Role of Cetrorelix and Highly Purified Human Menopausal Gonadotropin (HP-HMG) in Ovarian Stimulation in Assisted Reproductive Technique (ART)

Content

Background and Objective of the Survey

Controlled ovarian stimulation (COS) in in vitro fertilization (IVF) cycles is the crucial point from which good oocyte retrieval and couple's prognosis depend. Several protocols have been studied in order to find the therapy that ensures the best outcomes in terms of pregnancy and live birth, minimizing iatrogenic risks, and the risk of cycle cancellation due to poor response or ovarian hyperstimulation syndrome (OHSS).

Pulsatile secretion of gonadotropin-releasing hormone (GnRH) from the hypothalamus stimulates the synthesis and secretion of follicle-stimulating hormone (FSH) and LH from the anterior pituitary. Pituitary suppression during cycles of ART allows ovarian stimulation to be controlled by exogenous FSH and suppresses the mid-cycle LH surge. In modern reproductive medicine, this is achieved by the administration of GnRH analogues. Cetrorelix is a high-affinity GnRH antagonist that competitively blocks binding of GnRH to pituitary cell receptors. The binding affinity of cetrorelix for the GnRH receptor is ~20 times greater than that of native GnRH. Cetrorelix has a far greater effect on serum levels of LH than FSH.

Gonadotrophin products utilized in ovarian stimulation are derived from urinary or recombinant sources. The effectiveness of HP hMG in IUI patients treated with a mild stimulation protocol. The mild stimulation strategy seems a reasonable approach yielding an acceptable balance between ongoing and multiple pregnancy rates. In ART it is important to find a balance between high pregnancy rates and an acceptable multiple pregnancy rate.

The objective of the survey is:

To study the potential role of cetrorelix and highly purified human menopausal gonadotropin (HP-HMG) in ovarian stimulation in assisted reproductive technique (ART).

Methodology of the Survey

A survey was conducted the to study the potential role of cetrorelix and highly purified human menopausal gonadotropin (HP-HMG) in ovarian stimulation in assisted reproductive technique (ART). A total of 50 doctors from India participated in the survey.

Step 1: A literature search was done on the topic. Below topics were covered in the literature search

- Introduction
- Ovarian folliculogenesis
- Ovulation induction with low-dose gonadotrophins
- Ovarian stimulation for assisted reproduction cycles
- GnRH antagonists - Cetrorelix
- Gonadotropins
- Abstract

Step 2: A survey questionnaire was prepared based on the literature search. The survey form was shared through the digital medium with physicians across India.

Step 3: Their responses were analyzed and the findings are provided in this survey analysis booklet.

Literature Review

Introduction

Controlled ovarian stimulation (COS) in in vitro fertilization (IVF) cycles is the crucial point from which good oocyte retrieval and couple's prognosis depend. Several protocols have been studied in order to find the therapy that ensures the best outcomes in terms of pregnancy and live birth, minimizing iatrogenic risks, and the risk of cycle cancellation due to poor response or ovarian hyperstimulation syndrome (OHSS). In recent years, the concept of "one size fits all" has evolved into a concept of "individualization" in IVF. This should also reduce costs and the dropout rate of patients, mainly caused by the physical and psychological burden. Treatment individualization is based on ovarian reserve. The ovarian response to COS largely depends on a woman's ovarian reserve, the stimulation regimen itself is a secondary factor. Serum anti-Müllerian hormone (AMH) and ultrasound antral follicle count (AFC) in particular have been shown to be the most sensitive markers. Another strategy for the individualization of treatment is based on the response in previous IVF cycles (if a previous cycle had a good performance, the same protocol can be used).[1]

In the Italian scenario, a strong consensus exists among physicians on the importance of the prediction of ovarian response to treatment. Ovarian reserve markers are assessed in as many as 80% of women who enter IVF programs, and the majority of physicians agree that AMH and AFC are the most reliable factors for predicting ovarian response. The choice of therapy is a very important clinical point because of the possibility of using various kinds of drugs [gonadotrophin-releasing hormone (GnRH)-analogues or antagonists, different gonadotrophin preparations, adjuvant therapies]. Moreover, the selection of the follicle-stimulating hormone (FSH) starting dose is fundamental for IVF outcomes.[1]

Ovarian folliculogenesis

In women with adequate ovarian reserve, response to stimulation treatment only varies within a certain range. In these women, it has been shown that antral follicle count may be used as a predictor of ovarian response, as this determines the number of follicles that will grow in response to gonadotrophin stimulation in that particular cycle. However, in patients with

diminished ovarian reserve, there seems to be much more variability in response to gonadotrophin stimulation due to higher variability of antral follicles available. This varying number of antral follicles available is reflected by the change in FSH concentrations from cycle to cycle in these patients. This also explains why measuring FSH in two consecutive spontaneous cycles may predict the remaining ovarian reserve better than with a single measurement (Bancsi *et al*., 2004). However, even with two measurements, a substantial number of patients are still misdiagnosed with regard to their ovarian responsiveness.[2]

Repetitive stimulation procedures and cycle-to-cycle variability

Patients undergoing repetitive cycles of ovarian stimulation did not demonstrate a decrease in the number of oocytes produced. This was confirmed by the limited variation in consecutive cycles for intrauterine insemination treatment of regularly menstruating ovulating patients. The OMEGA project group, which is an ongoing prospective cohort study in the Netherlands, could not demonstrate a decrease in the number of oocytes retrieved in those patients, who underwent at least seven consecutive IVF cycles, from cycle number 1 to cycle number 6 after adjusting for patients' age. Similar observations have been published by other workers.[2]

Caligara *et al*. studied oocyte donors and could not see an impairment of oocyte number or quality in consecutive cycles. However, one has to be aware that oocyte donors would not have pre-existing impairment of ovarian function compared with infertile patients. Therefore, if ovarian function is already impaired before stimulation therapy, a decrease in the number of oocytes retrieved may occur, as was shown in patients with ovarian endometriomas. In conclusion, in patients with intact ovarian reserve, repetitive high-dose ovarian stimulation does not increase the risk of higher cycle-to-cycle variability. However, in patients with impaired ovarian function, it might decrease the number of oocytes retrieved after consecutive stimulation cycles. The number of oocytes retrieved is usually chosen as the primary end-point to look at cycle-to-cycle variability, as the majority of studies published so far lack the statistical power to look at clinical pregnancy rate or even live birth rate. Nonetheless, these would be the most relevant end-points to look at in order to find out whether cycle-to-cycle variability is just a phenomenon that might be observed during ovarian stimulation without any

clinical relevance, or whether this really impacts on the overall outcome of assisted reproduction treatment.[2]

Ovulation induction with low-dose gonadotrophins

Low-dose ovarian stimulation, especially in patients classified in WHO group II (patients with hypothalamic–pituitary dysfunction and oligo/amenorrhea) suffering from PCOS, has been evaluated extensively. These authors mainly concentrated on the predictability of the ovarian stimulation overall, and did not look for variation in the cycle-to-cycle response. It is recommended, however, that in a step-up protocol the last dose used should be chosen as the initial dose in a new treatment cycle with the step-down protocol after cancellation of the stimulation cycle for no response of follicular growth. This reflects the experience of an apparently relatively constant dose–response from cycle to cycle. Van Santbrink *et al*. (2002) described a group of 56 patients, who had been treated for ovulation induction with a step-down protocol. In 25% of the patients, the dose was changed initially from the standard of 150 IU daily to either a higher or a lower starting dose for the subsequent cycle, based on experiences with previous stimulation cycles. The authors observed three groups of patients during the actual treatment cycle: those who had an early step down of the urinary human menopausal gonadotrophin (uHMG) dose (group a), those with the standard procedure (group b) and those with a dose increase or no change of the initial dose (group c). Interestingly, those in group a had a mean of 28.5 IU above the predicted effective starting dose, those in group b of 13 IU above the predicted dose, and those in group c of 43 IU under the predicted starting dose in the subsequent cycle. This prediction was made using a previously established model.[2]

Ovarian stimulation for assisted reproduction cycles

In cases of an assisted reproduction cycle for IVF or intracytoplasmic sperm injection (ICSI), usually higher doses of gonadotrophins are chosen to achieve polyfollicular growth. The required maximum dose of gonadotrophins is limited by the physiological response possibilities of the ovary to these drugs. A cycle-to-cycle variation of gonadotrophin response is a well-established clinical fact, which sometimes cannot be sufficiently explained by clinical data. Even if sometimes dramatic changes in ovarian response from cycle to cycle are observed, the overall intraindividual variability seems to be small. Especially in long gonadotrophin-

releasing hormone (GnRH) agonist protocols, severe suppression of endogenous gonadotrophins may result in a higher dose of gonadotrophins required for follicular stimulation. This grade of suppression can be measured by the FSH to LH ratio. In GnRH antagonist cycles, the suppression of endogenous gonadotrophins is far less pronounced and only apparent over a short period of the stimulation cycle. Therefore overall gonadotrophin consumption in GnRH antagonist cycles is lower compared with the long agonist protocol.[2]

Controlled Ovarian Stimulation in Normo-Responders

An optimal response to COS cycles is considered as an oocyte yield between 10 and 15 oocytes. Pre-treatment with estrogen, progesterone or oral contraceptive pills (OCP) prior to COS do not offer any benefits in normo-responders. A recent meta-analysis showed a significantly lower ongoing pregnancy rate with antagonist compared to long agonist protocol. However, this outcome was noted only with the combination of oral hormonal pre-treatment and flexible antagonist protocol, while no such difference was evident between fixed antagonist and agonist protocol. Antagonist protocol is preferred in many IVF clinics worldwide considering convenience and safety aspects.[3]

Both recombinant follicles stimulating hormone (rFSH) and human menopausal gonadotropins (HMG) or highly purified HMG (HP-HMG) have been used for COS. A greater number of oocytes can be expected with rFSH compared to HMG. Non-inferiority of HP-HMG to rFSH has been established in both antagonist and long agonist protocols in terms of ongoing pregnancy rates. Thus, the choice of gonadotrophins in normo-responders is based on the availability, cost and clinician's discretion.[3]

GnRH antagonists

Pulsatile secretion of gonadotropin-releasing hormone (GnRH) from the hypothalamus stimulates the synthesis and secretion of follicle-stimulating hormone (FSH) and LH from the anterior pituitary. Pituitary suppression during cycles of ART allows ovarian stimulation to be controlled by exogenous FSH and suppresses the mid-cycle LH surge. In modern reproductive medicine, this is achieved by the administration of GnRH analogues. These analogues are synthetic versions of native GnRH and are available as either agonists or antagonists. GnRH

antagonists were first introduced to clinical practice in 1999 and have since replaced the use of agonists in many protocols worldwide.[4]

GnRH antagonists offer a therapeutic alternative to agonists for pituitary suppression. GnRH antagonists achieve an immediate and dose-dependent suppression of LH by competing with native GnRH to bind to pituitary cell membrane receptors. Third-generation GnRH antagonists, including cetrorelix and ganirelix, are commercially available for use in ART. The use of GnRH antagonists offers a number of advantages over agonists. GnRH antagonists produce a rapid and reversible suppression of LH and FSH, with no initial flare effect. Therefore, prolonged pretreatment to achieve pituitary down-regulation is not required. As GnRH antagonists are usually administered only when there is a risk of premature LH surge (usually from day 5 to 7 of stimulation), symptoms of hypo-estrogenemia are rare. Furthermore, lower total doses and fewer days of exogenous gonadotropin stimulation are reported with co-treatment with antagonists than agonists. Consequently, the total cycle duration is shorter and subsequent cycles can be initiated more quickly with antagonist than long agonist protocols. Last, the pituitary remains responsive to GnRH stimulation during antagonist co-treatment, and so a bolus dose of agonist can be administered (instead of human chorionic gonadotropin; hCG) to trigger final oocyte maturation while concurrently preventing OHSS.[4]

GnRH antagonists offer theoretical advantages over agonists for women with specific clinical conditions. Up to a quarter of all patients undergoing in vitro fertilization (IVF) respond poorly to gonadotropin stimulation. A poor response may be defined as the development of an inadequate number of follicles following COS. GnRH antagonist-based protocols could provide a useful option for patients with a (expected or proven) poor response to stimulation by avoiding prolonged pituitary suppression. The use of GnRH antagonists may also be beneficial during COS for women at high risk of OHSS, such as those with polycystic ovary syndrome (PCOS). GnRH antagonist co-treatment leads to the development of fewer ovarian follicles than in long agonist protocols. Moreover, there is early evidence to support a reduced incidence of moderate or severe OHSS in high-risk patients following GnRH agonist-induced final oocyte maturation and subsequent transfer of frozen–thawed oocytes. PCOS is characterized by chronic ovulatory dysfunction and hypersecretion of LH. It has been

suggested that the rapid and effective suppression of LH afforded by GnRH antagonists could be beneficial during cycles of ovulation induction (OI) for patients with PCOS. Avoidance of an untimely LH surge could also help to schedule intrauterine insemination (IUI) following OI. Indeed, a recent meta-analysis of data from six randomized controlled trials (RCTs) of OI, using recombinant human FSH (r-hFSH), followed by IUI indicates that, compared with conservative monitoring, GnRH antagonist co-treatment significantly increases the clinical pregnancy rate (odds ratio (OR) 1.56, 95% CI 1.05 – 2.33). Large studies are required to assess further the potential benefits of GnRH antagonists for women undergoing OI.[4]

Cetrorelix

Cetrorelix acetate was developed in the early 1990s and was the first GnRH antagonist to receive regulatory approval (in 1999 in the EU) for the inhibition of premature LH surges and ovulation in women undergoing COS. Cetrorelix is now approved for this indication in more than 80 countries worldwide. Cetrorelix is provided as lyophilized powder (0.25 mg/1 mL or 3 mg/3 mL) with a shelf-life of 2 years. Vials should be stored at ≤25°C within the outer carton to protect the peptide from light. The solution should be used immediately after reconstitution of the powder in water. Cetrorelix is administrated by s.c. injection during the early to mid-follicular phase of an ART cycle and is suitable for self-administration by the patient.[4]

Cetrorelix can be used in multiple- or single-dose protocols. In multiple-dose protocols, cetrorelix 0.25 mg is administered on day 5 or 6 of gonadotropin stimulation and, thereafter, is injected daily until hCG administration. In a single-dose protocol, cetrorelix 3 mg is injected when serum estradiol levels indicate an appropriate response to stimulation (usually between days 5 and 7). If hCG is not administered within 4 days after cetrorelix 3 mg, daily doses of cetrorelix 0.25 mg are injected until hCG administration. Cetrorelix is a decapeptide analog of native GnRH, with amino-acid substitutions at positions 1, 2, 3, 6, and 10. Acetyl and amide groups at the C- and N-terminals provide stability and full antagonistic activity .[4]

Pharmacodynamics

Cetrorelix is a high-affinity GnRH antagonist that competitively blocks binding of GnRH to pituitary cell receptors. The binding affinity of cetrorelix for the GnRH receptor is ~20 times

greater than that of native GnRH. Cetrorelix has a far greater effect on serum levels of LH than FSH. Serum levels of FSH are largely unaffected at the doses of cetrorelix used in COS. However, immediate suppression of serum LH levels (by 80% of baseline levels) occurs within 24 h of a single s.c. injection of cetrorelix 3 mg. Inhibition of the mid-cycle LH surge consequently delays ovulation. Repeated treatment with cetrorelix causes sustained suppression of LH (9 days after last administration) but reversal of the effects is observed after cessation of therapy.[4]

Pharmacokinetics and metabolism

Cetrorelix is rapidly absorbed after s.c. administration; in healthy women, maximal plasma concentrations occur within 1-2 h. The mean absolute bioavailability of cetrorelix is ~85% after s.c. injection, and 86% of cetrorelix is protein-bound. Cetrorelix has a plasma half-life of ~20 h, and at least 2-4% of the drug is excreted in urine and 5-10% in bile. Cetrorelix is excreted unchanged in urine, but four peptide metabolites are also present in bile. The effects of cetrorelix in cases of hepatic or renal impairment are yet to be determined.[4]

Clinical efficacy

Phase II studies

The efficacy of cetrorelix for prevention of the premature LH surge was assessed in a number of Phase II exploratory and dose-finding studies. An early Phase II study enrolled 20 women undergoing COS to receive treatment with cetrorelix at doses of 3 mg (n = 15) or 1 mg (n = 5) s.c. once daily from cycle day 7 until administration of hCG. None of the women experienced an endogenous LH surge following treatment with cetrorelix (irrespective of the dose administered). Furthermore, the total gonadotropin dose requirement was ~50% of the standard dose administered with a long GnRH agonist protocol, and yet oocyte quality was reported to be comparable. The results of a subsequent dose-finding study indicated that cetrorelix at 0.25 mg s.c. once daily was adequate to prevent an endogenous LH surge in women undergoing COS.[4]

One Phase II study was conducted to establish the optimum single-dose protocol in COS. Of the 65 women who received cetrorelix 3 or 2 mg s.c. on day 8 of the stimulation cycle, only 1

(in the 2 mg dose group) experienced an LH surge. There were no differences in the magnitude of change in LH levels between groups, but the LH surge was suppressed for a shorter duration in the 2 mg group. ART outcomes were similar in both groups.[4]

Phase III studies

As a result of the promising Phase II data, cetrorelix was investigated further in five prospective Phase III trials. Two dosing regimens of cetrorelix were evaluated: a multiple-dose protocol (0.25 mg s.c. once daily, starting on day 5 or 6 of gonadotropin stimulation) and a single-dose protocol (3 mg on day 7 of stimulation). Data on the efficacy and safety of cetrorelix versus GnRH agonist co-treatment for prevention of premature LH surges are available from two randomized, multi-center, Phase III clinical trials. A randomized study of cetrorelix 0.25 mg s.c. once daily (n = 188) versus buserelin (nasal spray; 0.15 mg four times daily; n = 85) in women aged up to 39 years was published in 2000. Pregnancy rates per started cycle were comparable between the cetrorelix and buserelin groups (22.3 versus 25.9%, respectively). However, cetrorelix was associated with the administration of significantly fewer hMG ampoules and fewer days of stimulation than buserelin ($p < 0.01$ for both). Cetrorelix co-treatment also involved fewer days of pituitary suppression (5.7 versus 26.6, respectively; $p < 0.001$) and led to the development of fewer small follicles (11-14 mm in diameter) that are associated with an increased risk of OHSS than did buserelin (3.2 versus 4.3, respectively; $p = 0.02$) .[4]

In another Phase III study, patients undergoing COS were randomized to co-treatment with cetrorelix 3 mg s.c. once daily (n = 115) or depot triptorelin 3.75 mg (n = 39). Although fewer oocytes were retrieved and the total number of embryos transferred was lower with cetrorelix than depot triptorelin, similar clinical pregnancy rates were obtained in both treatment groups. The single-dose cetrorelix protocol required a shorter duration of stimulation and administration of fewer ampoules of hMG. Additional data are available from one uncontrolled, open-label study and two randomized, single-center studies comparing the efficacy of cetrorelix with buserelin or leuprolide acetate. The findings of these studies were generally consistent with those of the multi-center Phase III trials, and showed that multiple- or single-dose protocols of cetrorelix achieved similar clinical outcomes as did GnRH agonists.

Two large Phase IIIb studies were subsequently conducted to compare the efficacy of cetrorelix in multiple- (n = 1066) and single-dose (n = 541) protocols in routine clinical practice. In these studies, multiple- and single-dose cetrorelix protocols were associated with similar efficacy and safety profiles. Although more oocytes were retrieved with single- than multiple-dose protocols (10.1 versus 9.0; p = 0.005), the total number of embryos obtained (IVF: 4.7 versus 4.7) and transferred (2.3 versus 2.5), and pregnancy rates per started cycle (24 and 23%, respectively) were similar using the two regimens. Cetrorelix was administered in combination with hMG in most clinical trials. However, limited experience suggests that cetrorelix has similar efficacy when used with r-hFSH.[4]

Experience in modified ART protocols

A number of studies have been conducted to evaluate the efficacy and safety of cetrorelix in oral contraceptive (OC)-programmed stimulation cycles. In a study of 182 patients who were randomized to receive an OC pretreatment cetrorelix (0.25 mg s.c. a day) regimen or buserelin (500 μg for ≥10 days, and 200 μg thereafter), similar outcomes were reported in both groups, including the mean number of oocytes retrieved (11.4 with cetrorelix versus 10.9 with buserelin). Body mass index was higher in the cetrorelix group but this did not affect the treatment outcomes. A randomized study of 185 infertile women receiving the combined OC pill for cycle programming showed similar efficacy of single-dose cetrorelix (3 mg s.c.) and daily doses of ganirelix 0.25 mg in preventing premature LH surges (LH <5 IU/L: 97.7 versus 96.6% of patients, respectively) and achieving a pregnancy (51.7 versus 48.9%, respectively). The single-dose cetrorelix protocol required a significantly lower median number of injections than did ganirelix co-treatment (1 versus 4 injections; p <0.001), which is expected to confer convenience benefits for patients.[4]

A prospective, randomized study of 120 women undergoing COS for ART was performed to identify the optimal starting dose of r-hFSH for COS for IVF or intracytoplasmic sperm injection when combined with cetrorelix (0.25 mg daily from day 6 of stimulation). A significantly greater mean number of oocytes were obtained using a starting dose of r-hFSH 225 IU compared with 150 IU (11.0 versus 9.1; p = 0.024), although this was judged to be of

minimal clinical significance. The authors concluded that cetrorelix simplifies ART treatment protocols and reduces the overall exposure to medication .[4]

Early studies have also produced promising data on the use of cetrorelix (at single or multiple doses of 0.25, 0.5 or 1 mg) administered in the late follicular phase of natural or mild IVF cycles. In a pilot study of 33 women who received cetrorelix (at a single dose of 0.5 or 1 mg) when plasma estradiol levels reached 100-150 pg/mL and a lead follicle of 12-14 mm in diameter was detected, only 4/44 natural cycles were cancelled and a pregnancy rate of 32% per embryo transfer was achieved.[4]

Experience in special patient populations

Specific groups of patients may benefit most from pituitary suppression with cetrorelix, including women who respond poorly to gonadotropin stimulation or those at high risk of developing OHSS. Data on pituitary suppression using cetrorelix compared with a GnRH agonist in patients with an expected or proven poor response to stimulation are available from four RCTs. Comparable implantation (15.1 versus 11.4%, respectively) and clinical pregnancy rates (26.3 versus 22.2% per embryo transfer, respectively) were demonstrated in an early study of 48 poor responders who received OC pretreatment plus leuprolide acetate (40 μg s.c. once daily) or cetrorelix alone (0.25 mg once daily during the late follicular phase). Similarly, pregnancy and implantation rates did not differ when cetrorelix (alone or as co-treatment with clomiphene citrate plus r-hFSH or hMG) or a short GnRH agonist protocol was administered to 90 patients with proven poor response.[4]

In a third randomized trial, 66 patients received cetrorelix (0.25 mg once daily starting on day 6 of stimulation) or buserelin (600 μg once daily starting in the mid-luteal phase of the previous cycle). The use of cetrorelix was associated with a higher mean number of embryos being transferred (2.32 versus 1.50; $p = 0.01$), although clinical pregnancy rates were not significantly different. The most recently published study compared a flexible cetrorelix protocol (0.125 mg for 2 days and thereafter 0.25 mg once daily) with a short GnRH agonist regimen (triptorelin 0.1 mg s.c. once daily) in 133 women at risk of a poor response. Pituitary suppression with

cetrorelix resulted in a significantly higher mean number of metaphase II oocytes than did triptorelin (5.73 versus 4.64; $p < 0.05$).[4]

Data are available from four RCTs comparing down-regulation with cetrorelix or a GnRH agonist in patients with PCOS who received OC pretreatment. A meta-analysis of data from these studies showed no significant differences in the number of oocytes retrieved or clinical pregnancy rate achieved with either cetrorelix or long GnRH agonist protocols in patients with PCOS. However, multiple-dose cetrorelix resulted in a significantly shorter stimulation period than did long GnRH agonist protocols (OR -8.6, 95% CI -1.14 to -0.59; $p < 0.01$).[4]

There is evidence to support a significant reduction in the incidence of OHSS in high-risk populations with cetrorelix compared with agonist protocols. Data from a prospective, multi-center, comparative study (using historical controls) suggest that cetrorelix may reduce the incidence of OHSS and, thus, the number of cancelled cycles among patients at high risk of an excessive response. A meta-analysis of data from eight comparative studies demonstrated a significantly lower incidence of OHSS with the use of cetrorelix (OR 0.23, 95% CI 0.10-0.54) compared with long agonist protocols. Whereas, the use of ganirelix or agonist co-treatment resulted in similar rates of OHSS (OR 1.13, 95% CI 0.24-5.31). Additional studies are required to evaluate prospectively the efficacy of cetrorelix with a GnRH agonist to trigger final oocyte maturation in high-risk patients .[4]

Safety and tolerability

Given the pharmacologic and physiologic effects of GnRH antagonists, their use has been postulated to reduce the risk of adverse effects associated with long GnRH agonist protocols, such as hormone withdrawal symptoms and OHSS. Clinical evidence shows that cetrorelix (in multiple- or single-dose protocols) is generally well tolerated in women undergoing COS. Hormone withdrawal symptoms such as tachycardia, hot flushes, headaches, vaginal bleeding, or decreased libido occur only rarely with cetrorelix. Few systemic adverse events were reported (by at least 1%) among 949 patients aged 19-40 years who received multiple or single doses of cetrorelix (0.1-5 mg) in key clinical trials. Overall, only 1.1% experienced headache, 1.3% nausea, and 3.5% moderate or severe OHSS. While the risk

of OHSS is not completely eliminated with the use of GnRH antagonists, the incidence of OHSS is significantly lower with the use of cetrorelix than GnRH agonists.[4]

As observed with other s.c. administered GnRH analogs, mild and transient injection-site reactions are commonly observed with cetrorelix. Hypersensitivity reactions are uncommon but cases have been reported. No adverse effects have been detected on the health of children conceived from oocytes collected during cycles of COS in which cetrorelix was used for pituitary suppression. Cetrorelix is contraindicated for use in patients who have experienced hypersensitivity reactions to cetrorelix, extrinsic peptide hormones or mannitol, or are pregnant, breast-feeding or postmenopausal, or have moderate or severe renal or hepatic impairment. Caution is advised for use in patients with hypersensitivity to GnRH; patients should be monitored carefully after the first injection.[4]

Gonadotropins

Gonadotrophin products utilized in ovarian stimulation are derived from urinary or recombinant sources. Urinary products include human menopausal gonadotrophins (hMG, highly purified [HP-hMG]), urinary follicle stimulating hormone (u-FSH) and human chorionic gonadotrophin (hCG).[5]

Effectiveness of HP hMG in IUI patient

The effectiveness of HP hMG in IUI patients treated with a mild stimulation protocol. The mild stimulation strategy seems a reasonable approach yielding an acceptable balance between ongoing and multiple pregnancy rates. In ART it is important to find a balance between high pregnancy rates and an acceptable multiple pregnancy rate. High pregnancy rates could be obtained only after excessive ovarian stimulation, which inevitably results in high numbers of twins and triplets. The overall success of IUI across different studies varies, with pregnancy rates ranging from 3% to 29%. This wide range may arise from different stimulation protocols and acceptance of higher multiple pregnancy rates, differences in treatment groups, diagnostic criteria, age of the cohort, techniques used, cause of subfertility and number of treatment cycles.[6]

With mild stimulation protocol, a high number of ongoing multiple pregnancies was prevented. Results on multifollicular growth reflect the mild stimulation protocol that was use; in the HP hMG group only 40.6% of the cycles showed multifollicular growth, resulting in a non-significant increase in multiple pregnancies in the HP hMG group when compared to the unstimulated IUI group. In IVF and ICSI, stimulation with HP hMG appears to be a milder way of stimulation than rFSH, resulting in comparable or even higher pregnancy rates at lower oocyte yields. As HP hMG contains LH activity, a number of advantages may be attributed to this LH activity.[6]

One has to bear in mind, however, that the advantages of HP hMG over rFSH derive from IVF studies applying FSH dosages as high as 225 IU. High dosages of FSH may lead to increased progesterone levels, which may have impaired endometrial receptivity and therefore led to impaired ART outcome. However, a study in which relatively low dosages of rFSH were applied, still showed advantages of HP hMG over rFSH. Therefore, in low dose treatments like those applied in IUI, there might still be clinical differences. In IUI treatment with (non-purified) hMG (Menogon1), increased LH activity has been associated with shorter treatment duration, lower menotropin consumption and reduced development of small ovarian follicles, while maintaining the development of larger and more mature follicles.[6]

The study is the first large study reporting on pregnancy rates after IUI treatment with HP hMG. One other study has investigated the effects of HP hMG in IUI. In that retrospective study 218 women were treated with either HP hMG or rFSH during one down-regulated IUI cycle (i.e., 218 cycles). Treatment with HP hMG resulted in higher ongoing pregnancy rates and higher perifollicular blood flow compared to rFSH. In retrospective study all patients started with three natural IUI cycles, followed by three mild stimulated cycles with HP hMG. It seems reasonable to assume that patients in the study became more subfertile along the six treatment cycles, which is supported by a study of Steures *et al*. That study demonstrated declining pregnancy rates in patients undergoing six subsequent unstimulated IUI cycles, except for an increase in the fourth cycle, probably due to the beneficial effect of a diagnostic laparoscopy performed after the third unstimulated IUI cycle.[6]

The significantly higher observed miscarriage rate in the patient group treated with HP hMG supports the assumption of negative patient selection. Despite this fact, pregnancy rates increased from 6 to 7.4% from natural to stimulated cycles. Furthermore, results demonstrated a significant increase in pregnancy rates in the fifth treatment cycle compared to natural cycles. This is probably caused by reaching a certain threshold for HP hMG, since dosages of HP hMG increased during subsequent stimulated cycles. In previous study, the lack of a control group continuing with three natural IUI cycles makes it difficult to explore the real effectiveness of HP hMG. The benefit of HP hMG might be underestimated. Future studies are required in which the investigation is performed in a prospective randomized setting.[6]

HP hMG seems to be most effective for patients with unexplained subfertility. In this group ongoing pregnancy rates increased from 13.8% in cycle 3 (natural) to 18.1% in cycle 4 (HP hMG). Other studies confirmed that stimulation is most effective in unexplained subfertility. Stimulated IUI is also most effective in younger women (age <37 years) compared to older women. In previous study, the ovarian stimulation with HP hMG seems to be effective for all age categories, but most effective for patients in the age category 30 to 34 years. In this age category ongoing pregnancy rates increased from 13.3% in cycle 3 (natural) to 16.8% in cycle 4 (HP hMG).[6]

Effectiveness of HP hMG in IVF patient

In modern medicine, IVF is a method that is actively used to treat couples with infertility. This process requires ovarian stimulation, and various medicines have been developed to achieve this. The gonadotrophin preparations for ovarian stimulation are classified as FSH only products and combinations, which possess FSH and LH activity. Among them, the menotrophin preparation is a gonadotrophin extracted from the urine of postmenopausal women, and contains FSH, LH and HCG, unlike the recombinant FSH preparations, and shows biological activity of FSH and LH in a 1:1 ratio. The advantages of LH activity have already emerged in ovarian stimulation. It is known that LH plays an important role in the follicular development process by acting on the theca cells to synthesize androgen in the early follicular phase. In addition, it plays an important role in estradiol synthesis, follicular growth and final maturation of oocytes in the mid-late follicular phase.[7]

During ovarian stimulation for IVF, the granulosa cells, which are hyperstimulated by FSH, produce a large amount of progesterone. Furthermore, insufficient enhancement of LH activity results in the non-activation of theca cells, which consequently causes some of the progesterone produced by the granulosa cells to enter the blood circulation and, thereby, increase the blood progesterone level. This may cause endometrial prematurity, which in turn may affect pregnancy and implantation. In such cases, the exogenous stimulation of LH activity further activates the theca cells and directs progesterone to the androgen synthesis pathway, which prevents progesterone absorption into the blood. As a result, the progesterone level is decreased while the estrogen level is increased in the body circulation, which would increase the endometrial receptivity and improve the quality of oocytes and embryos.[7]

Preparations with LH activity can be classified into HMG menotrophins and highly purified HP-HMG based on the manufacturing process. The advantage of HP-HMG is that it is available as a subcutaneous injection based on its improved purity. The enhanced purity is achieved by subjecting the HMG to two or three additional purification steps, which reduces the local reactions on injection sites. In addition, the LH activity in HP-HMG is mainly derived from HCG, which is concentrated during purification steps.[7]

Urine-derived gonadotrophin preparations, which had been used widely for ovarian stimulation before the development of recombinant FSH, induced LH activity inevitably. Controversies have arisen with the discovery of the negative effect of this induced LH activity on the quality of oocytes in animal models. Concerns have arisen, however, that the widespread use of the protocol using recombinant FSH alone under the GnRH analogs in ovarian stimulation may excessively suppress the endogenous LH level in some patients, possibly affecting clinical outcome. Therefore, a subsequent study reported that the exogenous LH induction might have a positive effect on the clinical efficacy and, consequently, it has been used actively in these procedures. According to published reports, the HMG group showed a higher level of serum estrogen and reduction in progesterone concentration on HCG administration day, although the number of oocytes was lower than that of the recombinant FSH-alone group. The LH activity can contribute to the improvement of oocyte quality by inducing the atresia of small follicles, and increase the implantation rate by improving endometrial receptivity.[7]

Several studies have reported that HP-HMG could be used properly under GnRH agonist as well as GnRH antagonist protocols in ovarian stimulation for the normoresponders. In randomized controlled trial results, the delivery rate was significantly increased in the HP-HMG group under the GnRH agonist compared with the group administered recombinant FSH alone by 3-4%. In the comparison of the efficacy of HP-HMG and recombinant FSH under the GnRH antagonist protocol, the delivery rate was higher by 2.9%, but not significantly so, which was similar to the level observed with the GnRH agonist. No clear conclusion, however, was derived owing to the inadequacy of the study on HP-HMG under the GnRH antagonist protocol. It has been reported that the exogenous enhancement of LH activity in ovarian stimulation improves IVF outcome in normoresponders and also poor responders, aged women, potential high responders and patients with highly suppressed LH activity or suboptimal response to the recombinant FSH-alone treatment.[7]

Previous studies have reported that potential high responders (initial AMH >5.2 ng/ml) showed a remarkable decrease in the ratio of high ovarian responses in the HP-HMG group, and especially under the GnRH antagonist, a remarkable increase in the live birth rate compared with the recombinant FSH-alone group. In addition, HMG preparations with LH activity can improve the number of oocytes retrieved and the pregnancy rate in patients with poor response to the initial recombinant FSH preparation compared with the recombinant FSH-alone preparation, by enhancing the LH activity in the mid-late follicular phase. It has been reported that women with infertility who show initial suboptimal responses to recombinant FSH alone during ovarian stimulation under GnRH agonist, have significantly increased serum estradiol levels and improvement in the oocyte quality after replacement therapy with HP-HMG at the stimulation day 8.[7]

The study has shown the clinical efficacy of HP-HMG with LH activity in women undergoing ovarian stimulation for assisted reproduction techniques under GnRH antagonist protocol. However, it does have some potential limitations: primary end-point (number of oocytes retrieved) for the non-inferiority may be affected by the dose of HP-HMG administered and the regimen triggering final oocyte maturation. In the present study, the initial dose was selected according to the judgment of investigators and clinical site's standard practice, and the

dose of HP-HMG after 5 days of stimulation could be adjusted in case of OHSS or low ovarian response in patients for the ethical reason in infertility treatment. Although total number of days and dose of administration between groups were not significantly different, the possibility cannot be ruled out that these limitations affected the primary outcome. Also, because this study was designed to demonstrate non-inferiority in number of oocytes retrieved, the cohort size was not sufficient to carry out a meaningful evaluation of the efficacy, such as implantation rate or clinical pregnancy rate. The live births, which would be an ultimate goal for the infertility treatment, were not confirmed in the study.[7]

Abstract

Highly purified hMG versus recombinant FSH in ovarian hyperstimulation with GnRH antagonists - a randomized study[8]

Background

Highly purified hMG (hp-hMG) has recently shown better cycle outcome than the recombinant FSH (rFSH) when compared in GnRH agonist long protocol cycles. However, they have not yet been compared in GnRH antagonist cycles.

Methods

A RCT comparing the ongoing pregnancy rate (primary end-point) in 280 patients undergoing IVF/ICSI after stimulation with hp-hMG or rFSH controlled with a GnRH antagonist.

Results

No significant differences were observed between hp-hMG and rFSH in terms of the ongoing pregnancy rate per started cycle (35.0 versus 32.1%, respectively; P = 0.61); relative risk: 1.09 (95% confidence interval: 0.78–1.51; risk difference: 2.9%). No differences were observed for implantation, clinical pregnancy and pregnancy loss rates. More oocytes were obtained from patients receiving rFSH then hMG (14.4 ± 8.1 versus 11.3 ± 6.0, respectively; P = 0.001). Estradiol was higher at the end of stimulation in the hp-hMG group (P = 0.02), whereas progesterone was higher in patients stimulated with rFSH (P < 0.001).

Conclusions

A similar outcome was observed for hp-hMG and rFSH when used for stimulation in GnRH antagonist cycles. However, some differences were found in ovarian response in terms of oocyte yield and hormonal profile.

The LHRH antagonist Cetrorelix: a review[9]

In those clinical situations in which an immediate and profound suppression of gonadotrophins is desired, LHRH agonists have the disadvantage of producing an initial stimulatory effect on hormone secretion. Therefore, the use of GnRH antagonists which cause an immediate and dose-related inhibition of LH and FSH by competitive blockade of the receptors is much more advantageous. One of the most advanced antagonists produced to date is Cetrorelix, a decapeptide which has been shown to be safe and effective in inhibiting LH and sex-steroid secretion in a variety of animal species and in clinical studies as well. Clinical trials in patients suffering from advanced carcinoma of the prostate, benign prostate hyperplasia, and ovarian cancer are currently in progress and have already shown the usefulness of this new treatment modality. In particular, the concept that a complete suppression of sex-steroids may not be necessary in indications such as uterine fibroma, endometriosis and benign prostatic hyperplasia represents a promising novel perspective for treatment of these diseases. Following completion of phase III trials in controlled ovarian stimulation for IVF regimens, Cetrorelix was given marketing approval and, thus, became the first LHRH antagonist available clinically.

References:

1. Sighinolfi G, Grisendi V, La Marca A. How to personalize ovarian stimulation in clinical practice. *J Turk Ger Gynecol Assoc*. 2017;18(3):148-53.
2. Keck C, Bassett R, Ludwig M. Factors influencing response to ovarian stimulation. Reproductive BioMedicine Online. 2005;11(5):562-9.
3. Jirge PR, Patil MM, Gutgutia R, *et al*. Ovarian stimulation in assisted reproductive technology cycles for varied patient profiles: An Indian perspective. *J Hum Reprod Sci*. 2022;15:112-25.

4. Ilan Tur-Kaspa, Ezcurra D. GnRH antagonist, cetrorelix, for pituitary suppression in modern, patient-friendly assisted reproductive technology. *Expert Opinion on Drug Metabolism & Toxicology*. 2009;5(10):1323-36.
5. Lehert P, Schertz JC, Ezcurra D. Recombinant human follicle-stimulating hormone produces more oocytes with a lower total dose per cycle in assisted reproductive technologies compared with highly purified human menopausal gonadotrophin: a meta-analysis. *Reproductive Biology and Endocrinology*. 2010;8(1):112.
6. Groeneveld E, Kouijzer IJE, Timmermans AJ, *et al*. Effectiveness of highly purified human menopausal gonadotropin in intra-uterine insemination. *European Journal of Obstetrics & Gynecology and Reproductive Biology*. 2011;154(2):182-6.
7. Koo HS, Kwon H, Choi DS, *et al*. Clinical utility of newly developed highly purified human menopausal gonadotrophins: a randomized controlled trial. Reproductive BioMedicine Online. 2017;34(5):499-505.
8. Bosch E, Vidal C, Labarta E, *et al*. Highly purified hMG versus recombinant FSH in ovarian hyperstimulation with GnRH antagonists--a randomized study. *Human Reproduction*. 2008;23(10):2346-51.
9. Reissmann T. The LHRH antagonist Cetrorelix: a review. *Human Reproduction Update*. 2000;6(4):322-31.

Survey Form

1. **In your practice, how commonly do you encounter cases of premature luteinization?**

A. 5-10%

B. 11-20%

C. 21-30%

D. 31-40%

E. 41-50%

F. >50%

2. **What according to you are the advantages of adjunctive use of a GnRH antagonist for IVF cycles?**

A. Reduce dose of HMG

B. Reduce cycle cancellation rate

C. Improved ovarian response

3. **What according to you are the advantages of using GnRH antagonist over GnRH agonist in IVF cycles?**

A. Shorter duration of stimulation

B. A reduced level of gonadotropin usage

C. Lower risk for ovarian hyper stimulation syndrome (OHSS)

D. Immediate suppression of serum LH levels unlike GnRH agonist

4. **An equivalent pregnancy rate is achievable using GnRH antagonist protocols as from long GnRH agonist protocols**

A. Yes

B. No

5. **What daily dose of cetrorelix do you prefer in your patients undergoing IVF treatment?**

A. 0.25 mg

B. 0.5 mg

C. 01 mg

6. **How do you evaluate response of cetrorelix in your patients?**

A. Monitoring serum LH levels

B. Monitoring serum P levels

C. Both A & B

7. **Which type of cetrorelix protocol do you prefer in your patients undergoing IVF treatment?**

A. Single-dose protocol

B. Multi-dose protocol

8. **Which of the following is your preferred technique using Cetrorelix along with HMG on IVF?**

A. Daily midcycle administration with concomitant HMG

B. Single dose administration at day 7 of HMG stimulation

9. **In what % of your infertility patients do you use HP-HMG in combination with cetrorelix?**

A. 20-30%

B. 31-40%

C. 41-50%

D. 51-60%

E. 61-70%

F. 71-80%

G. >80%

10. In the past, have you used ciscure in your practice?

A. Yes

B. No

11. If yes, how would you rate your experience (in terms of efficacy & safety) with ciscure on a scale of 1-10 (1 being worst, 10 being best)?

A. 1

B. 2

C. 3

D. 4

E. 5

F. 6

G. 7

H. 8

I. 9

J. 10

Survey Findings

1. In your practice, how commonly do you encounter cases of premature luteinization?

A. 5-10%

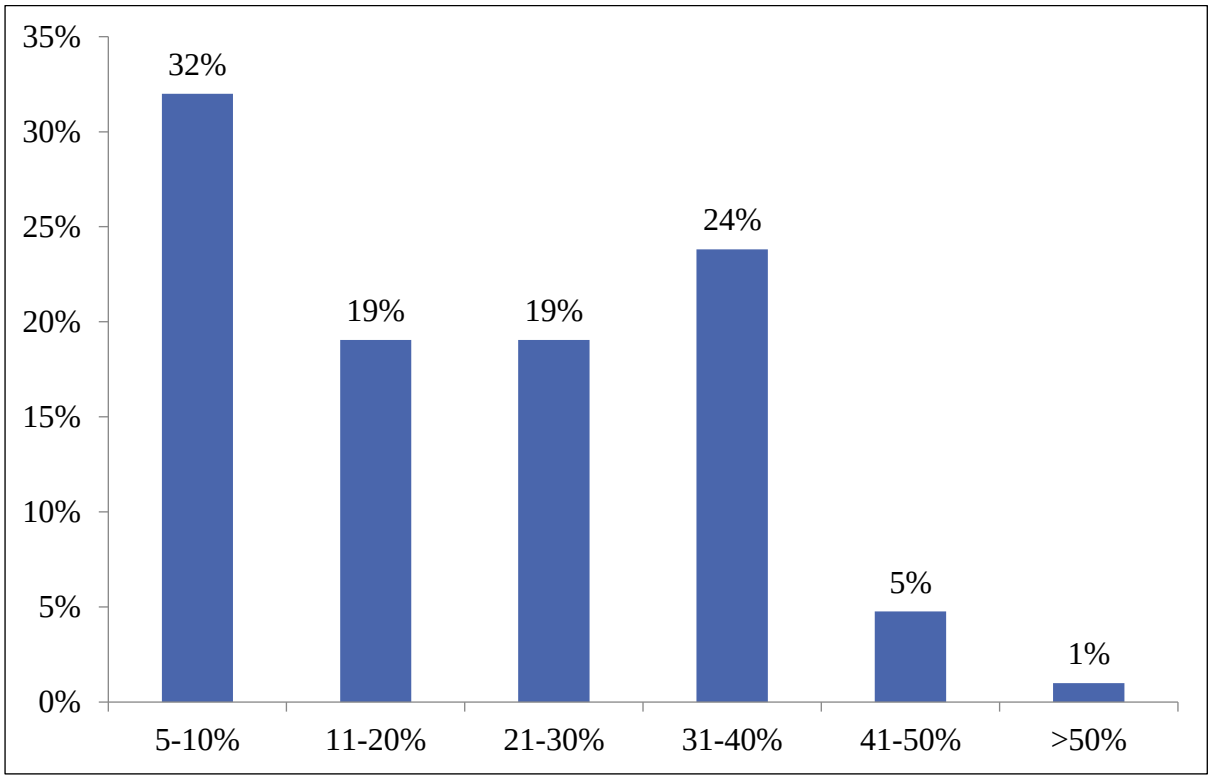
B. 11-20%

C. 21-30%

D. 31-40%

E. 41-50%

F. >50%



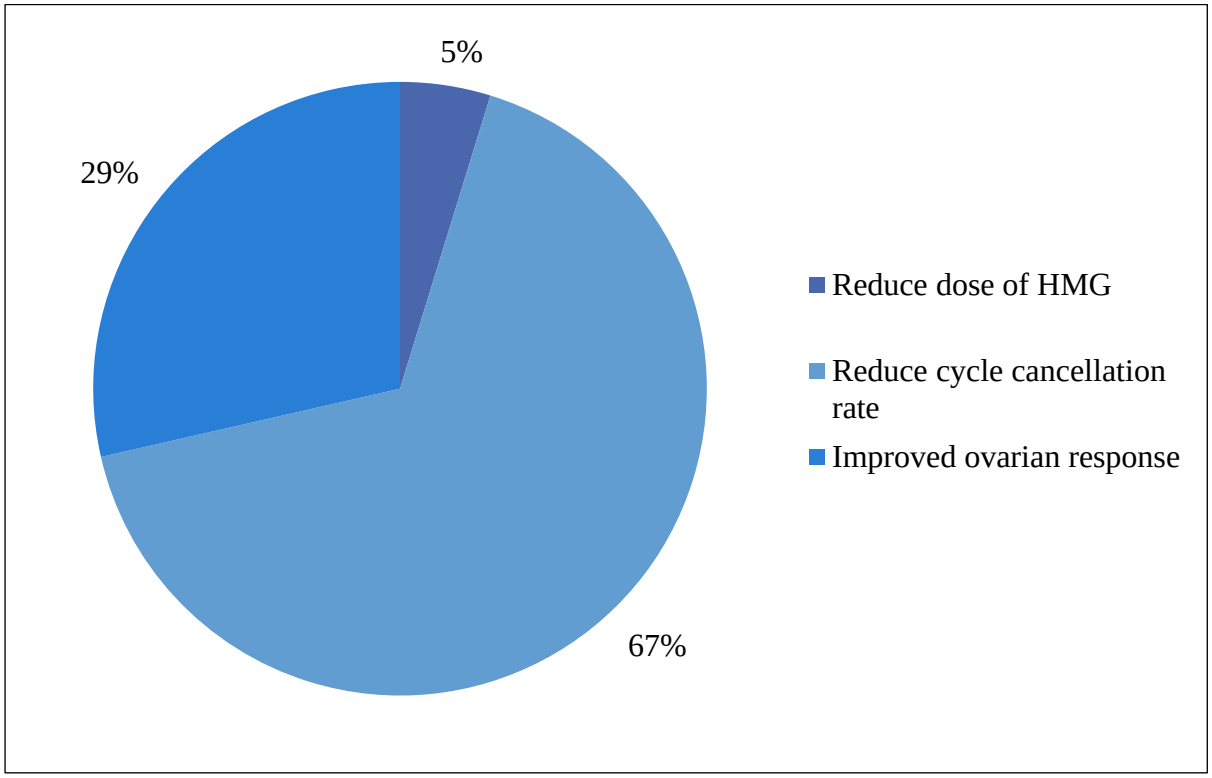
Around 32% of doctors encounter cases of premature luteinization in 5-10% of their patients.

2. What according to you are the advantages of adjunctive use of a GnRH antagonist for IVF cycles?

A. Reduce dose of HMG

B. Reduce cycle cancellation rate

C. Improved ovarian response



According to 67% of doctors, the advantages of adjunctive use of a GnRH antagonist for IVF cycles include reducing the cycle cancellation rate.

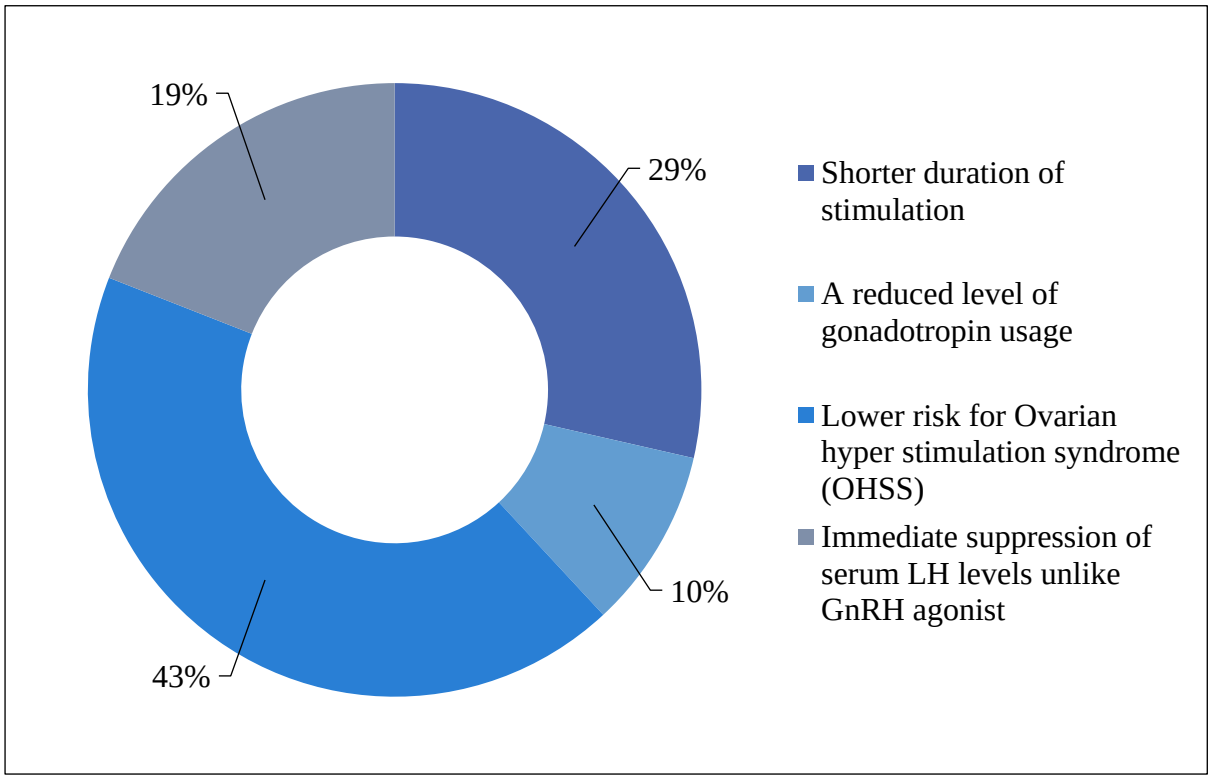
3. What according to you are the advantages of using GnRH antagonist over GnRH agonist in IVF cycles?

A. Shorter duration of stimulation

B. A reduced level of gonadotropin usage

C. Lower risk for ovarian hyper stimulation syndrome (OHSS)

D. Immediate suppression of serum LH levels unlike GnRH agonist

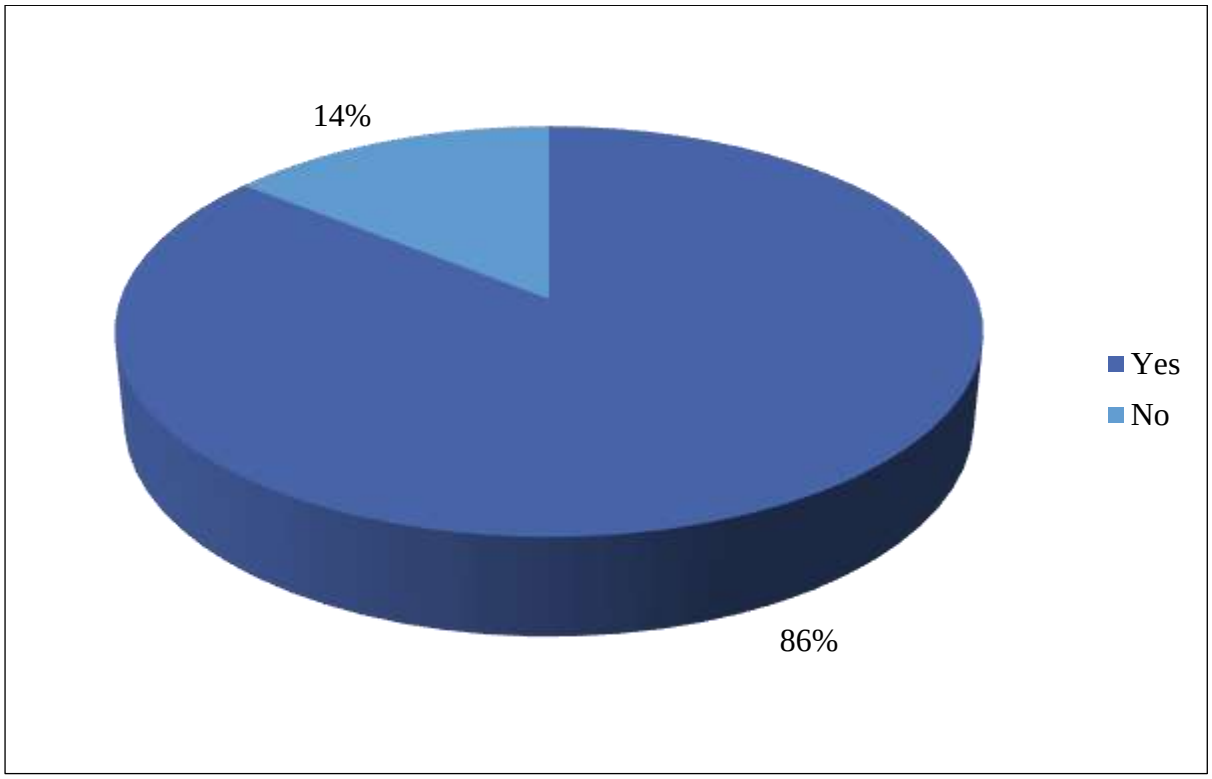


According to 43% of doctors, the advantages of using a GnRH antagonist over a GnRH agonist in IVF cycles include a lower risk for ovarian hyperstimulation syndrome (OHSS).

4. An equivalent pregnancy rate is achievable using GnRH antagonist protocols as from long GnRH agonist protocols

A. Yes

B. No



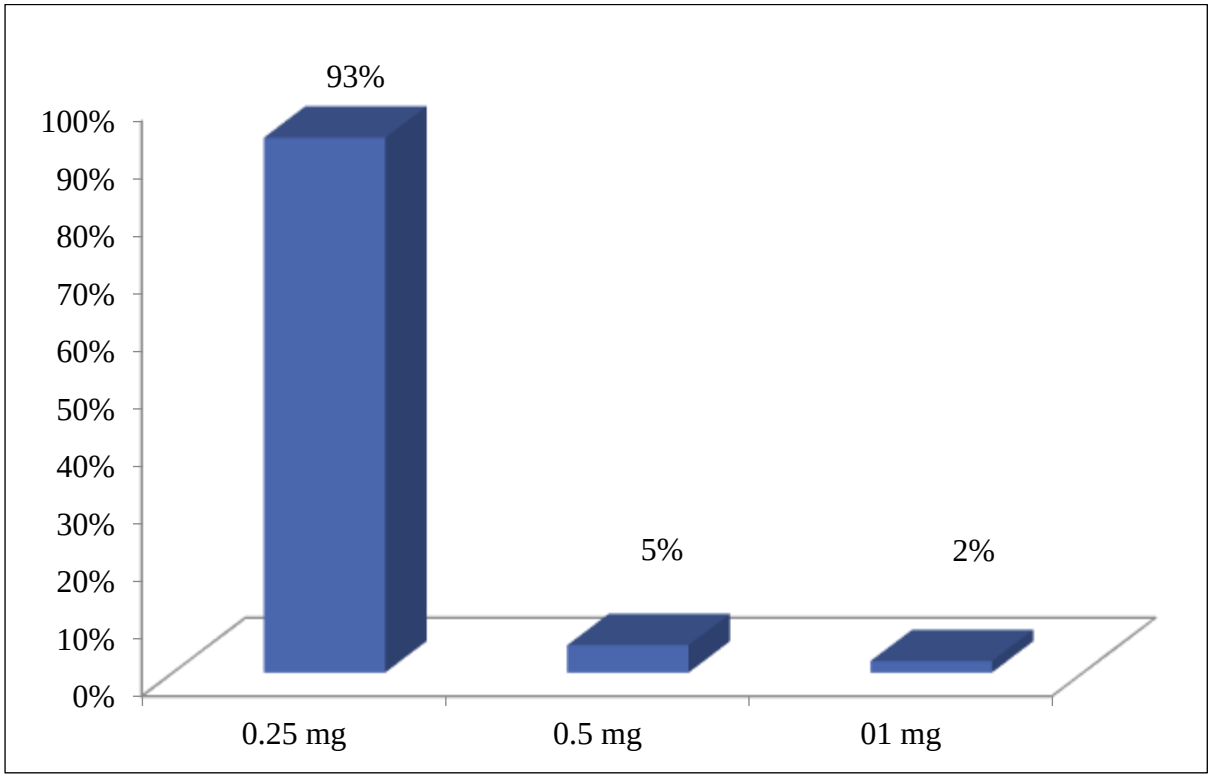
A majority of doctors, 86%, agree that an equivalent pregnancy rate is achievable using GnRH antagonist protocols as compared to long GnRH agonist protocols.

5. What daily dose of cetrorelix do you prefer in your patients undergoing IVF treatment?

A. 0.25 mg

B. 0.5 mg

C. 01 mg



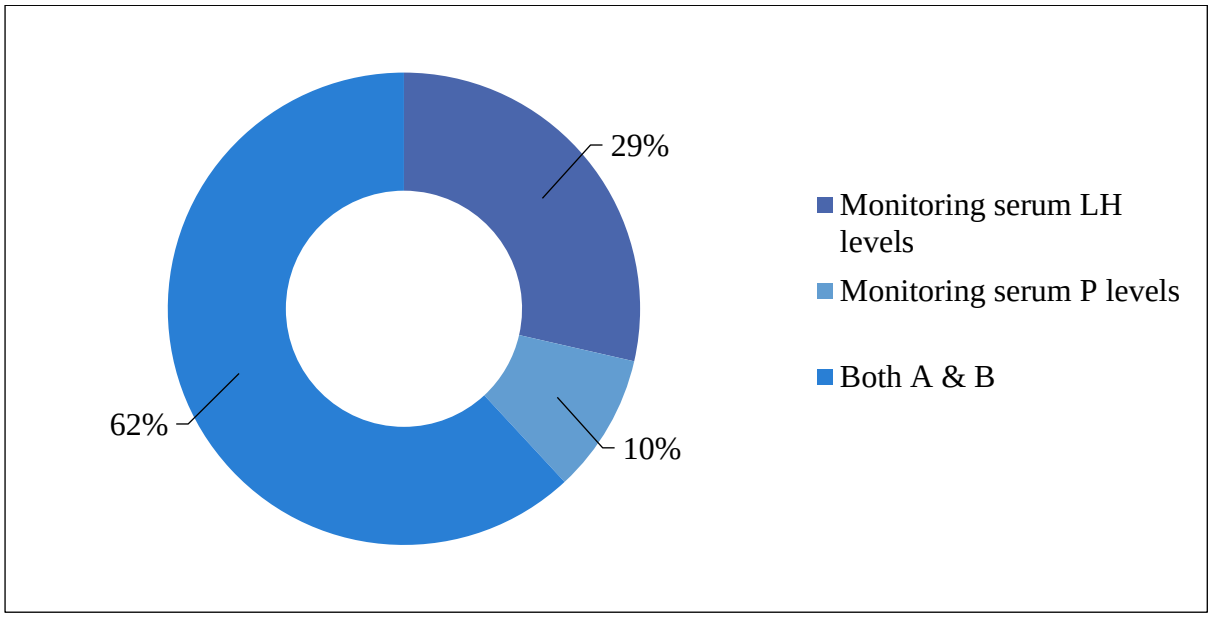
A majority of doctors, 93%, prefer a daily dose of 0.25 mg of cetrorelix for their patients undergoing IVF treatment.

6. How do you evaluate response of cetrorelix in your patients?

A. Monitoring serum LH levels

B. Monitoring serum P levels

C. Both A & B

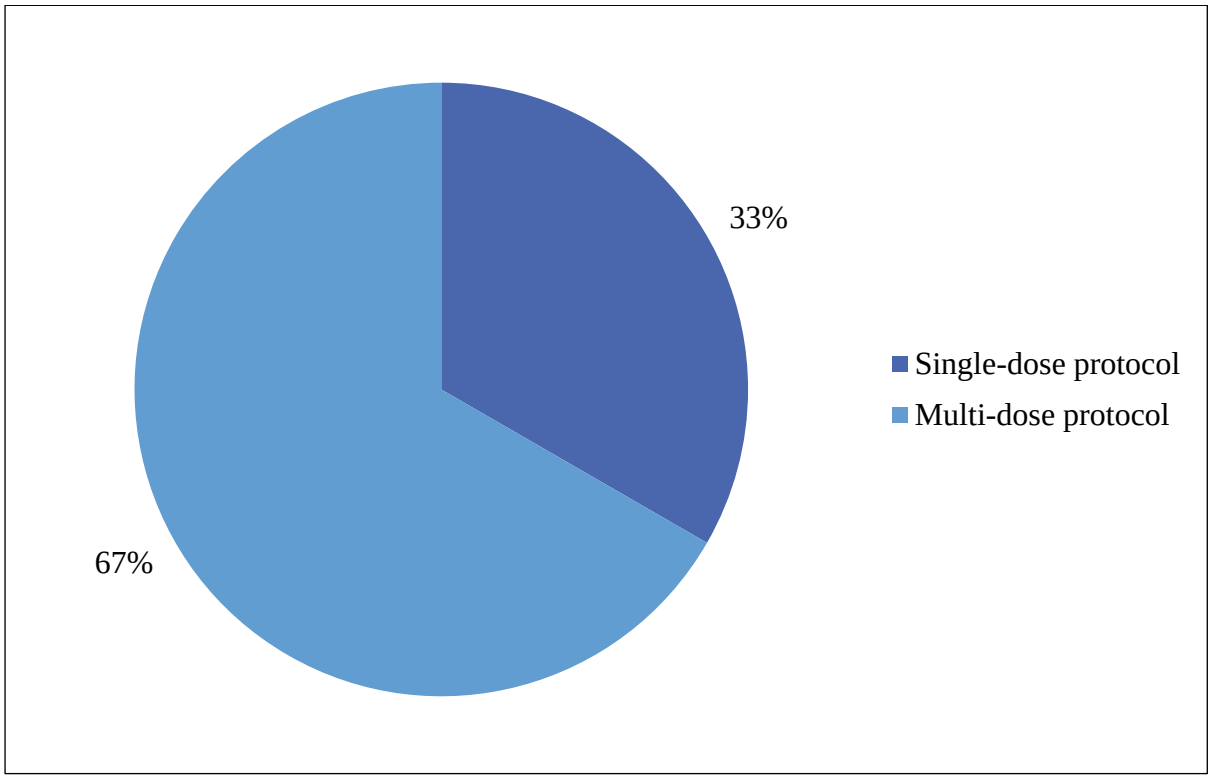


Sixty-two percent of doctors evaluate the response to cetrorelix in their patients by monitoring both serum LH levels and serum P levels.

7. Which type of cetrorelix protocol do you prefer in your patients undergoing IVF treatment?

A. Single-dose protocol

B. Multi-dose protocol

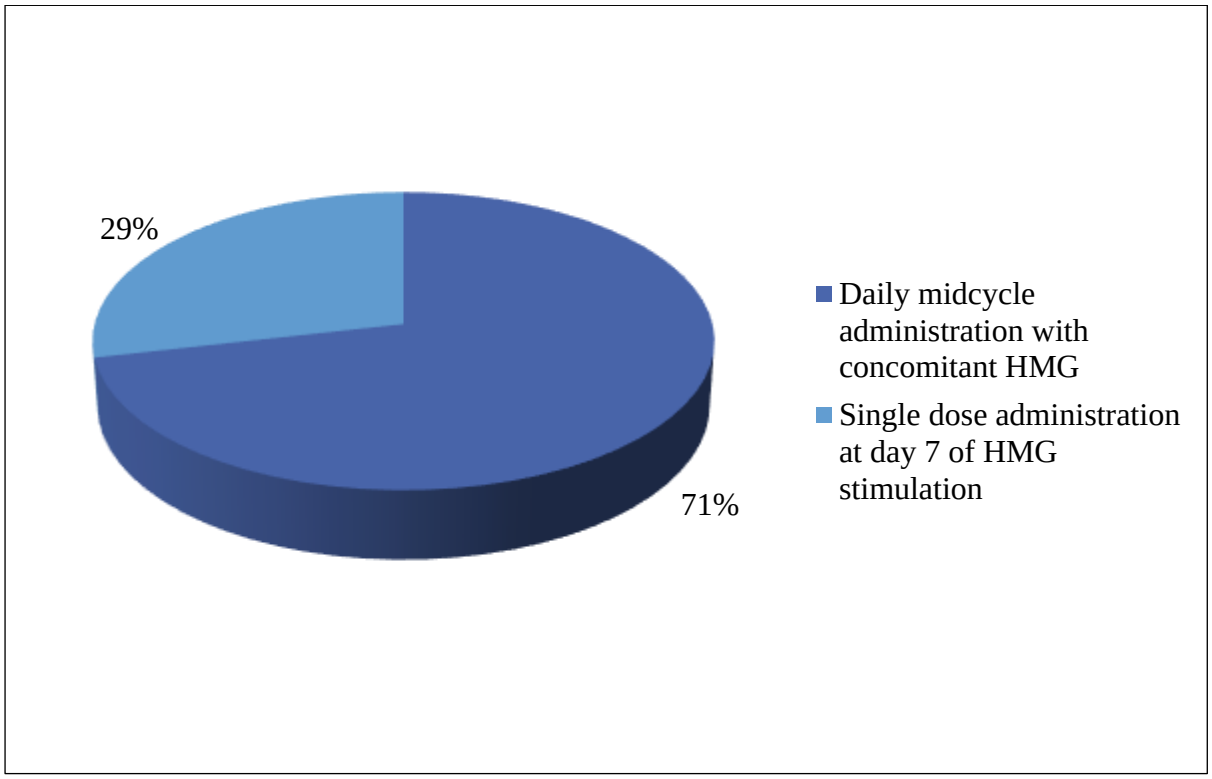


As per 67% of doctors, prefer multi-dose protocol of cetrorelix in their patients undergoing IVF treatment.

8. Which of the following is your preferred technique using Cetrorelix along with HMG on IVF?

A. Daily midcycle administration with concomitant HMG

B. Single dose administration at day 7 of HMG stimulation



According to 71% of doctors, daily midcycle administration with concomitant HMG is the preferred technique when using cetrorelix along with HMG on IVF.

9. In what % of your infertility patients do you use HP-HMG in combination with cetrorelix?

A. 20-30%

B. 31-40%

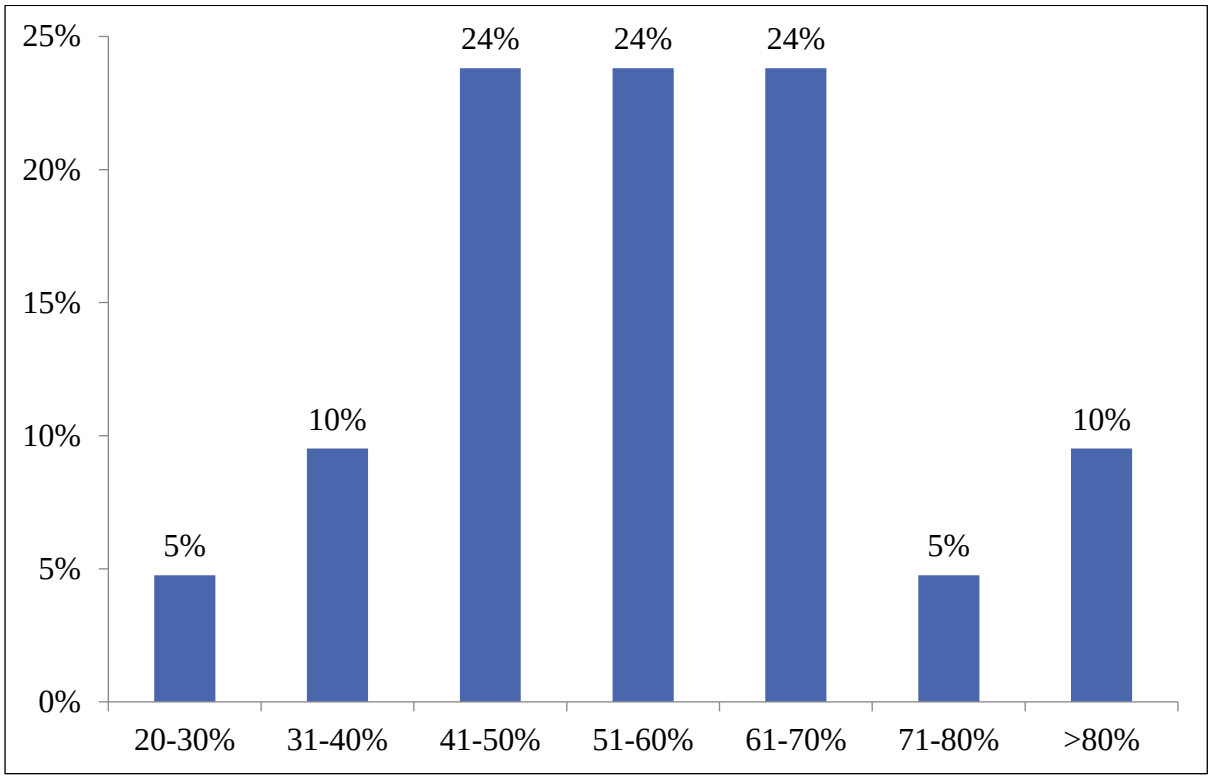
C. 41-50%

D. 51-60%

E. 61-70%

F. 71-80%

G. >80%

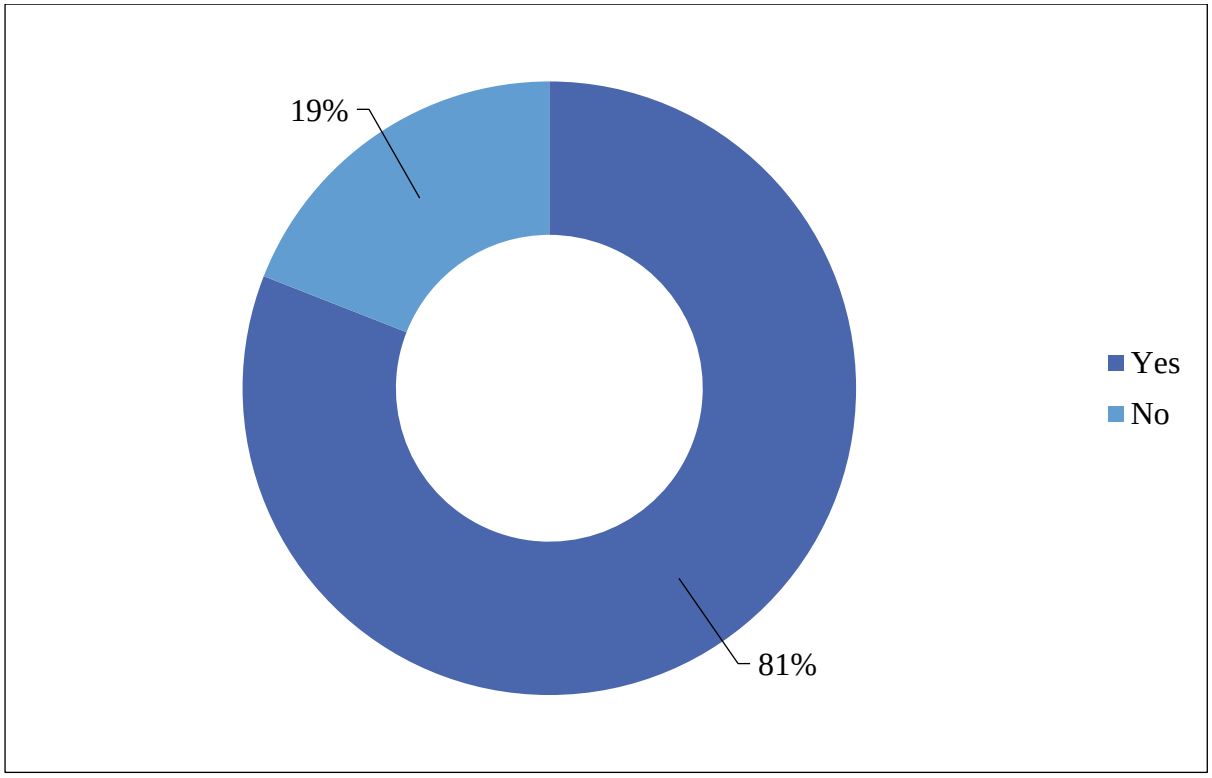


According to 24% of doctors, 41-50% of their infertility patients receive HP-HMG in combination with cetrorelix. Similarly, another 24% of doctors reported that 51-60% of their infertility patients receive this combination, while some other 24% stated that 61-70% of their patients are treated with this combination.

10. In the past, have you used ciscure in your practice?

A. Yes

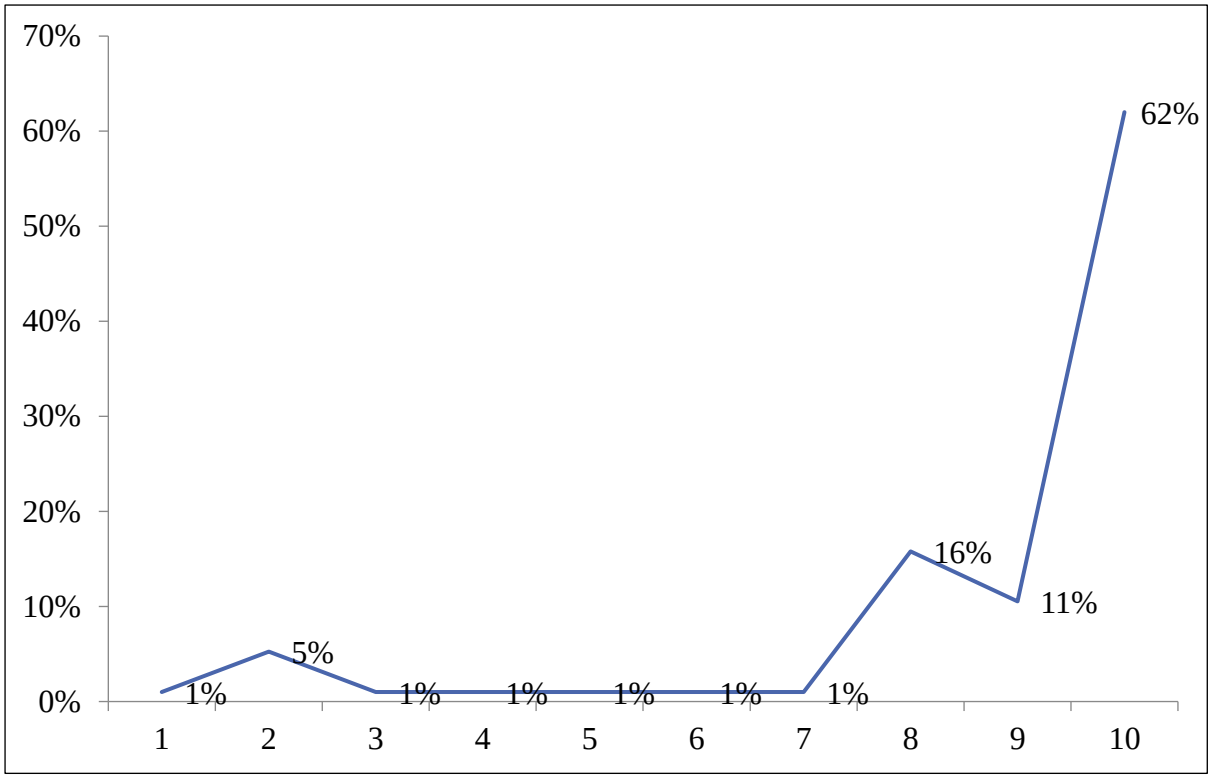
B. No



A majority of 81% of doctors, mentioned that they have used ciscure in their patients.

11. If yes, how would you rate your experience (in terms of efficacy & safety) with ciscure on a scale of 1-10 (1 being worst, 10 being best)?

A. 1
B. 2
C. 3
D. 4
E. 5
F. 6
G. 7
H. 8
I. 9
J. 10



Around 62% of doctors rated their experience with ciscure a 10 on a scale of 1-10, indicating the highest level of efficacy and safety.

Summary

- Around 32% of doctors encounter cases of premature luteinization in 5-10% of their patients.
- According to 67% of doctors, the advantages of adjunctive use of a GnRH antagonist for IVF cycles include reducing the cycle cancellation rate.
- According to 43% of doctors, the advantages of using a GnRH antagonist over a GnRH agonist in IVF cycles include a lower risk for ovarian hyperstimulation syndrome (OHSS).
- A majority of doctors, 86%, agree that an equivalent pregnancy rate is achievable using GnRH antagonist protocols as compared to long GnRH agonist protocols.
- A majority of doctors, 93%, prefer a daily dose of 0.25 mg of cetrorelix for their patients undergoing IVF treatment.
- Sixty-two percent of doctors evaluate the response to cetrorelix in their patients by monitoring both serum LH levels and serum P levels.
- As per 67% of doctors, prefer multi-dose protocol of cetrorelix in their patients undergoing IVF treatment.
- According to 71% of doctors, daily midcycle administration with concomitant HMG is the preferred technique when using cetrorelix along with HMG on IVF.
- According to 24% of doctors, 41-50% of their infertility patients receive HP-HMG in combination with cetrorelix. Similarly, another 24% of doctors reported that 51-60% of their infertility patients receive this combination, while some other 24% stated that 61-70% of their patients are treated with this combination.
- A majority of 81% of doctors, mentioned that they have used ciscure in their patients.
- Around 62% of doctors rated their experience with ciscure a 10 on a scale of 1-10, indicating the highest level of efficacy and safety.

Consultant Opinion

Market opportunities

Identify the increasing demand for effective ovarian stimulation protocols in assisted reproductive techniques (ART) as an opportunity for pharmaceutical companies to develop and market innovative combinations like Cetrorelix and Highly Purified Human Menopausal Gonadotropin (HP-HMG) to enhance fertility outcomes.

Value for healthcare professionals

Provide continued education and training for healthcare professionals on the optimal use of Cetrorelix and HP-HMG in ovarian stimulation protocols for ART, aiming to improve patient outcomes and success rates.

Adverse effect management

Conduct further research and development to minimize the adverse effects associated with ovarian stimulation protocols using Cetrorelix and HP-HMG, ensuring better tolerability and adherence to treatment in ART cycles.

Withdrawal management

Develop guidelines and protocols for the safe withdrawal of medications used in ovarian stimulation protocols, especially in patients experiencing adverse effects or those requiring adjustments in therapy to optimize ART outcomes.

Market positioning

Position the combination of Cetrorelix and HP-HMG as an effective and well-tolerated option for ovarian stimulation in ART, emphasizing its ability to provide optimal follicular development and improve oocyte quality compared to traditional stimulation regimens.

Personalized treatment decision

Encourage healthcare providers to consider individual patient characteristics and ovarian reserve when selecting ovarian stimulation protocols, thereby optimizing ART outcomes and minimizing the risk of complications.

Improving patient outcomes

Collaborate with healthcare providers to develop personalized treatment plans that address not only ovarian stimulation but also the overall ART process, including embryo transfer techniques and supportive care, to enhance pregnancy rates and patient satisfaction.

Innovation and research

Invest in ongoing research and development to explore novel approaches and technologies for ovarian stimulation in ART, including the development of targeted therapies based on patient-specific characteristics and underlying infertility etiologies.

By addressing these aspects, both healthcare professionals and pharmaceutical companies can work together to optimize ovarian stimulation protocols, improve ART success rates, and enhance the overall patient experience in assisted reproduction.

Notes

Notes

Notes

Weston Medical Education Foundation of India
Office No:- 99,9th Floor, Kalpataru Avenue, Opp. ESIC Hospital, Kandivali (East) , Mumbai -400101.
M: 9920154297 | W: www.wmefi.co.in