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GnRH agonist ovulation trigger and hCG-based, progesterone-free luteal support: a proof of concept study

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BACKGROUND: It is now well established that a GnRH agonist (GnRHa) ovulation trigger completely prevents ovarian hyperstimulation syndrome. However, early studies, using conventional luteal support, showed inferior clinical results following a GnRHa trigger compared with a conventional hCG trigger in normal responder IVF patients. We here present a novel approach for luteal support after a GnRHa trigger.

METHODS: Normal responder patients who failed at least one previous IVF attempt, during which a conventional hCG trigger was used, were consecutively enrolled in the study. A GnRH antagonist-based ovarian stimulation protocol was used in combination with a GnRHa trigger (Triptorelin 0.2 mg). The luteal phase was supported with a total of two boluses of 1500 IU hCG: on the day of oocyte retrieval and 4 days later. Neither progesterone nor estradiol was administered for luteal support.

RESULTS: The mean age was 33.8 years. The mean (±SD) numbers of oocytes and fertilized oocytes were 6.7 (±2.5) and 3.6 (±1.7), respectively. All 15 patients had embryo transfers and 11 patients conceived. On the day of pregnancy test (14 days after retrieval), the mean serum E_2 and progesterone levels were 6607 (±3789) and 182 (±50) nmol/l, respectively. Of the pregnancies, seven are ongoing, while four ended as miscarriages.

CONCLUSIONS: These preliminary results suggest that two boluses of 1500 IU hCG revert the luteolysis after a GnRHa trigger in the normo-responder patient. Importantly, no additional luteal support is needed. The novel concept combines the potential advantages of a physiological dual trigger (LH and FSH) with a simple, patient friendly, luteal support.

Key words: GnRH agonist / GnRH antagonist / luteal support / ovulation trigger

Introduction

Routine IVF practice employs hCG as a surrogate for the endogenous mid-cycle surge of LH to trigger final oocyte maturation. Due to the luteal LH insufficiency induced by supra-physiological steroid levels (estradiol and progesterone) after ovarian hyperstimulation (Tavaniotou *et al.*, 2001, 2003, 2006; Fauser *et al.*, 2003), luteal phase support with progesterone, either vaginally or intramuscularly, remains mandatory in all IVF protocols (Edwards *et al.*, 1980; Fatemi *et al.*, 2007). Moreover, once a pregnancy is achieved, many practitioners choose to continue the luteal support until the 9th or 10th week of gestation. This practice deviates significantly from physiology, in which a simultaneous surge of LH and FSH is responsible for triggering of final oocyte maturation and ovulation, whereas the continued pulsatile LH secretion from the pituitary during the luteal phase is pivotal for the steroidogenic activity of the corpus luteum (Casper and Yen 1979; Hoff *et al.*, 1983; Filicori *et al.*, 1984).

Following the introduction of the GnRH antagonist, a renewed interest in GnRH agonist (GnRHa) triggering opened new opportunities for a tailored approach to luteal phase support (Humaidan *et al.*, 2009, 2011). As previously described, a bolus of GnRHa will displace the GnRH antagonist from the GnRH receptors in the pituitary, eliciting a simultaneous surge of LH and FSH which reliably secures ovulation and final oocyte maturation (Itskovitz *et al.* 1988; Gonen *et al.*, 1990). However, the luteolysis followed by a GnRHa trigger necessitates a modified luteal support to rescue the luteal phase and to secure the reproductive outcome (Humaidan *et al.*, 2011). Thus, the GnRHa trigger allows the clinician to individualize the luteal phase according to the needs of the patient, supplementing with either hCG, LH or estradiol and progesterone (Engmann *et al.*,

2008; Castillo et al., 2010; Humaidan et al., 2010; Papanikolaou et al., 2011).

In this 'proof of concept' report we describe, for the first time, a protocol in which final oocyte maturation was induced with a bolus of GnRHa followed by an hCG-based luteal support, without any exogenous luteal progesterone or estradiol supplementation.

Materials and Methods

The observational period was from 1 December 2010 to 1 March 2011 and the study was a one center, prospective, observational, proof of concept study. Patients were thoroughly informed about alternative stimulation protocols before deciding to participate.

Patients

The inclusion criteria were IVF or ICSI treatment, age between 20 and 41 and normal response in a previous IVF trial defined as 5–12 follicles on the day of trigger.

Stimulation, GnRHa trigger, luteal phase support and pregnancy

The study included 15 normal responder patients who failed to conceive in at least one previous IVF attempt, during which a conventional hCG trigger was used. They were thoroughly counseled and agreed to partake in this proof of concept study. Stimulation commenced on Day 2 or 3 of the cycle with recombinant FSH or hMG in a daily dose of 150–300 IU FSH. Co-treatment with a GnRH antagonist (0.25 mg, either Orgalutran, MSD, Oss, The Netherlands or Cetrotide, Serono Geneva, Switzerland) was initiated at a follicle size of 13mm. As soon as ≥3 follicles had reached a size of 17 mm, a bolus of GnRHa (Triptorelin 0.2 mg) was administered. Oocyte pick-up (OPU) was performed 35 h later. Embryos were transferred on Day 2 or 3 post-OPU. The luteal phase was supported with a bolus of 1500 IU hCG following the OPU (Humaidan et al., 2010), and an additional bolus of 1500 IU hCG on day OPU+ 4. Neither progesterone nor estradiol luteal phase support was administered. All patients received embryo transfers. Blood sampling was performed on the day of the GnRHa trigger and on Day 14 after OPU. A clinical pregnancy was defined as a gestational sac with or without a fetal heart beat. An ongoing pregnancy was defined as a gestational sac with fetal heart beat 10 weeks after oocyte retrieval.

Results

Demographic data are given in Table I. The mean age was 33.8 years. The mean (±SD) numbers of oocytes and fertilized oocytes were 6.7 (±2.5) and 3.6 (±1.7), respectively. On the day of pregnancy test (14 days after retrieval), the mean serum E_2 and progesterone levels were 6607 (±3789) and 182 (±50) nmol/l, respectively (Table II). Of the 15 patients, 11 conceived (clinical pregnancy rate: 73%); 7 pregnancies are ongoing (ongoing pregnancy rate: 47%) while 4 pregnancies ended as miscarriages (miscarriage rate: 36%; Table III). The implantation rate (number of gestational sacs divided by number of embryos transferred) was 29%. None of the patients developed ovarian hyperstimulation syndrome (OHSS).

Table I Main demographic parameters of the study population.

Age	**33.8 ± 4.2**
Indication	
Male, No	8
Mechanical, No	2
Unexplained, No	5
Months in treatment, No	28 ± 13
Previous IVF attempts, No	3.75 ± 2.4
Primary infertility, No	7
Secondary infertility, No	8
Previous pregnancies, No	0.9 ± 1.2
Previous live birth, No	0.4 ± 0.7

Values are mean ± SD.

Table II Stimulation characteristics and embryology data.

Stimulation (days)	9.3 ± 2.0
GnRH antagonist (days)	3.8 ± 0.9
FSH (units)	2443 ± 925
E_2 day of trigger (pmol/l)	3764 ± 1227
P day of trigger (nmol/l)	2.4 ± 1.65
LH day of trigger (IU/l)	1.9 ± 1.3
Oocytes retrieved	6.7 ± 2.5
Embryos obtained	3.6 ± 1.7
Embryos transferred	2.9 ± 0.9
Embryos frozen	0.8 ± 1.5
Beta hCG (IU/l)	152 ± 86
E_2 (day of pregnancy test, pmol/l)	6607 ± 3789
P (day of pregnancy test, nmol/l)	182 ± 50

Values are mean ± SD.

Table III Reproductive outcomes.

Positive HCG/ cycle, *n* (%)	11/15 (73)
Clinical ongoing pregnancy, *n* (%)	7/15 (47)
Early pregnancy loss, *n* (%)	4/11 (36)

Discussion

During the last decade, the follicular phase has been the focus of intense efforts to make it as patient friendly as possible. Pen-like injecting tools have been developed to administer FSH, and a long-acting FSH molecule has been introduced for the purpose of cutting down on the number of injections necessary for ovarian stimulation

(Devroey *et al.*, 2009). In contrast, the luteal phase has received very little attention. Thus, luteal phase support with progesterone administered either intramuscularly or vaginally is the standard procedure in IVF treatment. This cannot be considered patient friendly as it involves daily dosing(s) for 7–10 weeks in case of the establishment of a pregnancy.

Unlike hCG triggering of final oocyte maturation, GnRHa triggering is a more physiological approach, eliciting a surge of gonadotrophins, similar to that of the natural mid-cycle surge. Thus, in contrast to hCG triggering, GnRHa triggering induces an endogenous surge of FSH as well as LH. However, after the GnRHa trigger, a modified luteal phase support including LH-like activity (hCG) or rLH is crucial (Humaidan *et al.*, 2011) and the challenge has been to titrate the amount of LH activity needed to sustain the function of only a few corpora lutea after GnRHa triggering (Castillo *et al.*, 2010; Humaidan *et al.*, 2010; 2011; Papanikolaou *et al.*, 2011). Until now, however, most studies employing GnRHa triggering also supplemented patients with progesterone either vaginally or intramuscularly.

Gonen *et al.* (1990) in a small study used no luteal support following a GnRHa trigger, and obtained three clinical pregnancies. However, Clomiphene Citrate (CC) was used for ovarian stimulation during the follicular phase. Due to the long half-life of CC, a higher pituitary secretion of LH during the luteal phase could be expected counteracting the luteolytic action following the GnRHa trigger. However, CC is rarely used in controlled ovarian stimulation nowadays.

Similarly, with another approach, Pirad *et al.* (2006) avoided exogenous progesterone supplementation by repeated (three times daily) applications of nasal Buserelin. The data suggest that maintaining LH secretion throughout the luteal phase by repeated administration of a GnRHa overcomes the GnRHa trigger-induced luteolysis.

In contrast, in the present report, since there was no risk of inducing OHSS as patients had a mean of 6.7 oocytes, the function of the corpora lutea was secured not only by a bolus of 1500 IU hCG on the day of aspiration but also by a second bolus given 4 days later. Importantly, no luteal progesterone or estradiol supplementation was administered. In patients with a higher ovarian response to stimulation, lower doses of hCG or repeated rLH administration may be used.

Once the functioning of a few corpora lutea has been secured, the endogenously secreted hCG from the implanting embryo(s) will take over their support until the luteo-placental shift. From the patients' perspective, a progesterone-free administration during the luteal phase after ovarian hyperstimulation would be an immense relief (Verhaak *et al.*, 2007). Moreover, the risk of developing acute eosinophilic pneumonia as seen after i.m. administration of progesterone would be abolished (Bouckaert *et al.*, 2004; Khan *et al.*, 2008). With this protocol, two injections of hCG would replace daily progesterone applications for many weeks.

In the present study, it was decided not to include patients with more than 12 developing follicles in previous IVF trials in order to minimize the risk for OHSS. Indeed, none of the patients developed early or late-onset OHSS, however, larger randomized controlled trials are needed to determine OHSS incidence with the proposed protocol. At present, patients at high OHSS risk should not be offered the described protocol.

Although long-term progesterone supplementation after a hCG trigger is still the conventional practice, there is intriguing evidence which questions this routine and suggests cessation of conventional luteal support during early pregnancy 14 days after embryo transfer (Nyboe-Andersen *et al.*, 2002), or after establishing one normal doubling of serum hCG levels (Kyrou *et al.*, 2011). The current proof of concept delineates a simple protocol that makes any progesterone supplementation totally redundant.

In this small series of 15 patients, with a high clinical pregnancy rate per patient started ($n = 11$, 73%), 7 pregnancies are ongoing (ongoing pregnancy rate: 47%) and 4 pregnancies ended as miscarriages of clinical pregnancies (miscarriage rate: 36%). Importantly, the serum progesterone levels in patients with missed abortions were comparable to that recorded for the ongoing pregnancies. No biochemical only pregnancies were seen. Although numbers are small, the results seem promising for a future progesterone and estradiol-free luteal support following ovarian stimulation in IVF.

In summary, we herein present, for the first time, a protocol for ovarian stimulation including a GnRHa trigger and two boluses of hCG during the luteal phase without any further luteal support. If the results are corroborated in a larger series of normo-responder patients, this protocol could lead to a paradigm shift in luteal support policy. Thus, we believe that the triggering property of hCG needs to be dissected from its luteal supportive properties. Moreover, we predict that in the near future GnRHa will be for triggering ovulation whereas LH-like activity (hCG or LH) will be for luteal support. This might significantly reduce, or even abolish, the risk of OHSS and at the same time secure a good reproductive outcome. Finally, cumbersome, leaky and painful luteal progesterone administration after IVF treatment would be history.

Authors' roles

All authors met the following criteria for authorship: Substantial contributions to conception and design, or acquisition of data, or analysis and interpretation of data. Drafting the article or revising it critically for important intellectual content. Final approval of the version to be published.

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