Assisted Reproductive Technology: Clinical Aspects

Pardis Hosseinzadeh, M. Blake Evans, and Karl R. Hansen

Contents

T. Falcone, W. W. Hurd (eds.), *Clinical Reproductive Medicine and Surgery*,
https://doi.org/10.1007/978-3-030-99596-6_17

Key Points

- Assisted reproductive technology (ART) is a complex series of procedures used to treat infertility in couples who have failed less invasive fertility treatments or wish to prevent certain genetic problems in the offspring.
- In vitro fertilization (IVF) is the most effective and commonly used form of modern ART, which involves collecting mature eggs from the ovaries and fertilizing them with sperm in a lab followed by transfer of the fertilized egg or embryo into the uterus.
- Most common indications for IVF include, but are not limited to, disorders of ovulation, damaged or blocked fallopian tubes, male factor infertility, unexplained infertility, same-sex couples, and fertility preservation for cancer or other medical problems.
- Risks associated with IVF include ovarian hyperstimulation syndrome, multiple gestation, preterm delivery, low birth weight, ectopic pregnancy, and complications associated with the egg retrieval procedure.

17.1 Introduction

17.1.1 Prevalence

Infertility is a significant public health problem in the USA that affects women, men, and couples. Even though perceived as a quality-of-life issue, both the World Health Organization and the American Society for Reproductive Medicine (ASRM) [1, 2] define infertility as a disease of the reproductive system. Infertility has public health consequences beyond the ability to have children, including psychological distress, social stigmatization, economic strain, and marital discord. Furthermore, infertility is associated with an increased risk of subsequent chronic health conditions [3–6].

According to data from the National Survey of Family Growth (NSFG) conducted from June 2006 through June 2010, 6% (or an estimated 1.5 million US couples) were infertile. Additionally, 12% of reproductive-aged women reported impaired fecundity [7]. Challenges to human fertility may arise from many conditions caused by genetic or structural abnormalities, infectious or environmental agents, and certain behaviors. Natural aging also limits human fertility. The recent decline in the US birth and fertility rates is mainly attributed to delayed childbearing age in women due to greater aspiration for advanced education and marriage later in life. These trends have underscored the limits to natural fertility, and today, Americans are increasingly aware of and are concerned about infertility. Efforts to treat tubal factor and later other causes of infertility lead to the development and refinement of assisted reproductive technology (ART), which has changed the course of human reproduction.

Based on data presented from the NSFG survey, 12% of women aged 15–44 in 2006–2010 (7.3 million women), or their partners, had ever used infertility services. Among women aged 25–44, 17% (6.9 million women) had ever used any infertility service [8]. Among these women, the most common services were advice on the timing of intercourse (29%), infertility testing (27%), and ovulation induction drugs (20%) [8]. Intrauterine insemination (IUI) was used by 7.4% of these women, 3.2% had undergone surgery or treatment for obstructed fallopian tubes, and 3.1% had ever used ART [2]. The NSFG report indicates that infertility treatment other than ART, such as ovarian stimulation followed by natural conception or IUI, is much more common than ART. Although the scientific literature indicates that the efficacy of these treatments is much lower than that of ART. While it is difficult to ascertain the denominator for patients where ART has been recommended, it is very likely that more patients would benefit from ART. Lower rates of IVF utilization have been correlated with a lack of insurance coverage and decreased

availability of physicians providing this service. Despite these social factors, approximately 248,000 cycles of in vitro fertilization (IVF) are performed each year in the USA [9].

Case Vignette

ART Clinical- A 30-year-old woman with a diagnosis of infertility and polycystic ovary syndrome consults you for in vitro fertilization (IVF). She has failed multiple cycles of ovulation induction with letrozole and clomiphene. The semen parameters are normal. Imaging for tubal disease was negative.

17.1.2 Indications for ART

It is important to understand which individuals or couples would most benefit from ART. Of note, the term "ART" has historically been used to describe all treatments involving the handling of sperm and oocytes, although currently more than 99% of ART procedures are in vitro fertilization (IVF) procedures. The term "IVF" will be used throughout the rest of this chapter. IVF was first developed as a method to overcome infertility resulting from irreversible tubal factor but now is applied much more broadly for the treatment of almost all causes of infertility. Currently, IVF is commonly used in the treatment of individuals or couples with severe male factor infertility or severe tubal factor infertility resulting from previous infection, severe endometriosis, or sterilization procedure. In addition, there are other situations in which IVF may be the first-line treatment, such as women or men who are single or homosexual-partnered males using a gestational carrier and any individual or couple using donor oocytes or previously frozen oocytes. Another indication for IVF as a first-line treatment is in couples who are carriers of autosomal or sex-linked genetic disorders or balanced chromosomal translocations. In these cases, IVF with preimplantation genetic testing (PGT) can decrease the risk of delivering an affected child. IVF is also often the best treatment for couples with multiple causes relating to their infertility in order to overcome all contributing causes at once. Furthermore, IVF may be recommended for patients with age-related or unexplained infertility diagnoses when other treatment options fail.

Some women or couples may have an indication for a fertility preservation procedure, which involves harvesting oocytes, with or without embryo creation, for later use. This may be performed prior to receiving gonadotoxic medications such as chemotherapy. This treatment is also gaining acceptance for purposes of deferred childbearing. In patients with a planned fertility-threatening treatment, the process can be expedited, requiring approximately 2–3 weeks from the time of medication initiation to oocyte retrieval [10, 11].

17.2 Evaluation Prior to IVF

17.2.1 History and Physical Exam

Prior to starting IVF, individuals and couples should be thoroughly evaluated to help maximize their chances for a healthy pregnancy. Ideally, the couple should both be present during the first office visit. Individual or each partner's detailed past medical, surgical, family, and social history should be reviewed, and any special considerations followed up as appropriate. Chronic diseases, including hypertension, diabetes, thyroid, and autoimmune disorders, should be optimally controlled. Lifestyle and environmental factors influence fertility and deserve consideration when relevant. For instance, substance abuse, particularly tobacco use, is one factor over which couples have control. Infertility is more prevalent in reproductive-aged women who smoke, and the time to conception is longer compared to nonsmoking women. Additionally, the effects of passive smoke exposure is only slightly less than active smoking [12]. Therefore, couples attempting to conceive should be encouraged to quit smoking. Other forms of substance abuse such as marijuana and cocaine use can adversely affect fertility. Marijuana use may interfere with ovulation in women [13] and also decrease fecundity [14]. In men, marijuana use has been associated with lower sperm concentration, motility, viability, morphology, and impaired

capacitation and fertilization [14, 15]. In both women and men, even modest alcohol consumption has been associated with lower pregnancy rates in IVF cycles [16]. Some studies have suggested that women ingesting greater than 200 mg caffeine per day may delay conception [17] or increase the risk of pregnancy loss [18].

If review of the history reveals conditions that may affect the patient or the pregnancy, a pre-pregnancy consultation with a maternal-fetal medicine specialist may be warranted to discuss the risks involved in becoming pregnant as well as management during pregnancy.

In general, the patient should be offered screening for conditions that could affect the health of the pregnancy. The patient's blood type should be confirmed, and if her blood type is Rh negative, she should be counseled on the indications for and benefits of RhoGam administration during pregnancy. She also should be screened for immunity to rubella and varicella and, if non-immune, offered vaccination prior to pregnancy. In addition, the patient and her partner (if applicable) should be tested for hepatitis B and C, human immunodeficiency virus (HIV), gonorrhea, chlamydia, and syphilis. A careful family history and review of ethnic background will also inform whether additional tests such as cystic fibrosis, Tay-Sachs disease, and hemoglobin electrophoresis (for sickle trait or thalassemia) may be recommended [19]. Another option is for couples to consider Universal Genetic Carrier Screening. This testing offers the additional advantages of identifying couples at risk for having children with genetic disease prior to pregnancy [20].

Ultimately, it is important to discuss and set achievable goals for the management of infertility personalized for each individual or couple. This can help increase patient compliance and manage expectations.

17.2.2 Ovarian Reserve Testing

Ovarian reserve testing is typically performed to estimate the expected response to gonadotropin stimulation. Ovarian reserve tests include both biochemical and ultrasonographic measures. An anti-Müllerian hormone (AMH) level is drawn, and in some patients, a basal (cycle day 3) follicle-stimulating hormone (FSH) and estradiol are obtained as well [21]. Exceptions to drawing "day 3" labs may include young patients with a high AMH and fertility preservation patients undergoing an expedited cycle. Measuring an antral follicle count with transvaginal ultrasound also gives an assessment of ovarian reserve. A selection of all or some of these tests assists with dosing of gonadotropins and protocol selection.

Since none of the mentioned ovarian reserve tests are perfect predictors of ovarian response, various combinations of ovarian reserve tests have been developed in the desire to improve diagnostic performance. However, as these tests are highly correlated, models combining tests do not perform significantly better than individual tests such as the AFC [22]. Thus, the use of combined tests will not only increase the cost of testing but generally will not improve clinical decision-making.

17.2.3 Uterine Evaluation

Abnormalities of the uterus are an uncommon cause of infertility, but a cavity evaluation is essential if an embryo transfer is anticipated. The anatomic uterine abnormalities that may adversely affect fertility include congenital malformations, leiomyomas, intrauterine adhesions, and endometrial polyps.

There are three commonly used methods for evaluation of the uterine cavity: hysterosalpingogram (HSG), transvaginal ultrasound (TVUS) with saline infusion sonogram (SIS), and hysteroscopy.

The best options include a SIS (injecting sterile saline into the uterine cavity under ultrasonographic guidance) and hysteroscopy (using a small lighted scope while using a distension medium such as normal saline, to look directly into the uterine cavity). Hysteroscopy serves as the gold standard method for both diagnosis and treatment of intrauterine pathology that may adversely

affect fertility. Although definitive, hysteroscopy has limited diagnostic advantages over SIS and generally can be reserved for treatment of abnormalities identified by less invasive and costly methods.

HSG is a procedure wherein radio-opaque contrast is injected through the cervix into the uterine cavity under fluoroscopy to evaluate the fallopian tubes and uterine cavity. It is less sensitive (50%) and has a lower positive predictive value (30%) compared to SIS and hysteroscopy [23]. Because HSG cannot reliably differentiate a septate from a bicornuate uterus, further evaluation with pelvic MRI or 3D ultrasonography may be necessary [24].

Although the benefit of optimizing the uterine cavity requires further study, this is generally considered a standard practice prior to IVF. Many offices also perform a "mock," or practice, embryo transfer prior to the actual embryo transfer in order to anticipate any difficulties and increase the chances for an atraumatic embryo transfer [20].

17.2.4 Sperm Testing

If a male infertility factor exists, it frequently will be revealed by an abnormal semen analysis. Semen parameters can vary widely over time, and if abnormal, another semen analysis should be obtained after at least 4 weeks [25]. Prior to IVF, a recent semen analysis is indicated to assess whether intracytoplasmic sperm injection (ICSI) is necessary and whether a sperm extraction technique may be needed.

17.2.5 Optimizing IVF Outcomes

The reason for extensive evaluation prior to IVF is that there are areas of a patient's health that can be optimized prior to IVF. One important example is the identification of a hydrosalpinx on HSG or TVUS. Hydrosalpinges adversely affect IVF outcomes and have been shown to decrease pregnancy, implantation, and delivery rates by approximately 50% compared to women without hydrosalpinges [26, 27]. Laparoscopic salpingectomy or a tubal transection prior to IVF significantly improves pregnancy rates in women with hydrosalpinges [28], and limited evidence suggests improved pregnancy rates in natural conception [29].

Another aspect is optimizing thyroid function prior to IVF. Suboptimal thyroid function is associated with adverse pregnancy outcomes, including an increased risk of miscarriage, preterm birth, and impaired neurological development in the offspring [30, 31]. Hypothyroidism denotes deficient production of thyroid hormones and can be overt or subclinical. Overt hypothyroidism is characterized by an elevated thyroid-stimulating hormone (TSH) concentration and a decreased free T4 and is often associated with clinical findings such as fatigue, constipation, cold intolerance, muscle cramps, weight gain, dry skin, hair loss, and prolonged deep tendon reflexes. Overt hypothyroidism decreases fertility, presumably due to ovulatory dysfunction, and thyroid hormone should be administered prior to pregnancy to normalize the thyroid axis. Subclinical hypothyroidism is defined as an elevated serum TSH concentration (TSH > 4.5–5 mIU/L depending on local standards) with free T4 level within the normal reference range [32]. Thyroid function should be optimized in infertile women with subclinical hypothyroidism.

17.3 Process of IVF

17.3.1 Ovarian Stimulation

Numerous regimens have been proposed for ovarian stimulation, ranging from no stimulation (natural cycle IVF) to minimal stimulation with clomiphene citrate or sequential treatment with clomiphene citrate and from low-dose exogenous gonadotropins to high-dose exogenous gonadotropins. Currently ovarian stimulation using exogenous gonadotropins in combination with a gonadotropin-releasing hormone (GnRH) analogue has almost entirely replaced the other regimens due to higher pregnancy rates.

There are three basic elements to a conventional ovarian stimulation protocol for IVF; the first is exogenous gonadotropins to stimulate multi-follicular growth, the second is GnRH antagonist or agonist to prevent premature ovulation, and the third is LH activity in the form of human chorionic gonadotropin (hCG) or GnRH agonist to trigger the final oocyte maturation.

17.3.2 Gonadotropins

Gonadotrophin preparations available for use include human menopausal gonadotrophin (hMG) (a urinary product with follicle-stimulating hormone (FSH) and luteinizing hormone (LH) activity), purified FSH, highly purified FSH, and various recombinant FSH and LH preparations.

In a natural cycle, FSH and LH act in concert to stimulate folliculogenesis and ovulation. FSH stimulates growth of follicles and upregulates aromatase activity (an enzyme that converts testosterone to estrogen). Administration of exogenous FSH prevents the physiologic decrease in FSH in a natural cycle when the dominant follicle is selected. This allows for multi-follicular growth during controlled ovarian hyperstimulation. LH acts on the theca cells to increase androgen production, which is the substrate for estradiol synthesis by the granulosa cells in the developing follicles. LH causes luteinization of the follicle(s) and the synthesis/secretion of a large amount of progesterone from the corpus luteum [33]. Therefore, these gonadotropins are used alone or in combination in the ovarian stimulation process. The data from a 2017 meta-analysis have shown that although the administration of FSH alone results in a higher number of oocytes retrieved than FSH + LH or hMG protocols, the embryo number, implantation, and pregnancy rates were higher in the FSH + hMG protocols [34]. In light of evidence suggesting that the use of hMG may increase live birth rates, many clinics favor the combined stimulation with FSH and hMG (or alternative form of LH activity) over stimulation with FSH alone. In the combination protocols, variations in the relative proportions of FSH and LH may have an impact on the outcomes of ovarian stimulation, with suggested optimal LH to FSH ratio of 0.30–0.60 to mitigate the risk of premature progesterone effect on the endometrium in fresh embryo transfer cycles [35]. Monitoring the response to ovarian stimulation is accomplished with a combination of frequent transvaginal ultrasound examinations, with serum estradiol and progesterone measurements.

17.3.3 Cycle Timing

Oral contraceptive pills (OCPs) are often used prior to an IVF cycle to control the onset of the menses and therefore allow optimized scheduling of the stimulation cycle. Once on OCPs, patients are typically instructed to stop at least 5 days before the scheduled start. This provides more flexibility with the timing of appointments, which is more convenient for both the patient and the provider as well as facilitating cycle batching for some clinics. Pretreatment with OCPs may also help synchronize the follicular cohort by attenuating the FSH rise before stimulation begins. However, as always, convenience and potential biological benefits should be weighed against any possible adverse effects from the intervention.

OCP priming in women undergoing ovarian stimulation with antagonist protocol has been suggested to be associated with longer duration of stimulation and higher gonadotropin administration without an increase in cumulus oocyte complexes and lower ongoing pregnancy and live birth rates [36]. However, data from a recent study indicates that OCP administration for an interval of 12- to 30-day treatment period with a 5-day washout period does not affect clinical pregnancy or live birth rates in patients undergoing IVF cycles using an antagonist protocol [37].

Patients over the age of 35 who undergo ovarian pretreatment with OCPs may require a longer duration of stimulation with gonadotropins [38]. In low responder patients, there is limited evidence to support that short-term suppression with OCPs may improve the response [39].

If women have contraindications to the use of combined OCPs or history of intolerance due to side effects, an IVF cycle may be started with menses or the use of progesterone-only pills. In women who are amenorrheic or oligomenorrheic, withdrawal bleeding may be induced using progesterone or a progestin. Additionally, fertility preservation patients in need of imminent cancer treatments may need to start stimulation as soon as possible. In this setting, gonadotropins can be administered as a "random start" protocol, regardless of where the patient is in her menstrual cycle [11]. There is good evidence that the timing of cycle start does not affect outcomes when embryo cryopreservation is planned, even when gonadotropins are started in the luteal phase [40].

17.3.4 Ovulation Prevention

As previously mentioned, the goal of ovarian stimulation for IVF is to harvest a cohort of mature oocytes before spontaneous ovulation takes place. If premature ovulation occurs, the oocytes cannot be harvested, and the cycle will be cancelled. Estradiol levels during controlled ovarian hyperstimulation usually far exceed the threshold that triggers an LH surge in a natural cycle. The introduction of long-acting GnRH agonists in the late 1980s transformed the approach to ovarian stimulation in ART by providing the means to suppress endogenous pituitary gonadotropin secretion and thereby prevent a premature LH surge during exogenous gonadotropin stimulation [41]. Prior to that time, more than 20% of stimulation cycles were cancelled because of a premature LH surge and ovulation [41]. There are three standard IVF protocols to physiologically prevent or delay an LH surge.

17.3.5 Gonadotropin-Releasing Hormone Agonists

GnRH agonists initially stimulate LH and FSH release, also known as the "flare" effect, and within 2 weeks will suppress gonadotropin release [42], owing to downregulation of gonadotropin-releasing hormone receptors at the level of the pituitary. In a typical cycle, GnRH agonist treatment starts during the mid-luteal phase, when endogenous gonadotropin levels are low and the "flare" effect is least likely to stimulate a new cohort of follicular growth.

In the USA, the most popular GnRH agonist is leuprolide acetate, given subcutaneously starting with 1.0 mg daily for approximately 10 days or until onset of menses or gonadotropin stimulation, decreasing to 0.5 mg daily thereafter until the ovulation trigger (Fig. 17.1).

17.3.6 Microdose, or "Flare," Protocols

Microdose Lupron is an alternative stimulation regimen that utilizes a smaller dose of the GnRH agonist leuprolide acetate just 2–3 days before the start of ovarian stimulation medications. In a typical standard microdose protocol, leuprolide acetate (usually 40 micrograms divided into twice daily) is admin-

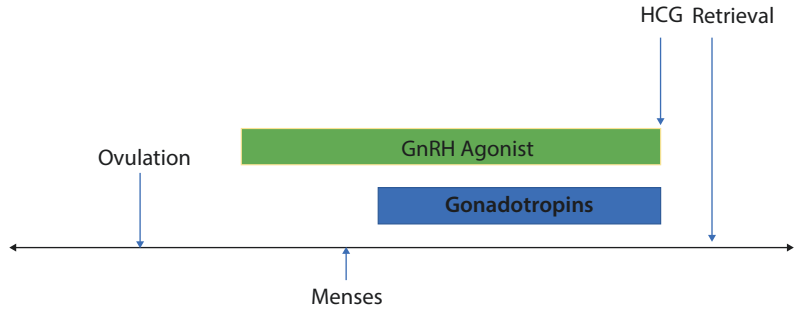

Fig. 17.1 GnRH agonist long protocol

istered starting on cycle day 1 or 2 and continuing daily until the trigger. The trigger must be an hCG as the GnRH receptors on the pituitary are fully occupied, and a GnRH agonist trigger would not work appropriately (Fig. 17.2). The goal is to minimize the suppressive effect and to take advantage of the initial "flare" phase to complement the exogenous gonadotropin injections. After 5–7 days of administration, longer-term treatment will result in pituitary suppression and thus prevents premature ovulation [43].

Minimizing excessive ovarian suppression while capitalizing on the initial stimulatory effect has led this regimen to gain popularity for poor responders. However, data from multiple studies including a meta-analysis regarding superiority of this protocol for poor responders are conflicting [44–46].

17.3.7 Gonadotropin-Releasing Hormone Antagonists

The development of GnRH antagonists provided another option for ovulation prevention in ART. Their main advantage is that antagonists block the GnRH receptor in a dose-dependent competitive fashion and have no flare effect. Since gonadotropin suppression is immediate, and because local GnRH receptors within the ovary may lead to decreased aromatase activity [47], treatment duration and total dose for gonadotropin usage are decreased [48]. A multicenter IVF trial compared the GnRH agonist protocol with the GnRH antagonist protocol and found a mean duration of 19 days of injections with the GnRH agonist, compared to only 4 days with the GnRH antagonist protocol [49]. The treatment protocol may be fixed and begin antagonist after 5–6 days of gonadotropin stimulation or individualized, starting antagonist when the lead follicle reaches approximately 13–14 mm in diameter [50, 51] (Fig. 17.3). The minimum usual dose to prevent ovulation is 250 μg/day.

A 2017 meta-analysis compared GnRH antagonist with long agonist protocols in couples undergoing IVF while accounting for patient diagnosis [52]. According to this study, in a general IVF population, GnRH antago-

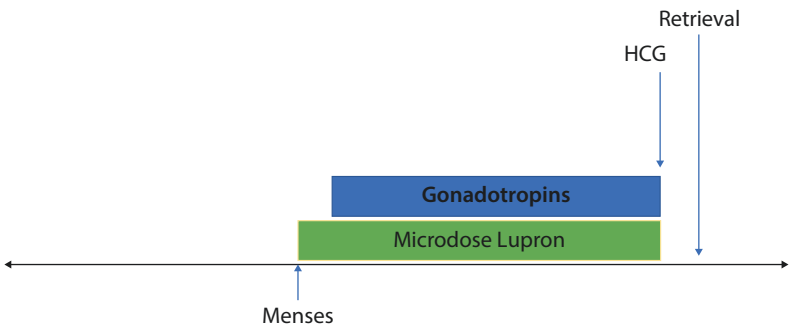

Fig. 17.2 Microdose Lupron or "flare" protocol

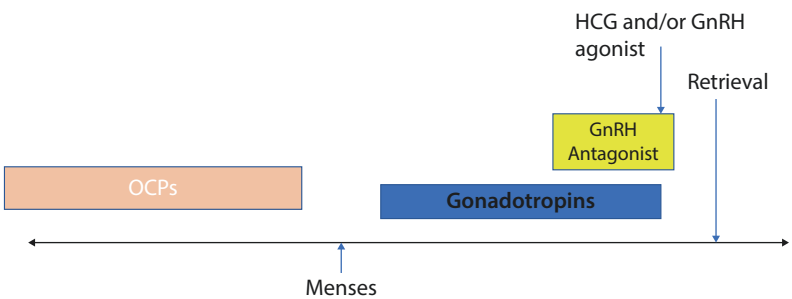

Fig. 17.3 Antagonist protocol

nist protocols are associated with lower ongoing pregnancy rates when compared to long protocol agonists but also with lower ovarian hyperstimulation syndrome (OHSS) rates [52]. The number needed to treat in the antagonist treatment group to prevent one case of OHSS was 40. In couples with PCOS and poor responders, GnRH antagonists do not seem to compromise ongoing pregnancy rates and are associated with less OHSS [52].

17.3.8 Ovulation Trigger

Once a cohort of follicles reaches maturity, urinary human chorionic gonadotropin (hCG, 5000–10,000 international units) is typically administered to mimic the LH surge and triggers the final steps of oocyte development. A total of 250 μg of recombinant hCG can also be used to trigger ovulation and has been shown to have comparable outcomes with urinary hCG [53]. Oocyte retrieval is performed prior to ovulation, 34–36 h after the trigger injection. hCG has a relatively long half-life and remains elevated in the serum for up to 6 days [54]. For this reason, it may exacerbate symptoms of ovarian hyperstimulation syndrome in patients at risk. Alternatively, a single bolus of GnRH agonist may be used to trigger ovulation in GnRH antagonist protocol cycles. GnRH agonist triggers an endogenous LH surge, which has a considerably shorter half-life than the endogenous LH surge that occurs in a natural cycle [55]. This short LH surge is inadequate to support the corpora lutea and limits the production of vascular endothelial growth factor, which is the key mediator leading to increased vascular permeability, the hallmark of OHSS [56]. Although initial studies reported decreased implantation and live birth outcomes with GnRH agonist trigger [57, 58] when compared to conventional hCG, most recent studies have reported comparable reproductive outcome and live birth rate in the presence of adequate luteal support [59, 60]. GnRH agonist trigger may also be ideal in situations when no fresh transfer is planned. GnRH agonist can also be used in combination with hCG trigger, known as a "dual trigger." Several studies, including a 2020 randomized clinical trial, suggests improved IVF outcomes, pregnancy, and live birth rates with the dual trigger compared to hCG alone [61–63].

17.3.9 Fertilization Methods

The two methods most commonly used to achieve fertilization are conventional IVF and intracytoplasmic sperm injection (ICSI). In conventional IVF, each oocyte is incubated with 50–100,000 motile sperm for an interval of 12–18 hours, whereas with ICSI, a single selected sperm is injected directly into an oocyte to attempt fertilization. Whether to fertilize the oocytes with conventional IVF or ICSI is a decision that should be made prior to the initiation of the IVF cycle. In general, ICSI is used when there is a known male factor or when concern for poor fertilization with conventional IVF exists. ICSI is the treatment of choice for male factor infertility, as it does not require sperm to undergo the acrosome reaction or to fuse with the oocyte membrane and may overcome the negative effects of abnormal semen characteristics and sperm quality on fertilization [64].

ICSI without male factor infertility may be of benefit for fertilization in selected patients undergoing IVF with preimplantation genetic testing for monogenic disease (PGT-M), coupled with previous failed fertilization with conventional fertilization, in vitro matured oocytes, and previously cryopreserved oocytes [65]. The number needed to treat to prevent one case of unexpected fertilization failure is approximately 30 cases of ICSI, when considering ICSI for non-male factor infertility [65]. However, regarding patients with unexplained infertility, a prior meta-analysis indicates that the number needed to treat to prevent one case of unexpected fertilization failure is 5 [66].

ICSI for unexplained infertility is associated with increased fertilization rates and decreased risk of failed fertilization; however, it has not been shown to improve live birth outcomes [65].

17.3.10 Luteal Phase Hormonal Support

Controlled ovarian stimulation cycles are associated with disruption of the luteal phase due to multiple mechanisms. Co-treatment with GnRH agonist or antagonist will result in suppressed endogenous LH levels during the luteal phase. Insufficient LH levels may be inadequate to stimulate and maintain estrogen and progesterone production by the corpora lutea required to promote endometrial development in preparation for implantation. Additionally, the disrupted follicle with the aspiration of granulosa cells may also limit the capacity of the new corpora lutea to synthesize and secrete hormones. In order to overcome these luteal deficiencies, exogenous progesterone and, in some cases, estradiol are administered in the luteal phase to support endometrial development, implantation, and early pregnancy.

Progesterone supplementation generally begins on the day of or day after oocyte retrieval [65, 67]. Progesterone can be administered orally, vaginally, or by intramuscular (IM) injections. Oral progesterone supplementation is the least common method due to intense hepatic metabolism during the first pass through the liver. This effect cannot be overcome by simply increasing the dose of progesterone administered, since it produces a degree of somnolence unacceptable to most patients. Although success rates with intramuscular versus vaginal route are reported to be similar in a fresh embryo transfer, recent data shows that exclusively using vaginal progesterone in frozen embryo transfer cycles is associated with a significant decrease in live birth rates and using intramuscular progesterone with or without vaginal progesterone supplementation is recommended [68]. Even though stopping luteal phase support after a positive pregnancy test does not seem to be associated with lower live birth rates [69], many clinics continue luteal support until 8–10 weeks of gestation [70].

Estradiol is also commonly administered for luteal phase support; however, there is no evidence that it improves outcomes compared to progesterone supplementation alone [71]. Although estrogen is commonly administered after a GnRH-only trigger due to rapid luteolysis, the optimal luteal support protocol is uncertain [72].

17.3.11 Embryo Transfer

Although embryos can be successfully transferred anytime during preimplantation development, the transfer is most commonly performed on day 3 or day 5 following the oocyte retrieval. In general, the goal of IVF is to maximize success rates while minimizing multiple gestation pregnancy rates. Transfer of a single day 5 blastocyst has been advocated as a method to minimize the risk of multiple gestation pregnancy while maintaining satisfactory pregnancy rates. However, for some patients, none of the day 3 embryos will continue to grow to the blastocyst stage. In a reputable lab, this is more reflective of embryo quality than the laboratory environment. The possibility of having no embryos to transfer is a risk with the decision to defer embryo transfer on day 3 and proceed to the blastocyst stage, and appropriate counseling is necessary.

The transfer of more than one embryo increases the chance of pregnancy but also increases the risk of multiple gestation to a much greater degree. This is especially important to consider in younger patients when the pregnancy rates are only increased by approximately 5% with more than one embryo transferred, but the risk of multiples can be as high as 40% [73]. ASRM has issued guidelines in 2017 for the number of embryos to be transferred based on the age of the woman, presence or absence of favorable characteristics, stage of embryo development, and if known, euploid status [74] (Table 17.1). The following criteria have been characterized as favorable prognosis: first cycle of IVF, good embryo quality, excess embryos for cryopreservation, or previous successful IVF. Additional favorable criteria for FET cycles include the availability of vitrified, euploid, day 5, or day 6 blastocysts for transfer [74].

Table 17.1 Recommended limits on the number of embryos to transfer

Prognosis	<35 years	35–37 years	38–40 years	41–42 years
Cleavage stage				
Euploid	1	1	1	1
Favorable	1	1	≤3	≤4
All others	≤2	≤3	≤4	≤5
Blastocysts				
Euploid	1	1	1	1
Favorable	1	1	≤2	≤3
All others	≤2	≤2	≤3	≤3

Reproduced from "Recommendations on the limits to the number of embryos to transfer: a committee opinion" with permission [74]

17.3.12 Embryo Transfer Technique

Embryo transfer is the final, and one of the most important, step in the process of IVF. The basic steps of an embryo transfer include placing a speculum in the vagina, inserting a catheter into the uterus under ultrasound guidance versus blind placement, and injecting the embryo in the upper or middle third of the uterine cavity. The optimal technique for embryo transfer has been studied, as there is large variation in techniques. In an effort to standardize the embryo transfer process, the ASRM has published guidelines and a standard embryo transfer protocol template [75, 76].

Table 17.2 summarizes various common practice techniques and whether their efficacy is supported by the literature.

Table 17.2 Interventions that are supported by the literature to improve pregnancy rates versus interventions without clear benefit [75]

Interventions supported by the literature for improving pregnancy rates	Interventions have been shown not to be beneficial for improving pregnancy rates
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	Acupuncture Analgesics, massage, general anesthesia, whole-systems traditional Chinese medicine Prophylactic antibiotics to improve embryo transfer outcomes Waiting after expulsion of embryos for any specific period of time before withdrawing the embryo transfer catheter

17.3.13 Cryopreservation of Embryos

Cryopreservation of embryos is now an integral aspect of modern ART. Cryopreservation allows for future use of the embryos that are not being transferred in a fresh cycle and thus significantly increases cumulative pregnancy rate per egg retrieval. Additionally, cryopreservation is essential for cycles with preimplantation genetic testing (PGT) as results typically take up to 2 weeks to return and is an effective strategy to reduce the risk of ovarian hyperstimulation syndrome (OHSS) [77].

Cryopreservation has two main steps: freezing and thawing/warming. Two cryo-

preservation methods are routinely used: the slow-freeze technique and vitrification. Slow-freezing is the process of gradual decrease in temperature to −30° C to –110° C before storage in liquid nitrogen. In the vitrification technique, embryos are immersed in liquid nitrogen and are flash frozen following treatment with high concentration cryoprotectants [78]. The improvement obtained with the introduction of verification has several important clinical implications in ART. Vitrification is associated with consistently higher embryo survival rates (90–100%) and yields higher implantation and pregnancy rates compared to slow-freezing protocols [79].

17.4 Special Considerations

17.4.1 Disposition of Embryos

Cryopreservation of embryos has produced important legal, ethical, and practical considerations regarding the disposition of embryos. The disposition of embryos must be clear before initiation of the IVF process and embryo creation. Generally, there are five choices for embryo disposition: warm and transfer for intended parent's pregnancy attempt, donate to another individual/couple, donate for research (or clinical training), warm and discard, and maintain indefinitely in cryostorage (this option is no longer typically offered).

In particular, the wishes of a patient or couple must be clear regarding the possible situation of death of one or both of the intended parents, divorce, separation, failure to pay storage charges, inability to agree on disposition in the future, or prolonged lack of contact with the program. For this reason, the ASRM has a committee opinion dedicated to this subject, stating that programs should develop written policies to optimize disposition dilemma and reduce potential liabilities [80].

17.4.2 Third-Party Reproduction

Third-party reproduction, which is the use of oocytes, sperm, embryos, or a gestational carrier from individuals other than the intended parent/parents, enables an infertile individual or couple to become a parent/parents. Patients who are intending to use third-party reproduction will need to go through additional testing and counseling, including a psychological evaluation. Because laws regarding third-party reproduction are different from one state to another, all couples are advised to consult with an attorney knowledgeable in reproductive and family law in their individual state(s). Additionally, potential donors and recipients should be made aware that laws may change and anonymity cannot be guaranteed for the future.

17.4.3 Oocyte Donation

The first pregnancy achieved with oocyte donation was reported in 1984, 6 years after the first human IVF baby [81]. Oocyte donation is now commonly utilized in IVF using oocytes retrieved from a healthy young donor after ovarian stimulation. Donor oocytes may be obtained from a donor undergoing a fresh stimulation cycle or from a cryobank. Following fertilization with the sperm of the recipient's partner, an appropriate number of the resulting embryos are transferred to the uterus of the recipient. Oocyte donors may be anonymous or known to the recipients. Fresh oocyte donors are typically recruited through an agency or are a known friend or family member of the intended parents.

17.4.4 Indications for Oocyte Donation

Oocyte donation is often used for women with ovarian insufficiency, genetically transmitted disease, significantly decreased ovarian reserve, advanced reproductive age, and persistent poor oocyte quality in prior IVF cycles [82]. Oocyte donation is also an option for single men or same-sex male couples who choose to build their families through assisted reproduction.

17.4.5 Evaluation of the Oocyte Donor

Donors, both anonymous and known, are screened for eligibility with extensive testing according to FDA and ASRM guidelines [82]. Psychological evaluation and counseling by a qualified mental health professional is strongly recommended for the donor and her partner (if applicable). A complete personal, medical, and family history as well as their sexual and substance abuse history should be obtained. Oocyte donors should be of legal age and preferably between the ages of 21 and 34. Proven fertility in the donor is desirable but not required. Appropriate genetic screening should be performed based on ethnic background and current recommendations.

In the USA, the Federal Drug Administration (FDA) requires following tests be performed within 30 days of oocyte collection:

(a) HIV-1 antibody as well as nucleic acid test (NAT) (spell out the first time listed)
(b) HIV-2 antibody
(c) Hepatitis C antibody and NAT
(d) Hepatitis B surface antigen
(e) Hepatitis B core antibody (IgG and IgM)
(f) Serologic test for syphilis
(g) *Neisseria gonorrhoeae* and *Chlamydia trachomatis* on urine or a swab obtained from the cervix, urethral meatus, or vagina

If all parties have satisfactory evaluations, the fresh oocyte donor undergoes ovarian stimulation. The recipient's uterus is typically prepared with exogenous estrogen and progesterone to receive the embryo(s). Many regimens for endometrial preparation have been described, and successful pregnancies have also been reported in natural cycles where donor oocytes were used to create the embryos. The use of donor oocytes for IVF consistently results in high pregnancy rates when young, healthy, fertile women donate their oocytes, with pregnancy rates from 51 to 58% per IVF cycle [83].

17.4.6 Sperm Donation

Donor insemination has been practiced since the early 1900s. Sperm donation may also be used in the context of IVF. Current FDA and ASRM guidelines recommend that in any case of therapeutic sperm donation, the sperm be quarantined for 6 months before being used, with FDA testing performed at the time of specimen collection and following the quarantine, as this decreases the risk of transmission of communicable diseases such as human immunodeficiency virus [83].

17.4.7 Indications for Sperm Donation

In heterosexual couples, therapeutic donor sperm is indicated when severe male factor (azoospermia or severe oligospermia) exists and sperm cannot be successfully recovered via invasive sperm retrieval methods. In addition, sperm donation may be indicated in cases of severe male factor infertility with history of fertilization failure in prior IVF cycles or if the male partner is a carrier of a genetic disease [83]. Single women and same-sex female couples may choose to use sperm donation to achieve pregnancy.

17.4.8 Evaluation of the Sperm Donor

Similar to oocyte donation, the sperm donor can be known or anonymous to the intended parent/parents. ASRM recommends the anonymous donors to be in the age range of 18–40 years. The ASRM also recommends that all donors be tested for communicable diseases similar to oocyte donors, although the FDA requires that only anonymous donors be tested. A thorough medical history is reviewed, with focus on sexual history, genetic issues, or psychological factors that would preclude them from being donors. A semen analysis is performed, and a test sam-

ple will evaluate the post-freezing/thawing parameters. The sperm donor is again screened for communicable diseases 6 months after the semen sample is frozen to ensure that the results of screening are negative [83].

17.4.9 Embryo Donation

Embryo donation entails transfer of embryos created by infertile couples who underwent IVF previously to another infertile patient once they achieve a pregnancy and do not desire another pregnancy or have other reasons for choosing not to use their embryos. Indications include untreatable infertility or genetic disorders that involve one or both of the partners. The evaluation process is similar to recipients of oocyte or sperm donation. Pregnancy rates following embryo donation depends on a number of factors, such as age of the women at the time of the oocyte retrieval, the quality of the embryos, and the number of embryos transferred [83].

17.4.10 Gestational Carriers

A gestational carrier is a woman who carries a pregnancy for the intended parent/parents. It is both medically and emotionally complex and involves legal and ethical issues as well. Common indications include women who lack a uterus (either due to congenital absence or surgical removal), patients with recurrent pregnancy loss related to uterine factor (such as severe Asherman syndrome), or patients with a medical contraindication to pregnancy [83].

17.4.11 IVF Outcomes

17.4.11.1 Success Rates

IVF outcomes have improved throughout the years since its introduction. Although many endpoints have been used to express IVF success rates, the most common are clinical pregnancy and live birth rates and, more recently, singleton live birth rate. Pregnancy or live birth rates may be calculated as a percentage of per cycle starts, per retrieval, or per embryo transfer. This statistic for a specific fertility clinic as well as cumulative national data can be accessed on the Society for Assisted Reproductive Technology (SART) website and is available to the public [84]. In the USA, these data are collected on an annual basis and reported by the CDC.

Table 17.3 highlights the most recent National Data Summary statistics for fresh cycles, cycles of previously frozen embryos, fresh and frozen donor oocyte, as well as frozen donor embryo cycles. The recent SART report is useful in that it notes the available data as well as limitations in the reporting process, such as cancelled cycles and delay to having outcome data (e.g., in embryo banking and fertility preservation treatments).

On the basis of long-term national data, SART has also developed an online calculator that provides individualized estimated chance of live birth rate based on patient's age, height, weight, and infertility diagnosis [75]. This calculator also provides information on the live birth rates and the chance of a multiple birth (twins, triplets, or quadruplets) when two embryos are transferred as well as cumulative live birth rate following one, two, and three fresh IVF cycles. It also provides estimated success rates if a patient uses her own oocytes versus donor oocytes.

17.4.12 Potential Adverse Outcomes

17.4.12.1 Ovarian Hyperstimulation Syndrome

One of the most serious side effects of controlled ovarian stimulation is ovarian hyperstimulation syndrome (OHSS). The incidence of mild to moderate OHSS is estimated to be 3–6% [85], while the severe form may occur in 0.9–1.4% of all cycles [86]. The pathophysiology of OHSS is not fully understood, but increased capillary permeability (as a result of hCG-induced vasoendothelial growth factor production) with the resulting third space

Table 17.3 Society for assisted reproductive technology 2016 live birth rates per intended retrieval[a] [84]

Oocyte source	Age of woman				
	<35	35–37	38–40	41–42	>42
Fresh, patient's own oocytes	40.5%	24%	15.5%	8%	3.2%
Frozen, patient's own oocytes	45.7%	37.5%	42.7%	37.9%	27.7%
Fresh, donor oocytes	54.6% (all ages)				
Frozen, donor oocytes	44.9% (all ages)				
Frozen, donor embryo	42.7% (all ages)				

[a]The primary outcome is the outcome for the first embryo transfer following an oocyte retrieval (fresh or frozen) within a year of the oocyte retrieval cycle start

fluid shift is its main feature. OHSS may be characterized as early onset (1–9 days after oocyte retrieval, usually as a result of the hCG trigger) or late onset (10 or more days after oocyte retrieval due to endogenous hCG production if pregnancy occurs) [72]. Symptoms can include increased ovarian size, nausea, vomiting, bloating, accumulation of fluid in the abdomen, breathing difficulties, hemoconcentration, and in the most severe cases, venous thromboembolic disease, kidney failure, and death. Options to decrease the risk of OHSS include cycle cancellation, administration of the dopamine agonist, cabergoline, 0.5 mg for 8 days following the trigger, coasting (withholding the gonadotropins for 1–3 days), GnRH antagonist "rescue" [87–89], use of GnRH agonist only or dual trigger (GnRHa combined with low-dose hCG) in GnRH antagonist cycles, and freeze-only cycles [90]. The relative risk of severe complications is higher if pregnancy occurs, which is why a fresh transfer is usually not performed in patients at high risk. Mild or moderate cases of OHSS may be managed outpatient with supportive therapy that involves oral hydration with electrolyte rich fluids, moderate ambulation, with daily weight, abdominal circumference, and urinary output measurements while also monitoring liver and kidney function and assessing for hemoconcentration [87]. For patients who develop severe symptoms, hospitalization may be necessary for close fluid and electrolyte management, antiemetics, anticoagulation, and occasionally paracentesis [90].

17.4.13 Risk of Cancer in Women Undergoing IVF

Infertility and nulliparity are risk factors for breast, ovarian, and endometrial cancer [91]. Although some reports have suggested an increased risk of cancer in patients who use fertility medications, most recent studies have reported no relationship between cancer and ovarian stimulation in women who underwent IVF treatment [92, 93]. Larger studies with appropriate follow-up are needed to examine the long-term effect that fertility medications may have on cancer rates, although to date there are no compelling data.

17.4.14 Obstetrical Complications

Pregnancies that occur through IVF are associated with an increased risk of certain complications, including gestational hypertension, gestational diabetes, placental abnormalities,

preterm delivery, low birth weight, and congenital malformations [94]. Some of these risks may be related to the older age of women undergoing IVF or the underlying cause of subfertility. Some risks may also be due to the IVF procedure itself, although further studies are needed to determine whether these risks are associated with the process of IVF per se or rather the underlying cause of subfertility. IVF also increases the risk of multiple gestation pregnancy. SART data from 2017 reported that, of live births in women under 35 years undergoing IVF with their own eggs, 86.9% had a singleton birth, 12.8% had a twin birth, and 0.3% had triplets or more [9]. Multiple gestation also imposes additional risks of pregnancy, both to the patient and the offspring.

17.4.15 Risks to the Offspring

Numerous studies have been conducted to assess the overall health of children conceived via IVF, and the majority of studies have been reassuring. A major problem with studies done thus far has been comparing a group of infertile couples with pregnancies achieved through IVF to a group of normally fertile couples with unassisted conceptions. Interestingly, a recent study has addressed this issue [95]. This study compared health of children of fertile couples with children of subfertile couples with and without ART treatment. Results from this study showed that prematurity was more common in subfertile couples with or without ART treatments when compared to fertile couples [95]. When stratified by gestational age (GA), infants of subfertile mothers with or without ART were at greater risk for congenital malformations [95]. Additionally, when comparing infants born to subfertile mothers without ART treatments, infants born to ART-treated mothers were at lower risk for being small for GA and having congenital malformations and cardiovascular conditions and at higher risk for infectious disease conditions [95].

A number of studies also suggest that ART may be associated with an increased risk of imprinting disorders, theoretically due to laboratory manipulations that occur during meiosis [96]. Because imprinting disorders are quite rare, a causal relationship with IVF is difficult to determine.

Another important question is whether the ICSI procedure, in particular, affects the risk of congenital malformations and long-term health of the offspring. The invasive nature of ICSI circumvents natural selection mechanisms, and the ICSI process, as well as the underlying related infertility conditions that lead to the need of ICSI, has raised concerns regarding this matter. There have been some reports of the possible increased risk of congenital and urogenital malformations, epigenetic disorders, chromosomal abnormalities, infertility, cancer, delayed psychological and neurological development, and impaired cardiometabolic profile compared with naturally conceived children [97, 98]. Taken together, the literature suggests that infertile couples may have a higher risk of having children with congenital malformations. It is unclear whether the process of IVF itself increases this risk. Indeed, if additional risk is present, the effect size is small.

The vast majority of risks to the offspring are related to multiple gestation pregnancy and preterm delivery and the comorbidities that are associated with prematurity. It has been shown that singletons conceived with IVF tend to be born slightly earlier than naturally conceived babies (39.1 weeks compared to 39.5 weeks) and IVF twins are not born earlier than naturally conceived twins. There may be a slight increase in low birth weight of IVF singletons conceived following fresh embryo transfer compared to naturally conceived singletons [94].

Monochorionic twinning occurs in approximately 2–3% of IVF pregnancies, which is higher than the spontaneous rate of 0.4% for in vivo conceptions [99]. This further increases the risk to the pregnancy, as complications such as twin-twin transfusion may occur (in up to 20% of monochorionic diamniotic gestations) as well as umbilical cord entanglement (in monochorionic monoamniotic gestations).

17.4.16 Controversies

17.4.16.1 Freeze-Only with Cryopreserved Transfer Versus Fresh Embryo Transfer

As stated earlier, with the development and refinement of vitrification techniques and improved embryo survival, the pregnancy rates following frozen embryo transfer (FET) have approached, if not exceeded, fresh transfer. According to the most recent national SART report in 2016, the live birth rate following FET was higher in all age groups than the corresponding rates following fresh embryo transfer among patients undergoing an autologous cycle [84]. Accordingly, there has been a shift in practice toward favoring IVF with FET in the recent years the USA [100]. As FET has become more common, freeze-only protocols have emerged in which all good-quality embryos are electively frozen and transferred in a later natural or medicated cycle.

Improved outcomes resulted from FET are attributed to reduction in OHSS rates, allowing for preimplantation genetic testing (PGT) of embryos, as well as overcoming embryo-endometrial asynchrony, due to either delayed blastulation or premature progesterone elevation. In addition, there is increasing evidence that FET may lead to more favorable perinatal and live birth outcomes, including a lower risk of preterm birth, low birth weight, placenta previa, and placental abruption. However, this may be at the cost of increased rates of pre-eclampsia, large for gestational age, and high birth weight [101, 102]. The risk for these complications may be related to the method of endometrial preparation (i.e., natural cycle versus programed) [103]. It should be considered that the use of a freeze-only strategy may be more expensive due to the costs of embryo cryopreservation, endometrial priming, extra medication use, and ultrasound monitoring for FET. However, there is increasing evidence that in a planned freeze-only cycle, pituitary suppression with progesterone in lieu of a GnRH antagonist is more cost-effective by approximately $2000 and with comparable outcomes [104, 105]. In addition, it is not yet clear which specific groups of patients might specifically benefit from the use of a freeze-only versus fresh transfer.

Data from a recent systematic review and meta-analysis suggests that a significantly higher probability of live birth occurs in high but not normal responders after FET when compared to fresh embryo transfer [106]. A cost-effective analysis performed alongside an RCT, with the effectiveness measure being live birth rate, showed that there is low probability of the freeze-only strategy being more cost-effective than fresh transfer for women, except those with PCOS [107].

Based on the available literature, common scenarios to consider implementing a freeze-only strategy include patients at risk of OHSS, premature progesterone elevation (discussed later), and in those undergoing PGT. Otherwise, the use of FET for other clinical scenarios is unlikely to offer meaningful improvements in outcomes that outweigh the added cost of treatment.

17.4.16.2 Preimplantation Genetic Testing

Preimplantation genetic testing (PGT) involves the removal of one or more cells from the dividing embryo to test for genetic content. PGT is subdivided into PGT-M (for monogenic disorder), PGT-SR (structural rearrangements), and PGT-A (aneuploidy screening). For the purpose of this section, we will focus on the clinical outcomes following embryo biopsy and selection with PGT-A.

The high incidence of chromosomal abnormalities observed with increasing maternal age is the major cause of miscarriage and IVF failure [84]. Most aneuploidies take place in the process of meiosis in the female partner and contribute to rapidly declining IVF success and live birth rates in women over the age of 35. In contrast, IVF cycles with the transfer of a euploid embryo have similar implantation rates regardless of maternal age [84]. Thus, PGT-A has been proposed as a method to select embryos with the highest potential of ongoing implantation.

In contemporary IVF practice, trophectoderm biopsy with single nucleotide polymor-

phism (SNP) array, array comparative genomic hybridization (aCGH), quantitative real-time polymerase chain reaction (qPCR), or next-generation sequencing (NGS) are routinely used for aneuploidy detection. Several, mainly single-center, RCTs performed in the past decade have shown significant improvement in ongoing pregnancy rates per embryo transfer procedure following PGT-A [108–110]. However, data from a recent multicenter randomized controlled trial (the STAR study) demonstrated that the use of trophectoderm biopsy at the blastocyst stage and NGS-based PGT-A to select euploid embryos for single vitrified-warmed blastocyst transfer significantly improved ongoing pregnancy rates and live birth rates per transfer in women aged 35–40 years, but not in women aged 25–34 years [111]. Additionally, there was no difference in outcomes in any age group on an intent to treat basis.

Possible explanations for the lack of improvement in outcomes following transfer of euploid embryos in the STAR trial include the following: (1) trophectoderm biopsy may reduce an embryo's potential for implantation more than previously realized, or (2) testing results from the trophectoderm may not be representative of the embryo as a whole, thus resulting in discarding embryos that have the potential for pregnancy due to being deemed abnormal. PGT is inherently imperfect. It is an invasive procedure, and errors may occur during the genetic analysis of a small amount of DNA collected. This may lead to indeterminate results or, in some instances, discarding of a normal embryo following erroneous abnormal results. More importantly, finding trophectoderm mosaicism with limited data on implantation potential may create more questions than answers and lead to lengthy consultation with the patient (and a genetic counselor) on whether to consider those embryos for transfer [112]. Ultimately, it is important to counsel patients about the use of PGT and its clinical efficacy in an individualized manner regarding potential benefits and cost-effectiveness of PGT in a case-by-case basis rather than a universal approach.

17.4.16.3 Management of Prematurely Elevated Progesterone in IVF Cycle

Despite the utilization of GnRH analogues and GnRH antagonists to suppress the pituitary and prevent ovulation, premature progesterone elevation has been reported to still occur in 5–38% of IVF cycles [112–115]. Premature progesterone elevation can lead to asynchrony between the embryo and the endometrium and thus adversely affect implantation rates [116–119]. In the late 1980s, Hamori et al. and Feldberg et al. first reported a concern of prematurely elevated progesterone in IVF cycles leading to decreased pregnancy rates [120, 121]. Shortly thereafter, Schoolcraft et al. and Fanchin et al. found that a prematurely elevated progesterone level on the day of hCG trigger led to decreased pregnancy rates in IVF cycles [122, 123]. Furthermore, preliminary evidence continued to support that prematurely elevated progesterone levels on the day of hCG trigger led to decreased pregnancy rates as demonstrated by Bosch et al. (>4000 IVF cycles) using a progesterone threshold of 1.5 ng/mL [114] and Xu et al. (>10,000 IVF cycles) [124] using a progesterone threshold of 1.75 ng/mL and confirmed by a large meta-analysis by Venetis et al. (>60,000 IVF cycles) [125].

Recently, several studies have clearly shown that a progesterone level using thresholds of 1.5 and 2.0 ng/mL on the day of trigger leads to decreased pregnancy rates, and a freeze-all approach of the embryos should be considered without performing a fresh embryo transfer [126–130]. Although the evidence supports that premature progesterone elevation decreases pregnancy rates, the definition of the progesterone threshold has not been clearly defined and has changed over the years [104]. A publication by Hill et al. performed a threshold and cost analysis of 7608 IVF cycles demonstrating that freezing embryos in lieu of a fresh embryo transfer is cost-effective, if the progesterone on the day of trigger is 1.5 ng/mL or above, with a number needed to treat of 13 [131]. At these thresholds, elevated

progesterone on the day of trigger showed a decrease in live birth rate of up to 20% when the progesterone was at 2.0 ng/mL on the day of trigger. In conclusion, an elevated progesterone on the day of trigger demonstrates a clinically significant negative impact in IVF cycles, and those at risk for fresh transfer failure should be counseled of the risks versus benefits in proceeding with a transfer.

17.5 Review Questions

1. About what percentage of US couples have impaired fertility?
   A. 10%
   B. 12%
   C. More than 25%
   D. Less than 5%
2. What is the recommended number of cleavage stage embryos to be transferred in a 34-year-old female?
   A. 2
   B. 3 or less
   C. 1
   D. Up to 4
3. Which of the following obstetrical complications has *not* been associated with IVF?
   A. Gestational hypertension
   B. Abnormalities of the placenta
   C. Low birth weight
   D. Oligohydramnios
4. Which of the following is *not* the role of GnRH analogues in ovarian stimulation protocols?
   A. Prevention of OHSS
   B. Induction of oocyte maturation
   C. Prevention of premature ovulation
   D. Improving oocyte quality

17.6 Answer

1. B
2. C
3. D
4. D

References

1. Practice Committee of the American Society for Reproductive M. Definitions of infertility and recurrent pregnancy loss: a committee opinion. Fertil Steril. 2013;99(1):63.
2. Zegers-Hochschild F, et al. International Committee for Monitoring Assisted Reproductive Technology (ICMART) and the World Health Organization (WHO) revised glossary of ART terminology, 2009. Fertil Steril. 2009;92(5):1520–4.
3. Eisenberg ML, et al. Relationship between semen production and medical comorbidity. Fertil Steril. 2015;103(1):66–71.
4. Kurabayashi T, et al. Ovarian infertility is associated with cardiovascular disease risk factors in later life: a Japanese cross-sectional study. Maturitas. 2016;83:33–9.
5. Meczekalski B, et al. Functional hypothalamic amenorrhea and its influence on women's health. J Endocrinol Investig. 2014;37(11):1049–56.
6. Tobias DK, et al. History of infertility and risk of type 2 diabetes mellitus: a prospective cohort study. Diabetologia. 2015;58(4):707–15.
7. Chandra A, Copen CE, Stephen EH. Infertility and impaired fecundity in the United States, 1982–2010: data from the National Survey of Family Growth. Natl Health Stat Report. 2013(67):1–18, 1 p following 19.
8. Chandra A, Copen CE, Stephen EH. Infertility service use in the United States: data from the National Survey of Family Growth, 1982-2010. Natl Health Stat Report. 2014;73:1–21.
9. Society for Assisted Reproductive Technology. SART national summary report 2017. 2017; Available from: https://www.sartcorsonline.com/rptCSR_PublicMultYear.aspx?ClinicPKID=0.
10. Sonmezer M, et al. Random-start controlled ovarian hyperstimulation for emergency fertility preservation in letrozole cycles. Fertil Steril. 2011;95(6):2125 e9-11.
11. Cakmak H, et al. Effective method for emergency fertility preservation: random-start controlled ovarian stimulation. Fertil Steril. 2013;100(6):1673–80.
12. Hull MG, et al. Delayed conception and active and passive smoking. The Avon Longitudinal Study of Pregnancy and Childhood Study Team. Fertil Steril. 2000;74(4):725–33.
13. Mueller BA, et al. Recreational drug use and the risk of primary infertility. Epidemiology. 1990;1(3):195–200.
14. Mumford SL, et al. Cannabis use while trying to conceive: a prospective cohort study evaluating associations with fecundability, live birth and pregnancy loss. Hum Reprod; 2021;36(5): 1405−15.
15. Payne KS, et al. Cannabis and male fertility: a systematic review. J Urol. 2019;202(4):674–81.

16. Klonoff-Cohen H, Lam-Kruglick P, Gonzalez C. Effects of maternal and paternal alcohol consumption on the success rates of in vitro fertilization and gamete intrafallopian transfer. Fertil Steril. 2003;79(2):330–9.
17. Caan B, Quesenberry CP Jr, Coates AO. Differences in fertility associated with caffeinated beverage consumption. Am J Public Health. 1998;88(2):270–4.
18. Cnattingius S, et al. Caffeine intake and the risk of first-trimester spontaneous abortion. N Engl J Med. 2000;343(25):1839–45.
19. Hussein N, et al. Preconception risk assessment for thalassaemia, sickle cell disease, cystic fibrosis and Tay-Sachs disease. Cochrane Database Syst Rev. 2018;3:CD010849.
20. Society for Assisted Reproductive Technology. Patient Evaluation. 2021; Available from: https://www.sart.org/patients/sart-patient-evaluation/.
21. Practice Committee of the American Society for Reproductive Medicine. Electronic address, a.a.o. and M. Practice Committee of the American Society for Reproductive. Testing and interpreting measures of ovarian reserve: a committee opinion. Fertil Steril. 2020;114(6):1151–7.
22. Verhagen TE, et al. The accuracy of multivariate models predicting ovarian reserve and pregnancy after in vitro fertilization: a meta-analysis. Hum Reprod Update. 2008;14(2):95–100.
23. Soares SR, Barbosa dos Reis MM, Camargos AF. Diagnostic accuracy of sonohysterography, transvaginal sonography, and hysterosalpingography in patients with uterine cavity diseases. Fertil Steril. 2000;73(2):406–11.
24. Practice Committee of the American Society for Reproductive M. Diagnostic evaluation of the infertile female: a committee opinion. Fertil Steril. 2015;103(6):e44–50.
25. Schlegel PN, et al. Diagnosis and treatment of infertility in men: AUA/ASRM guideline part II. Fertil Steril. 2021;115(1):62–9.
26. Zeyneloglu HB, Arici A, Olive DL. Adverse effects of hydrosalpinx on pregnancy rates after in vitro fertilization-embryo transfer. Fertil Steril. 1998;70(3):492–9.
27. Camus E, et al. Pregnancy rates after in-vitro fertilization in cases of tubal infertility with and without hydrosalpinx: a meta-analysis of published comparative studies. Hum Reprod. 1999;14(5):1243–9.
28. Melo P, et al. Surgical treatment for tubal disease in women due to undergo in vitro fertilisation. Cochrane Database Syst Rev. 2020;10:CD002125.
29. Sagoskin AW, et al. Salpingectomy or proximal tubal occlusion of unilateral hydrosalpinx increases the potential for spontaneous pregnancy. Hum Reprod. 2003;18(12):2634–7.
30. Hollowell JG Jr, Garbe PL, Miller DT. Maternal thyroid deficiency during pregnancy and subsequent neuropsychological development of the child. N Engl J Med. 1999;341(26):2016–7.
31. Taylor PN, et al. TSH levels and risk of miscarriage in women on long-term levothyroxine: a community-based study. J Clin Endocrinol Metab. 2014;99(10):3895–902.
32. Thyroid disease in pregnancy: ACOG practice bulletin, number 223. Obstet Gynecol. 2020;135(6):e261–74.
33. Hillier SG. Paracrine control of follicular estrogen synthesis. Semin Reprod Endocrinol. 1991;9(4):332–40.
34. Santi D, et al. Efficacy of follicle-stimulating hormone (FSH) alone, FSH + luteinizing hormone, human menopausal gonadotropin or FSH + human chorionic gonadotropin on assisted reproductive technology outcomes in the "personalized" medicine era: a meta-analysis. Front Endocrinol (Lausanne). 2017;8:114.
35. Werner MD, et al. Defining the "sweet spot" for administered luteinizing hormone-to-follicle-stimulating hormone gonadotropin ratios during ovarian stimulation to protect against a clinically significant late follicular increase in progesterone: an analysis of 10,280 first in vitro fertilization cycles. Fertil Steril. 2014;102(5):1312–7.
36. Farquhar C, et al. Oral contraceptive pill, progestogen or oestrogen pretreatment for ovarian stimulation protocols for women undergoing assisted reproductive techniques. Cochrane Database Syst Rev. 2017;5:CD006109.
37. Montoya-Botero P, et al. The effect of type of oral contraceptive pill and duration of use on fresh and cumulative live birth rates in IVF/ICSI cycles. Hum Reprod. 2020;35(4):826–36.
38. Schmitz C, et al. Does the degree of hypothalamic-pituitary-ovarian recovery after oral contraceptive pills affect outcomes of IVF/ICSI cycles receiving GnRH-antagonist adjuvant therapy in women over 35 years of age? J Assist Reprod Genet. 2012;29(9):877–82.
39. Tarlatzis BC, et al. Clinical management of low ovarian response to stimulation for IVF: a systematic review. Hum Reprod Update. 2003;9(1):61–76.
40. Alexander VM, et al. Ovarian stimulation for fertility preservation in women with cancer: a systematic review and meta-analysis comparing random and conventional starts. J Gynecol Obstet Hum Reprod. 2021;50(8):102080.
41. Porter RN, et al. Induction of ovulation for in-vitro fertilisation using buserelin and gonadotropins. Lancet. 1984;2(8414):1284–5.
42. Kumar P, Sharma A. Gonadotropin-releasing hormone analogs: understanding advantages and limitations. J Hum Reprod Sci. 2014;7(3):170–4.
43. Padilla SL, et al. Use of the flare-up protocol with high dose human follicle stimulating hormone and human menopausal gonadotropins for in vitro fertilization in poor responders. Fertil Steril. 1996;65(4):796–9.
44. Pandian Z, et al. Interventions for 'poor responders' to controlled ovarian hyper stimulation

(COH) in in-vitro fertilisation (IVF). Cochrane Database Syst Rev. 2010;1:CD004379.
45. Xiao J, Chang S, Chen S. The effectiveness of gonadotropin-releasing hormone antagonist in poor ovarian responders undergoing in vitro fertilization: a systematic review and meta-analysis. Fertil Steril. 2013;100(6):1594–601 e1–9.
46. Pu D, Wu J, Liu J. Comparisons of GnRH antagonist versus GnRH agonist protocol in poor ovarian responders undergoing IVF. Hum Reprod. 2011;26(10):2742–9.
47. Maggi R, et al. GnRH and GnRH receptors in the pathophysiology of the human female reproductive system. Hum Reprod Update. 2016;22(3):358–81.
48. Olivennes F, et al. The use of GnRH antagonists in ovarian stimulation. Hum Reprod Update. 2002;8(3):279–90.
49. Barmat LI, et al. A randomized prospective trial comparing gonadotropin-releasing hormone (GnRH) antagonist/recombinant follicle-stimulating hormone (rFSH) versus GnRH-agonist/rFSH in women pretreated with oral contraceptives before in vitro fertilization. Fertil Steril. 2005;83(2):321–30.
50. Kolibianakis EM, et al. Fixed versus flexible gonadotropin-releasing hormone antagonist administration in in vitro fertilization: a randomized controlled trial. Fertil Steril. 2011;95(2):558–62.
51. Escudero E, et al. Comparison of two different starting multiple dose gonadotropin-releasing hormone antagonist protocols in a selected group of in vitro fertilization-embryo transfer patients. Fertil Steril. 2004;81(3):562–6.
52. Lambalk CB, et al. GnRH antagonist versus long agonist protocols in IVF: a systematic review and meta-analysis accounting for patient type. Hum Reprod Update. 2017;23(5):560–79.
53. Youssef MA, Abou-Setta AM, Lam WS. Recombinant versus urinary human chorionic gonadotrophin for final oocyte maturation triggering in IVF and ICSI cycles. Cochrane Database Syst Rev. 2016;4:CD003719.
54. Casper RF. Basic understanding of gonadotropin-releasing hormone-agonist triggering. Fertil Steril. 2015;103(4):867–9.
55. Turkgeldi E, et al. Gonadotropin-releasing hormone agonist triggering of oocyte maturation in assisted reproductive technology cycles. Turk J Obstet Gynecol. 2015;12(2):96–101.
56. Wang TH, et al. Human chorionic gonadotropin-induced ovarian hyperstimulation syndrome is associated with up-regulation of vascular endothelial growth factor. J Clin Endocrinol Metab. 2002;87(7):3300–8.
57. Humaidan P, et al. GnRH agonist (buserelin) or hCG for ovulation induction in GnRH antagonist IVF/ICSI cycles: a prospective randomized study. Hum Reprod. 2005;20(5):1213–20.
58. Kolibianakis EM, et al. A lower ongoing pregnancy rate can be expected when GnRH agonist is used for triggering final oocyte maturation instead of HCG in patients undergoing IVF with GnRH antagonists. Hum Reprod. 2005;20(10):2887–92.
59. Haahr T, et al. GnRH agonist trigger and LH activity luteal phase support versus hCG trigger and conventional luteal phase support in fresh embryo transfer IVF/ICSI cycles-a systematic PRISMA review and meta-analysis. Front Endocrinol (Lausanne). 2017;8:116.
60. Yilmaz N, et al. GnRH agonist versus HCG triggering in different IVF/ICSI cycles of same patients: a retrospective study. J Obstet Gynaecol. 2020;40(6):837–42.
61. Haas J, et al. GnRH agonist and hCG (dual trigger) versus hCG trigger for final follicular maturation: a double-blinded, randomized controlled study. Hum Reprod. 2020;35(7):1648–54.
62. Ehrenreich H, Goebel FD. The role of opioids in the endocrine function of the pancreas. Diabetes Res. 1986;3(2):59–66.
63. Li S, et al. Dual trigger of triptorelin and HCG optimizes clinical outcome for high ovarian responder in GnRH-antagonist protocols. Oncotarget. 2018;9(4):5337–43.
64. Practice Committee of American Society for Reproductive M. Intracytoplasmic sperm injection (ICSI). Fertil Steril. 2008;90(5 Suppl):S187.
65. Practice Committees of the American Society for Reproductive M. and a.a.o. the Society for Assisted Reproductive Technology. Electronic address, Intracytoplasmic sperm injection (ICSI) for non-male factor indications: a committee opinion. Fertil Steril. 2020;114(2):239–45.
66. Johnson LN, et al. Does intracytoplasmic sperm injection improve the fertilization rate and decrease the total fertilization failure rate in couples with well-defined unexplained infertility? A systematic review and meta-analysis. Fertil Steril. 2013;100(3):704–11.
67. Practice Committee of the American Society for Reproductive M. Progesterone supplementation during the luteal phase and in early pregnancy in the treatment of infertility: an educational bulletin. Fertil Steril. 2008;89(4):789–92.
68. Devine K, et al. Vitrified blastocyst transfer cycles with the use of only vaginal progesterone replacement with Endometrin have inferior ongoing pregnancy rates: results from the planned interim analysis of a three-arm randomized controlled noninferiority trial. Fertil Steril. 2018;109(2):266–75.
69. Pan SP, et al. Early stop of progesterone supplementation after confirmation of pregnancy in IVF/ICSI fresh embryo transfer cycles of poor responders does not affect pregnancy outcome. PLoS One. 2018;13(8):e0201824.
70. Shoham G, Leong M, Weissman A. A 10-year follow-up on the practice of luteal phase support using worldwide web-based surveys. Reprod Biol Endocrinol. 2021;19(1):15.

71. van der Linden M, et al. Luteal phase support for assisted reproduction cycles. Cochrane Database Syst Rev. 2015;7:CD009154.
72. Jerome S, Robert B. Medical approaches to ovarian stimulation for infertility. In: Yen & Jaffe's reproductive endocrinology. Elsevier; 2018.
73. Stillman RJ, et al. Elective single embryo transfer: a 6-year progressive implementation of 784 single blastocyst transfers and the influence of payment method on patient choice. Fertil Steril. 2009;92(6):1895–906.
74. Practice Committee of the American Society for Reproductive Medicine. Electronic address, A.a.o. and T. Practice Committee of the Society for Assisted Reproductive, Guidance on the limits to the number of embryos to transfer: a committee opinion. Fertil Steril. 2017;107(4):901–3.
75. Practice Committee of the American Society for Reproductive Medicine. Electronic address, A.a.o. and M. Practice Committee of the American Society for Reproductive, Performing the embryo transfer: a guideline. Fertil Steril. 2017;107(4):882–96.
76. Practice Committee of the American Society for Reproductive Medicine. Electronic address, A.a.o., et al. ASRM standard embryo transfer protocol template: a committee opinion. Fertil Steril. 2017;107(4):897–900.
77. D'Angelo A, Amso N. Embryo freezing for preventing ovarian hyperstimulation syndrome. Cochrane Database Syst Rev. 2007;3:CD002806.
78. Kuwayama M. Highly efficient vitrification for cryopreservation of human oocytes and embryos: the Cryotop method. Theriogenology. 2007;67(1):73–80.
79. Rienzi L, et al. Oocyte, embryo and blastocyst cryopreservation in ART: systematic review and meta-analysis comparing slow-freezing versus vitrification to produce evidence for the development of global guidance. Hum Reprod Update. 2017;23(2):139–55.
80. Ethics Committee of the American Society for Reproductive M. Disposition of abandoned embryos: a committee opinion. Fertil Steril. 2013;99(7):1848–9.
81. Lutjen P, et al. The establishment and maintenance of pregnancy using in vitro fertilization and embryo donation in a patient with primary ovarian failure. Nature. 1984;307(5947):174–5.
82. Practice Committee of the American Society for Reproductive M. and T. the Practice Committee of the Society for Assisted Reproductive, Recommendations for gamete and embryo donation: a committee opinion. Fertil Steril. 2013;99(1):47–62 e1.
83. Myers ER. Outcomes of donor oocyte cycles in assisted reproduction. JAMA. 2013;310(22):2403–4.
84. Centers for Disease Control and Prevention. 2014 assisted reproductive technology national summary report. US Dept of Health and Human Services. 2016.
85. Delvigne A, Rozenberg S. Epidemiology and prevention of ovarian hyperstimulation syndrome (OHSS): a review. Hum Reprod Update. 2002;8(6):559–77.
86. Tomas C, et al. Annual incidence of severe ovarian hyperstimulation syndrome. Dan Med J. 2021;68(2):A12190738.
87. Gustofson RL, Segars JH, Larsen FW. Ganirelix acetate causes a rapid reduction in estradiol levels without adversely affecting oocyte maturation in women pretreated with leuprolide acetate who are at risk of ovarian hyperstimulation syndrome. Hum Reprod. 2006;21(11):2830–7.
88. Aboulghar MA, et al. A prospective randomized study comparing coasting with GnRH antagonist administration in patients at risk for severe OHSS. Reprod Biomed Online. 2007;15(3):271–9.
89. Hill MJ, et al. GnRH antagonist rescue in high responders at risk for OHSS results in excellent assisted reproduction outcomes. Reprod Biomed Online. 2012;25(3):284–91.
90. Practice Committee of the American Society for Reproductive Medicine. Electronic address, A.a.o. and M. Practice Committee of the American Society for Reproductive, Prevention and treatment of moderate and severe ovarian hyperstimulation syndrome: a guideline. Fertil Steril. 2016;106(7):1634–47.
91. Britt K, Short R. The plight of nuns: hazards of nulliparity. Lancet. 2012;379(9834):2322–3.
92. van den Belt-Dusebout AW, et al. Ovarian stimulation for in vitro fertilization and long-term risk of breast cancer. JAMA. 2016;316(3):300–12.
93. Yli-Kuha AN, et al. Cancer morbidity in a cohort of 9175 Finnish women treated for infertility. Hum Reprod. 2012;27(4):1149–55.
94. Hayashi M, et al. Adverse obstetric and perinatal outcomes of singleton pregnancies may be related to maternal factors associated with infertility rather than the type of assisted reproductive technology procedure used. Fertil Steril. 2012;98(4):922–8.
95. Hwang SS, et al. Health of infants after ART-treated, subfertile, and fertile deliveries. Pediatrics. 2018;142(2):e20174069.
96. Hattori H, et al. Association of four imprinting disorders and ART. Clin Epigenetics. 2019;11(1):21.
97. Massaro PA, et al. Does intracytoplasmic sperm injection pose an increased risk of genitourinary congenital malformations in offspring compared to in vitro fertilization? A systematic review and meta-analysis. J Urol. 2015;193(5 Suppl):1837–42.
98. Tararbit K, et al. The risk for four specific congenital heart defects associated with assisted reproductive techniques: a population-based evaluation. Hum Reprod. 2013;28(2):367–74.
99. Busnelli A, et al. Risk factors for monozygotic twinning after in vitro fertilization: a systematic review and meta-analysis. Fertil Steril. 2019;111(2):302–17.

100. Shapiro BS, et al. Clinical rationale for cryopreservation of entire embryo cohorts in lieu of fresh transfer. Fertil Steril. 2014;102(1):3–9.
101. Maheshwari A, et al. Is frozen embryo transfer better for mothers and babies? Can cumulative meta-analysis provide a definitive answer? Hum Reprod Update. 2018;24(1):35–58.
102. Maheshwari A, Raja EA, Bhattacharya S. Obstetric and perinatal outcomes after either fresh or thawed frozen embryo transfer: an analysis of 112,432 singleton pregnancies recorded in the Human Fertilisation and Embryology Authority anonymized dataset. Fertil Steril. 2016;106(7):1703–8.
103. Ginstrom Ernstad E, et al. Neonatal and maternal outcome after frozen embryo transfer: increased risks in programmed cycles. Am J Obstet Gynecol. 2019;221(2):126 e1–126 e18.
104. Evans MB, et al. Adverse effect of prematurely elevated progesterone in in vitro fertilization cycles: a literature review. Biol Reprod. 2018;99(1):45–51.
105. Giles J, et al. Medroxyprogesterone acetate is a useful alternative to a gonadotropin-releasing hormone antagonist in oocyte donation: a randomized, controlled trial. Fertil Steril. 2021;116(2):404−12.
106. Bosdou JK, et al. Higher probability of live-birth in high, but not normal, responders after first frozen-embryo transfer in a freeze-only cycle strategy compared to fresh-embryo transfer: a meta-analysis. Hum Reprod. 2019;34(3):491–505.
107. Le KD, et al. A cost-effectiveness analysis of freeze-only or fresh embryo transfer in IVF of non-PCOS women. Hum Reprod. 2018;33(10):1907–14.
108. Rubio C, et al. In vitro fertilization with preimplantation genetic diagnosis for aneuploidies in advanced maternal age: a randomized, controlled study. Fertil Steril. 2017;107(5):1122–9.
109. Scott RT Jr, et al. Blastocyst biopsy with comprehensive chromosome screening and fresh embryo transfer significantly increases in vitro fertilization implantation and delivery rates: a randomized controlled trial. Fertil Steril. 2013;100(3):697–703.
110. Harton GL, et al. Diminished effect of maternal age on implantation after preimplantation genetic diagnosis with array comparative genomic hybridization. Fertil Steril. 2013;100(6):1695–703.
111. Munne S, et al. Preimplantation genetic testing for aneuploidy versus morphology as selection criteria for single frozen-thawed embryo transfer in good-prognosis patients: a multicenter randomized clinical trial. Fertil Steril. 2019;112(6):1071–1079 e7.
112. Viotti M, et al. Using outcome data from one thousand mosaic embryo transfers to formulate an embryo ranking system for clinical use. Fertil Steril. 2021;115(5):1212–24.
113. Silverberg KM, et al. Elevated serum progesterone levels on the day of human chorionic gonadotropin administration in in vitro fertilization cycles do not adversely affect embryo quality. Fertil Steril. 1994;61(3):508–13.
114. Bosch E, et al. Circulating progesterone levels and ongoing pregnancy rates in controlled ovarian stimulation cycles for in vitro fertilization: analysis of over 4000 cycles. Hum Reprod. 2010;25(8):2092–100.
115. Edelstein MC, et al. Progesterone levels on the day of human chorionic gonadotropin administration in cycles with gonadotropin-releasing hormone agonist suppression are not predictive of pregnancy outcome. Fertil Steril. 1990;54(5):853–7.
116. Van Vaerenbergh I, et al. Progesterone rise on HCG day in GnRH antagonist/rFSH stimulated cycles affects endometrial gene expression. Reprod Biomed Online. 2011;22(3):263–71.
117. Li R, et al. MicroRNA array and microarray evaluation of endometrial receptivity in patients with high serum progesterone levels on the day of hCG administration. Reprod Biol Endocrinol. 2011;9:29.
118. Labarta E, et al. Endometrial receptivity is affected in women with high circulating progesterone levels at the end of the follicular phase: a functional genomics analysis. Hum Reprod. 2011;26(7):1813–25.
119. Shulman A, et al. The significance of an early (premature) rise of plasma progesterone in in vitro fertilization cycles induced by a "long protocol" of gonadotropin releasing hormone analogue and human menopausal gonadotropins. J Assist Reprod Genet. 1996;13(3):207–11.
120. Feldberg D, et al. The impact of high progesterone levels in the follicular phase of in vitro fertilization (IVF) cycles: a comparative study. J In Vitro Fert Embryo Transf. 1989;6(1):11–4.
121. Hamori M, et al. Premature luteinization of follicles during ovarian stimulation for in-vitro fertilization. Hum Reprod. 1987;2(8):639–43.
122. Fanchin R, et al. Premature progesterone elevation spares blastulation but not pregnancy rates in in vitro fertilization with coculture. Fertil Steril. 1997;68(4):648–52.
123. Schoolcraft W, et al. Lower pregnancy rate with premature luteinization during pituitary suppression with leuprolide acetate. Fertil Steril. 1991;55(3):563–6.
124. Xu B, et al. Serum progesterone level effects on the outcome of in vitro fertilization in patients with different ovarian response: an analysis of

more than 10,000 cycles. Fertil Steril. 2012;97(6):1321–7.e1–4.
125. Venetis CA, et al. Progesterone elevation and probability of pregnancy after IVF: a systematic review and meta-analysis of over 60 000 cycles. Hum Reprod Update. 2013;19(5):433–57.
126. Healy MW, et al. Does a frozen embryo transfer ameliorate the effect of elevated progesterone seen in fresh transfer cycles? Fertil Steril. 2016;105(1):93–9.e1.
127. Hill MJ, et al. Are good patient and embryo characteristics protective against the negative effect of elevated progesterone level on the day of oocyte maturation? Fertil Steril. 2015;103(6):1477–84.e1–5.
128. Healy M, et al. Does premature elevated progesterone on the day of trigger increase spontaneous abortion rates in fresh and subsequent frozen embryo transfers? Gynecol Endocrinol. 2017;33(6):472–5.
129. Hill MJ, et al. Does elevated progesterone on day of oocyte maturation play a role in the racial disparities in IVF outcomes? Reprod Biomed Online. 2017;34(2):154–61.
130. Connell MT, et al. Is the effect of premature elevated progesterone augmented by human chorionic gonadotropin versus gonadotropin-releasing hormone agonist trigger? Fertil Steril. 2016;106(3):584–589.e1.
131. Hill MJ, et al. Defining thresholds for abnormal premature progesterone levels during ovarian stimulation for assisted reproduction technologies. Fertil Steril. 2018;110(4):671–679.e2.