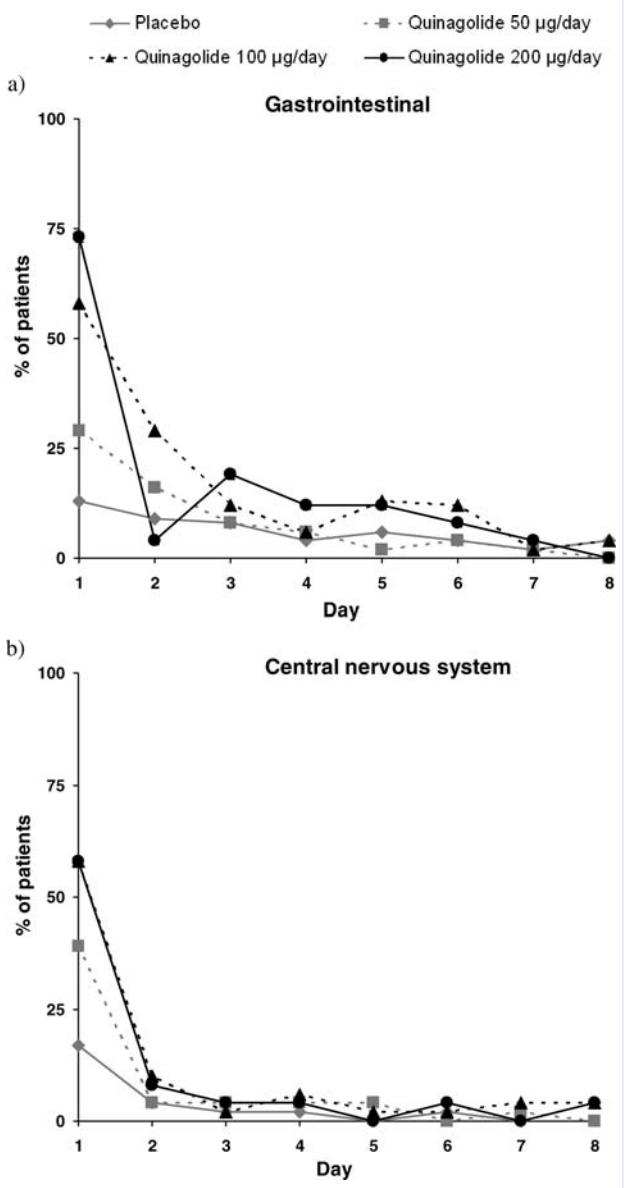

Figure 6 Percentage of patients with onset of a gastrointestinal adverse event (**a**) and percentage of patients with onset of a central nervous system adverse event (**b**) [Day 1 corresponds to hCG administration, Day 3 to OR and Day 6 to OR + 3].

or other pathophysiological mechanisms could be activated in patients that achieve pregnancy, resulting in a cascade of events leading to increased vascular permeability that cannot be avoided by the doses of oral dopamine agonist preparations tested so far. There is, however, evidence in the literature that using intravenous dopamine in severely hyperstimulated women increased urinary output and improved the OHSS symptoms (Ferraretti *et al*., 1992). Although the mechanism of dopamine action was not established in this pioneer report, it is now known from studies in rodents and humans that dopamine agonists are potent inhibitors of VEGF-mediated increased vascular permeability by inhibition of VEGF-R2 phosphorylation and specifically act at the ovarian level (Gomez *et al*., 2006; Alvarez *et al*., 2007b). Future studies should establish whether intravenous dopamine is the treatment of choice in late OHSS that appears in pregnant patients.

The major concern with the use of dopamine agonists in ART is that therapies which could reduce vascular permeability may also decrease embryo implantation as angiogenesis would be reduced. In line with reports that cabergoline does not appear to affect treatment outcome (Alvarez *et al*., 2007b; Carizza *et al*., 2008), clinical pregnancy rates were comparable in the quinagolide and placebo groups, indicating that administration of quinagolide for 3 weeks after hCG administration does not have a detrimental impact on treatment success. It is reassuring that there was no detrimental dose–response in pregnancy or live birth rates and that even the highest dose of quinagolide did not negatively influence the patients' probability of conceiving. The neonatal health data from the offspring in this trial did not raise any safety concerns associated with the use of quinagolide during the initial 3 weeks after OR. There was no dose–response pattern in the congenital abnormalities reported as part of the post-trial safety follow-up; and they were considered random occurrences with very unlikely biological plausibility. The short half-life of quinagolide (~17 h) compared with other dopamine agonists should minimize exposure to the compound during organogenesis. Non-clinical reproductive toxicology studies with quinagolide have provided no evidence of embryotoxic or teratogenic potential and the clinical data from post-marketing surveillance on exposure to quinagolide during pregnancy have suggested incidences of congenital abnormalities of around 4% which are in line with the 4% estimated for the general population (Pregnancy Outcomes Working Group of the FDA Pregnancy Labeling Taskforce in the Center for Drug Evaluation and Research and the Center for Biologics Evaluation and Research at the Food and Drug Administration, 2005).

Nausea, vomiting, dizziness and somnolence were more frequent in quinagolide-treated patients than controls with placebo, and they appeared in a dose-dependant manner. However, most events reflecting an effect of quinagolide on the gastrointestinal and central nervous systems occurred within 1–3 days of administration, and mainly after the first dose. The 200 µg dose of quinagolide was the most effective in preventing moderate/severe early OHSS, but this dose was associated with poor tolerability when administered without dose-titration. Future studies should investigate if high doses of quinagolide when administered with dose-titration have an acceptable patient tolerability or if lower doses in a fixed regimen would be suitable strategies for prevention of early onset of OHSS.

In summary, the present study suggests that quinagolide in a fixed regimen reduces the frequency of moderate/severe early OHSS without compromising pregnancy or treatment outcome. The treatment effect is more marked in patients who did not obtain a clinical pregnancy. When administered without dose-titration, quinagolide at high doses is associated with poor tolerability, although the incidence of adverse events declines after the initial days of treatment. The findings from this trial may be especially relevant for oocyte donation programs and for patients with postponed embryo transfer.

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