Objectives:
- Name the mechanisms and barriers involved in sperm transport along the female genital tract
- State the events occurring in the spermatozoa immediately prior to fertilization
- State the events occurring in the oocyte immediately after sperm penetration
- Outline the procedures involved in in-vitro fertilization
- Name other types of assisted conception

Fertilization
Includes those mechanisms where by:
- A sperm approaches to
- Becomes attached to and then penetrates the surface of an ovum
- The early series of changes which follow

Sperm Transport
From their storage site in the epididymis (mainly in its tail) the sperms are rapidly transported to the urethra by peristaltic contractions of the thick muscular coat of the ductus deferens.

The accessory sex glands—seminal glands (vesicles), prostate, and bulbourethral glands—produce secretions that are added to the sperm-containing fluid in the ductus deferens and urethra.

The ejaculate
Its volume averages 3.5 mL (a range of 2 to 6 mL) with approximately 100 million sperm per milliliter.

The sperms move 2 to 3 mm per minute, but the speed varies with the pH of the environment.
They are nonmotile during storage in the epididymis, but become motile in the ejaculate.
They move slowly in the acid environment of the vagina, but move more rapidly in the alkaline environment of the uterus.

vagina
200 to 600 million sperms are deposited around the external os of the uterus and in the fornix of the vagina during sexual intercourse.

Cervix
Only 1% of sperm deposited in the vagina enter the cervix, where they may survive for many hours.
- consistency and viscosity of cervical mucus, under hormonal control, play an important role in the process of fertilization.
- Prior to ovulation—watery cervical mucus
- Luteal phase—viscous and disorganized cervical mucus

Passage of sperms through the uterus and uterine tubes by
muscular contractions of the uterus and uterine tube. Prostaglandins in the semen are thought to stimulate uterine motility at the time of intercourse and assist in the movement of sperms to the site of fertilization in the ampulla of the tube.

their own propulsion. Fructose, secreted by the seminal glands, is an energy source for the sperms in the semen.

The trip from cervix to oviduct requires a minimum of 2 to 7 hours.

Oviduct

Uterotubal junction a significant barrier
after reaching the isthmus, sperm become less motile and cease their migration
Spermatozoa are not able to fertilize the oocyte immediately upon arrival in the female genital tract
They must undergo
(1) capacitation and
(2) acrosome reaction to acquire this capability

Oviduct

Capacitation
It is a period of conditioning in the female reproductive tract, associated with
1. removal of glycoprotein coat and seminal plasma proteins from the plasma membrane that overlies the acrosomal region of the spermatozoa.
2. reorganization of plasma membrane lipids and proteins to prepare the sperm for acrosome reaction.

Much of this conditioning occurs in the uterine tube
In the human, it lasts approximately 7 hours.
Sperm can be capacitated by incubation in certain fertilization media.

Oviduct

Oocyte transport
During ovulation, the fimbriated end of the uterine tube becomes closely applied to the ovary.
The fingerlike processes of the tube, fimbriae, move back and forth over the ovary

It is thought that the oocyte surrounded by some granulosa cells is carried into the tube by

- The sweeping action of the fimbriae and
- fluid currents produced by the cilia of the mucosal cells of the fimbriae

Once in the tube,
cumulus cells withdraw their cytoplasmic processes from the zona pellucida and lose contact with the oocyte.

The oocyte passes into the ampulla of the tube mainly as the result of peristalsis (alternate contraction and relaxation) movements of the wall of the tube.

At ovulation, sperm
- again become motile, perhaps because of chemoattractants produced by cumulus cells surrounding the egg, and
- swim to the ampulla, where fertilization usually occurs.

The oviduct provides the appropriate environment
not only for fertilization but for early embryonic development (the embryo remain there for a period of 3 days).

The fertilizable lifespan of gametes
The **oocyte** can be fertilized for up to 24 h after ovulation. Some sperm cells remain viable in the female reproductive tract for up to 6 days, although most of them have degenerated after 24 h. For fertilization to occur successfully, sexual intercourse must, therefore, occur between 5 days before and one day after ovulation.

**Attaching & penetrating the surface of an ovum by capacitated sperm**

A spermatozoon has to penetrate four layers before it fertilizes the oocyte:

1. penetration of the **corona radiata**
2. penetration of the **zona pellucida**
3. penetration of vitelline membrane

1. **Passage of a sperm through the corona radiata.**
   - The corona radiata is a barrier to the sperm cells reaching the oocyte.
   - The sperm cells are propelled through the loose matrix between the follicular cells of corona radiata by the action of their flagella.
   - Of the 200 to 300 million spermatozoa deposited in the female genital tract, only 300 to 500 reach the site of fertilization. Only one of these fertilizes the egg.
   - Only capacitated sperm pass freely through corona cells.

   **Dispersal of the follicular cells of the corona radiata appears to result mainly from**
   - the enzyme **hyaluronidase** released from the acrosome of the sperm
   - **Tubal mucosal enzymes**
   - Movements of the tail of the sperm

2. **penetration of the zona pellucida**
   - The zona pellucida is an extracellular membrane, comprised mostly of glycoproteins, between corona radiata and the oocyte.
   - One particular zona pellucida glycoprotein called **zp3** which is species-specific sperm cell receptor to which molecules on the acrosomal cap of the sperm cell bind.
   - This binding initiates the acrosomal reaction.

   **The acrosome reaction**
   - Is stimulated by sperm-binding glycoprotein molecules in the zona pellucida (ZP3, ZP1 and ZP2 ligands).
   - Is accompanied by Ca\(^{2+}\) influx into sperm.
   - Culminates in the release of hydrolytic enzymes needed to penetrate the zona pellucida, including acrosin- and trypsin-like substances.

3. **Fusion of the Oocyte and Sperm Cell Membranes**
   - **The initial adhesion** of sperm to the oocyte is mediated in part by the interaction of **integrins** on the oocyte and their ligands, **disintegrins**, on sperm.
   - Because the plasma membrane covering the acrosomal head cap disappears during the acrosome reaction, actual fusion is accomplished between the oocyte membrane and the membrane that covers the posterior region of the sperm head.
   - In the human, both the head and tail of the spermatozoon enter the cytoplasm of the oocyte, but the plasma membrane is left behind on the oocyte surface.

**Three changes occur in the oocyte after penetration of vitelline membrane:**

- **Polyspermy**
- penetration of more than one spermatozoon into the oocyte

**Prevention of polyspermy**
- Fast block to polyspermy
- Slow block to polyspermy
Fast block to polyspermy
Once the first sperm cell attaches to the integrin (alpha 6 beta 1) on the surface of the oocyte plasma membrane, depolarization of the oocyte plasma membrane occurs within 2-3 seconds. This depolarization (fast block to poly sperm) prevents additional sperm from attaching to the oocyte plasma membrane.

Slow block to poly sperm
Depolarization causes the intracellular release of Ca^{+2}, which in turn causes the exocytosis of water and other molecules from secretory vesicles referred to as cortical granules on the inner surface of the oocyte plasma membrane. The released fluid causes the oocyte to shrink and the zona pellucida to denature and expand away from the oocyte. As a result of denaturation of the zona pellucida, zp3 is inactivated and no additional sperm cells can attach. This reaction is referred to as the slow block to poly sperm.

The early series of changes which follow “egg activation”
Prior to fertilization, the egg is in a quiescent state. Upon binding of a sperm, the egg rapidly undergoes a number of metabolic and physical changes collectively called “egg activation”
  o Cortical and zona reactions
  o Resumption of the second meiotic division
  o Metabolic activation of the egg – The activating factor is probably carried by the spermatozoon.

After penetration of the oocyte by the sperm, the oocyte completes meiosis, the female pronucleus and the larger male pronucleus approach each other as DNA is doubled in maternal and paternal chromosomes to initiate the first mitotic division.

Pronuclear membranes then break down and maternal and paternal chromosomes assemble on the metaphase plate.

Centromeres then replicate, and homologous chromosomes are distributed to the first two cells of the embryo.

  o The cytoplasmic organelles of the zygote are almost entirely maternal
  o Mitochondrial DNA is almost entirely maternal
  o Mitochondrial genetic diseases are generally inherited through the mother but may affect both sons and daughters
  o Most mitochondrial diseases affect muscle and nerve
  o Examples of mitochondrial inheritance are:
    – mitochondrial myopathy (affects muscle)
    – Leber’s optic atrophy (affects optic nerve)

The main results of fertilization
  o Restoration of the diploid number of chromosomes, the zygote contains a new combination of chromosomes different from both parents.
  o Determination of the sex of the new individual. An X-carrying sperm produces a female (XX) embryo, and a Y-carrying sperm produces a male (XY) embryo. Hence, the chromosomal sex of the embryo is determined at fertilization.
  o Initiation of cleavage.
Clinical correlates

Contraceptive Methods

Barrier techniques of contraception include
- the male condom,
- the female condom,
- the diaphragm,
- the cervical cap,
- and the contraceptive sponge.

Prevention of ovulation
- The contraceptive pill is a combination of estrogen and the progesterone which together inhibit ovulation but permit menstruation.
- Depo-Provera is a progestin compound that can be implanted subdermally or injected intramuscularly to prevent ovulation for up to 5 years or 23 months.
- A male "pill"
  - has been developed and tested in clinical trials.
  - It contains a synthetic androgen that prevents both LH and FSH secretion
  - It either stops sperm production (70% to 90% of men) or
  - reduces it to a level of infertility.

The intrauterine device (IUD)
- is placed in the uterine cavity.
- Its mechanism for preventing pregnancy is not clear but
  - may entail direct effects on sperm and oocytes or
  - inhibition of preimplantation stages of development.

Vasectomy and tubal ligation
- are effective means of contraception, and
- both procedures are reversible, although not in every case.
- Infertility
- Infertility is a problem for 15% to 30% of couples.

Male Infertility
- be a result of insufficient numbers of sperm and/or poor motility.

Female Infertility
- occluded uterine tubes (most commonly caused by pelvic inflammatory disease),
- hostile cervical mucus,
- immunity to spermatozoa,
- absence of ovulation, and others.

Assisted reproductive technology (ART)
is a group of fertility treatments that involve both the sperm and the egg.
- In vitro fertilization (IVF)
- intracytoplasmic sperm injection (ICSI)
- gamete intrafallopian transfer (GIFT)
- zygote intrafallopian transfer (ZIFT).

In vitro fertilization (IVF)
is the most common type of ART.
the sperm fertilizes the egg outside the body, and doctors implant it into the woman’s uterus in hopes of a successful pregnancy.
IVF cycle takes four to six weeks to complete and usually costs about $12,000. In vitro fertilization and embryo transfer procedures.

In vitro Fertilization (IVF)

The risk of producing malformed offspring by in vitro procedures is low because preimplantation-stage embryos are resistant to teratogenic insult, A disadvantage of IVF is its low success rate. Only 20% of fertilized ova implant and develop to term. Therefore, to increase chances of a successful pregnancy, four or five ova are collected, fertilized, and placed in the uterus. This approach sometimes leads to multiple births.

The following methods of ART require patent uterine tubes.

- Gamete intrafallopian transfer (GIFT) oocytes and sperm into the ampulla of the fallopian (uterine) tube, where fertilization takes place.

- Zygote intrafallopian transfer (ZIFT) fertilized oocytes are placed in the ampullary region.

**Intracytoplasmic sperm injection (ICSI)**

(Oligozoospermia) or even (azoospermia), can be overcome by using intracytoplasmic sperm injection (ICSI).

**Intracytoplasmic sperm injection (ICSI)**

With this technique, a single sperm, which may be obtained from any point in the male reproductive tract, is injected into the cytoplasm of the egg to cause fertilization. The technique carries an increased risk for fetuses to have Y chromosome deletions but no other chromosomal abnormalities.