Introduction

Anovulation is a relatively common cause of infertility, accounting for about 25% of all cases. Ovulatory disorders may be either due to hypothalamic–pituitary ovarian axis dysfunction or other endocrine diseases. Anovulation has been classified by the World Health Organization (WHO) based on the circulating concentrations of gonadotropin and oestrogen into three categories:

- Hypogonadotrophic hypogonadism (WHO group I)
- Normogonadotrophic anovulation (WHO group II)
- Hypergonadotrophic hypogonadism (premature ovarian failure), (WHO group III) ***(Saad, 2007)***.

By far, the most common cause of anovulation is polycystic ovarian syndrome (PCOS, WHO group II), which affects about 6–8% of women of reproductive age and accounts for approximately 80% of all cases of anovulation. Other endocrine causes of WHO Group II ovulatory disorders include obesity, hyperprolactinaemia, hypothyroidism and pituitary–adrenal diseases such as Cushing's syndrome and adult-onset congenital adrenal hyperplasia (CAH) ***(Imani et al., 2002)***.

Successful ovulation induction in anovulatory patients depends on the correct identification of the underlying cause of the ovulatory disorder. Several drugs are available for ovulation induction, the choice of which depends on the cause of anovulation ***(Saad, 2007)***.

Clomiphene citrate (CC) is the traditional drug of choice for ovulation induction in anvoulatory infertile women with normal thyroid functions, prolactin level and no galactorrhea who also exhibit evidence of endogenous estrogen production (oligomenorrhea, estrogenic cervical mucus), serum estradiol > 40 pg/ml, or a normal menstrual response to progestational challenge (WHO group II) ***(Speroff and Fritz, 2005).***

Considering its hypothalamic site of action, it is not surprising that clomiphene is typically ineffective in women with hypogonadotrophic hypogonadism (WHO group I) ***(Speroff and Fritz, 2005).***

Being similar to estrogen, it binds to estrogen receptors for long periods of time, for weeks rather than hours, depleting, receptor concentrations interfering with receptor recycling. It modifies hypothalamic activity by affecting the concentration of intracellular estrogen receptors. So the hypothalamic-pituitary axis will be blind to endogenous level of estrogen so GnRH secretion is activated (negative feedback is diminished), leading to FSH and LH pulse frequency increases in women with normal cycles ***(Speroff and Fritz, 2005).***

At the level of the pituitary, it has direct effect on stimulation of gonadotropin release. At the level of the ovary, it has a direct effect, in the absence of estrogen, acting as estrogen agonist, it enhances FSH stimulation of LH receptors in granulosa cells ***(Speroff and Fritz, 2005)***.

human chorionic gonadotrophin (hCG) is a glycoprotein hormone produced during pregnancy that is made by the developing embryo after conception and later by the syncytiotrophoblast, a part of the placenta ***(Williams et al., 2009).***

Human chorionic gonadotropin promotes the maintenance of the corpus luteum during the beginning of pregnancy, causing it to secrete the hormone progesterone which enriches the uterus with a thick lining of blood vessels and capillaries so that it can sustain the growing fetus***.(Schmidt and Michael, 2009).***

Due to its highly-negative charge, hCG may repel the immune cells of the mother, protecting the fetus during the first trimester ***(McClain, 2010).***

It has also been hypothesized that hCG may be a placental link for the development of local maternal immunotolerance and may facilitate the trophoblast invasion, which is known to expedite fetal development in the endometrium ***(McClain, 2010).***

Because of its similarity to LH, Exogenous hCG has been used commonly as surrogate LH surge to trigger ovulation in clomiphene induced cycles ***(Speroff and Fritz, 2005).***

As ovulation will happen between 38 and 40 hours after a single HCG injection, procedures can be scheduled to take advantage of this time sequence, such as intrauterine insemination or sexual intercourse ***(Homburg and Insler, 2002).***

Also, patients that undergo IVF, in general, receive hCG to trigger the ovulation process, but have a oocyte retrieval performed at about 34 to 36 hours after injection by a few hours before the eggs actually would be released from the ovary ***(Phillips, 2011).***

The addition of an ovulatory dose of hCG has been used as an adjuvant to CC treatment to trigger ovulation when size of the preovulatory follicle reaches 16–25 mm, but there are no randomized controlled clinical trials documenting the efficacy of this approach ***(Homburg and Insler, 2002)***.

Aim of the Work

To evaluate the efficacy of triggering ovulation by Human Chorionic Gonadotropin in patients using clomiphene citrate.

Ovulation Induction

Definition

Ovulation induction refers to the triggering of ovulation then rupture of the preovulatory follicle and release of the oocyte. Ovarian stimulation for anovulatory women aims at restoring normal fertility by generating normo-ovulatory cycles (i.e., to mimic physiology and induce single dominant follicle selection and ovulation ***(Fauser and Macklon, 2004).***

Table (1): Indications for medical induction of ovulation

1- Oligo/anovulation.
2- Empiric therapy.
   - Endometriosis
   - Male factor
   - Unexplained infertility
3- Assisted reproductive technology (ARF)
   - In vitro fertilization (IVF)
   - Gamete intrafallopian transfer (GIFT)
   - Zygote intrafallopian transfer (ZIFT)

(Sozos and Neri, 2003)

Principles of ovulation induction

The aim of induction of ovulation in anovulatory women is to stimulate a single follicle to develop up to the preovulatory stage and subsequently ovulate. This therapeutic goal should be clearly distinguished from two other forms of ovarian stimulation.

First ovulatory women with unexplained infertility may undergo a mild form of ovarian stimulation aimed at producing two or three follicles and an increased chance of fertilization in a given cycle. This treatment, which is frequently combined with intrauterine insemination (IUI). Second, ovarian hyperstimulation may be applied in ovulatory women undergoing IVF treatment, where multi-follicular development is required to produce multiple oocytes. Surgical techniques, such as laparoscopic ovarian drilling, offer an alternative to medical therapies in this context ***(Rizk, 2007)***.

Adverse effects and concerns regarding ovulation induction agents

1. Multiple gestations

The significant increase in the incidence of multiple births in most countries is almost entirely the result of the use of gonadotropins and other agents for induction of ovulation. Among the strategies used are increasing the number of ovarian follicles matured by the administration of exogenous gonadotropin during induction of ovulation and increasing the number of embryos or gametes transferred in cases of in vitro fertilization or gamete transferred in cases of IVF. Each of these strategies is aimed at increasing the percentage of successful pregnancies but also substantially increases the likelihood of multiple gestations. That increase, in turn, drives obstetrical and neonatal charges higher. Many fear that decreasing the number of follicles stimulated or the number of embryos transferred in order to reduce the number of multiple gestations will lead to

lower success rates and an increase in the number of menstrual cycles of treatment needed to achieve a pregnancy. In conclusion, none of the methods suggested to reduce the risk of multiple pregnancies after ovarian stimulation and IUI proved to be effective without reducing the success rate significantly or adding significant cost by conversion to IVF ***(Ratts et al., 2007).***

2. Mild OHSS: (moderate ovarian enlargement is relatively common but does not require active management) ***(Delvigne and Rozenberg, 2002).*** Severe OHSS (massive ovarian enlargement, progressive weight gain, severe abdominal pain, nausea and vomiting, hypovolemia, ascites, and oliguria) ***(Nargund, 2007).***

Table (2): Risk factors for OHSS

- Young age (< 35 years)
- Asthenic habitus
- Polycytic ovary syndrome
- High serum E2 (> 4000 pg/ml)
- Multiple immature and intermediate follcioles
- Necklace" sign on ultrasonography
- Luteal hCG supplementation
- GnRHa protocol
- pregnancy

(Sozos and Neri, 2003)

3. Ovulation induction and cancer

The question of whether treatment with ovulation-inducing drugs increases risk of ovarian tumors or cancer remains unsettled and cannot be summarily dismissed. Certainly, no causal relationship between ovulation-inducing drugs and ovarian cancer has been established***(Calderon-Margalit et al.,2009)***Carrying a

pregnancy to term and using oral contraceptives for 2 or more years have both been demonstrated to significantly reduce overall ovarian cancer risk and could offset any potential increase in risk from fertility therapy ***(Mahdavi et al.,2006)***.

Anti-estrogens

The most widely used anti-estrogen for treating anovulation is clomiphene citrate. The principal indication for clomiphene citrate is the treatment of anovulatory infertility in women with an intact hypophseal-pituitary-ovarian axis. In this role it remains the first-line therapy ***(Usadi and Fritz, 2008)***.

Tamoxifen is, like clomiphene, a nonsteroidal selective estrogen receptor modulator. Primarily developed for and used in the treatment of breast cancer, it has also been used in ovulation induction for many years. In contrast to clomiphene, tamoxifen only contains the zu-isomer and appears to be less anti-estrogenic at the uterine level ***(De paula et al., 2011)***. The possible advantages of tamoxifen over clomiphene include beneficial effects on cervical mucus and an agonistic effect at the endometrium. Although endometrial thickening may be observed on ultrasound monitoring, histologic studies indicate that this may be due to edema and enlargement of stromal cells, rather than a purely estrogenic proliferative effect. Uncontrolled studies have suggested that tamoxifen may be a safe and efficacious alternative to clomiphene ***(Steiner et al., 2005)***.

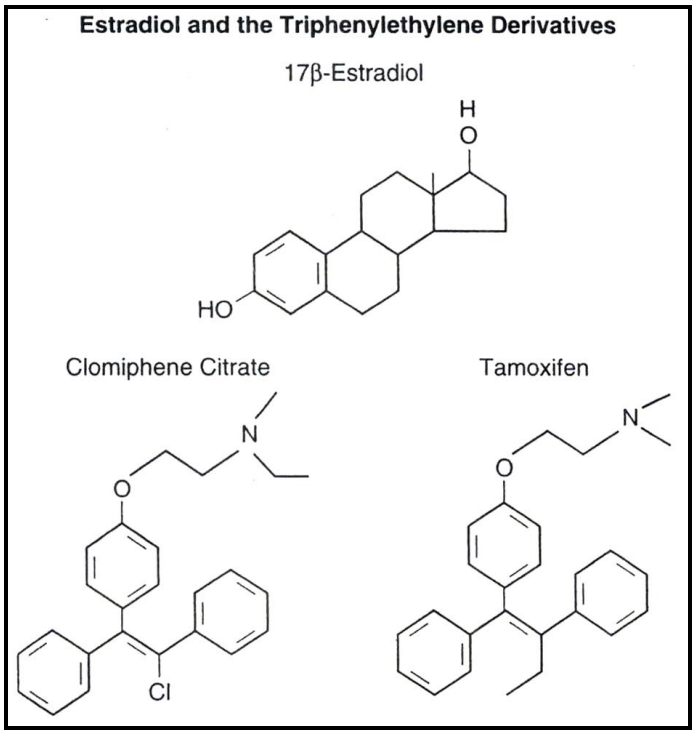

Figure (1): Estradiol, Clomiphene citrate and Tamoxifen
(Fauser and Macklon, 2004)

Insulin sensitizing drugs

Insulin resistance and hyperinsulinemia are recognized features of PCOS. Hyperinuslinemia contributes significantly to hyperandrogenism and chronic anovulation. It is logical that agents that improve insulin action in the body (insulin sensitizers) will help infertile PCOS women with insulin resistance to achieve ovulation by lowering their hyperinsulinemia ***(Glueck et al., 2002).***

The most extensively studied insulin-sensitizing drug used in the treatment of anovulation is metformin, metformin (dimethylbiguanide) is an orally administered drug used to lower blood glucose concentrations in patients with type 2 diabetes mellitus. It is antihyperglycemic in action and increases sensitivity

to insulin by inhibiting hepatic glucose production and by increasing glucose uptake and utilization in muscle. These actions result in reduced insulin resistance, lower insulin secretion, and reduce serum insulin levels ***(Barbeiri, 2007)***.

Other insulin-sensitizing agents such as the thiazolidinediones (troglitazone, rosiglitazone, and pioglitazone) and chiroinositol have been used in small clinical trials to achieve ovulation in PCOS women with insulin resistance. However, because of the higher safety profile, longer clinical experience, and lower cost of metformin, the other insulin sensitizers should be reserved for cases resistant to metformin treatment ***(Mitwally and Casper, 2011)***.

It is necessary to stress the importance of other non-pharmacologic approaches to improve insulin action and reduce insulin resistance, including dietary factors, weight loss and exercise. The use of insulin-sensitizing agents should be as an adjuvant to these nonpharmacologic approaches ***(Mitwally and Casper, 2005)***.

Insulin sensitizers may work through other mechanism to achieve ovulation. One such mechanism is a direct effect on ovarian steroidogenesis. There is evidence that the insulin sensitizers, metformin and thiazolidinediones, can modulate steroid production through direct suppression of steroidogenesis enzymes. Troglitazone was found to inhibit progesterone production by human luteinized granulosa cells and to reduce

androgen production by rat theca cells ***(Mitwally and Casper, 2011)***.

Recent suggestions that metformin therapy continued into early pregnancy may reduce the risk of miscarriage in women with PCOS have not been substantiated. Recent studies have suggested that metformin induces a rise in circulating levels of glycodelin. This protein is thought to play an important role in implantation, possibly by inhibiting endometrial immune response to the embryo ***(Nestler, 2008)***.

Researchers recommend that oral metformin be started at 500 mg daily, titrating up to 500 mg three times daily over seven to 10 days. Depending on response, this dose may be increased, to 1000 mg twice daily the optimal duration of treatment has not been established; however, most studies reporting a beneficial effect from metformin have shown this outcome within two to four months ***(Fauser and Macklon, 2004)***.

Adverse effects and complications:

Metformin has been used for many years for the treatment of diabetic patients and appears to be safe for long-term use. The drug has not been associated with an increased risk of congential abnormalities in diabetic women who subsequently became pregnant ***(Fauser and Macklon, 2004)***.

Rarely lactic acidosis may occur, if hepatic or renal disease is present. The main adverse effects of metformin are nausea and diarrhea, which may occur in 10% to 25% of patients and contribute to the weight loss effects observed with metformin.

However, the relatively greater costs per cycle compared to clomiphene citrate, as well as the complexity of metformin treatment and the frequency of significant gastrointestinal side effects (e.g., nausea, vomiting, diarrhea) made others prefer to reserve metformin treatment for those who first prove resistant to clomiphene citrate ***(Palomba et al., 2009)***.

Troglitazone is a thiazolidinedione, a group of insulin lowering oral drugs. A large randomized study suggested that troglitazone may be an effective ovulation induction agent. However, reports of fatal liver toxicity have led to its withdrawal by the U.S. Food and Drug Administration ***(Fauser and Macklon, 2004)***.

Aromatase inhibitors

Aromatase description

Aromatase catalyzes the rate-limiting step in the production of estrogens, that is, the conversion of androstenedione and testosterone via three hydroxylation steps to estrone and estradiol. Aromatase is a good target for selective inhibition because estrogen production is a terminal step in the biosynthetic sequence ***(Mitwally and Casper, 2000).***

Types of aromatase inhibitors

Aromatase inhibitors have been classified in different ways: into steroidal and nonsteroidal and according to the stage of development, into first-, second-, and third-generation groups ***(Mitwally and Casper, 2000)***.

Aromatase inhibitors block the conversion of androstenedione and testosterone to E3 and E2, respectively. This increases gonadotropin secretion, resulting in stimulation of ovarian follicles. Aromatase inhibitors have been in clinical use for more than 20 years, primarily in the treatment of post-menopausal patients with advanced breast cancer ***(Lee and Ledger, 2011)***.

Local effect at the ovary has also been proposed, in which sensitivity to FSH is increased by blocking the conversions of androgens to estrogens because an accumulating intraovarian androgens may increase FSH-receptor gene expression ***(Sozos and Neri, 2003)***.

The first aromatase inhibitor to be used clinically was aminoglutethimide. The third-generation aromatase inhibitors commercially available include two nonsteroidal preparations, anastorozole and letrozole, and a steroidal agent, exemestane ***(Mitwally and Casper, 2000).***

And at doses of 1 to 5 mg/day, inhibit estrogen levels by 97% to more than 99% down. Aromoatase inhibitors are completely absorbed after oral administration, with a mean terminal half-life of approximately 45 hours (with clearance from the systemic circulation mainly by the liver ***(Mitwally and Casper, 2005)***.

Letrozole has been studied at doses of 2.5-7.5 mg per day, and anastrozole at doses of 1 mg, both given for 5 days early in the cycle, and both have been used to induce ovulation

in normogonadotropic, normoprolactinemic anovulation ***(Abu Hashim et al., 2010)***.

Table (3): Potential advantages of using an aromatase inhibitor for induction of ovulation

➢ High pregnancy rates
➢ Monofollicular ovulation in most anovulatory patients.
➢ High safety due to short half-life and few adverse effects.
➢ Reduced rate of multiple pregnancy
➢ Reduced risk of severe ovarian hyperstimulation syndrome.
➢ Reduced FSH dose required for controlled ovarian stimulation.
➢ Improved response to FSH in poor responders.
➢ Low cost of treatment (average, $ 30-$100 per cycle)
➢ Convenience of administration oral route, different regimens, including single-dose regimen

(Sozos and Neri, 2003)

Nonsteroidal aromatase inhibitors are generally well tolerated. The main adverse events observed are hot flushes, gastrointestinal events (nausea and vomiting), and leg cramps.These adverse effects were observed in older women with advanced breast cancer who were given the aromatase inhibitors on a daily basis over several months. Fewer adverse effects would be expected in the usually healthy younger women administered a short course of aromatase inhibitor for induction of ovulation ***(Mitwally and Casper, 2005)***.

Recent data has raised concern that letrozole may be associated with an increased risk of congenital abnormalities ***(American Society for Reproductive Medicine, 2006).***