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INFERTILITY AND NEW TREATMENT MODALITIES

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Infertility and Current Treatment Modalities

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Abstract

This book is aim to give new information about infertility and new treatment modalities. In the fi rst part of the book, infertility reasons are also discussed. In the second part, the new treatments of infertility and the parts of assisted reproductive technology are explained. And fi nally, recurrent implantation failure and recurrent pregnancy loss are described. This book off ers new strategies to reproductive endocrinology doctors for their training.

Keywords

Infertility; Assisted reproductive technology; Reproduction; Endocrinology

Introduction

Infertility and Reasons

Infertility is defined as one year of regular unprotected intercourse without conception because fertility in women is known to decline dramatically with age, we advice to evaluate and treat women aged 35 years or older after 6 months of unprotected sex. Couples have approximately 85%-90% conceive within 1 year. Therefore, infertility affects approximately 10%-15% of couples and represents an important part of infertility clinics. Cycle fecundability is the probability that a cycle will result in pregnancy and fecundity is the probability that a cycle will result in a live birth [1]. Fecundity is the result of a process that has many steps.

The steps to get pregnant:

- An egg from one ovary (quality and reserve of ovary)
- Sperm must fertilize the egg (concentration, motility, morphology of sperm)
- The embryo must go through fallopian tube (normal tubal physiology)
- The embryo must attach to the inside of the uterus (implantation)

Infertility may result from a problem with any or several of these steps.Therefore, pregnancy needs functioning of ovary, sperm, fallopian tubes and uterus. We analyzes the infertile couples according to this parameters.

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So, main analysis of the infertile couples are:

- Transvaginal ultrason and hormonal parameters of men and women
- Semen analysis
- Hysterosalpingography (HSG)

The most important factor to affect the fertility is the age of women. Advanced age is associated with decreased fertility. Fertility rates are 4%-8% lower in women aged 25-29 years, 15%-19% lower in those aged 30-34 years, 26%-46% lower in women aged 35-39 years, and as much as 95% lower for women aged 40-45 years [2]. The risk of miscarriage was lowest among women aged 25-29 (9.8%), with the absolute lowest risk at age 27 (9.5%), and the highest risk at age 45 and over (53.6%) [3,4] Figure 1.

The most important factor of miscarriages in Advanced Maternal Age (AMA) women is increased the prevelance of aneuploid oocytes and this prevelance is especially increase more than 40 years and reaching 100% after age 45 [4] Figure 2.

Additionally ovarian reserve is important for the reproduction and aging is negatively effect on ovarian reserve. So ovarian reserve is declined with aging and serum ovarian reserve tests are used as the marker of Diminished Ovarian Reserve (DOR).

Basal FSH and Estradiol (E2) concentrations are simple indicators that suggest ovarian reserve with the lowest sensitivity. Especially more than 10 IU/L levels of FSH predicts DOR. Decreased E2 levels with increased FSH levels is significant. Elevated levels of E2 more than 80 pg/mL shows decreased ovarian response but diagnostic performance is low. Therefore, AMH and antral follicle count is important to diagnose DOR [6].

AMH is produced by the granulosa cells of preantral and small antral follicles, beginning when primordial follicles start developing into primary follicles and ending when early antral follicles reach a diameter of 2 mm-6 mm. The fertility of women may be determined by the ovarian reserve tests including counting the number of ovarian

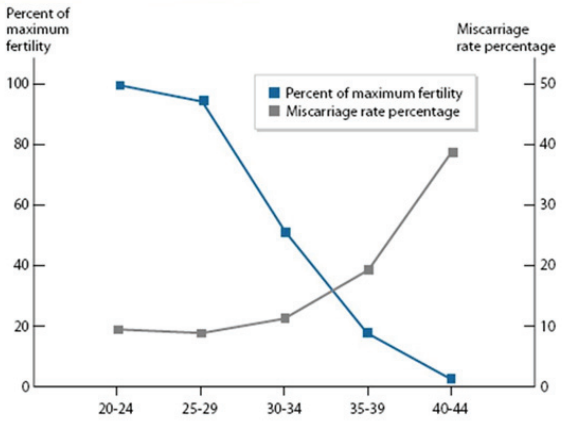

Figure 1: Correlation of women age to fertilty and miscarriage.

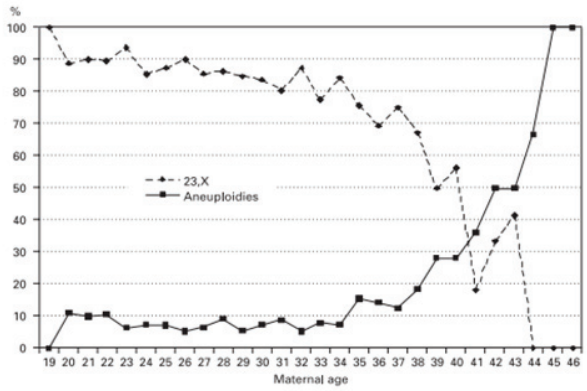

Figure 2: Ovarian aging and aneuploidy rates [5].

follicles and measuring the level of AMH. The tests are the most widely used parameters in infertility clinics [7].

Normal AMH levels are between 1.5 ng/mL and 4.0 ng/mL (Table 1). AMH has been examined in the general IVF population and in populations of women at low or high risk for DOR. Decreased AMH levels have been associated with poor response to ovarian stimulation and low oocyte yield, embryo quality, and pregnancy rates [1].

Table 1: AMH levels and prediction of ovarian reserve.

Interpretation (Women under age 35)	AMH Blood Level
High (Often PCOS)	Over 4 ng/ml-0 ng/ml
Normal	1.5 ng/ml-4.0 ng/ml
Low Normal Range	1.0 ng/ml-1.5 ng/ml
Low	0.5 ng/ml-1.0 ng/ml
Very Low	Less than 0.5 ng/ml

AFC is total number of antral follicles measuring 2 mm-10 mm in both ovaries and provides an indirect but useful measure of ovarian reserve. A low AFC has high specificity for predicting poor response to ovarian stimulation and treatment failure [1].

Ovarian reserve also negatively effects with endometriosis, smoking, excessive alcohol use, extreme weight gain or loss, excessive physical or emotional stress, chemotherapy or radiotherapy history, and ovarian surgery history [6].

Most common, objective, and reliable test of ovulatory function test is serum progesterone measurement. A progesterone concentration less than 3 ng/mL implies anovulation. The serum progesterone level should be drawn approximately 1 week before the expected onset of menses, when the concentration is at or near its peak. However, popular practice in the clinics is the measurement on cycle day 21 is not always the best time to measure the serum progesterone concentration.

The probability for achieving a live birth without treatment decreases with increasing age and duration of infertility. So the prognosis for success without treatment is relatively poor more than 3 years of infertility duration.

Infertility reasons summarized in (Figure 3). Causes of infertility are 35% tubal and pelvic pathology, 35% male problems, 15% unexplained problems and 15% ovulatory dysfunction.

Infertility reasons of women are also summarized in Figure 4. Infertility reasons are 25% Ovulatory disorders, 15% endometriosis, 12% pelvic adhesions, 11% tubal blockage, 11% other tubal abnormalities, 10% unexplained, 7% hormonal problems.

Polycystic Ovary Syndrome (PCOS)

Polycystic Ovary Syndrome (PCOS) is the most common endocrinopathy affecting reproductive aged women, with a prevalence of between 8%-13% depending on the population and as high as 15% when the broader Rotterdam criteria are applied. In PCOS, ovaries contain many cystic follicles that are associated with chronic anovulation and overproduction of androgens. The cystic follicles exist presumably because the eggs are not expelled at the time of ovulation. Symptoms may include irregular menstrual periods, obesity, hirsutism, and infertility [8].

According to the Rotterdam criteria, a clinical diagnosis of PCOS requires that a patient present with two of the following symptoms: Oligo-ovulation or anovulation. Hyperandrogenism, clinical (including signs such as hirsutism) or biological (including a raised free androgen index or free testosterone).

They determined that an ovarian volume >10 mL provided excellent specificity for PCOS in a majority of studies and used 12 or more follicles of 2 mm-9 mm as the follicle number with the best sensitivity and specificity to distinguish PCOS [8]. However, with improvement in ultrasound technology it was recognized that the criteria defining PCOM needed to evolve to better distinguish the

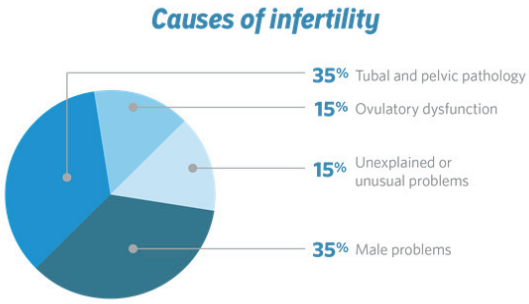

Figure 3: Infertility causes.

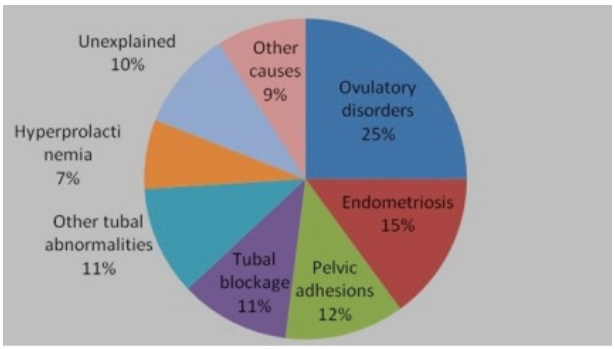

Figure 4: Infertility reasons of women.

ovarian morphology in women with PCOS from that in women who do not have irregular menses and hyperandrogenism [8]. A more recent review of the literature increased the follicle number threshold to 20 follicles to provide better specificity for PCOS [9].

For the infertility treatment:

- Diet with reduced glycemic load may be beneficial in alleviating hyperinsulinemia and its metabolic consequences. 5% decrease of body weight might be clinically meaningful
- Several studies have examined combination therapy of diet and exercise
- Obesity adversely affects reproduction and is associated with anovulation, pregnancy loss, and late-pregnancy complications
- Obesity within PCOS is associated with failure of infertility treatment. Weight loss before infertility treatment improves ovulation rates in women with PCOS but there are limited data that it improves fecundity or lowers pregnancy complications
- Experience from other areas of medicine suggests lifestyle modifications as the first-line treatment of obesity in PCOS [10]
- Clomiphene Citrate (CC) remains the treatment of first choice for induction of ovulation in anovulatory women with PCOS. The starting dose of CC should be 50 mg/day (for 5 days) and the recommended maximum dose is 150 mg/day. Treatment generally should be limited to six (ovulatory) cycles
- Aromatase inhibitors, letrozole appears to be as effective as CC for induction of ovulation
- There is good evidence that metformin alone versus placebo increases the ovulation rate in women with PCOS. There is insufficient evidence to suggest that metformin alone increases pregnancy rates or live-birth rates compared with placebo [11]
- There is insufficient evidence to suggest that metformin alone increases pregnancy or live-birth rates compared with letrozole alone. However, there is fair evidence based on one well-designed trial in support of letrozole for ovulation induction. Therefore, letrozole is a reasonable first-line agent for ovulation induction in PCOS patients [11]
- There is good evidence that metformin in combination with CC improves ovulation and clinical pregnancy rates but does not improve live-birth rates compared with CC alone in women with PCOS. There is fair evidence from one RCT that pretreatment with metformin for at least 3 months followed by the addition of another ovulation-inducing drug increases live-birth rate
- There is fair evidence that overall pregnancy rates are not different with CC-metformin, CC-LOD, or LOD alone in women with CC-resistant PCOS
- Ovulation induction using gonadotropins is effective with cumulative live birth rates of approximately 70%. Low-dose protocols (37.5 IU/day-75 IU/day), which have essentially replaced the original conventional protocol is firstly recommended and hCG is administered in the presence of one or two follicles >16 mm to minimize the risk of multiple pregnancies in women with PCOS
- Laparoscopic Ovarian Drilling (LOD) should be performed when laparoscopy is indicated, this procedure is typically effective in approximately 50% of cases
- Finally, *In Vitro* Fertilization (IVF) or Intracytoplasmic Sperm Injection (ICSI) is the third-line treatment and is recommended when the previous interventions fail

In summarized:

First line infertility treatment; CC or CC+metformin or letrozole, second line infertility treatment; gonadotropins or LOD, third line infertility treatment; IVF or ICSI.

In a recent study group; according to the international evidence-based guidelines, including 166 recommendations and practice points, addressed prioritized questions to promote consistent, evidence-based care and improve the experience and health outcomes of women with PCOS [12]. According to this study we only added the evidence based recommendations in this chapter.

- Calculated free testosterone, free androgen index or calculated bioavailable testosterone should be used to assess biochemical hyperandrogenism in the diagnosis of PCOS
- Serum AMH levels should not yet be used as an alternative for the detection of PCOM or as a single test for the diagnosis of PCOS
- The Combined Oral Contraceptive Pills (COCP) alone should be recommended in adult women with PCOS for management of hyperandrogenism and/or irregular menstrual cycles. The COCP alone should be considered in adolescents with a clear diagnosis of PCOS for management of clinical hyperandrogenism and/or irregular menstrual cycles
- In combination with the COCP, metformin should be considered in women with PCOS for management of metabolic features where COCP and lifestyle changes do not achieve desired goals. In combination with the COCP, metformin could be considered in adolescents with PCOS and BMI ≥ 25 kg/m2 where COCP and lifestyle changes do not achieve desired goals. In combination with the COCP, antiandrogens should only be considered in PCOS to treat hirsutism, after 6 months or more of COCP and cosmetic therapy have failed to adequately improve symptoms
- Metformin in addition to lifestyle, could be recommended in adult women with PCOS, for the treatment of weight, hormonal and metabolic outcomes. Metformin in addition to lifestyle, should be considered in adult women with PCOS with BMI ≥ 25 kg/m2 for management of weight and metabolic outcomes. etformin in additional to lifestyle, could be considered in adolescents with a clear diagnosis of PCOS or with symptoms of PCOS before the diagnosis is made
    - ◊ Adverse effects including gastrointestinal side effects that are generally dose dependent and self-limiting need to be the subject of individualized discussion
    - ◊ Starting at a low dose with 500 mg increments 1-2 weekly and extended release preparations may minimize side effects

◊ Metformin use appears safe long term based on use in other populations, however ongoing requirement needs to be considered and use may be associated with low vitamin B12 levels

- Where COCPs are contraindicated or poorly tolerated, in the presence of other effective forms of contraception, antiandrogens could be considered to treat hirsutism and androgen-related alopecia
- Inositol should currently be considered an experimental therapy in PCOS but further studies needed
- Letrozole should be considered first line pharmacological treatment for ovulation induction in women with PCOS with anovulatory infertility and no other infertility factors to improve ovulation, pregnancy and live birth rates
- Clomiphene citrate could be used alone in women with PCOS with anovulatory infertility and no other infertility factors to improve ovulation and pregnancy rates
- Metformin could be used alone in women with PCOS with anovulatory infertility and no other infertility factors to improve ovulation, pregnancy and live birth rates although women should be informed that there are more effective ovulation induction agents
- Clomiphene Citrate could be used in preference when considering Clomiphene Citrate or Metformin for ovulation induction in women with PCOS who are obese (BMI is ≥ 30 kg/m2) with anovulatory infertility and no other infertility factors
- If Metformin is being used for ovulation induction in women with PCOS who are obese (BMI ≥ 30 kg/m2) with anovulatory infertility and no other infertility factors. Clomiphene Citrate could be added to improve ovulation, pregnancy and live birth rates
- Clomiphene Citrate could be combined with Metformin rather than persisting with clomiphene citrate alone in women with PCOS, who are Clomiphene Citrate-Resistant (CCR) (Figure 5) with anovulatory infertility and no other infertility factors to improve ovulation and pregnancy rates

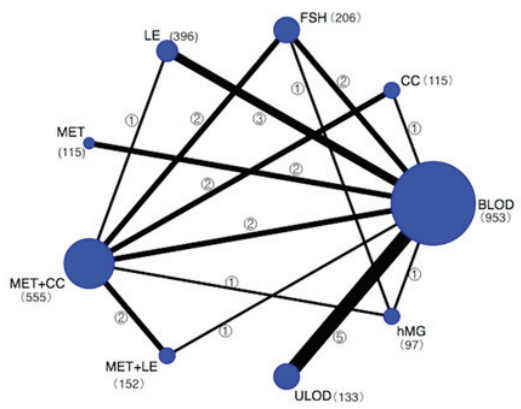

Figure 5: Clomiphene Citrate-resistant alternative treatments (LE:Letrozole, FSH or hMG: Gonadotropins, MET: Metformin, BLOD: Bilateral LOD, ULOD: Unilateral LOD) [13].

- Clinical consensus suggests that gonadotropin therapy (hMG or FSH), aromatase inhibitors (usually letrozole), LOD and adding metformin are common options for CCR patients. It is still challenging to induce mono-ovulation with gonadotropin therapies even under careful monitoring, in order to avoid multiple pregnancies and OHSS [13]. Although there was still low-quality evidence based the results of the ranking probabilities showed that apart from hMG and FSH, metformin+letrozole was potentially more effective than other treatments [13]
- Gonadotrophins could be used as second line pharmacological agents in women with PCOS who have failed first line oral ovulation induction therapy and are anovulatory and infertile with no other infertility factors. Gonadotrophins could be considered as first line treatment in the presence of ultrasound monitoring following counseling on cost and potential risk of multiple pregnancy in women with PCOS with anovulatory infertility and no other infertility factors. Gonadotrophins, where available and affordable should be used in preference to clomiphene citrate combined with metformin therapy for ovulation induction in women with PCOS with anovulatory infertility, clomiphene citrate-resistance and no other infertility factors to improve ovulation pregnancy and live birth rates
- Gonadotrophins with the addition of metformin could be used rather than gonadotrophin alone in women with PCOS with anovulatory infertility, clomiphene citrate-resistance and no other infertility factors to improve ovulation, pregnancy and live birth rates. Either gonadotrophins or laparoscopic ovarian surgery could be used in women with PCOS with anovulatory infertility, clomiphene citrate-resistance and no other infertility factors, following counseling on benefits and risks of each therapy
- Aparoscopic ovarian surgery could be second line therapy for women with PCOS, who are clomiphene citrate resistant with anovulatory infertility and no other infertility factors
- A gonadotrophin releasing hormone antagonist protocol is preferred in women with PCOS undergoing an IVF ± ICSI cycle to reduce the duration of stimulation total gonadotrophin dose and incidence of Ovarian Hyperstimulation Syndrome (OHSS)

Ovarian Hyperstimulation Syndrome (OHSS)

In our clinic all embryo freeze protocol is firstly chosen for all of the PCO women undergoing an ICSI cycle and also Gonadotrophin Releasing Hormone antagonist protocol is preferred in women with PCOS undergoing an ICSI cycle and a Gonadotrophin Releasing Hormone aganist trigger to reduce the duration of stimulation, total gonadotrophin dose and incidence of Ovarian Hyperstimulation Syndrome (OHSS).

Ovarian Hyperstimulation Syndrome (OHSS) is an uncommon but serious complication associated with controlled ovarian stimulation during Assisted Reproductive Technology (ART). Moderate-to-severe OHSS occurs in approximately 1%-5% of cycles [14].

OHSS classification is summarized in Table 2.

Table 2: OHSS classification.

OHSS Stage	Clinical Feature	Laboratory Feature
Mild	Abdominal Distension/Discomfort Mild nausea/Vomiting Mild dyspnea Diarrhea Enlarged Ovaries	No important alterations
Moderate	Mild Feature Ultrasonographic Evidence of ascites	Hemoconcentration (Hct>41%) Elevated WBC (>15,000 ml)
Severe	Mild and moderate Feature Clinical evidence of ascites Hydrothorax Severe dyspnea Oliguria/anuria Intractable nausea/Vomiting Low blood/central venous pressure Pleural effusion Rapid Weight gain(>1 kg in 24 h) Syncope Severe Abdominal Pain Venous thrombosis	Severe Hemoconcentration (Hct>55%) WBC>25,000 ml CrCl<50 ml/min Cr>1.6 mg/dL Na^+<5 mEq/L K^+<5 mEq/L Elevated Liver enzymes
Critical	Anuria/acute renal failure Arrhythmia Thromboembolism Pericardial effusion Massive hydrothorax Arterial thrombosis Adult respiratory Distress syndrome Sepsis	Worsening of findings
Note: Hct: Hematocrit; WBC :White Blood Cell; CrCl: Creatinine Clearance; Cr: Creatinine; Na: Sodium; K: Potassium		

Prevention of OHSS:

- There is good evidence to support the use of ovarian stimulation protocols using GnRH antagonists in order to reduce the risk of OHSS
- There is good evidence that metformin decreases the risk of OHSS risk in PCOS patients
- There is insufficient evidence to recommend coasting for the prevention of OHSS. Coasting is the practice of withholding gonadotropins at the end of controlled ovarian stimulation for up to 4 days to decrease OHSS risk
- There is good evidence to recommend the use of a GnRH agonist to trigger oocyte maturation prior to oocyte retrieval in order to reduce the risk of OHSS. There is good evidence that live-birth rates are lower in fresh autologous cycles after GnRH trigger
- There is good evidence that dopamine agonist administration starting at the time of hCG trigger for several days reduces the incidence of OHSS. Cabergoline 0.5 mg/day after the day of trigger for 8 days in women at high risk (estradiol >4,000; >20 follicles)
- Frozen-thawed embryo transfer is one of the important ways to prevent OHSS in high-risk patients
- In patients with PCOS after the day of Oocyte Pick Up (OPU) GnRH antagonist was administered in the 3 or 5 days to prevent early OHSS

Early OHSS presented 3-7 days due to controlled ovarian induction, whereas late OHSS was identified 12-17 days after the ovulatory dose of hCG in women due to pregnancy.

Ovulation Induction Protocols

This diagram (Figure 6) illustrates a basic ovulation induction cycle using Clomiphene or Gonadotropin to increase follicular production. Note that the medications to increase follicular development are given early in the cycle; clomiphene on cycle days 3 through 7, 4 through 8 or 5 through 9 or gonadotropins beginning day 3 through day 10 or 11 until the leading follicle measures at least 16 mm. Ovulation release occurs when Human Chorionic Gonadotropin (hCG) or recombinant (a synthetic) hCG is given. Sperm exposure by Intrauterine Insemination (IUI) or intercourse could occur 36 hours-40 hours after hCG. Progesterone can be given to help the uterus be more receptive to the embryo.

Low dose step-up protocol is also prefered in our clinic especially for PCOS (Figure 7).

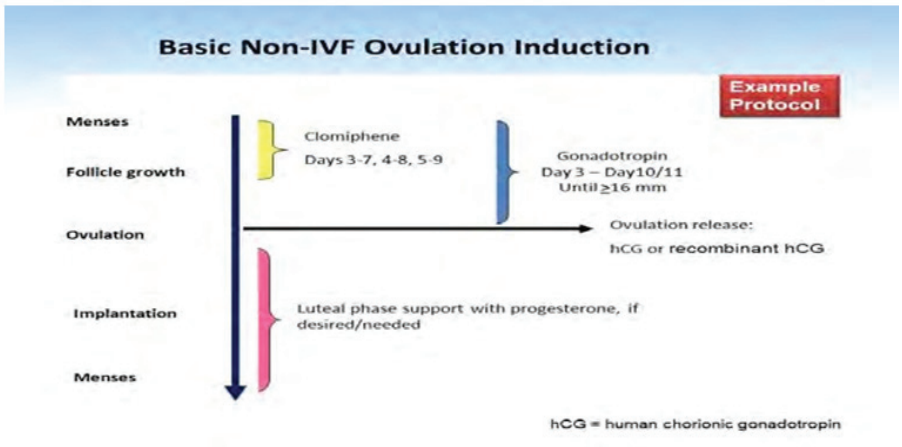

Figure 6: Basic ovulation induction for non-IVF.

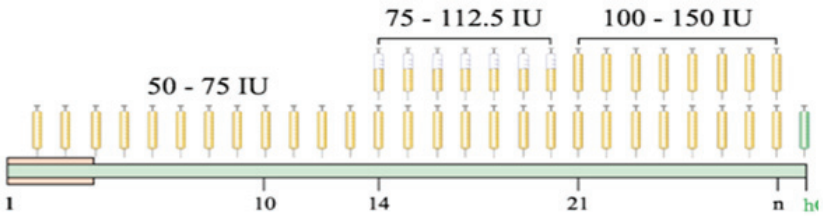

Figure 7: Low dose step up protocol.

Endometriosis

Endometriosis is a common chronic disease. Although women may be asymptomatic most women typically present with pelvic pain, infertility or an adnexal mass. 30% to 50% of women with endometriosis are infertile and infertile women are 6 to 8 times more likely to have endometriosis than fertile women [14].

Endometriosis negatively affected the ovarian reserve and distorted pelvic anatomy. Endometriosis also altered pertioneal function and IgG and IgA antibodies and lymphocytes may be increased in the endometrium of women with endometriosis. These abnormalities may alter endometrial receptivity and embryo implantation. Reduced endometrial expression of $\alpha_v\beta_3$ integrin (a cell adhesion molecule) during the time of implantation has been described in some women with endometriosis [14]. Infertility in women with endometriosis may be related to alterations within the follicle, poor oocyte quality and decreased endometrial receptivity. ASRM staging system is also summarized in Table 3. When endometrioma has been seen on ultrasonography, the stage is IV.

ASRM's classification of endometriosis bases the determination of the stage or degree of endometrial involvement on a weighted point system at the time of laparoscopy. The number, size and location of endometrial implants, endometriomas and/or adhesions are noted.

Table 3: Endometriosis staging according to ASRM [15].

Site	Endometriosis	Lession<1 cm, points	Lession=1 cm-3 cm, points	Lession>1 cm-3 cm, points
Peritoneum	Superficial Deep	1 2	2 4	4 6
Ovary Right	Superficial Deep	1 4	2 16	4 20
Left	Superficial Deep	1 4	2 16	4 20
Posterior culdesac Obliteration	Partial Obliteration 4 Points Complete Obliteration 40 Points			
Site	**Adhesion Type**	**Site<1/3 Enclosed, points**	**Site=1/3-2/3 Enclosed, points**	**Site>2/3 Enclosed, points**
Ovary Right	Filmy Dense	1 4	2 8	4 16
Left	Filmy Dense	1 4	2 8	4 16
Tube Right	Filmy Dense	1 4	2 8	4 16
Left	Filmy Dense	1 4	2 8	4 16

Note: Stage of endometriosis is determined by the total no. of points assigned to endometriotic lesions and adhesion. Stge I (Minimal) 1-5 points, Stge II (Mild) 6-15 points, Stage III (Moderate) 16-40 points, Stage IV (severe)>40 points.

Infertility treatment of Endometriosis:

- Medical therapy is also effective for relieving pain associated with endometriosis, there is no evidence that medical treatment of endometriosis improves fertility
- Hormonal treatment does not improve the fecundity of infertile women with Stage I/II endometriosis. In stage I/II endometriosis, laparoscopic ablation of endometrial implants has been associated with a small but significant improvement in live birth rates
- Several studies report success with Superovulation (SO)/ Intrauterine Insemination (IUI) in the treatment of endometriosis-associated infertility
- In younger women under age 35 years with stage I/II endometriosis-associated infertility expectant management or SO/IUI can be considered as first-line therapy. For women 35 years of age or older more aggressive treatment such as SO/IUI or IVF may be considered. In women with stage III/IV endometriosis-associated infertility conservative surgical therapy with laparoscopy or possible laparotomy may be beneficial. Surgical management of an endometrioma should include resection or ablation rather than drainage with resection preferred. For women with stage III/IV endometriosis who fail to conceive following conservative surgery or because of advancing reproductive age IVF-ET is an effective alternative

Surgical treatment of endometriosis is associated with the possibility of ovarian injury. This has been a major concern regarding surgery as a treatment modality and various attempts have been made to minimize the associated damage to ovarian tissue. The stripping technique for endometrioma excision may cause surgical injury to normal ovarian tissue. Removal of excessive ovarian tissue along with the walls of ovarian cysts may cause follicle loss. Electrical coagulation of the remaining parenchyma of the ovary after excision of the cyst wall may cause thermal damage resulting in a decrease in AMH levels immediately after surgery [16]. Therefore we can't recommend surgical treatment for infertility.

Surgical treatment and subsequent IUI with ovarian stimulation should be considered in women with minimal to mild endometriosis with infertility. However, in women with moderate to severe endometriosis surgical treatment can reduce ovarian reserve and adversely affect subsequent IVF outcomes. According to the recent European Society of Human Reproduction and Embryology

guidelines surgical excision is not routinely recommended before considering IVF.

IUI indications:

- Male subfertility TPMSS >5 × 106 and <10 × 106
- Endometriosis
- Unexplained infertility
- ASA (cervical factor)

Success rates for IUI depend a great deal on the etiology of infertility, a woman's age, sperm count, use of ovarian stimulation and number of pre-ovulatory ovarian follicles.

Hypogonadotropic Hypogonadism

Hypogonadotropic hypogonadism refers to suppression of Gonadotropin Releasing Hormone (GnRH) pulsatility from the hypothalamus resulting in lack of production of FSH and LH from the pituitary and lack of production of hormones from the ovaries. Presenting symptoms include amenorrhea and vaginal dryness but usually not hot flushes or night sweats. Hormonal assessment reveals FSH and LH in the low-normal range or very suppressed less than 3 mIU/mL. Estradiol is also suppressed to less than 30 pg/mL. Longstanding hypogonadotropic hypogonadism is associated with low bone density due to prolonged hypoestrogenism.

Causes of Hypogonadotropic Hypogonadism include eating disorders such as anorexia, bulimia and disordered eating. Extreme exercise especially in combination with disordered eating is also associated with hypothalamic suppression as are hyperprolactinemia and hypothyroidism.

Central Nervous System (CNS) lesions such as craniopharyngioma can lead to Hypogonadotropic Hypogonadism. These lesions can be detected by magnetic resonance imaging. Kallmann syndrome is caused by failure of the GnRH neurons to migrate during fetal development and is associated with anosmia (inability to smell) and primary amenorrhea. Idiopathic causes are common. Treatment is based on correcting the underlying pathology, treating eating disorders, central nervous system lesions etc. If hypogonadotropic hypogonadism persists then treatment involves the use of gonadotropins to induce ovulation.

Prolactinomas are the most common pituitary adenoma accounting for 40%. Prolactinomas are considered microadenomas if the size is less than 10 mm and macroadenomas if greater than 10 mm. Macroadenomas can cause visual symptoms due to their size and compression of the optic chiasm. Secretion of high levels of prolactin suppresses production of GnRH leading to decreased FSH and LH and hypoestrogenism. Prolactinemia results in galactorrhea and suppresses gonadotropin secretion leading to amenorrhea. Presenting symptoms are therefore Galactorrhea, Amenorrhea or both.

The goal of treatment for women with a prolactinoma is to normalize prolactin and therefore restore gonadotropin levels to normal in order to facilitate menstrual cyclicity. Medical therapy involves the use of dopamine agonist to suppress prolactin secretion. The most commonly used dopamine agonist is bromocriptine. Cabergoline is a newer option and has fewer side effects. Once the prolactin level is normalized ovulation will be restored within a few months. Macroadenomas can be treated medically but surgery is often the preferred method for large masses. The macroadenoma is resected transsphenoidally.

Tubal disease is one of the most common causes of infertility accounting for 14%-30% of couples with infertility. Tubal disease is caused by insult to the fallopian tubes directly from an ascending infection or indirectly to the area around the tubes by conditions such as pelvic peritonitis, prior surgery or endometriosis.

Pelvic Inflammatory Disease (PID) is the major cause of tubal factor infertility. The risk of tubal factor infertility increases with the number and severity of episodes 10%-12% after one episode, 23%-35% after two episodes and 54%-75% after three episodes. Unlike acute pelvic inflammatory disease ascending infection with chlamydia is often asymptomatic.

Tubal occlusion can occur at the distal or proximal end of the fallopian tube. Distal occlusion is usually a result of ascending infection. Distal occlusion can be complete with the fimbriae fused together resulting in a large sac-like appearance to the tube or partial with minimal to varying degrees of fimbrial adhesion. Complete obstruction leads to damage of the internal structure of the tube and inability of sperm and egg to meet. Partial obstruction is a risk factor for ectopic pregnancy. Proximal tubal occlusion can be due to tubal muscle spasm that occurs during tests of tubal patency or true obstruction due to damage from ascending infection or trauma.

Adnexal adhesions can impair tubal function preventing oocyte pickup. Diagnosing tubal disease is most easily performed by Hysterosalpingogram (HSG), which involves injection of radio-opaque dye through the cervical canal into the uterine cavity and through the fallopian tubes under fluoroscopic observation. A sonohysterogram is more sensitive than the HSG for evaluation of intrauterine pathology. Although a sonohysterogram can determine patency of at least one tube, this technique gives no information regarding tubal anatomy. Laparoscopy is the definitive test for evaluation of tubal factors.

Surgical treatment of distal tubal disease can be performed by laparoscopy or laparotomy. Using a neosalpingostomy procedure, an incision is made to create a new opening in the fallopian tube. The success of opening the fallopian tubes is inversely related to the severity of the disease. Surgical correction of mild distal tubal obstruction yields pregnancy rates of 40%-60% with a risk of ectopic pregnancy of 4%-10%. Therefore, ART is the most common treatment option for tubal pathologies.

However, surgical correction of severe hydrosalpinges is associated with a much lower pregnancy rate (<15%) and a significant risk of ectopic pregnancy (>20%). For severe distal disease, IVF is the preferred treatment. There are two proposed mechanisms for this:

- The fluid within the hydrosalpinx is toxic to the embryo
- The fluid within the hydrosalpinx flows in a retrograde fashion and flushes the embryo out of the uterus prior to implantation

Therefore, significant hydrosalpinges should be removed prior to doing IVF. This can be accomplished by laparoscopic salpingectomy with care to avoid damage to the ovarian blood supply.

Alternatively, if there are significant pelvic adhesions and salpingectomy is technically not feasible, occlusion of the proximal fallopian tube can be performed instead. This procedure effectively

disrupts flow of fluid from the tube to the uterus thereby negating the effect of the hydrosalpinx.

Unexplained Infertility

Unexplained infertility affects up to 30% of couples with infertility. The diagnosis is made after the basic evaluation fails to reveal a cause for the infertility. The basic evaluation involves confirmation of ovulation, patent fallopian tubes with no suggestion of pelvic adhesions, confirmation of normal pelvic anatomy and proof of adequate sperm production. If the basic evaluation is normal and history and physical examination reveal no abnormalities, then laparoscopy is not necessary to make the diagnosis of unexplained infertility.

Gonadotropin with intrauterine insemination has been shown to have a cumulative pregnancy rate per couple of 33% for up to 4 cycles. IVF is not only a treatment, but also a diagnostic test as information about ovarian response, fertilization and embryo development can be ascertained. Live-birth delivery rates following IVF for a diagnosis of unexplained infertility are comparable to rates for tubal factor.

Intrauterine Insemination (IUI) is often considered first-line treatment for couples with unexplained or mild male-factor infertility because it is less invasive and less costly than IVF. IUI can be performed with Ovarian Stimulation (OS).

There is good evidence that Clomiphene Citrate or Letrozole or Gonadotropins with timed intercourse is no more effective than expectant management. Therefore ASRM is not recommended to use Clomiphene Citrate or Letrozole or Gonadotropins with timed intercourse as a treatment for unexplained infertility as it is no more effective than expectant management.

It is also recommended to use Clomiphene Citrate with IUI in the treatment of couples with unexplained infertility. Letrozole with IUI is the alternative treatment. It is not recommended to use low-dose gonadotropins with IUI in the treatment of unexplained infertility as it is more complex and expensive and likely no more effective than OS with oral medications with IUI.

It is recommended that a single IUI be performed between 0 and 36 hours relative to hCG injection in OS with IUI treatments.

There is good evidence that immediate IVF in women >38 years of age may be associated with a higher pregnancy rate and shorter time to pregnancy as compared to a strategy consisting of OS with IUI treatments with either oral medications or Gonadotropins prior to IVF.

It is recommended that couples with unexplained infertility initially undergo a course (typically 3 or 4 cycles) of OS and IUI with oral agents. For those unsuccessful with OS and IUI treatments with oral agents, IVF is recommended rather than OS and IUI with gonadotropins [17].

Male Infertility

A male factor is solely responsible in about 20% of infertile couples and informative in another 30%-40% [18]. A male factor is almost always defined by abnormal semen parameters (Table 4). However, a male factor may also be involved when the semen analysis is normal. A male factor may be due to hormonal abnormalities resulting in decreased sperm production, abnormalities of testicular function or abnormal sperm transport or delivery. Causes of male infertility associated with normal sperm production include poor coital technique, erectile dysfunction and ejaculatory disorders.

Table 4: WHO 2010 Semen parameters.

Variable	Cut-off Value
Sperm Volume	>1.5 ml
Sperm concentration	>15 million/ml
Total Sperm Count	>39 million
Sperm Progressive Motility (A+B)	>32%
Sperm Morphology	>4%
Sperm DNA Fragmentation	<30%
Non-Sperm Cells	<1 million/ml
Note: WHO loboratory manual for the examination and processing of human semen; Geneva: World Health Organization; 2010	

Chromosomal abnormalities are a congenital cause of male infertility. Y-chromosome microdeletions are found in 10% to 15% of men with azoospermia, when there are no sperm present in semen and severe oligospermia defined as less than 5 million sperm per milliliter. This condition involves the long arm (Yq11) of the Y chromosome. There are 3 regions: Azoospermia Factor a (AZFa), Azoospermia Factor b (AZFb), Azoospermia Factor c (AZFc). Deletion of the entire AZFa or AZFb region results in azoospermia. Men with AZFc deletions may have azoospermia or produce very limited numbers of sperm, resulting in oligospermia (Table 5).

Table 5: ASRM evaluation [18].

Etiology	Semen Volume	T	FSH
Pre-Testicular: Hypogonadotropic hypogonadism Exogenous Androgens	 N/↓ N/↓	 ↓ ↑/ N/ ↓	 ↓ ↓
Testicular: Primary Testicular Failure, Genetic etiology, Varicocele	N	↓	↑
Post-Testicular: Vasectomay, Epididymal obstruaction Ejaculatory duct obstruction , ejaculatory dysfunction	 N ↓	 N N	 N N/↑
Note: FSH: Follicle Stimulating Horomone; N: Normal, T: Testosterone			

The most common sex chromosome disorder is Klinefelter Syndrome which is due to the presence of an extra X chromosome (47,XXY) and occurs in about 1 out of every 1000 males. Men with Klinefelter syndrome have high Follicle-Stimulating Hormone (FSH) levels and may have low testosterone levels. They characteristically have small, firm testes and azoospermia. While some men have bilateral gynecomastia (breast enlargement), delayed puberty and a female body shape with relatively long legs many men appear normal but only have small testes and infertility.

Some men have a mosaic pattern on karyotype (47, XXY/46, XY) and may have oligospermia. Many other genetic abnormalities such as chromosome deletions and translocations may also cause infertility.

A number of genetic abnormalities result in male infertility. In congenital bilateral absence of the vas deferens (CBAVD) does not develop. Many men with CBAVD also lack much of the epididymis. This means sperm cannot pass from the testis into the ejaculate. However, there is normal testicular function. Congenital bilateral absence of the vas deferens is caused by a mutation in the cystic fibrosis gene on chromosome 7.

Developmental abnormalities include Sertoli-cell-only syndrome also known as germ cell aplasia. In this condition, there is congenital absence of germ cells in the seminiferous tubules of the testes due to failure of migration during embryonic development. As a result only Sertoli cells line the seminiferous tubules of the testes. A pattern of Sertoli-cell-only can also be acquired as a result of exposure to chemotherapy or radiation therapy that destroys the germ cells. Some men have small pockets of sperm production within portions of the testicle, which can be retrieved by testicular sperm extraction.

In cryptorchidism, there is a failure of the testes to descend into the scrotum. It is the most common birth defect of the male genitalia. In boys born at full term, the cause is usually unknown. About 30% of men with unilateral cryptorchidism and 50% of men with bilateral cryptorchidism will have oligospermia. Fertility rates are worse in men with bilateral cryptorchidism.

A varicocele is an abnormal dilation of the veins of the pampiniform plexus in the scrotum. It is usually left-sided but may be bilateral. Varicocele is present in 15% of all males and in 40% of infertile males. Varicoceles have a progressive deleterious effect on spermatogenesis. This is the most common correctable cause of male infertility.

Ejaculatory duct obstruction can be congenital or acquired. Congenital duct obstruction is due to compression and obstruction of the ejaculatory ducts by cysts within the prostate such as müllerian duct cysts or ejaculatory duct cysts.

Acquired obstruction may be due to infections such as prostatitis or epididymitis, or related to prior urethral surgery. The obstruction can be partial or complete with azoospermia. Ejaculatory volume is low. Ejaculatory duct obstruction can be diagnosed by transrectal ultrasound and is treated by transurethral resection of the ejaculatory ducts or unroofing of the prostatic cyst.

Hormonal abnormalities also contribute to male infertility. Isolated Gonadotropin-Releasing Hormone (GnRH) deficiency will result in infertility. Some patients lack GnRH production and lack the nerves enabling their sense of smell called anosmia. Hypogonadotropic Hypogonadism associated with anosmia is referred to as Kallmann syndrome. Patients with GnRH deficiency do not produce LH and FSH with resulting azoospermia. Gonadotropin replacement therapy can reverse the azoospermia.

Medications and drugs of abuse also are factors in male infertility and include alkylating agents used in Chemotherapy, ionizing radiation, ketoconazole, exogenous testosterone supplementation, illegal substances including marijuana, opiates, and heroin and legal substances such as alcohol and tobacco, which have effects on male fertility.

Excessive alcohol use can interfere with testicular function and male hormone production, resulting in impotence, infertility and reduction of male secondary sex characteristics such as facial and chest hair. Tobacco use adversely affects sperm. The effect is worse in men who also have a varicocele [19].

According to AUA criterias, evaluation of the male infertility [20]:

- Reproductive history and one or more semen analyses are the initial evaluation of the male
- Discuss risk factors especially lifestyle, medication usage, cigarette smoking, alcohol, environmental exposures associated with male infertility and patients should be counseled that the current data on the majority of risk factors are limited
- Look at the hormonal evaluation including Follicle-Stimulating Hormone (FSH) and testosterone for infertile men with impaired libido, erectile dysfunction, oligozoospermia or azoospermia, atrophic testes or evidence of hormonal abnormality on physical evaluation
- Azoospermic men should be initially evaluated with semen volume, physical exam and FSH levels to differentiate genital tract obstruction from impaired sperm production
- Karyotype and Y-chromosome microdeletion analysis should be recommended for men with primary infertility and azoospermia or severe oligozoospermia (<5 million sperm/mL) with elevated FSH or testicular atrophy or a presumed diagnosis of impaired sperm production as the cause of azoospermia
- Sperm DNA fragmentation analysis is not recommended in the initial evaluation of the infertile couple
- For couples with recurrent pregnancy loss men should be evaluated with karyotype and sperm DNA fragmentation
- Scrotal or Transrectal Ultrasonography (TRUS) should not be performed as part of the initial evaluation. Clinicians should recommend TRUS in men with SA suggestive of ejaculatory duct obstruction
- Surgical varicocelectomy should be considered in men attempting to conceive who have palpable varicocele (s), infertility and abnormal semen parameters except for azoospermic men
- Clinicians should not recommend varicocelectomy for men with non-palpable varicoceles detected solely by imaging
- For men with clinical varicocele and NOA, couples should be informed of the absence of definitive evidence supporting varicocele repair prior to ART
- For men with NOA undergoing sperm retrieval, Microdissection Testicular Sperm Extraction (TESE) should be performed
- In men undergoing surgical sperm retrieval, either fresh or cryopreserved sperm may be used for ICSI
- In men with azoospermia due to obstruction undergoing surgical sperm retrieval, sperm may be extracted from either the testis or the epididymis
- Clinicians may use Aromatase Inhibitors (AIs), hCG, Selective Estrogen Receptor Modulators (SERMs) or a combination thereof for infertile men with low serum testosterone. Additionally, testosterone monotherapy should not be prescribed
- The infertile male with hyperprolactinemia should be evaluated for the etiology and treated accordingly
- Clinicians should counsel patients that the benefits of

supplements (e.g. antioxidants, vitamins) are of questionable clinical utility in treating male infertility

- For men with idiopathic infertility, a clinician may consider treatment using an FSH analogue with the aim of improving sperm concentration, pregnancy rate and live birth rate
- A clinician may advise an infertile couple with a low total motile sperm count on repeated SA that IUI success rates may be reduced and treatment with ART (IVF/ICSI) may be considered. In our clinic, we advice ART according to total progressive motile sperm count (TPMSC) (<5 million).
- The TPMSC between 5 million-10 million was performed in an Intrauterine Insemination (IUI) program and morphology did not have a predictive value for the pregnancy rate [21]
- Based on these cut-off values the WHO uses a descriptive nomenclature including Oligozoospermia (O), Asthenozoospermia (A) and Teratozoospermia (T) and combinations of these factors to classify the different forms of male factor infertility. The pathological terms of semen analysis are summarized in Table 6.

Table 6: The terms of semen analysis.

	WHO 1999	WHO 2010	Nomenclature if below cut-off value
Volume	2 ml	1.5 ml	Hypospermia
Sperm Concentration	20 × 10^6 spermatozoa/ml	15 × 10^6 spermatozoa/ml	Oligozoospermia
Motility (A+B)	50%	32%	Asthenozoospermia
Morphology	30% normally formed	4% normally formed	Teratozoospermia

Antisperm antibodies are present in 9%-13% of infertile couples and 1%-2.5% of fertile males and females. Many antisperm antibodies have been described both in men and women, in blood, testes and cervical fluid. Antisperm antibody testing should not be a routine test for infertile couples. Men who have undergone testicular surgery such as vasectomy or vasectomy reversal have a high rate of antisperm antibodies. Effective treatments include IUI and IVF with Intracytoplasmic Sperm Injection (ICSI), which are common treatments for most causes of infertility and therefore obviate the need for testing. With IUI, washed sperm are deposited directly into the uterus, thereby avoiding the adverse effect of cervical mucus. IVF with ICSI is the most effective treatment as it bypasses almost all potential adverse effects of antisperm antibodies. The presence of antisperm antibodies does not reduce the effectiveness of IVF-ICSI as a treatment (Figure 8).

The management of Azoospermia according to ASRM (Figure 9)

Finally, Optimal care for men with NOA requires a multidisciplinary clinical team that includes a reproductive urologist or other specialist in male reproductive medicine. Preimplantation genetic testing may be helpful to minimize the risks to offspring of affected men. Men who harbor complete AZFa or AZFb Y-chromosome microdeletions

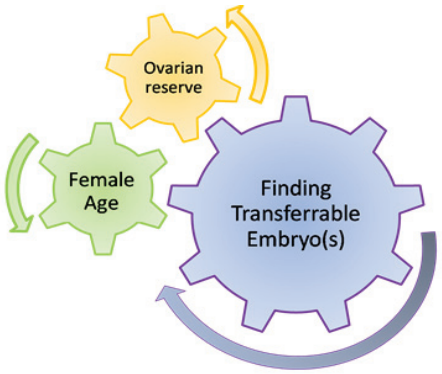

Figure 8: IVF success strategy.

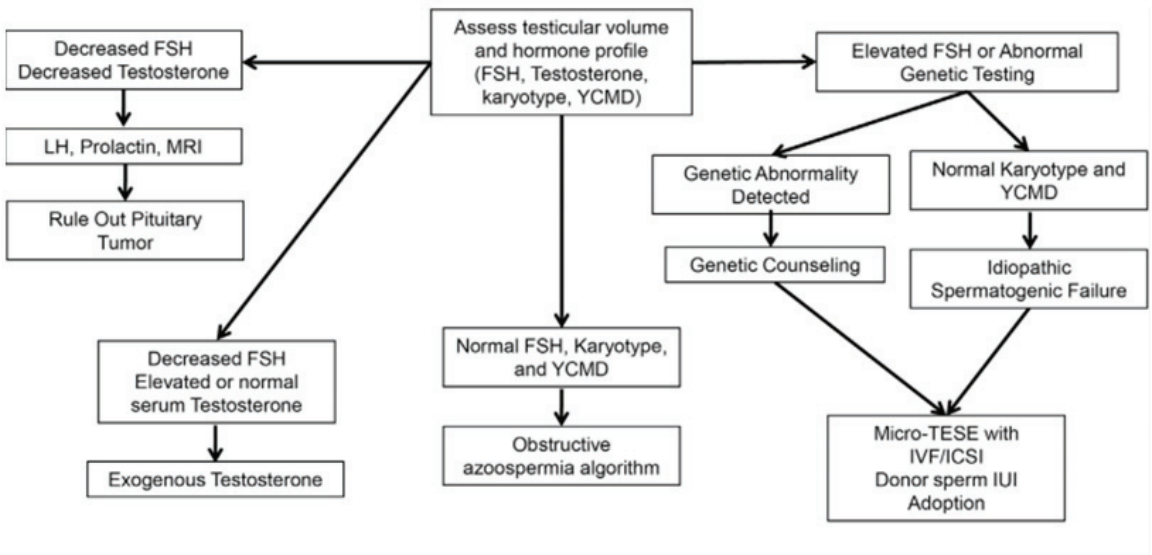

Figure 9: Evaluation of Azoospermia.

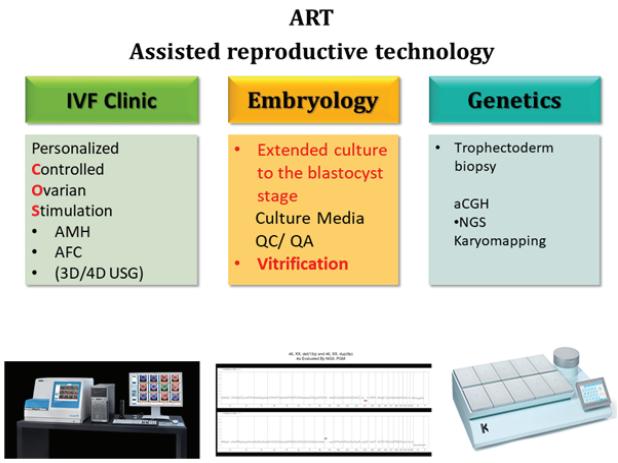

Figure 10: Assisted Reproductive Technology (ART).

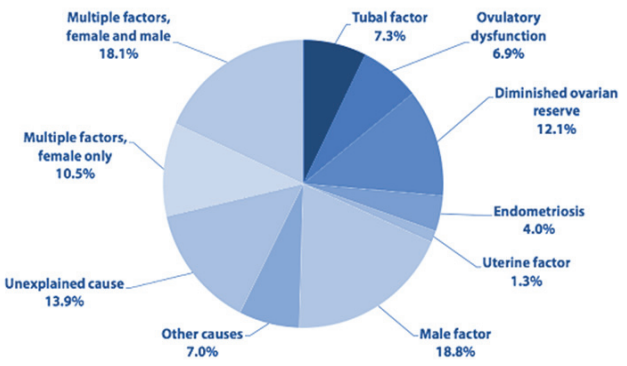

Figure 11: ART reasons.

should be counseled to consider donor sperm. Varicocelectomy should be considered in men with varicocele-associated NOA prior to sperm retrieval [22].

Assisted Reproduction Technique

The most common causes of female infertility include tubal factor, unexplained, ovulatory dysfunction, decreased ovarian reserve, endometriosis and multiple female factors. Data from the Centers for Disease Control describes the rates of various factors found in couples who had assisted reproductive technology (Figure 10) cycles with fresh, nondonor eggs or embryos: tubal factor, ovulatory dysfunction, diminished ovarian reserve, endometriosis, uterine factor, male factor, other cause, unexplained cause, multiple factors, female only, and multiple factors, female and male [17].

The following causes of infertility will be discussed in this presentation (Figure 11): Ovulatory failure or Dysfunction, including Primary Ovarian insufficiency, hypogonadotropic hypogonadism, polycystic ovary syndrome and prolactin disorders, tubal disease, endometriosis, unexplained infertility and autoimmune disorders.

Females are born with a set number of oocytes that decreases with age due to atresia of the follicle/oocyte, a process that is independent of the menstrual cycle. The maximum number of oocytes is 7 million and occurs at 7 months' fetal gestation. At birth, puberty and menopause, the number decreases to 1 million, 300,000 and essentially zero, respectively.

Premature Ovarian Failure (POF) is now referred to as primary ovarian insufficiency. This occurs when there are no more viable oocytes remaining in the ovary. Causes of primary ovarian insufficiency include deletion of one X chromosome such as with Turner Syndrome 45X, Turner mosaic 45X/46XX and in women with a deletion of a portion of the distal long arm of the X chromosome. Recently, abnormal CGG repeats in the FMR1 gene have been associated with decreased ovarian reserve and ovarian failure. Chemotherapy especially alkylating agents and pelvic radiation are also associated with premature ovarian insufficiency as both these treatments destroy follicles.

Surgery associated with removal or destruction of ovarian tissue such as ovarian cystectomy and laser ablation of endometriosis are also associated with ovarian insufficiency.

Autoimmune causes of ovarian insufficiency have been described, but the association with other autoimmune disease, such as Addison's disease and autoimmune thyroiditis, is statistically unsubstantiated. Recently, Fragile X premutation has been associated with primary ovarian insufficiency. As this condition is associated with severe mental retardation in offspring, it is important to identify this mutation in a woman desiring pregnancy. Other genetic mutations are being investigated.

Idiopathic causes of primary ovarian insufficiency make up a significant portion of patients with this condition. Women with primary ovarian insufficiency present with irregular or absent menses. Unlike other causes of amenorrhea, primary ovarian insufficiency is usually accompanied by "menopausal" symptoms including hot flushes, night sweats, vaginal dryness and irritability and mood changes. The diagnosis is determined when the level of FSH is greater than 20 mIU/mL and a simultaneous estradiol level is less than 30 pg/mL. Primary Ovarian İnsufficiency can often be intermittent with symptoms coming and going in association with fluctuating levels of FSH and estradiol. Reproductive aging reflects the decreasing number and quality of oocytes in the ovaries that occurs with age. Oocytes in women in their 40s have been shown to have a higher rate of mitotic spindle abnormalities, single chromatid abnormalities and aneuploidy in preimplantation embryos compared to younger women. Therefore, preimplantation genetic screening may be adviced.

Ovarian reserve is the ability of the ovaries to respond to gonadotropins with adequate follicular development leading to ovulation, fertilization and implantation. This process is independent of age, young women can have poor ovarian reserve and older women may have adequate ovarian reserve. However, there is no one reliable test available to assess the number and quality of competent oocytes remaining. AFC ve AMH is more reliable test to choose the starting dosage of COS. Antral follicle count reflects the number of antral follicles measuring 2 mm to 10 mm seen on transvaginal ultrasound on cycle day 3. This test is not predictive of ovarian reserve but rather is used to determine who will respond poorly to gonadotropin therapy. Anti-müllerian hormone is produced by antral follicles and promoted as a cycle-independent assessment of ovarian reserve. It is comparable to other ovarian reserve tests.

Poor ovarian reserve has been shown to correlate with decreased fertility in general. Specifically, poor ovarian reserve correlates with decreased pregnancy rates in a general infertility population, lower live birth rates with ovulation induction and intrauterine insemination and decreased pregnancy rates with IVF. Ovarian reserve can be used to counsel patients regarding the success of their fertility options.

Indications for IVF-ET include the absence or disease of bilateral fallopian tubes, severe male factor infertility (defined as less than 5 million-10 million total motile sperm in ejaculate or surgically retrieved sperm) and couples requiring donor oocytes, a gestational carrier, or preimplantation genetic diagnosis.

Use of donated oocytes is indicated for women who have age-related ovarian failure, premature ovarian insufficiency, a transmissible genetic abnormality or poor quality oocytes. Use of a gestational carrier is indicated for women who do not have a uterus, who have a uterine abnormality leading to infertility, recurrent pregnancy loss, or who have a medical contraindication to pregnancy and for gay men who wish to parent a genetically-related child.

Preimplantation genetic diagnosis is indicated when there is the potential for transmission of an undesired genetic defect.

Failure of previous infertility treatment is another indication for IVF-ET. Issues such as female age and presence of multiple infertility factors can influence decisions about proceeding to IVF but general indications include the following: no success after 1 year of treatment or no success after 3 to 6 cycles of IUI (for male factor infertility) or no success after 6 cycles of ovulation induction and IUI (for unexplained infertility or a combination of male and female infertility factors) or overstimulation with gonadotropin ovulation induction leading to concerns about multiple gestation.

Controlled ovarian stimulation is frequently used with IVF-ET cycles, as it does improve pregnancy rates. Exact protocols vary by clinic but involve the use of injectable gonadotropins. The use of an oral contraceptive prior to treatment may minimize cycle cancellation and allow for greater control of cycle management. A commonly-used stimulation protocol uses a combination of GnRH agonists and gonadotropins. Natural cycle IVF (using no medications) and minimal stimulation protocols (using oral ovulation-induction agents) are other possibilities but the yield of oocytes and embryos and thus pregnancy rates are significantly lower. These alternatives are useful for avoiding ovarian hyperstimulation syndrome and other side effects of high hormone levels and for decreasing the chances of a multiple pregnancy.

Specialized assessments of the uterine cavity are required depending on the clinical situation. For a cycle using cryopreserved embryos, uterine assessment should show an endometrial lining thickness of ≥ 6 mm to 8 mm on ultrasound at time of frozen-thawed embryo transfer. While more controversial in fresh IVF-ET treatment cycles, endometrial lining thickness (and potentially lining appearance) appears to predict pregnancy and live birth rates. In general, a thicker endometrium with a trilaminar pattern is preferred as shown in the ultrasound image. This is a sagittal transvaginal image of a retroverted uterus with a periovulatory endometrial pattern. The dotted line indicates the endometrial thickness. A three-layer endometrium is noted: the endometrial lumen is demonstrated by the central echogenic line, the hypoechogenic layer representing the edematous endometrial functionalis and an outer echogenic area representing the endometrium basalis.

There are very few successful pregnancies when endometrial lining thickness is <6 mm at the time of progesterone supplementation and frozen-thawed embryo transfer. In addition, ovaries should have no active corpus luteum and the progesterone level must be <3 ng/mL. Endogenous progesterone production will alter the window of implantation for a frozen-thawed embryo and such cycles must be delayed or cancelled.

For fresh embryo cycles, prior to starting gonadotropin stimulation, ovaries should have no productive ovarian follicles i.e. no baseline follicles measuring 15 mm or larger. The estradiol level must be less than 50 pg/mL to 80 pg/mL. Large dominant ovarian follicles will prevent stimulation of other ovarian follicles by the gonadotropin medications during a fresh IVF-ET cycle, thus the cycle must be delayed or cancelled if these are present.

Controlled Ovarian Stimulation Protocols

Controlled Ovarian Stimulation (COS) (Figure 12) is the most important part of ART involving the use of fertility medications to induce ovulation by multiple ovarian follicles.

First of all ovarian response and optimal dose for ovarian response is the pivotal part of COS. AFC and AMH both have a high accuracy in the prediction of an ovarian response. Basal FSH have some predictive value for ovarian response especially FSH more than 15 IU/L is lower sucess rate. Therefore pretreatment may be adviced more than 15 IU/L. Several studies have now demonstrated that women with modest FSH elevations will make fewer eggs and have a high risk of cycle cancellation.

Age also has some predictive value, however assessment of expected ovarian response by age alone is not sufficiently reliable. Especially Advanced Maternal Aged (AMA) should be careful to prevent premature ovulation of the follicles. Follicular dynamics, illustrated by follicular size and homogeneity of follicular development, influence early human embryo development. Patterns of follicular growth have an impact on embryo quality and viability which is reflected in morphokinetic variables. So pretreatment may be need for the follicular synchronicity.

Basal Oestradiol (E2) and Body Mass İndex (BMI) alone are not predictors of ovarian response but the dosage of gonadotropin is adjusted according to the BMI. Additionally, E2 more than 80 pg/mL is not recommended to start COS. Assessment of progesterone prior to initiation of stimulation on cycle day 2 appears to have some predictive value for the probability of pregnancy.

The GnRH antagonist protocol is recommended for predicted normal responder women with regards to improved safety [23]. GnRH antagonists and GnRH agonists are equally recommended for predicted poor responders. Clomiphene citrate alone or in combination with gonadotrophins and gonadotropin stimulation alone are equally recommended for predicted poor responders. The addition of letrozole to gonadotropins in stimulation protocols is probably not recommended for predicted poor responders.

It is unclear whether a higher gonadotropin dose is recommended over 150 IU for predicted poor responders.

A gonadotropin dose higher than 300 IU is not recommended for predicted poor responders.

The GnRH antagonist protocol is recommended over the GnRH

agonist protocols given the comparable efficacy and higher safety in the general IVF/ICSI population.

The use of recombinant FSH (rFSH) and Human Menopausal Gonadotropin (hMG) for ovarian stimulation is equally recommended.

Final oocyte maturation is triggered at sizes of several of the leading follicles between 16 mm-22 mm.

A single measurement of the endometrium during ultrasound assessment on the day of triggering or oocyte pick-up to counsel patients is important for thin endometrium <7 mm to affect the implantation.

The use of recombinant hCG and urinary hCG is equally recommended for triggering final oocyte maturation during ovarian stimulation protocols. According to OHSS risk, hCG or GnRH agonist trigger may be chosen. The use of GnRH agonist for final oocyte maturation with conventional luteal support and fresh transfer is not recommended in the general IVF/ICSI population. If the GnRH agonist trigger with triptorelin is applied, dosages ranging of 0.1 mg-0.4 mg can be chosen. A freeze-all strategy is recommended to eliminate the risk of late-onset OHSS and is applicable in both GnRH agonist and GnRH antagonist protocols.

The addition of a GnRH agonist to hCG as a dual trigger for final oocyte maturation is probably not recommended for predicted normal responders but may be prefered for poor responders.

Progesterone is recommended for luteal phase support after IVF/ICSI.

The dosing of natural progesterone has evolved empirically, usually dosages used include: 50 mg once daily for intramuscular progesterone, 25 mg once daily for subcutaneous progesterone, 90 mg once daily for vaginal progesterone gel, 200 mg three times daily for micronized vaginal progesterone, in-oil capsules 100 mg two or three times daily for micronized vaginal progesterone, in starch suppositories 400 mg two times daily for vaginal pessary.

Starting of progesterone for luteal phase support should be in the window between the evening of the day of oocyte retrieval and day 3 post oocyte retrieval and be administered at least until the day of the pregnancy test.

A normal response to standard IVF stimulation indicates normal ovarian function and generally a good prognosis, although this is still limited by a woman's age. Some women are "hyper" responders and are at risk for ovarian hyperstimulation syndrome. While certain characteristics do increase the risk for hyperresponse such as PCOS, hyperstimulation generally cannot be predicted and vigilance is key. A low response suggests "ovarian aging" common in women of advanced maternal age and may be associated with poor IVF outcome.

No single protocol is perfect for every patient. A long protocol (Figure 13) using prestimulation ovarian suppression, or downregulation, is effective for certain patients, such as those with ovarian cysts, but this protocol also decreases androgens and estrogens. Long protocols may over-suppress poor responders, requiring a reduced downregulation dosage in the prestimulation phase. Another disadvantage is the extended length of stimulation and increased amount of gonadotropin used during the stimulation phase. A long protocol does however, prevent a premature LH surge during the stimulation phase allowing better control of ovulation timing.

The short protocol does not require lengthy suppression. With the short protocol there may be effects from ovarian androgens and cysts.

There is a potential for a better response, but a premature LH surge may occur if a Gonadotropin-Releasing Hormone (GnRH) antagonist or agonist is not given.

A long stimulation protocol for a normal responder is shown here. Oral contraceptives may be given during the prior cycle. Beginning at day 21, leuprolide is given daily for prestimulation downregulation and the dosage is decreased once menses begins on cycle day 1. Beginning on day 3, gonadotropins are given daily. When monitoring indicates follicles are ready, hCG is administered, oocyte retrieval is done 36 hours later, and luteal phase support begins.

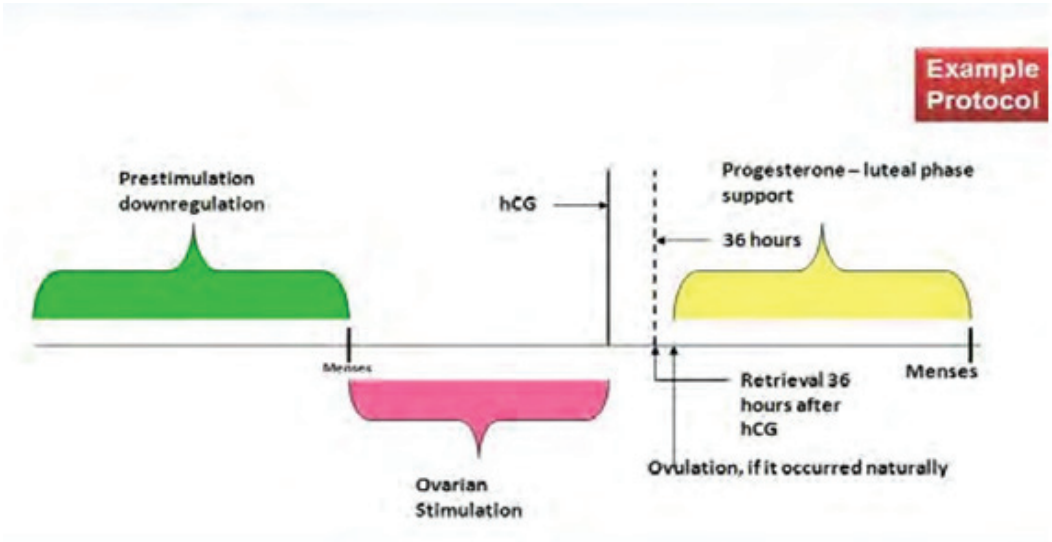

Figure 12: Standart COS protocol.

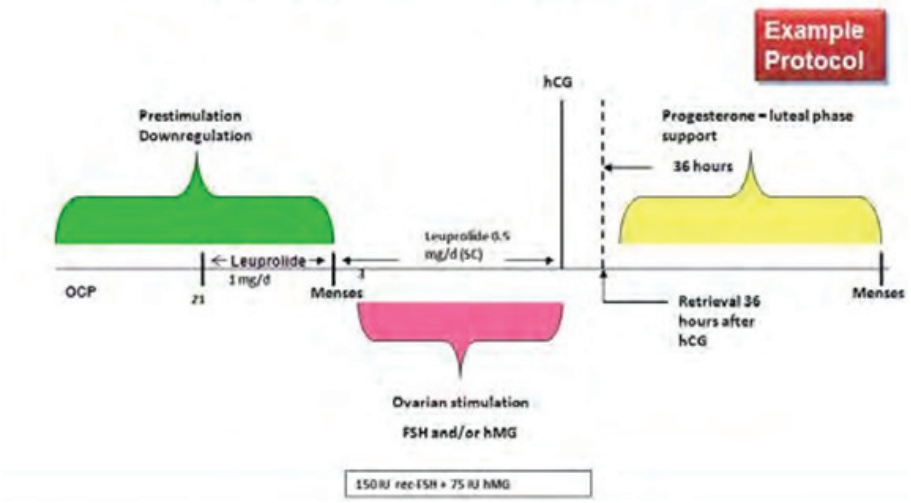

Figure 13: Long protocol.

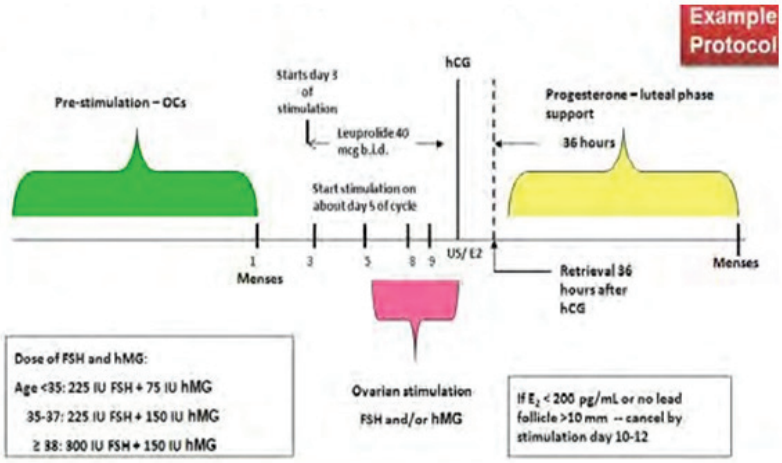

Figure 14: Poor responder COS protocol.

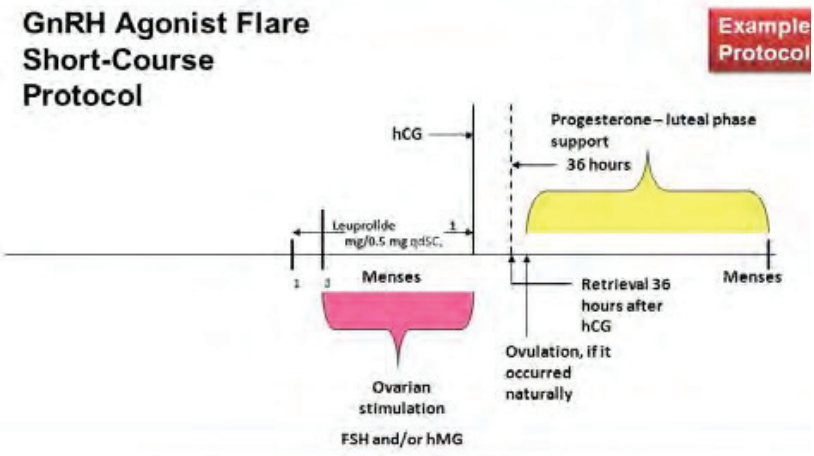

Figure 15: Agonist protocol of COS.

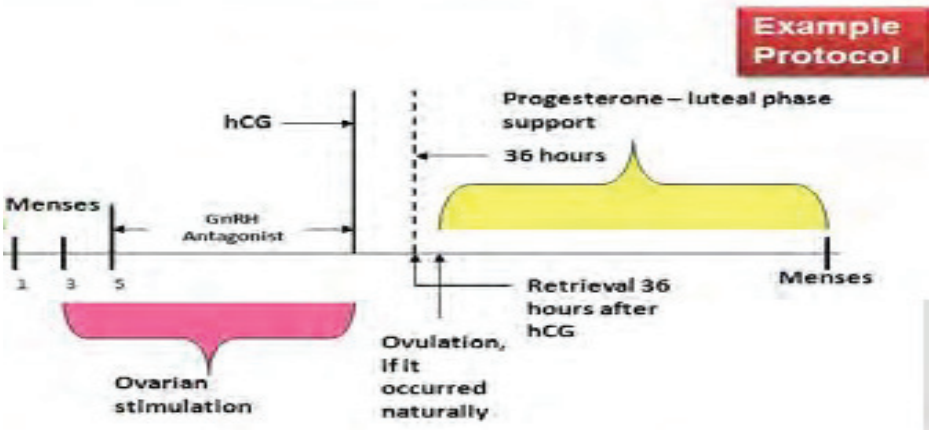

Figure 16: Fresh cycle after COS.

Expansion status	1 =	Early blastocyst; blastocoel less than half the volume of the embryo, little or no expansion in overall size, zona pellucida (ZP) still thick
	2 =	Blastocyst; blastocoel more than half the volume of the embryo, some expansion in overall size, ZP beginning to thin
	3 =	Full blastocyst: blastocoel completely fills the embryo
	4 =	Expanded blastocyst, blastocoel volume now larger than that of the early embryo. ZP very thin
	5 =	Hatching blastocyst; trophoectoderm has started to herniate through the ZP
	6 =	Hatched blastocyst; the blastocyst has evacuated the ZP
ICM grading	A =	ICM prominent, easily discernable and consisting of many cells, cells compacted and tightly adhered together
	B =	Cells less compacted so larger in size, cells loosely adhered together, some individual cells may be visible
	C =	Very few cells visible, either compacted or loose, may be difficult to completely distinguish from trophoectoderm
	D =	Cells of the ICM appear degenerate or necrotic
	E =	No ICM cells discernable in any focal plane
Trophoectoderm	a =	Many small identical cells forming a continuous trophoectoderm layer
	b =	Fewer, larger cells, may not form a completely continuous layer
	c =	Sparse cells, may be very large, very flat or appear degenerate

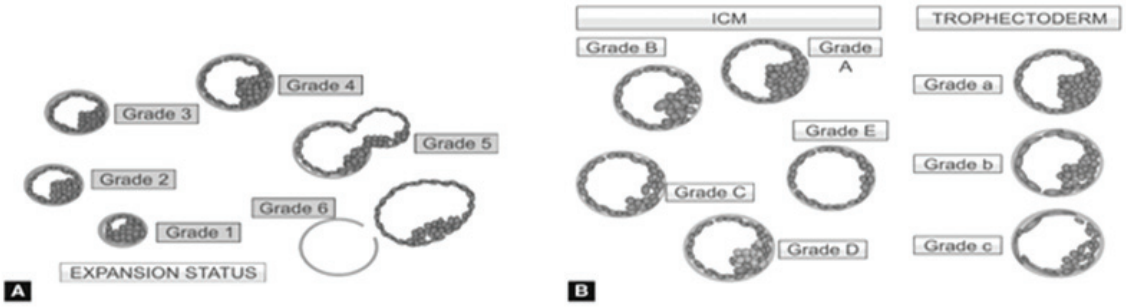

Figure 17: Embryo grading.

The cycle for poor responders begins (Figure 14) with downregulation using oral contraceptives in the prestimulation phase. At a time that can be adjusted, the continuous oral contraceptives may be stopped. After 2 days, leuprolide is started in a twice-daily dosing. After 2 additional days, gonadotropins are begun. The approximate starting doses of gonadotropins are related to age but could be adjusted for clinical history as well. The goal is steady growth and development of follicles.

This diagram (Figure 15) illustrates the principles of the GnRH agonist flare short-course protocol with no downregulation prior to the cycle. The leuprolide dosage is reduced from 1.0 mg to 0.5 mg daily on cycle day 3.

Gonadotropins are given beginning day 2 or 3 and the GnRH antagonist is started at day 6 (fixed protocol) or E2>200 pg/mL or follicle more than 11 mm (flexible protocol) continued until hCG is given. In our clinic, after 5 days of r-hFSH or hMG stimulation. A daily dose of 0.25 mg of GnRH antagonist was initiated when the leading follicle size was >13 mm or the serum E2 level reached >200 pg/mL (Figure 16).

In vitro fertilization and embryo transfer (Figure 17) is an ART in which sperm and eggs are retrieved and combined outside of the body in a laboratory dish where fertilization occurs. The fertilized eggs or embryos are then transferred to the woman's uterus with the hope one will implant and develop into a viable fetus

New Treatments in Art and Implantation

Past and future perceptions of an ART treatment cycle [24] is summarized in Table 7

Table 7: Yesterday and today and tomorrow of the ART.

Yesterday	Today and Tomorrow
GnRH agonist protocol	GnRH antagonist protocol
hCG for final oocyte maturation	GnRH agonist for final oocyte maturation
Fresh embryo transfer-slow freezing of supernumerary embryos	Vitrification of all embryos-'freeze all'
Embryo transfer as SET, DET, TET, QET	SET in subsequent natural or artificial cycle
OHSS 2%-5%	OHSS 0%
Multiple pregnancies 20%	Multiple pregnancies 0%

To increase pregnancy rates in ART, we have to focus on;

- Maternal age
- Ovarian reserve: (AMH level and AFC)
- Ovarian response
- No of Cumulus Oocyte Complexes (COCs)
- No of mature oocytes (MII)

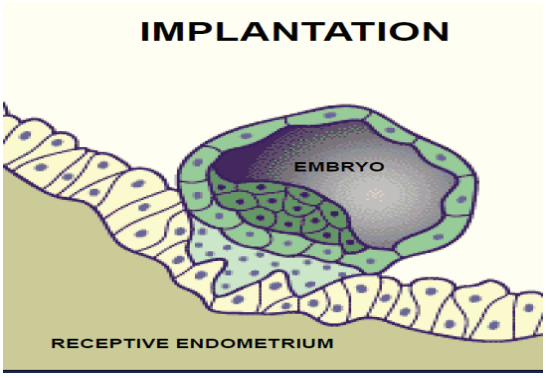

Figure 18: Implantation and endometrial receptivity [25].

- No of embryos (top quality and euploid)
- How to transfer fresh or frozen-thawed
- Euploid ratio of embryo (PGD)

The second critical step is euploid embryonic implantation (Figure 18) into a synchronous endometrium in achieving pregnancy.

Implantation consists of three stages:

- the blastocyst contacts the implantation site of the endometrium (apposition)
- trophoblast cells of the blastocyst attach to the receptive endometrial epithelium (adhesion)
- invasive trophoblast cells cross the endometrial epithelial basement membrane and invade the endometrial stroma (invasion)

So some authors discuss frozen thawed embryo transfer strategy get high pergnency rates because of hCG effect on implantation in this figure 19.

Pregnancy success is dependent on a complex pre- and postimplantation harmony between the embryo and the maternal endometrium. Implantation window opens owing to P after appropirate E2 priming (figure 20).

Implantation failure according to early progesteron elevation and supraphysiological E2 especially high responders by reducing endometrial reseptivity (E2>3000 pg/ml).

Endometrial Changes in COS (Figure 21) :

- Supraphysiologic steroid hormone concentrations have detrimantal effects on endometrial reseptivity according to reduced alfa and beta integrins
- Early elevated progesteron levels dysregulated miRNA and mRNA expressions and reduced implantation in genomic levels
- Precocious elevation in progesterone levels on the day of hCG

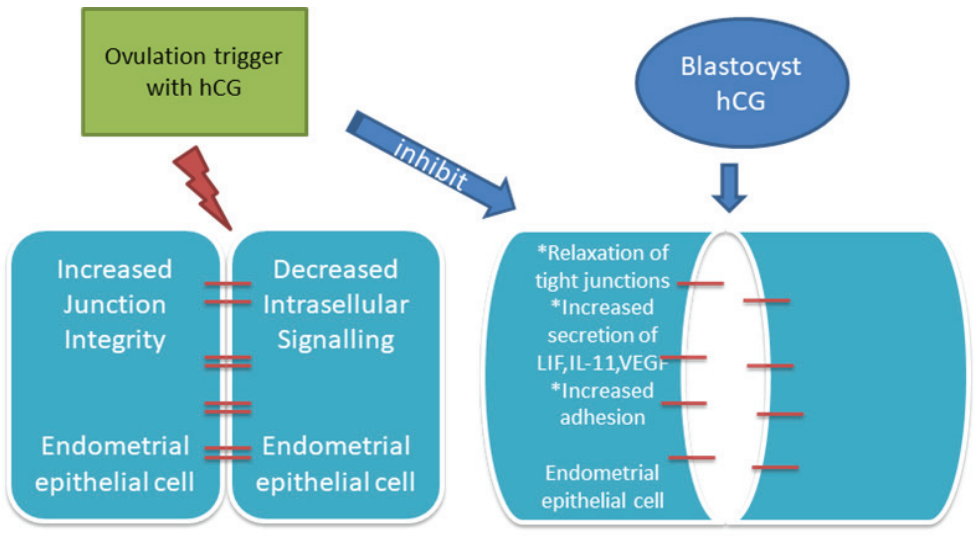

Figure 19: hCG effect on endometrium.

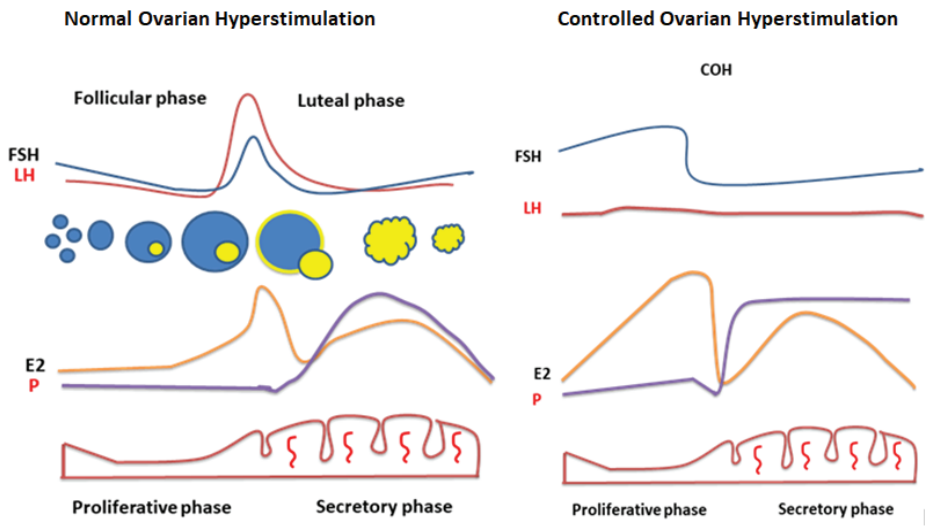

Figure 20: Normal versus controlled ovarian hyperstimulation effects on hormon and endometrium.

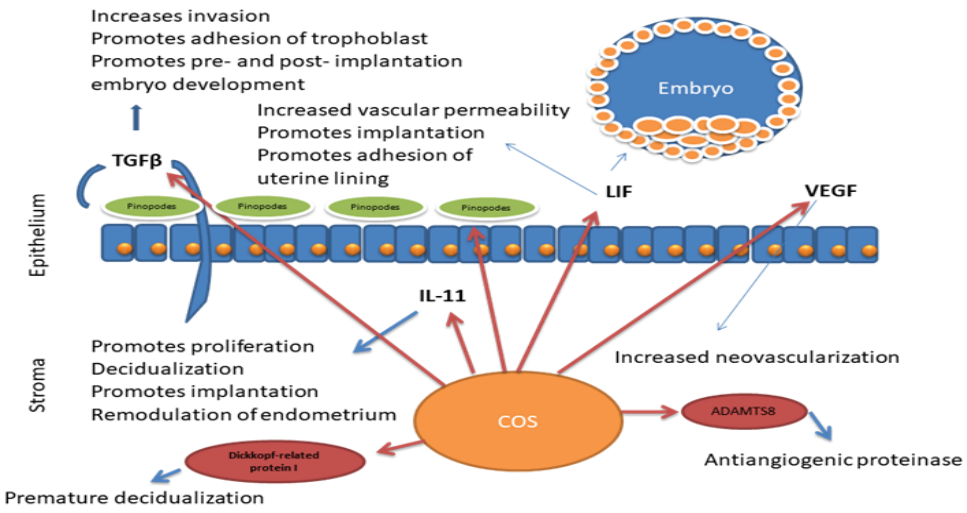

Figure 21: Implantation of the embryo.

administration is associated with reduced clinical pregnancy

- GnRH agonist or antagonist protocols significantly altered endometrial histology according to Noyes criteria
- Pinopodes have role in implantation and endometrial reseptivity appear 1-2 days earlier in cycles with COS and altered implantation failure
- Progesterone receptor down-regulated 1-2 days earlier in cysles with COS
- Advanced endometrial histology has been correlated with implantation failure in fresh siklus

Endometrial preparing of frozen thawed Embryo transfer

A Frozen Embryo Transfer (FET) cycle (Figure 22) is when one or more embryos (frozen during a previous treatment cycle) are thawed and transferred to the uterus. Some women undergo fresh Embryo Transfer (ET) cycles with embryos derived from donated oocytes. In both situations the endometrium is primed with oestrogen and progestogen in different doses and routes of administration. There is insufficient evidence on the use of any particular intervention for endometrial preparation in women undergoing fresh donor cycles and frozen embryo transfers. In frozen embryo transfers, low-quality evidence showed that clinical pregnancy rates may be improved in a stimulated cycle compared to a programmed one and we are uncertain of the effect when comparing a programmed cycle to a natural cycle. Cycle cancellation rates are probably reduced in a natural cycle. Although administering a GnRH agonist compared to without may improve live birth rates clinical pregnancy rates will probably be improved in a GnRH antagonist cycle over an agonist cycle. In fresh

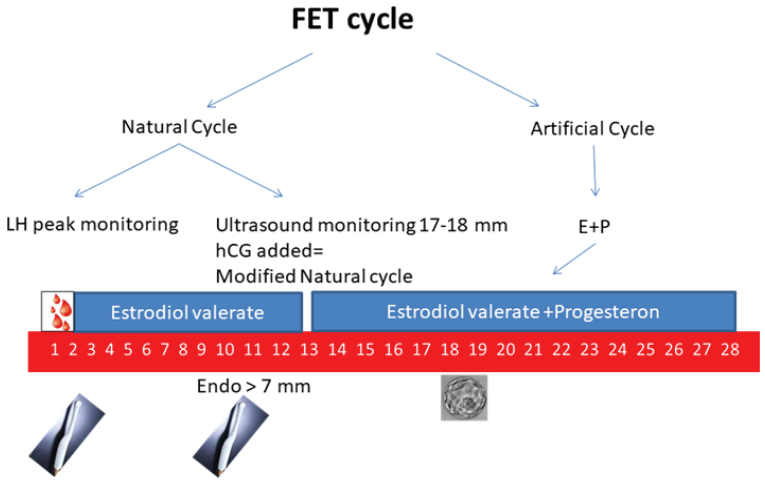

Figure 22: Endometrial preparing for frozen thawed cycles.

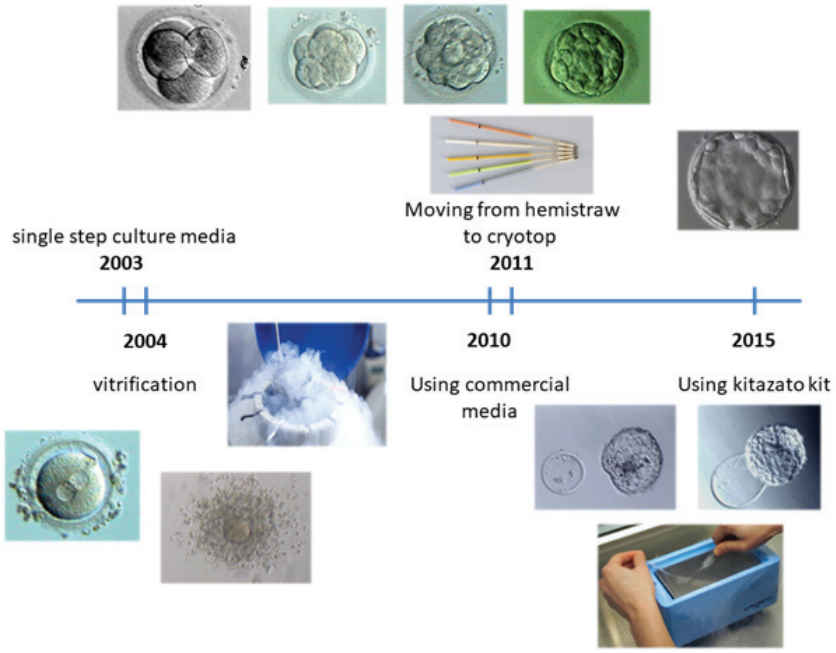

Figure 23: Embryo laboratories are also so important in ART.

synchronised oocyte donor cycles the clinical pregnancy rate is probably improved and cycle cancellation rates are probably reduced when starting progestogen the day of or day after donor oocyte retrieval. Adequately powered studies are needed to evaluate each treatment more accurately (Figure 23).

OHSS is one of the most feard iatrogenic complication of an ART treatment and the most devastating consequence is its threat to the patients life. Therefore the prevention of OHSS should remain the number one priority in reproductive medicine. Final oocyte maturation and ovulation triggering with a GnRH agonist is currently the most accepted strategy and it has been the focus of much research. However it causes the luteal phase disfunction By inducing multifollicular development, ovarian stimulation is an essential step in assisted reproduction. In recent years, a growing concern has been the potential adverse affect that ovarian stimulation could have on endometrial receptivity. Therefore ovarian stiuation may interfere with embryo endometrium interaction The hypothesis is that introduction of a cryopreserved-embro into a potentially more favourable intrauterin environment could avoid possible adverse effects os supra-physiological hormonal levels related to ovarian stimulation on endometrial receptivity.

Single embryo transfer:

- To prevent risk of multiple pregnancy

- To reduce the risk of OHSS
- Preimplantation genetic diagnosis

During cycles of COS, the traditional focus to maximize oocyte yield may inadverently diminish expected outcomes by creating a suboptimal endometrial environment. A growing body of eviedence suggest tat patient undergoing COS may potentially experience suboptimal endometrial development. Such effects uavoidably diminish the liklihood of embryo implantation and consequently lower pregnancy rate. It is theorised that possible mechanism that may alter embryo-endometrium synchrony may include a premature evaluation of progesterone that could alter the normal endometrial window of implantation.

FET is recommended:

- To reduce the risk of OHSS
- Preimplantation Genetic Testing (PGT)
- Endometrial pathologies may be seen during COS
- Thin endometrium
- A negative result after fresh ET

PGT is recommended:

- AMA>38 years
- recurrent implantation failure
- inherited disease history
- Pregnancy loss due to chromosomal deficiency

PG Diagnosis involves testing an embryo before it implants for a specific, known genetic disorder. PGD is used so that embryos unaffected by the disorder can be returned to the uterus.

PG Screening involves testing an embryo for chromosomal abnormalities. Many embryos with chromosomal abnormalities will not lead to a pregnancy or will result in a miscarriage. All women will have some eggs that are chromosomally abnormal. All men will have some sperm that are chromosomally abnormal. The percentage of embryos that are abnormal can be affected by many factors, including the age and health history of the parents.

With more recent and sensitive assays such as Next-Generation Sequencing NGS, it has become increasingly common to report identification and quantification of mosaicism within a trophectoderm biopsy sample [26].

Embryo Transfer

Embryo transfer steps:

- Abdominal ultrasound guidance for embryo transfer
- Removal of cervical mucus
- Use of soft embryo transfer catheters
- Placement of embryo transfer tip in the upper or middle (central) area of the uterine cavity greater than 1 cm from the fundus, for embryo expulsion
- Immediate ambulation once the embryo transfer procedure is completed

There is good evidence to recommend transabdominal ultrasound guidance during embryo transfer to improve clinical pregnancy rate and live-birth rate [27].

There is good evidence to recommend the use of a soft embryo transfer catheter to improve IVF-embryo transfer pregnancy rates.

There is good evidence not to recommend bed rest after embryo transfer.

There is fair evidence to placement of the catheter tip in the upper or middle (central) area of the uterine cavity greater than 1 cm from the fundus for embryo expulsion optimizes pregnancy rates.

Recurrent pregnancy loss is a disease distinct from infertility defined by the spontaneous loss of two or more pregnancies.

Patients preparing to undergo gonadotoxic medical therapy, radiation therapy or gonadectomy should be provided with prompt counseling regarding available options for fertility preservation for iatrogenic infertility. Embryo, oocyte and ejaculated or testicular sperm cryopreservation remain the principle established modalities for fertility preservation.

Adjuvant immunotherapy treatments in IVF aim to improve the outcome of ART in both the general ART population as well as subgroups such as patients with recurrent miscarriage or implantation failure.

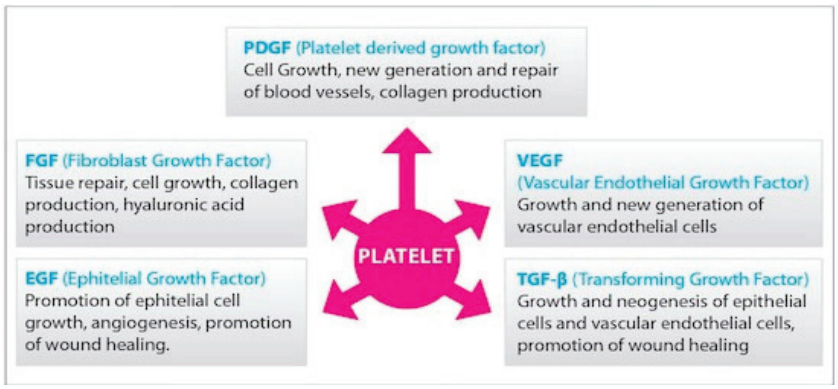

Figure 24: PRP effects.

Aspirin has been investigated as a means to increase blood flow to the ovaries and thereby improve oocyte yield and/or quality and increase blood flow to the uterus which may improve endometrial thickness and receptivity.

Uterine receptivity is regulated by locally acting growth factors and cytokines that have important roles in mediating immunological and non-immunological activity within the endometrium. Natural Killer (NK) cells play an important role in maintaining maternal-fetal immune tolerance and regulating trophoblast invasion. Aberrant cytokine activity and overactivity of NK cells have been associated with implantation failure and early pregnancy loss. The administration of corticosteroids around the time of implantation has been proposed as a strategy to normalize NK cell activity and cytokine expression and suppress inflammatory mediators to improve endometrial receptivity and the odds of successful implantation.

There is insufficient evidence to routinely recommend intravenous fat emulsions for infertile women pursuing IVF.

There is insufficient evidence to recommend for or against local G-CSF to improve endometrial thickness in women with thin endometrium or clinical pregnancy rates with IVF.

There is insufficient evidence to recommend IVIG administration as part of IVF to improve IVF outcomes.

The establishment of a healthy pregnancy requires maternal immune tolerance to the invading trophoblast to ensure successful implantation and adequate placentation and fetal growth. Th1 and Th2 mediate immune rejection and tolerance with recurrent implantation failure being associated with a high peripheral blood Th1/Th2 ratio. A Th1 immune response is associated with allograft as well as embryo rejection. Based on this rationale, a prospective study evaluated the effect of treating patients with recurrent implantation failure with tacrolimus-an immunosuppressive drug that inhibits antigen-induced lymphocytic proliferation, cytotoxic T-cell formation, IL-2 receptor expression and the production of IL-2 and interferon-gamma. The study included patients with a history of at least five prior failed IVF cycles and elevated peripheral blood Th1/Th2 ratios and compared the outcomes of patients who received 1 mg to 3 mg tacrolimus 2 days prior to ET.

There is fair evidence that seminal plasma insemination as part of IVF improves clinical pregnancy rate.

In DOR, platelet rich plasma injection to the ovaries is a new technique (Figure 24). Also PRP (Figure 25) is used for thin endometrium.

Thin endometrium (fresh cycles <7 mm and forzen-thawed cycles <8 mm)

- High doses of E2 (oral or patch)

  Prolonged E2 (patch)

  Local E2 (vaginal)
- hCG for

  VEGF expression

  Tissue remodelling

  Plays role in receptivity

  ◊ Poor dose hCG (Figure 26)

  ◊ High dose hCG

  750 IU hCG injection every 3 days concomitant
- Acetyl salisilic acid (ASA 80 mg or 100 mg orally)

  ◊ Might enhance endometrial growth and embryo implantation *via*

  ◊ Reducing sub-endometrial contractility

  ◊ Minimizing inflammation by inhibiting cyclooxygenase and prostaglandin biosynthesis

  ◊ Improving uterine endometrial blood flow
- Local PRP or Hysteroscopic PRP
- Vitamin E ± pentoxifylline: 800 mg PTX and 1000 IU vit-E per day till ET
- Hysteroscopic adhesiolysis is a safe and effective procedure for restoring the normal menstrual pattern and fertility.

Premature ovarian insufficiency is a clinical syndrome defined by

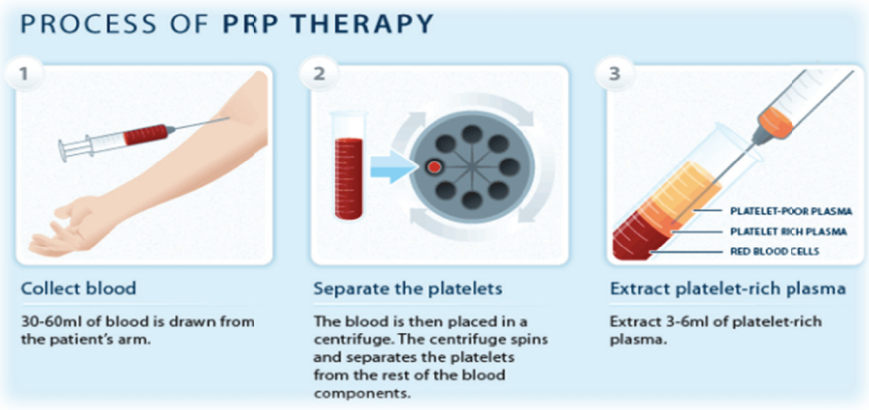

Figure 25: How to prepare PRP.

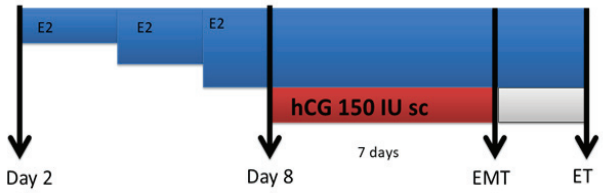

Figure 26: hCG supplementation in luteal phase.

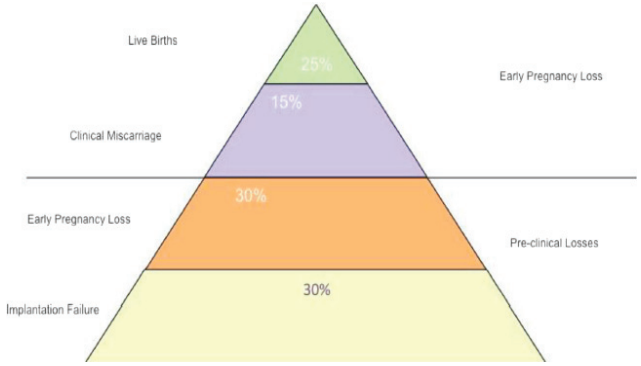

Figure 27: Pregnancy loss.

depletion of follicular activity before the age of 40 [28].

POI is characterised by menstrual disturbance (amenorrhea or oligomenorrhea), raised gonadotropins and low estradiol. The prevalence of POI is approximately 1%.

The diagnosis of POI is defined

- Oligo/amenorrhea for at least 4 months, and
- An elevated FSH level >25 IU/l on two occasions >4 weeks apart

Chromosomal analysis should be performed in all women with Premature Ovarian Insufficiency. Gonadectomy should be recommended for all women with detectable Y chromosomal material.

Fragile-X premutation testing is indicated in POI women.

Screening for thyroid (TPO-Ab) antibodies should be performed in women with POI.

In patients with a positive TPO-Ab test, Thyroid Stimulating Hormone (TSH) should be measured every year.

Women with POI should be informed that HRT has not been found to increase the risk of breast cancer before the age of natural menopause.

Women with POI with an intact uterus are advised to use a progestogen in combination with estrogen therapy for endometrial protection.

17-β estradiol is preferred to ethinylestradiol or conjugated equine estrogens for estrogen replacement.

Recurrent Implantation Failure

Recurrent Implantation Failure (RIF) (Table 8) refers to cases in which women have had three failed *In Vitro* Fertilization (IVF) attempts with good quality embryos. The definition should also take advanced maternal age and embryo stage into consideration. The failure of embryo implantation can be a consequence of uterine, male or embryo factors or the specific type of IVF protocol. These cases should be investigated to determine the most likely etiologies of the condition as this is a complex problem with several variables. There are multiple risk factors for recurrent implantation failure including advanced maternal age, smoking status of both parents, elevated body mass index and stress levels. Immunological factors such as cytokine levels and presence of specific autoantibodies should be examined as well as any infectious organisms in the uterus leading to chronic endometritis. Uterine pathologies such as polyps and myomas as well as congenital anatomical anomalies should be ruled out. Sperm analysis, pre-implantation genetic screening and endometrial receptivity should be considered [29]. The incidence varies from 8 to 33% (Figure 27)

Implantation rates/transferred embryo are still low (20%-40%). The limiting factor in achieving pregnancy for most couples is implantation and still poorly understood the reasons.

Table 8: Table of recurrent implantation failure.

Decreased Endometrial Receptivity
Uterine cavity abnormalities
Thin endometrium
Altered expression of adhesive molecules
Immunological factors
Thrombophilias
Defective Embryonic Development
Genetic abnormalities (male/female/gametes/embryos)
Zona hardening
Suboptimal culture conditions
Multifactorial Effectors
Endometriosis
Hydrosalpinges
Suboptimal ovarian stimulation

In 18%-27% of women with a normal initial hysteroscopy or hysterosalpingogram repeated hysteroscopic visualization after RIF revealed uterine abnormalities mainly hyperplasia, polyps, endometritis, synechiae and leiomyomata [30] (Figure 28).

The treatment strategies of RIF [30,31] (Table 9):

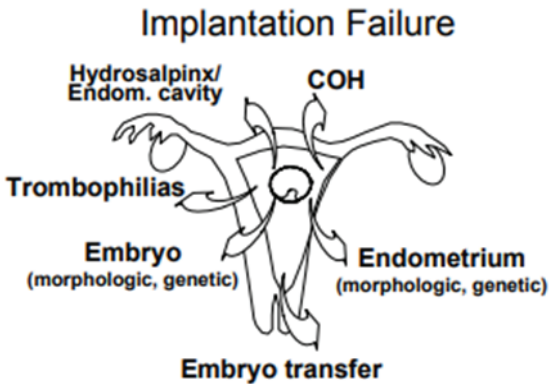

Figure 28: Reasons of implantation failure.

- Uterine interventions (e.g. intentional endometrial injury, Hysteroscopy, Endomerial sampling for histology and microbiological investigations and Endometritis treatment, Atosiban administration; Copper intrauterine device placement)
- Laboratory and procedural technologies and interventions i.e. sequential ET (i.e. sequential ET on day 2/3 and on day 5), ET medium enriched with hyaluronic acid, Autologous embryo-cumulus cells co-culture, Intracytoplasmic Morphologically Selected Sperm Injection (IMSI); blastocyst stage ET, Zygote Intrafallopian Tube Transfer (ZIFT), Assisted Hatching (AH); Preimplantation Genetic Testing for Aneuploidies (PGT-A)
- Immunomodulatory therapies e.g. Intravenous Immunoglobulin (IVIG), Intrauterine Peripheral Blood Mononuclear Cell (PBMC) infusion, Tacrolimus, Subcutaneous or Intrauterine Granulocyte Colony Stimulating Factor (G-CSF) administration, Intrauterine Autologous Platelet-Rich Plasma (PRP) Infusion, Intravenous Intralipid, Intrauterine Human Chorionic Gonadotropin (hCG) injection, Low-Molecular-Weight Heparin (LMWH), Aspirin, Prednisolone
- Treatments enhancing endometrial receptivity or technologies aimed at identifying the Endometrial Window of Implantation (WOI), e.g. Intramuscular Growth Hormone (GH), Vaginal Sildenafil, Endometrial Receptivity Array (ERA)

Table 9: Suggested methods for treatment of Repeated Implantation Failure (RIF).

Improving Endometrial Receptivity
Hysteroscopic correction of cavity pathology
Myomectomy
Treatment of thin endometrium
Endometrial stimulation (biopsy)
Immunotherapy (intravenous immunoglobulin, steroids, aspirin and heparin)
Treatment of the Embryos
Preimplantation genetic screening
Assisted hatching
Zygote intra-Fallopian transfer
Co-culture
Blastocyst transfer
Cytoplasmic transfer
Improving ET technique
Multifactorial Treatment Options
Treating endometriosis
Danazol
Salpingectomy in case of hydrosalpinges
Tailoring the stimulation protocols
Psychological assistance

The strategies of RIF is summarized (Figure 29):

Results of treatment options with regard to implantation rate, clinical pregnancy rate and live birth rate (Table 10) follows as:

Pregnancy loss (Table 11) is common ccurring in approximately 15%-25% of pregnancies. The majority of sporadic losses before 10 weeks' gestation result from random numeric chromosome errors specifically trisomy, monosomy and polyploidy. In contrast Recurrent Pregnancy Loss (RPL) is a distinct disorder defined by two or more failed clinical pregnancies. RPL have examined factors related to genetics, age, antiphospholipid syndrome, uterine anomalies, thrombophilias, hormonal or metabolic disorders, infection, autoimmunity, sperm quality, and lifestyle issues [32].

- Genetic Problems, In the evaluation of RPL parents should undergo peripheral karyotyping in order to detect any balanced structural chromosomal abnormalities. Balanced reciprocal translocations and Robertsonian translocations are observed in about 2%-5% of couples with recurrent miscarriage. Genetic counseling is important when a structural genetic factor is identified. PGD may be adviced.

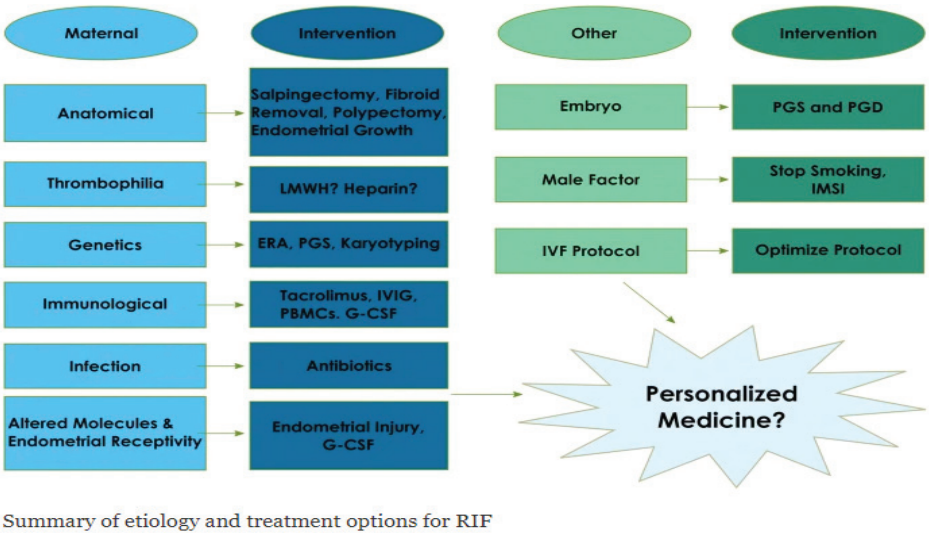

Figure 29: Treatment of Recurrent Implantation Failure (RIF).

Table 10: Summary of results of treatment options with regard to implantation rate, clinical pregnancy rate and live birth rate.

	Implantation Rate	Clinical Pregnancy Rate	Live Birth Outcome
	(treatment *vs.* control)	(treatment *vs.* control)	(treatment *vs.* control)
Tacrolimus	45.7% *vs.* 0%	64% *vs.* 0%	60% *vs.* 0%
	(p<0.0001)	(p<0.0001)	(p<0.0001)
IVIG	34.4% *vs.* 13.7%	60.2% *vs.* 39.3%	49.8% *vs.* 31.6%
	RR 2.708	RR 1.475	RR 1.616
	(95% CI: 1.302-5.629, I2=65.0%)	(95% CI: 1.191-1.825, I2=65.7%)	(95% CI: 1.243-2.101, I2=58.2%)
PBMC	22% *vs.* 4.88%	39.58% *vs.* 14.29%	33.33% *vs.*9.58%
	(p=0.014)	(p=0.038)	(p=0.038)
G-CSF	31.5% *vs.* 13.9%	48.1% *vs.* 25%	33.3% *vs.* 17.3%
	(p<0.01)	(p=0.01)	NS
Antibiotics for CE	37% *vs.* 17%	65.2% *vs.* 33%	60.8% *vs.* 13.3%
	NS	(p=0.039)	(p=0.02)
Salpingectomy	25.6% *vs.* 12.3%	45.7% *vs.* 22.5%	40% *vs.* 17.5%
	(p=0.038)	(p=0.029)	(p=0.038)
Endometrial Biopsy	27.7% *vs.* 14.2%	66.7% *vs.* 30.3%	48.9% *vs.* 22.5%
	(p=.00011)	(p=.00009)	(p=.016)
IMSI procedure	19.2% *vs.* 7.8%	43.1% *vs.* 10.5%	34.7% *vs.* 0%
	(p=0.042)	(p=0.02)	(p=0.003)

Table 11: Evaluation of recurrent pregnancy loss.

Cause	Contribution to RPL(%)	Recommended Screening	Supportive Scientific Evidence	Controversial Scientific evidence	Not recommended
Cytogenetic	2-5	Balanced reciprocal Translocations			
Apl syndrome	8-42 (mean, 15)	Lupus anticoagulant, Anticardiolipin IgG or IgM antibody. Anti-β_2 glycoprotein I	IgG and IgM antibodies, apL testing for other phospholipids and β_2 glycoprotein I	IgG and IgM Anti-annexin AS, anti-factor XII, anti-prothrombin, IgA aPLs	ANA, antithyroid antibodies
Anatomic	1.8-37.6 (mean,12.6)	Hysterosalpingography Sonohysterography	Congenital uterine abnormalities	Uterine Fibroids, polyps	Cervical incompetence
Hormonal or Metabolic		Prolactin TSH Hemoglobin A1c	Uncontrolled diabetes or thyroid disease, prolactin	Polycystic ovary syndrome and insulin resistance, luteal phase progesterone.	
Infectious		None		Bacterial vaginosis, endocervical infections	
Male factors		None		Abnormal sperm DNA	
Psychological		None		Psychological effects on uterine receptivity	
Alloimmune		None		Mucosal CD16-NK cells, embryotoxic factor, cytokine profiles, blocking antibodies, HLA typing, anti-paternal leukocyte antibodies, circulating CD16-NK cells	Circulating CD16-NK cells
Environmental, occupational or personal habits		History			Not related to recurrent pregnancy loss.

Table 12: LWMH treatment strategies for thrombophilia.

Anticoagulation Regimen	Anticoagulation Dosage
Prophylactic LMWH	Enoxaparin, 40 mg SC once daily Dalteparin, 5,000 units SC once daily Tinzaparin 4,500 units SC once daily Nadroparin 2,85units SC once daily
Intermediate-dose LMWH	Enoxaparin 40 mg SC every 12 hours Dalteparin 5,000 units SC every 12 hours
Adjusted-dose (therapeutic) LMWH	Enoxaparin, 1 mg/Kg every 12 hours Dalteparin, 200 units/Kg once daily Tinzaparin, 175 units/Kg once daily Dalteparin 100 units/Kg every 12 hours Target an anti-Xa level in the therapeutic range of 0.6 units/ml-10 units/ml 4 hours after last injection for twice- daily regimen; slightly higher doses may be needed for a once-daily regimen.

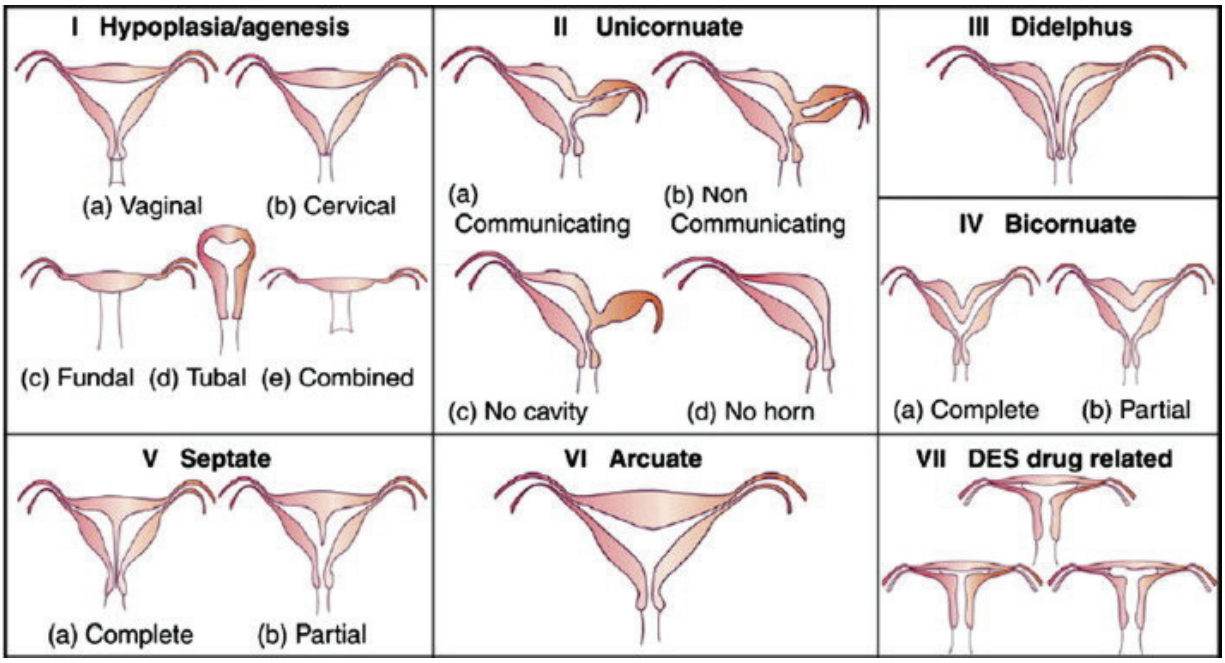

Figure 30: ASRM classification of uterine anomalies.

- Antiphospholipid Syndrome The antiphospholipid syndrome is associated with recurrent pregnancy loss.

International Consensus Classification criteria for the antiphospholipid syndrome (APS):

APS is present if one of the following clinical criteria and one of the laboratory criteria are met. Clinical criteria

- Vascular thrombosis
- Pregnancy morbidity
- One or more unexplained deaths of morphologically normal fetuses after the 10th week of gestation by ultrasound or direct examination of the fetus.

◊ One or more premature births of a morphologically normal neonate before the 34th week of gestation because of eclampsia or severe pre-eclampsia or recognized features of placental insufficiency.

◊ Three or more unexplained consecutive spontaneous abortions before the 10th week of gestation with maternal anatomic or hormonal abnormalities and paternal and maternal chromosomal causes excluded.

◊ Laboratory criteria

◊ Lupus anticoagulant present in plasma on two or more occasions at least 12 weeks apart, or

◊ Anticardiolipin antibody of IgG or IgM isotype in serum or plasma present in medium or high titer (>40 GPL or MPL or >99th percentile) on two or more occasions at least 12 weeks apart or

◊ Anti-b2 glycoprotein-I antibody of IgG and/or IgM isotype in serum or plasma (in titer greater than the 99th percentile) present on two or more occasions at least 12 weeks apart.

The standard treatment for documented antiphospholipid syndrome consists of low-dose aspirin and heparin.

- Congenital uterine abnormalities (Figure 30) associated with second trimester pregnancy loss in addition to other complications including preterm labor, fetal malpresentation and increased rates of cesarean delivery. Although the role of uterine malformations in first-trimester RPL is debatable, assessment of uterine anatomy is widely recommended. Potentially relevant congenital Mullerian tract anomalies include unicornuate, didelphic, bicornuate, septate, or arcuate uteri. Hysteroscopy is standart treatment.
- Inherited Thrombophilias, Screening for inherited thrombophilias (factor V Leiden and the prothrombin gene mutations, protein C, protein S and antithrombin deficiencies) may be clinically justified when a patient has a personal history of venous thromboembolism in the setting of a non-recurrent risk factor (such as surgery) or a first-degree relative with a known or suspected high-risk thrombophilia. LWMH is adviced for the treatment (Table 12).
- Hormonal parameters
- Maternal endocrine disorders such as diabetes, thyroid dysfunction should be evaluated and treated. As long as Thyroid Stimulating Hormone (TSH) levels are in the normal range, there is insufficient evidence to recommend routine Thyroxine (T4) testing or screening for anti-thyroid antibodies and consensus is emerging that TSH values above 2.5 mIU/L are outside the normal range.

Prolactin is commonly measured because elevated prolactin levels are associated with ovulatory dysfunction. Hyperprolactinemia may be associated with recurrent pregnancy loss through

alterations in the hypothalamicpituitary-ovarian axis, resulting in impaired folliculogenesis and oocyte maturation.

- Male Factors, Sperm aneuploidy and DNA fragmentation have been associated with RPL. Abnormal DNA fragmentation may be seen in the setting of advanced paternal age or may result from correctable environmental factors, such as exogenous heat, toxic exposures, varicoceles, or increased reactive oxygen species in semen.
- Lifestyle, Cigarette smoking has been suggested to have an adverse effect on trophoblastic function and is linked to an increased risk of sporadic pregnancy loss. Obesity has also been shown to be associated with an increased risk of RPL in women who conceive naturally. Other lifestyle habits such as cocaine use, alcohol consumption (3 to 5 drinks per week), and increased caffeine consumption (>3 cups of coffee) have been associated with risk of miscarriage.

Acknowledgment

It is an honor for me to editor “Infertility and Current Treatment Modalities”. The aim of this book is to learn the infertility reasons and current treatment modalities. I focused on changes and new guidelines in infertility. This book contains information about female infertility, male infertility, ovulation induction, assisted reproductive technologies, recurrent implantation failure. Advances in the new treatment of reproductive disorders are accelerating. This book is best demonstrated by the tables that summarized reproductive treatments. I dedicate this book to my wife “Cansu Oner” who is always help and encourage me to prepare my scientific studies. And thank you my mother “Nurel Oner” and my father “Gazi Oner”. I can’t forget my hero “Mustafa Kemal Atatürk” said “Science is the most real guide for civilization, for life, for success in the world. To search for a guide other than science is absurdity, ignorance and heresy” to say so glad to be your scientist. The authors and publishers have also attempted to trace the copyright holders of all material reproduced in this publication and apologize to copyright holders if permission to publish in this form has not been obtained. If any copyright material has not been acknowledged please write and let us know so we may rectify in any future reprint. I hope that readers of this open access book will gain new perspective and treatment options.d.

References

1. Taylor HS, Pal L, Sell E (2019) Speroff’s Clinical Gynecological Endocrinology and Infertility. (9th edtn), Wolters Kluwer, Netherlands.
2. Maroulis GB (1991) Effect of aging on fertility and pregnancy. Semin Reprod Endocrinol 9:165.
3. Magnus MC (2019) Role of maternal age and pregnancy history in risk of miscarriage: prospective register based study. BMJ 364: 1-8.
4. Wilcox AJ, Weiberg CR, O’Connor JF, Baird DD, Schlatterer JP, et al. (1988) Incidence of early loss of pregnancy. N EnglJ Med 319:189-194.
5. Pellestor F, Anahory T, Hamamah S (2005) Effect of maternal age on the frequency of cytogenetic abnormalities in human oocytes. Cytogenet Genome Res 111:206-212.
6. Oner G (2013) Assessment of ovarian reserve based on hormonal parameters, ovarian volume, and antral follicle count in women with familial Mediterranean fever. Eur J Obstet Gynecol Reprod Biol 170:449-451.
7. Tas M, Oner G, Ulug P, Yavuz A, Ozcelik B (2019) Evaluation of protective effects of GnRH agonist or antagonist on ovarian reserve with anti-Müllerian hormone and histological analysis in a rat model using cisplatin. Arch Med Sci.
8. Fauser BC, Tarlatzis BC, Rebar RW, Legro RS, Balen AH, et al. (2012) Consensus on women’s health aspects of polycystic ovary syndrome (PCOS): the Amsterdam ESHRE/ASRM-Sponsored 3rd PCOS Consensus Workshop Group. Fertil Steril 97:28-38.
9. Kim JJ, Hwang KR, Chae SJ, Yoon SH, Choi YM (2020) Impact of the newly recommended antral follicle count cutoff for polycystic ovary in adult women with polycystic ovary syndrome. Hum Reprod 35:652-659.
10. The Thessaloniki ESHRE/ASRM-Sponsored PCOS Consensus Workshop Group (2008) Consensus on infertility treatment related to polycystic ovary syndrome. Hum Reprod 23:462-477.
11. Practice Committee of the American Society for Reproductive Medicine (2017) Role of metformin for ovulation induction in infertile patients with polycystic ovary syndrome (PCOS): a guideline. Fertil Steril 108:426-441.
12. Teede HJ, Misso ML, Costello MF, Dokras A, Laven J, et al. (2018) Recommendations from the international evidence-based guideline for the assessment and management of polycystic ovary syndrome. Hum Reprod 33: 1602-1118.
13. Yu Y, Fang L, Zhang R, He J, Xiong Y, et al. (2017) Comparative effectiveness of 9 ovulation-induction therapies in patients with clomiphene citrate-resistant polycystic ovary syndrome: a network meta-analysis. Sci Rep 7:3812.
14. Practice Committee of the American Society for Reproductive Medicine (2016) Prevention and treatment of moderate and severe ovarian hyperstimulation syndrome: a guideline Fertil Steril 106:1634-1647.
15. The Practice Committee of the American Society for Reproductive Medicine (2012) Endometriosis and infertility: a committee opinion Fertil Steril 98:591-598.
16. Lee D, Kim SI, Lee JR, Jee BC (2020) Management of endometriosis-related infertility: Considerations and treatment options Clin Exp Reprod Med. 47: 1-11.
17. Practice Committee of the American Society for Reproductive Medicine (2020) Evidence-based treatments for couples with unexplained infertility: a guideline. Fertil Steril 113 :305-322.
18. American Society for Reproductive Medicine (2018) Evaluation of the azoospermic male: a committee opinion. Fertil Steril 109:777-782.
19. American Society for Reproductive Medicine (2008) Varicocele. Patient Fact Sheet.
20. ASRM Practice Committee Publishes New Guidelines with AUA and Updates Opinions, ASRM (2020)
21. Hamilton JA, Cissen M, Brandes M, Smeenk JM, de Bruin J , et al. (2015) Total motile sperm count: a better indicator for the severity of male factor infertility than the WHO sperm classification system. Hum Reprod 30:1110-1121.
22. The Practice Committee of the American Society for Reproductive Medicine (2018) Management of nonobstructive azoospermia: a committee opinion. Fertil Steril 110:1239-1245.
23. ESHRE COS Guideline 2019.
24. ESHRE Focus 2016.
25. Glujovsky D, Romina R, Sueldo C, Quinteiro Retamar AM, Hart RJ (2020) Endometrial preparation for women undergoing embryo transfer with frozen embryos or embryos derived from donor oocytes. Cochrane Database Syst Rev 10: CD006359.
26. Practice Committee of the American Society for Reproductive Medicine (2018) The role of immunotherapy in in vitro fertilization: a guideline. Fertil Steril 110:387-400.
27. Practice Committee of the American Society for Reproductive Medicine (2017) Performing the embryo transfer: a guideline. Fertil Steril 107:882-896.

28. ESHRE 2018 POI

29. Bashiri A, Halper KI, Orvieto R (2018) Recurrent Implantation Failure-update overview on etiology, diagnosis, treatment and future directions. Reprod Biol Endocrinol 16:121.

30. Simon A, Laufer N (2012) Repeated implantation failure: clinical approach. Fertil Steril 97:1039-1043.

31. Busnelli A, Somigliana E, Cirillo F, Baggiani A, Levi-Setti PE (2021) Efficacy of therapies and interventions for repeated embryo implantation failure: a systematic review and meta-analysis. Sci Rep 11:1747.

32. Ford HB (2009) Recurrent Pregnancy Loss: Etiology, Diagnosis and Therapy. Rev Obstet Gynecol. 2: 76-83.

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