

Assisted Reproductive Technology: Techniques and Limitations

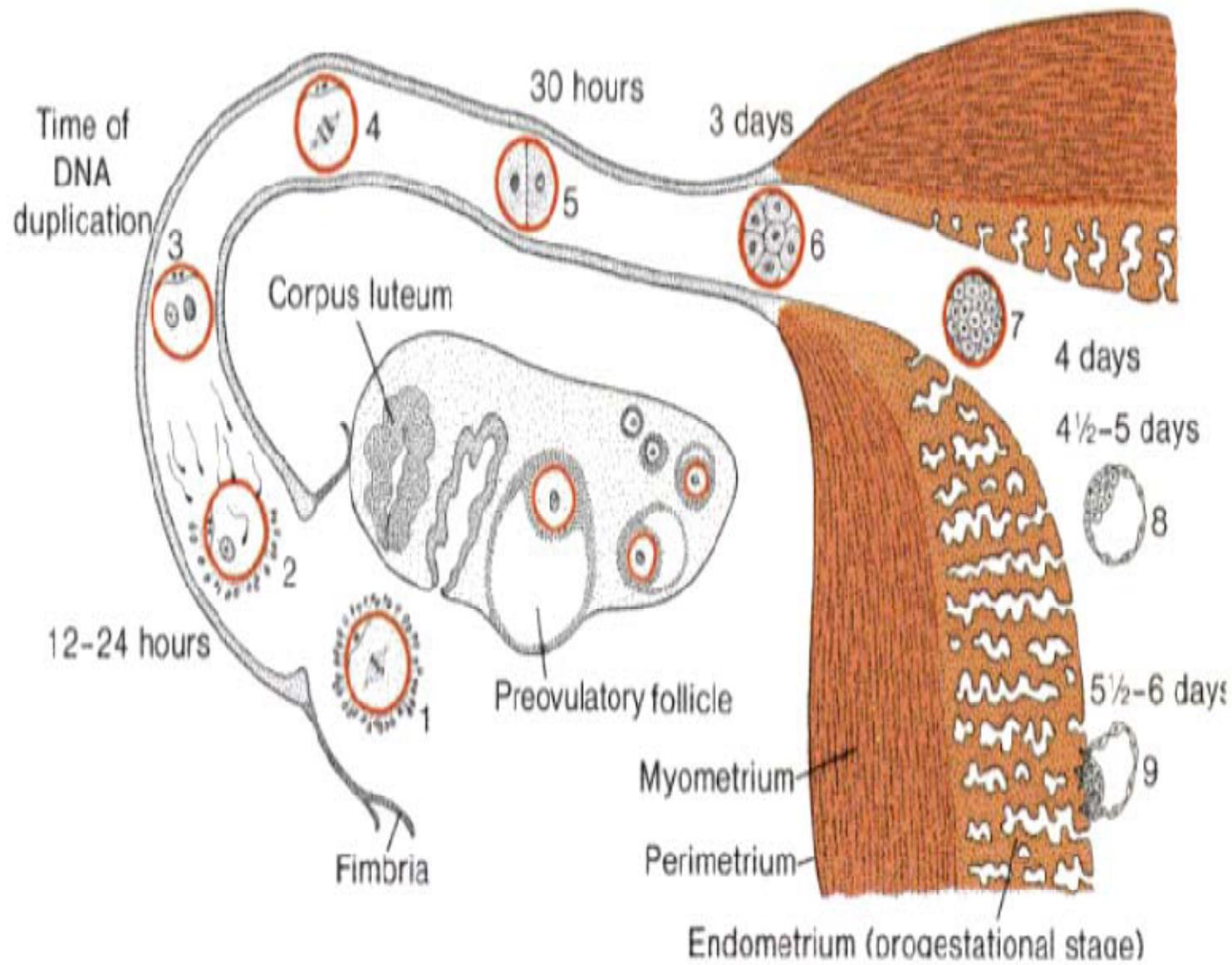
Dr. Da'ad A. Hussain

**Institute of Genetic Engineering and Biotechnology for Post
Graduate Studies , University of Baghdad**

Introduction

Infertility is the central issue in the lives of the individual who suffers from it. It is a source of social and psychological suffering for both men and women and can place great pressure on the relationship within the couple. Based on the note from UN declaration of human rights, it is said that men and women of full age, without any limitation due to race, nationality or religion have the right to marry and to find a family¹. But unfortunately 1 in 6 couples of any society remains infertile, and 10% of them need the help of assisted reproductive technology.

Assisted reproductive technology (ART) defined as a diverse group of medical procedures that allow infertile couples to be pregnant. Or refers to all technology where gametes are manipulated outside the body. On the other hand, infertility defined as inability to become pregnant after 1 year of unprotected coitus.



Incidence of Infertility

- 15-20% of all couples of reproductive age are infertile
- 30-40% of infertility due to male factors
- 40-50% of infertility due to female factors
- 10-15% of infertility unknown

The Major Etiologies

1- Ovarian causes (30%)

Abnormal ovulation, hormone production

Premature ovarian failure

2- Tubal causes (30%)

Occlusion

Inflammation

Adhesion

3- Uterine causes (10%)

Uterine abnormalities

Leiomyoma

4- Cervical causes (15%)

Stenosis of cervix

Hostile cervical mucus

5- Male factors (40%)

Oligozoospermia

Asthenozoospermia

Teratozoospermia

Endocrine disorders

Vas obstruction

6- Other causes (5%)

Pituitary disorders

Psychological disorders

Metabolic disorders

Unknown causes

Major ART Procedures

- In-vitro fertilization and embryo transfer (IVF & ET)
- Gamete intrfallopian transfer (GIFT)
- Pronucleate or Zygote intra-fallopian transfer (PROT, ZIFT)
- Intracytoplasmic sperm injection (ICSI)
- Round nuclei injection (ROSNI) or spermatid injection
- Assisted hatching

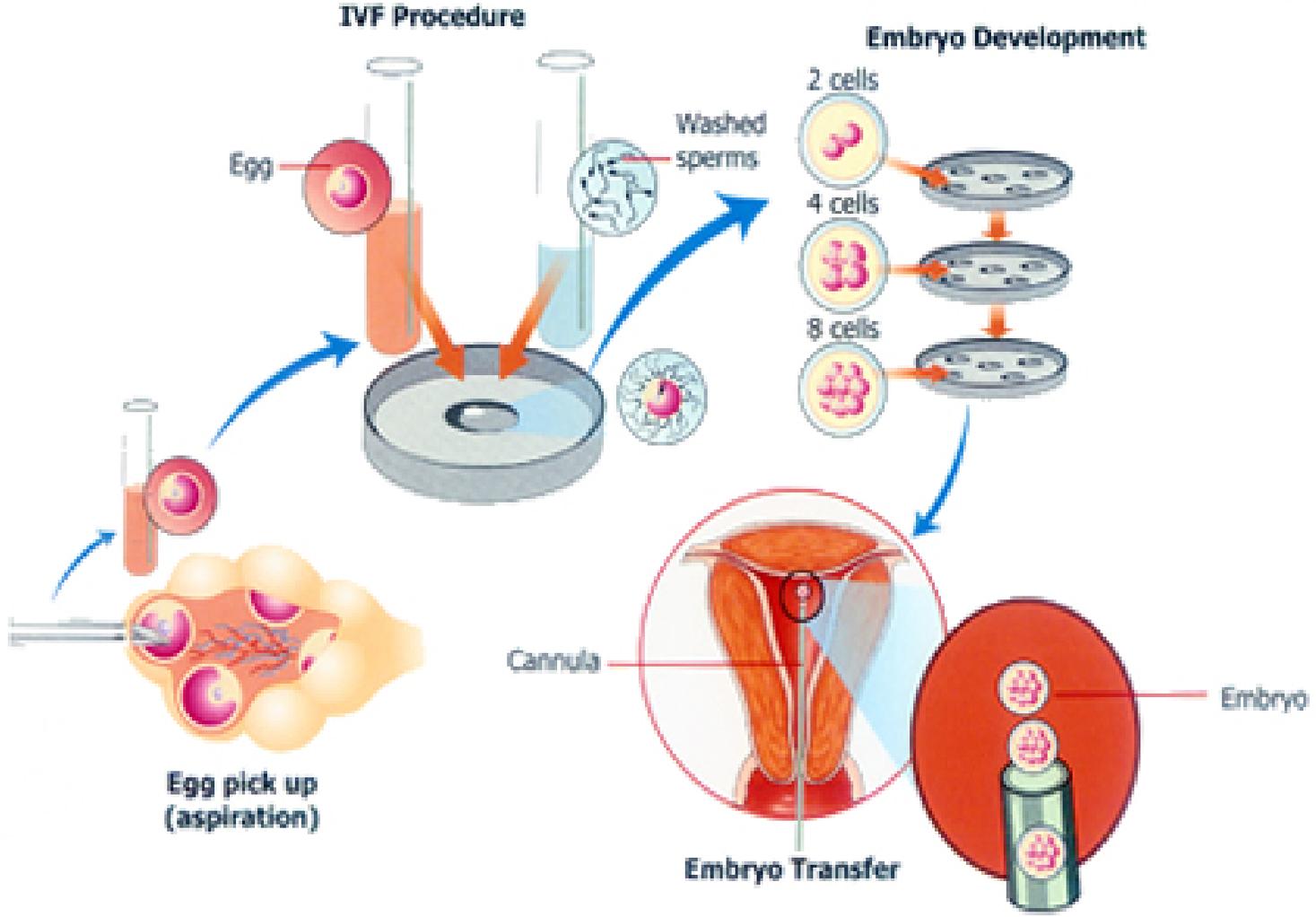
Among this list most commonly practiced procedures are IVF &ET and ICSI.

1- In-vitro fertilization (IVF):

IVF stands for *in vitro fertilization*. *In vitro* means “in the lab” and *fertilization* refers to conception. Usually, IVF involves taking many eggs (retrieved via a transvaginal ultrasound-guided needle) and placing them in a petri dish with specially washed sperm cells (retrieved via masturbation.) If all goes well, some of the retrieved eggs will become fertilized by the sperm cells and become embryos. One or two of those healthy embryos will be transferred to the uterus.



In Vitro Fertilization (IVF)



Indications of IVF

- Absent fallopian tubes or bilateral tubal block or disease that can not be treated successfully by surgery.
- Endometriosis that has not responded to surgical or medical treatment.
- A male factor contributing to infertility, in which sperm counts or motility are low but there are enough active sperm to allow fertilization in the laboratory.
- Unexplained infertility that has not responded to other treatments.
- Infertility secondary to sperm antibodies.
- Genetic disease that result in miscarriage or abnormal births.

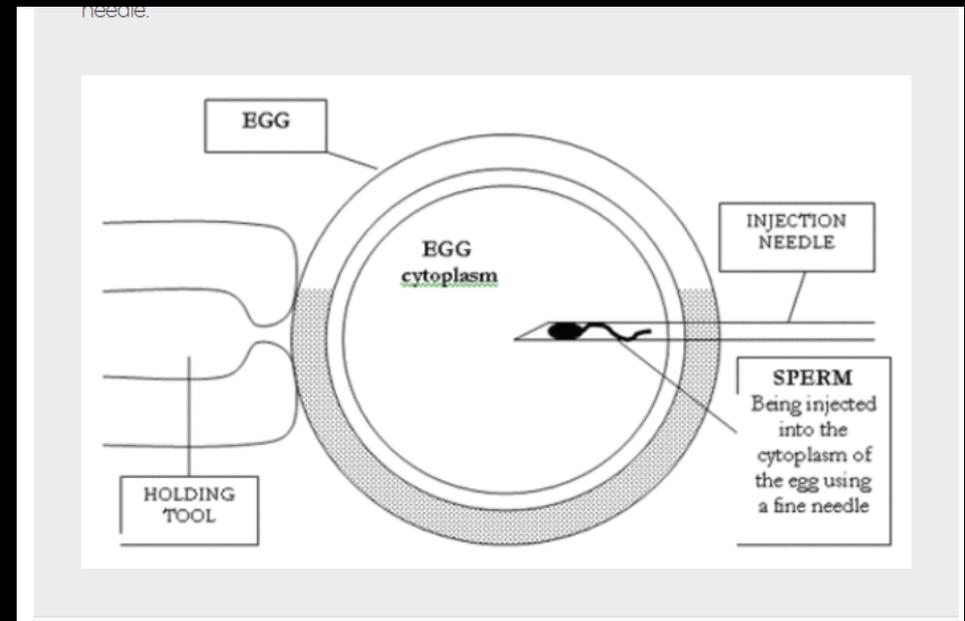
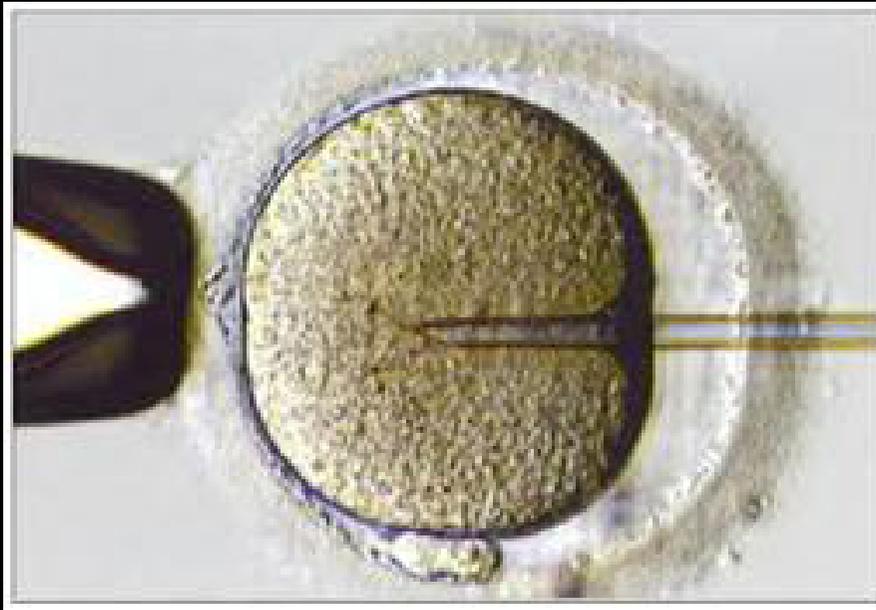
IVF Success Rates

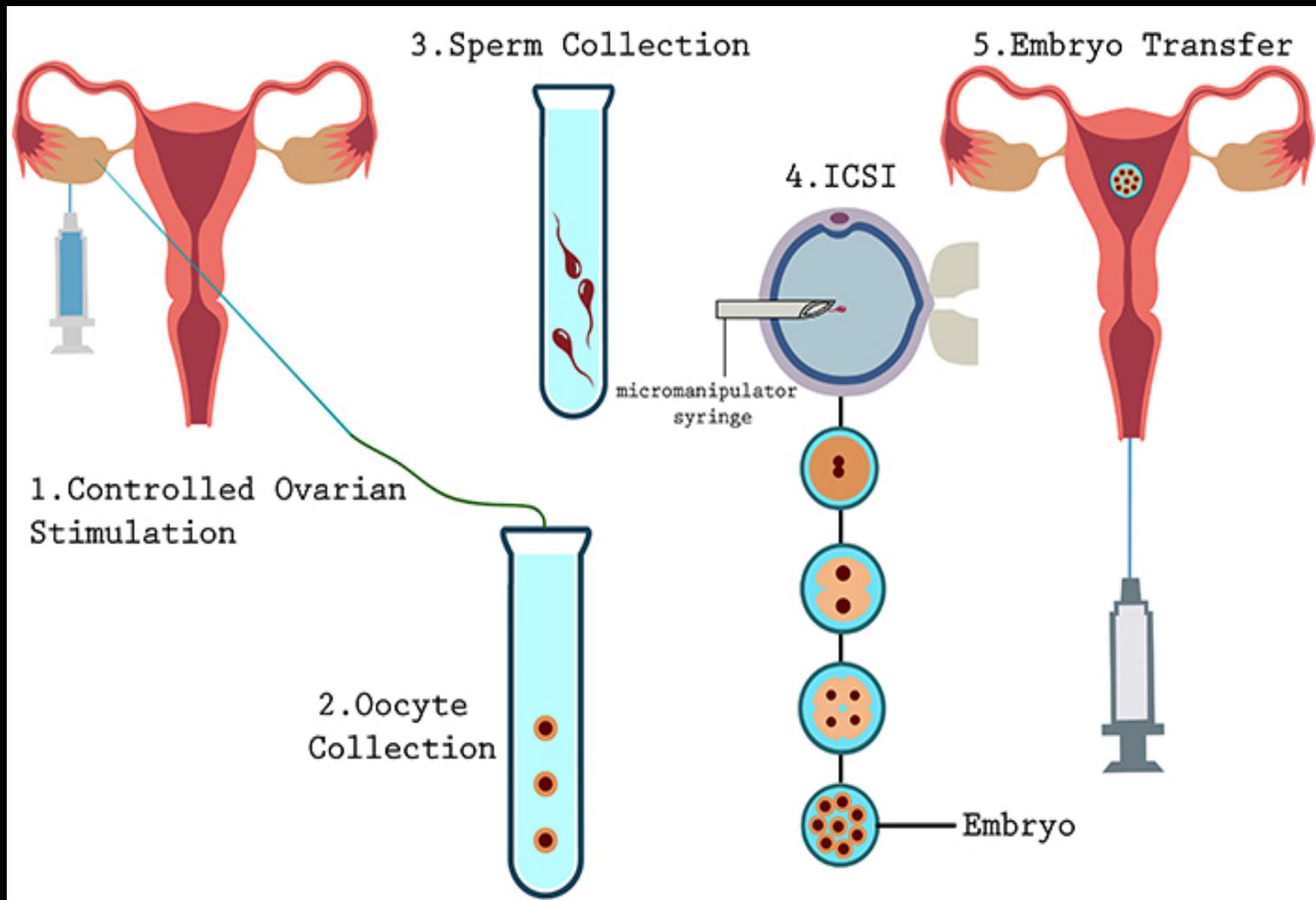
IVF is pretty successful. According to a study of approximately 156,000 women, the average live-birth rate for the first cycle was 29.5 percent. This is comparable to [the success rates for a natural cycle in couples with healthy fertility](#).

The best odds for success may come from repeated treatment cycles. This same study found that after six IVF cycles, the cumulative live-birth rate was 65.3 percent. These six cycles usually took place over two years.

Age does play an important role in your success, as does the reason for your infertility. [Using an egg donor](#) will also affect your success.

2- Intracytoplasmic Sperm Injection (ICSI):



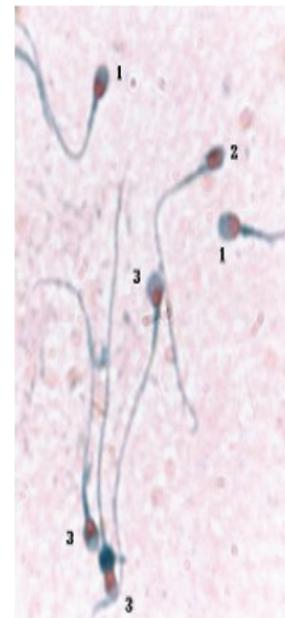
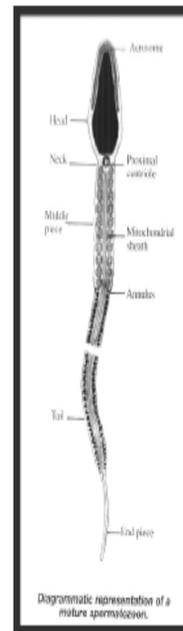


ICSI: INTRACYTOPLASMIC SPERM INJECTION



Normal Sperm Morphology

- Head – 4.6 μ length, 2.6 μ wide, 1.5 μ thick.
- Midpiece - ~ 5 μ
- Tail - ~ 50 μ long
- Acrosome - ~ 40%
- Nucleus - ~ 60% DNA, protamines



Indications of intracytoplasmic sperm injection (ICSI):

- Very low sperm count (also known as oligospermia)
- Abnormally shaped sperm (also known as teratozoospermia)
- Poor sperm movement (also known as asthenozoospermia)
- Per epididymal sperm aspiration (PESA): It is done using a small needle under local anaesthesia to aspirate sperm from proximal to the obstruction.
- Testicular sperm aspiration (TESA): Testicular sperm is aspirated from non obstructive azoospermic men by a syringe or butterfly needle.
- Testicular sperm extraction (TESE): If spermatozoa is unavailable after PESA or TESA testicular tissue is taken under local or general anaesthesia.

Previous IVF cycle had few or no fertilized eggs: Sometimes, a good number of eggs are retrieved, and sperm counts look healthy, but no eggs get fertilized. In this case, during the next IVF cycle, ICSI may be tried.

Frozen sperm are being used: If the thawed sperm don't appear especially active, ICSI-IVF may be recommended.

Frozen oocytes are being used: Vitrification of eggs can sometimes cause hardening of the egg's shell. This may complicate fertilization, and IVF with ICSI may help overcome this hurdle.

PGD is being done: PGD (preimplantation genetic diagnosis) is an IVF technology that allows for genetic screening of embryos. There is concern that regular fertilization techniques may cause sperm cells (who have not fertilized the egg) to "hang around" the embryo, and that this may interfere with accurate PGD results.

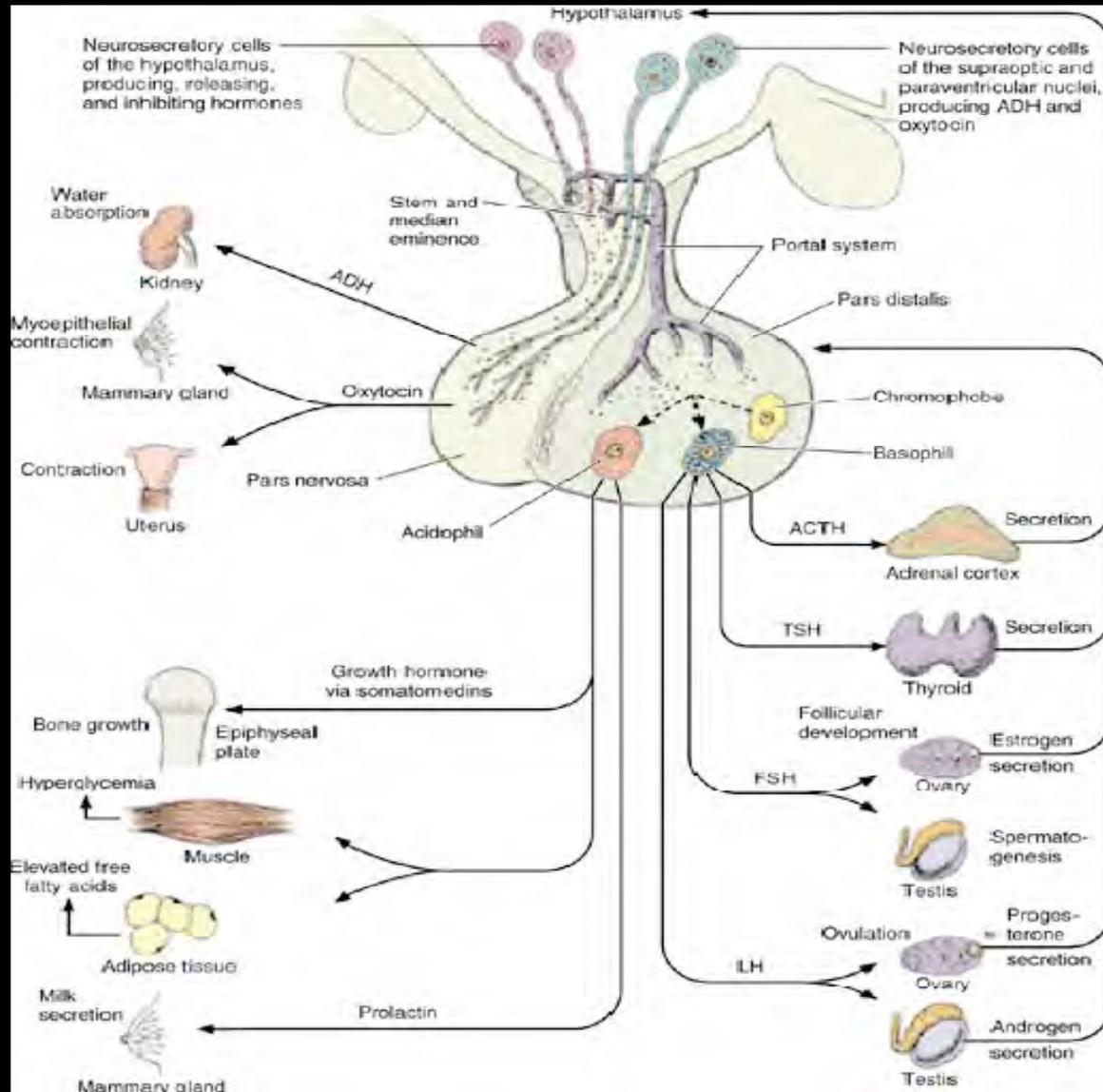
IVM (in vitro maturation) is being used: IVM is an IVF technology where eggs are retrieved from the ovaries before they completely mature. They go through the final stages of maturation in the lab. Some research has found that IVM eggs may not become fertilized by sperm cells at rates comparable to traditional IVF. More research is needed, but it may be that IVM with ICSI is a good option.

ICSI Success Rates:

The ICSI procedure fertilizes 50 to 80 percent of eggs. You might assume all eggs get fertilized with ICSI-IVF, but they don't. Fertilization isn't guaranteed even when a sperm is injected into the egg.

Remember that fertilization rates don't tell you the clinical pregnancy or live birth rates. Once fertilization happens, the [success rates for a couple using](#) ICSI with IVF is the same as a couple using regular IVF treatment.

Steps of ART



Steps of ART

Preparation of female partner

Step 1 (Down regulation protocols)

Agonist protocol / Antagonist protocol:

For GnRH agonist there are mainly two different protocols

1. Long protocol
2. Short protocol

In the long protocol the basic principal is to conduct the complete period of folliculogenesis with the lowest possible LH. The GnRH agonist is given either at D2 or D21st of cycle. After 2-3 weeks of administration when hypophyseal desensitization is complete, follicular growth and maturation are induced by exogenous gonadotrophins while GnRH agonist is continued to prevent any premature LH rise. The administration of GnRH discontinued at the same time as gonadotrophin administration is stopped.

Step II

- ❑ **Ovarian stimulation:** Either HMG or rFSH can be used. According to need of the patient 150-450 IU daily for 10-12 days is required to get mature eggs. When 3 or more follicles attain a size of 18 mm or more it indicates follicular maturity. At that point both GnRH agonist and gonadotropins stopped and injection hCG 5000 – 10000 IU is injected to trigger ovulation.
- ❑ **Monitoring of ovarian response:** Steady synchronous increase of at least three follicles with diameter increasing roughly 2mm per day. Steady increase in serum E₂ level leading to approximately 200pg/ml per follicle larger than 14mm in diameter and thickness of the endometrial bed 8mm or more on the day of hCG administration generally denote appropriate response to stimulation. Cancellation and avoidance of hCG injection are to be considered if the ovaries are markedly hyperstimulated (more than 25 follicles and /or E₂ more than 4000pg/ml on day of hCG).

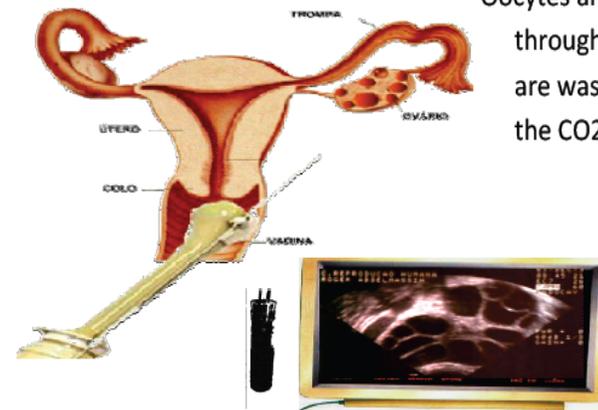
Step III

Egg retrieval

After 36 hours of hCG injection the ripe eggs are collected. This is done usually under deep sedation or general anaesthesia, which takes between 10-30 minutes depending upon the number of follicles that have grown in response to the drugs.



Collection of Oocytes



Oocytes are collected by U/S guided collection through the vagina. The collected oocytes are washed in culture media and placed in the CO₂ incubator until insemination.

Step IV

Insemination/ICSI and fertilization:

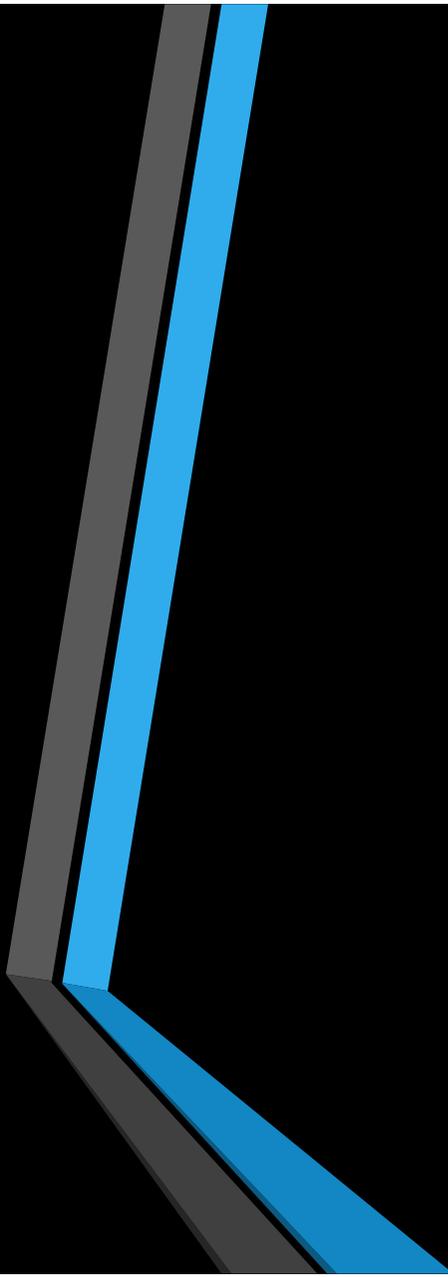
Step V

Embryo transfer:

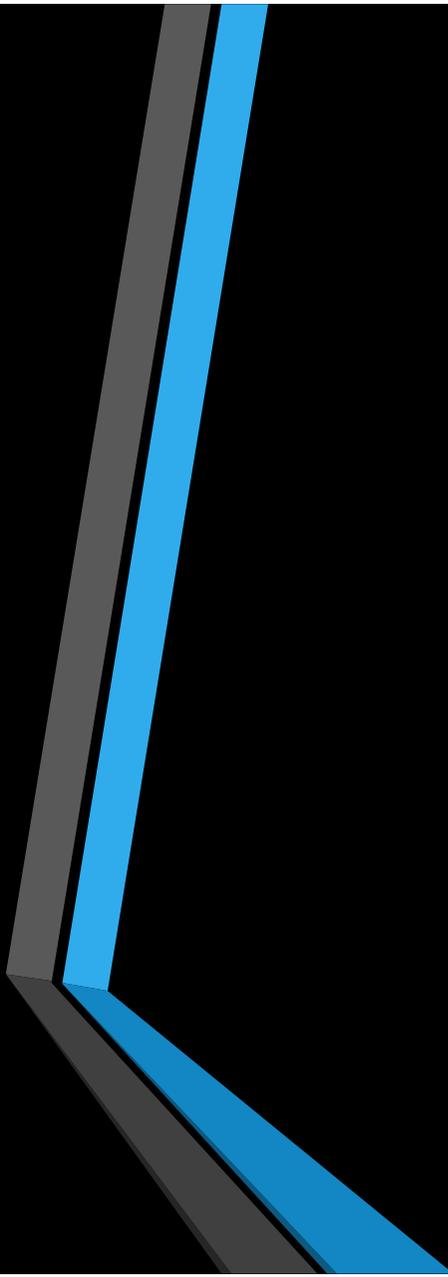
Complications:

Ovarian hyperstimulation syndrome (OHSS) is a rare complication of controlled ovarian stimulation. The incidence of OHSS is usually between 0.1 and 6.1% of all controlled ovarian hyperstimulation cycles and the severe form is seen in about 0.4% of cases. The syndrome is more common in young patient and in PCOS patients.

The basic pathology is hyper permeability of capillaries leading to loss of fluid and protein from intravascular compartment. The net effect is hypovolemia and hypoproteinaemia. The syndrome may partly be prevented by withholding hCG for triggering for ovulation and for luteal support when E2 level is more than 4000 pg/ml and too many follicles have appeared.



Religious and social bar:



Thank You