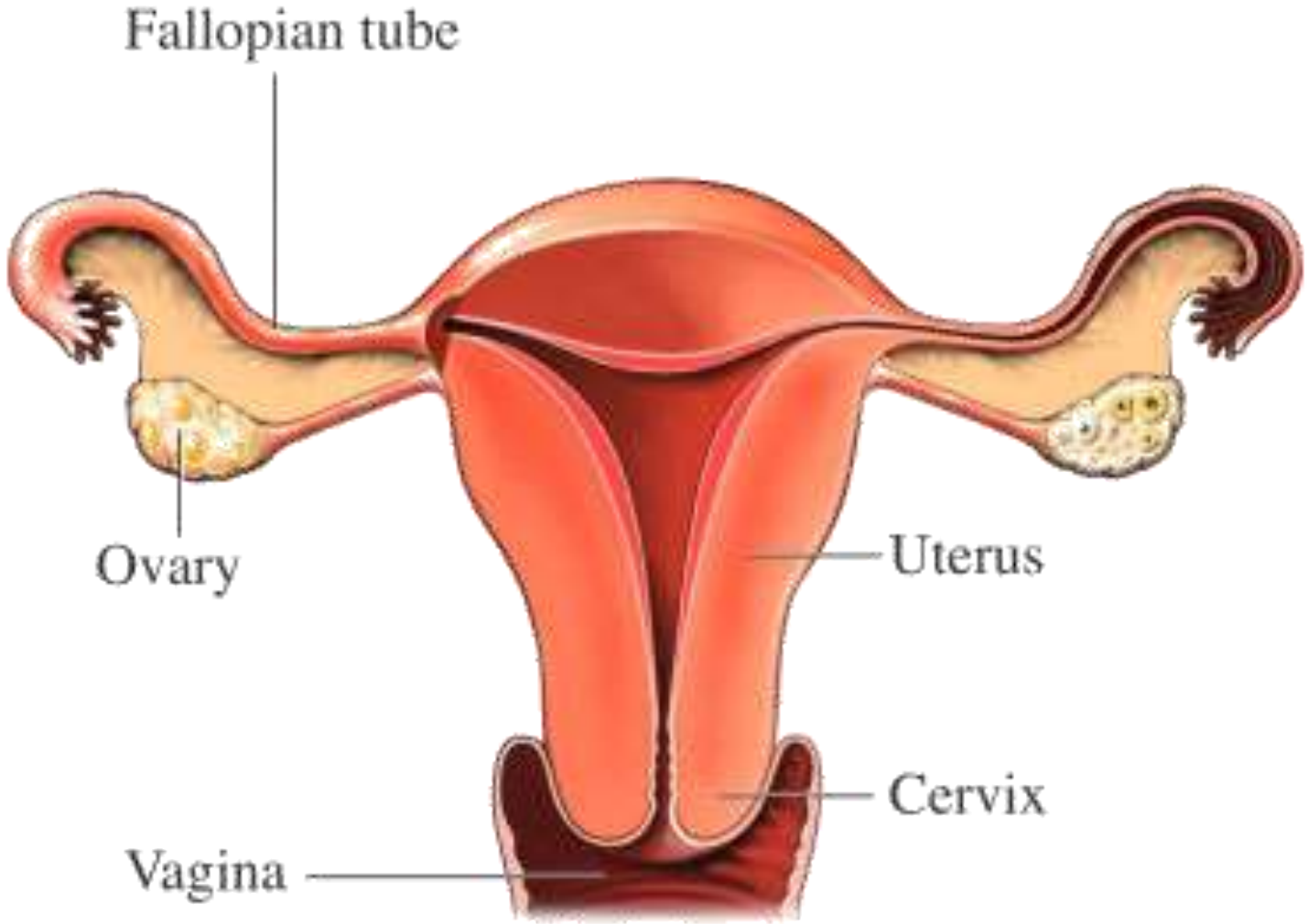
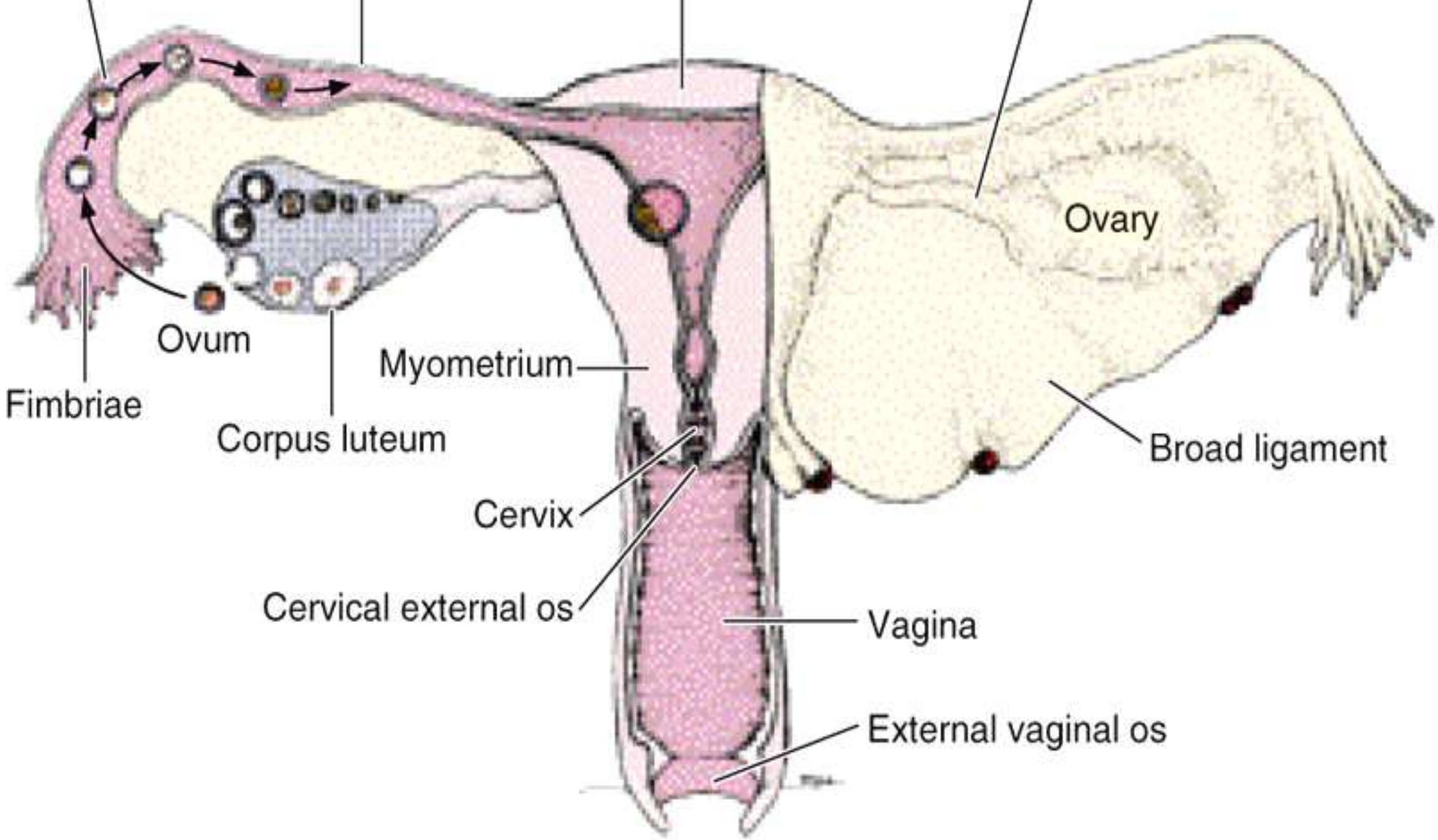
OVULATION, FERTILIZATION AND IMPLANTATION

DR KIBE


Fallopian tube
Ovary
Uterus
Cervix
Vagina


Ovary
Ovum
Myometrium
Fimbriae
Corpus luteum
Broad ligament
Cervix
Cervical external os
Vagina
External vaginal os

Ovulation

- At the onset of puberty, a female starts undergoing regular monthly cycles.
- These cycles are called sexual cycles / ovarian cycles and are controlled by gonadotrophic hormones which are secreted by the pituitary gland.
- These hormones are; follicle stimulating hormone (FSH) and Luteinizing hormone (LH).
- The production of gonadotrophic hormones is under the influence of gonadotrophic-releasing hormone which is produced by hypothalamus.

- At the beginning of each ovarian cycle, 15 to 20 primary

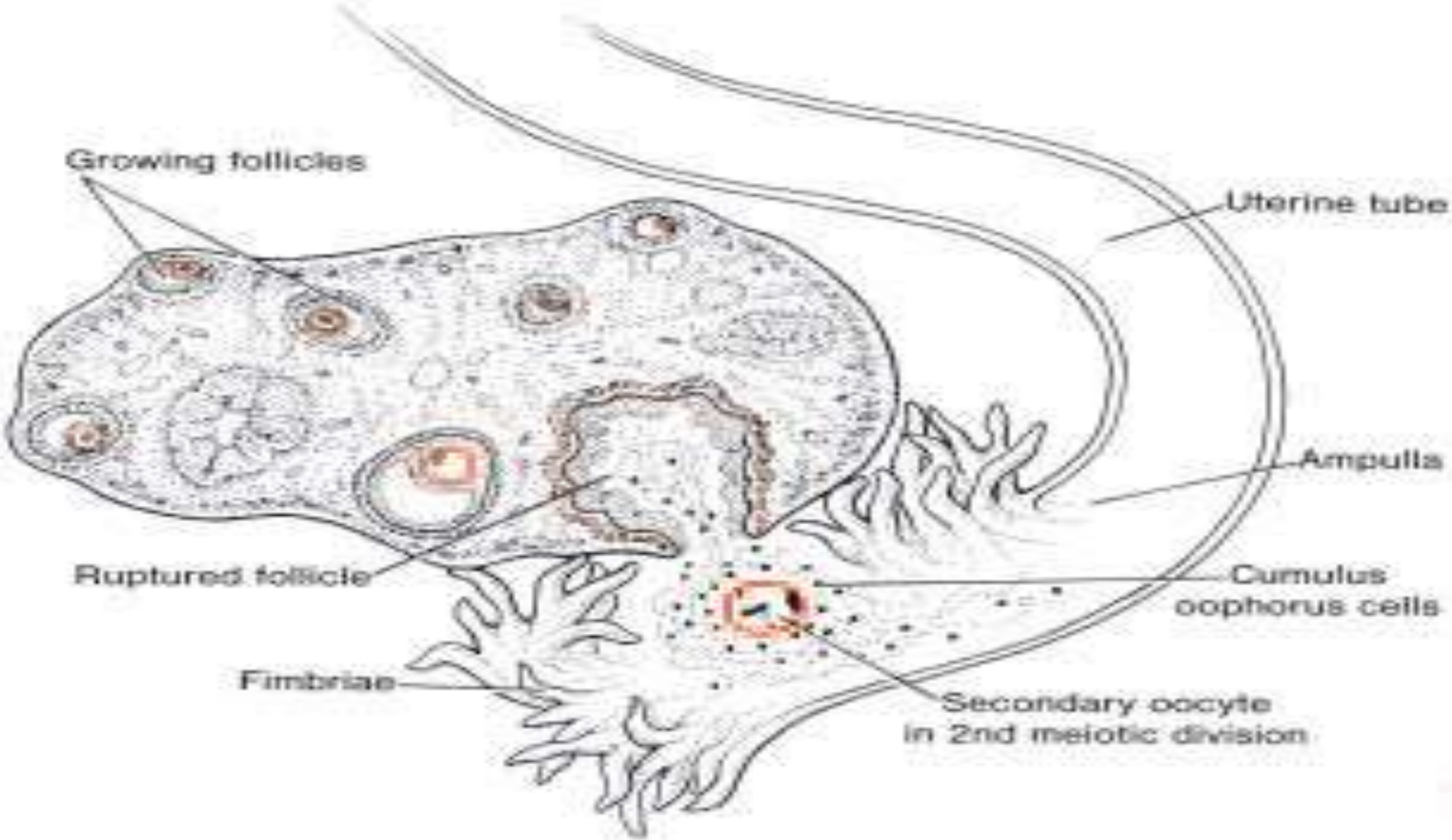
(preantral) stage follicles are stimulated to grow under the

influence of FSH.

- The hormone is not necessary to promote development of primordial follicles to the primary follicle stage,

but without it, these primary follicles die and become atretic.)

- Only one follicle reaches fully maturity while the others degenerate. Also only one oocyte is discharged.


Growing follicles
Uterine tube
Ampulla
Ruptured follicle
Cumulus oophorus cells
Fimbriae
Secondary oocyte in 2nd meiotic division

- As the follicles grow, large numbers of follicullar and theca cells are produced.
- These cells produce estrogen which stimulates the pituitary gland to produce luteinizing hormone
- LH is necessary for the final stages of follicle maturation, and for ovulation.
- LH facilitates the oocyte to complete 1st meiotic division and to start 2nd meiotic division which get arrested in the metaphase.
- LH also stimulates production of progesterone by follicular cells and proliferation of endometrium.

- Consequently, most follicles degenerate without ever reaching full maturity.
- When a follicle becomes atretic, the oocyte and surrounding follicular cells degenerate and are replaced by connective tissue, forming a **corpus atreticum.**
- FSH also stimulates maturation of **follicular (granulosa)** cells surrounding the oocyte.
- In turn, proliferation of these cells is mediated by growth differentiation factor-9 (GDF-9),

- granulosa and thecal cells produce estrogens that;

(*a*) cause the uterine endometrium to enter the follicular or **proliferative phase;**

(*b*) Cause thinning of the cervical mucus to allow passage of sperm; and

(*c*) stimulate the pituitary gland to secrete LH

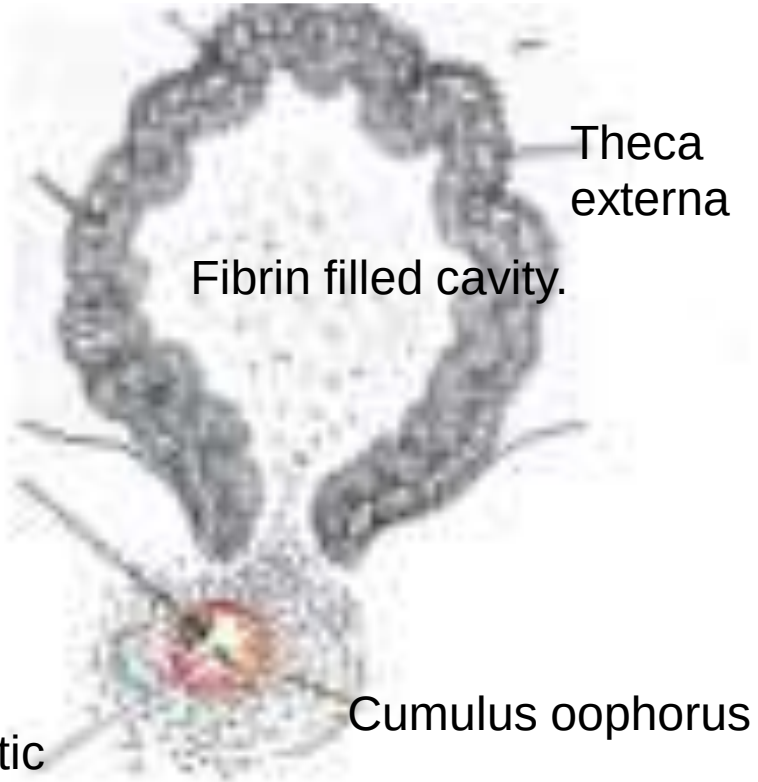
- At mid-cycle, there is an **LH surge** that
- (*a*) elevates concentrations of maturation-promoting factor, causing oocytes to complete meiosis I and initiate meiosis II;
- (*b*) stimulates production of progesterone by follicular stromal cells **(luteinization);** and
- (*c*) causes follicular rupture and ovulation.

ovulation

- In the days immediately preceding ovulation, under the influence of FSH and LH, the secondary follicle grows rapidly to a diameter of 25 mm
- Ovulation is the release from the ovary of a mature oocyte which has finished its 1st meiotic division and has started its 2nd meiotic division and arrested at metaphase approximately 3 hours before ovulation.
- In the meantime, the surface of the ovary begins to bulge locally, and at the apex, an avascular spot, the **stigma,** appears.
- The high concentration of LH increases collagenase activity, resulting in digestion of collagen fibers surrounding the follicle.
-

- Prostaglandin levels also increase in response to the LH surge and cause local muscular contractions in the ovarian wall.
- Those contractions extrude the oocyte, which together with its surrounding granulosa cells from the region of the cumulusoophorus, breaks free (**ovulation**) and floats out of the ovary.
- Some of the cumulus oophorus cells then rearrange themselves around the zona pellucida to form the **corona radiata**

Granulosa cells



Theca externa

Fibrin filled cavity.

Oocyte in 2nd meiotic division

Cumulus oophorus

C L I N I C A L C O R R E L A T E S

- During ovulation, some women feel a slight pain, known as **middle pain** because it normally occurs near the middle of the menstrual cycle.
- Ovulation accompanied by a rise in basal temperature,.
- Some women fail to ovulate because of a low concentration of gonadotropins.
- administration of an agent to stimulate gonadotropin release and hence ovulation can be employed.
- such drugs are often produce multiple ovulations, the risk of multiple pregnancies is 10 times higher in these women than in the general population.

Corpus luteum

- After the oocyte is discharged, the follicular cells + theca interna proliferate & become vascularised.
- LH transforms these cells into yellow mass= luteal cells, which mature to **corpus luteum**.
- Corpus luteum produces progesterone which prepares the uterus in readiness for implantation

- Progesterone, together with estrogenic hormones, causes the uterine mucosa to enter the **progestational** or **secretory stage** in preparation for implantation of the embryo

Fate of corpus luteum

- the corpus luteum reaches maximum development approximately 9 days after ovulation
- If fertilization fails, the corpus luteum degenerates and forms a mass of fibrotic scars called **corpus albicans.**
- Then there is reduction in progesterone production, thus precipitating the menstrual bleeding
- If fertilization occurs, the corpus luteum continues to grow and forms the **corpus luteum of pregnancy (corpus luteum graviditatis).** The corpus luteum of pregnancy continues to produce progesterone until the end of fourth month when cells of the placenta takes over the secretion of progesterone.

- If the oocyte is fertilized, degeneration of the corpus luteum is prevented by **human chorionic gonadotropin** (hCG), a hormone secreted by the syncytiotrophoblast of the developing embryo.
- By the end of the third month, this structure may be one-third to one-half of the total size of the ovary
- After 4^{th} month luteum regress slowly as secretion of progesterone by the trophoblastic component of the placenta becomes adequate for maintenance of pregnancy.
- Removal of the corpus luteum of pregnancy before the fourth month usually leads to abortion

OOCYTE TRANSPORT

- Shortly before ovulation, fimbriae of the oviduct begin to sweep over the surface of the ovary, and the tube itself begins to contract rhythmically.
- It is thought that the oocyte surrounded by some granulosa cells is carried into the tube by these sweeping movements of the fimbriae and by motion of cilia on the epithelial lining.
- Once in the tube, cumulus cells withdraw their cytoplasmic processes from the zona pellucida and lose contact with the oocyte.
- Once the oocyte is in the uterine tube, it is propelled by cilia

fertilization

- Fertilization, the process by which male and female gametes fuse, occurs in the **ampullary region of the uterine tube.** This is the widest part of the tube
- Spermatozoa are deposited in the vaginal tract during sexual intercourse.
- They pass rapidly into the uterus, and then into the fallopian (uterine) tube.
- This movement is aided by contractions of the musculature of the uterus and uterine tube.
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- Has to be fertized within 12-24 hrs

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- Spermatozoa may remain viable in the female reproductive tract for several days.
- Only 1% of sperm deposited in the vagina enter the cervix, where they may survive for many hours.
- Movement of sperm from the cervix to the oviduct is accomplished primarily by their own propulsion, although they may be assisted by movements of fluids created by uterine cilia.
- The trip from cervix to oviduct requires a minimum of 2 to 7 hours, and after reaching the isthmus, sperm become less motile and cease their migration. Motility resumes after ovulation

- Fertilization takes place in the ampullary of the uterine tube
- Without fertilization, the oocyte degenerates 24hrs after ovulation
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- Spermatozoa undergo some changes in the female genital tract in order to be able to fertilize in order to fertilize
- Changes; 1)**Capacitation.** A glycoprotein coat and seminal plasma proteins are removed from the plasma membrane covering the acrosomal region of the nucleus. Only capacitated sperm penetrate corona radiate & undergo acrosomal rxt
- The process takes around 7 hrs.

- 2) **Acrosomal reaction**. The **acrosome reaction,** which occurs after binding to the zona pellucida, is induced by zona proteins.
- This reaction culminates in the release of enzymes needed to penetrate the zona pellucida, including acrosin and trypsin-like substances
- The enzymes help the spermatozoa to penetrate the barriers surrounding the oocyte. Barriers= **corona radiate, zona pellucida**, **and oocyte cell membrane**

- Penetration of corona radiate: Not all spermatozoa deposited in the female genital tract reach the site of fertilization. Of those that reach the site, only one is needed for fertilization. A few spermatozoa penetrate corona radiate

- Once spermatozoon touches zona pellucida, it penetrates rapidly.
- As the head of spermatozoon touches the oocyte cell membrane, changes occur in zona pellucida (zona reaction) which results in the inactivation of the spermatozoa receptor sites, and therefore only one sperm is able to penetrate the oocyte= prevents polysperms
- Fusion of oocyte-sperm cell membrane. As the spermatozoon touches the oocyte cell membrane, their plasma membranes fuse while the head and tail of the spermatozoon enter the cytoplasm of the oocyte. Once it has entered, a reaction follows that make the oocyte membrane impenetrable to other spermatozoa

- The phases of fertilization include phase 1, penetration of the corona radiata;
- phase 2, penetration of the zona pellucida; and
- phase 3, fusion of the oocyte and sperm cell membranes

PENETRATION OF THE CORONA RADIATA

- Of the 200 to 300 m spermatozoa deposited in tract, only 300 to 500 reach the site of fertilization.
- Only one of these fertilizes the egg. It is thought that the others aid the fertilizing sperm in penetrating the barriers protecting the female gamete.
- Capacitated sperm pass freely through corona cells

PENETRATION OF THE ZONA PELLUCIDA

- The zona facilitates and maintains sperm binding and induces the acrosome reaction
- Releaseof acrosomal enzymes (acrosin) allows sperm to penetrate the zona,
- Permeability of the zona pellucida changes when the head of the sperm comes in contact with the oocyte surface.
- This contact results in release of lysosomal enzymes from plasma membrane
- In turn, these enzymes alter properties of the zona pellucida **(zona reaction)** to prevent other sperm penetration and inactivate s receptor sites for spermatozoa on the zona surface.

FUSION OF THE OOCYTE AND SPERM CELL MEMBRANES

- As soon as the spermatozoon has entered the oocyte, the egg responds in three ways:
- **Cortical and zona reactions.**
- As a result of the release of cortical oocyte granules, which contain lysosomal enzymes,

(*a*) the oocyte membrane

- becomes impenetrable to other spermatozoa, and
- (*b*) the zona pellucida alters its structure and composition to prevent sperm binding and penetration.
- These reactions prevent polyspermy

Results of fertilization

- **Resumption of the second meiotic division.**

The oocyte finishes its second meiotic division immediately after entry of the spermatozoon

- **Metabolic activation of the egg.** The activating factor is probably carried by the spermatozoon
- Restoration of diploid

- The oocyte finishes its 2nd meiotic division immediately the spermatozoon enters, with only one mature daughter cell.
- The spermatozoon nucleus move closer to the oocyte nucleus.
- Eventually, the oocyte and spermatozoon lose their nuclear envelop and come into close contact.
- Zygote is formed

Results of fertilization

- 1) Restoration of diploid numbers of chromosomes,
- 2) the determination of the sex of the new individual.
- X carrying sperm=female embryo=XX, Y carrying sperm=male embryo=XY,
- 3) Initiation of cleavage = Mitotic division

IMPLANTATION

- Implantation is when a developing embryo is attached to the wall of the uterus. After fertilization, a zygote is formed.
- The zygote grows and moves at the same time in the uterine tube until it is implanted in the body of the uterus as a blastocyst
- **Cleavage.** Normal mitotic division of the zygote results in a two-cell stage, which takes approximate 30 hrs after fertilization.
- A series of mitotic divisions follow resulting in a rapid increase in the number of cells. These cells are called blastomeres and they become smaller with each cleavage division

- A four- cell stage takes approximate 40 hrs.
- A 16-cells stage zygote is known as the **morula** and is reached approximately three days after fertilization.
- At this stage, the morula consists of a group of centrally located cells=the **inner cell mass** and a surrounding layer, the **outer cell mass.**
- The inner cell mass give rise to **embryoblasts** and the outer cell mass give rise to **trophoblasts**

C L I N I C A L C O R R E L A T E S

- **Contraceptive Methods**

Barrier techniques of contraception include the male condom &the female condom,.

Other barriers placed in the vagina include the diaphragm, the cervical cap, and the contraceptive sponge.

- The **contraceptive pill** is a combination of estrogen and the progesterone analogue progestin, which together inhibit ovulation but permit menstruation
- Both hormones act at the level of FSH and LH, preventing their release from the pituitary

- **Depo-Provera** is a **progestin** compound that can be implanted subdermally or injected intramuscularly to prevent ovulation for up to 5 years
- 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

Cleavage

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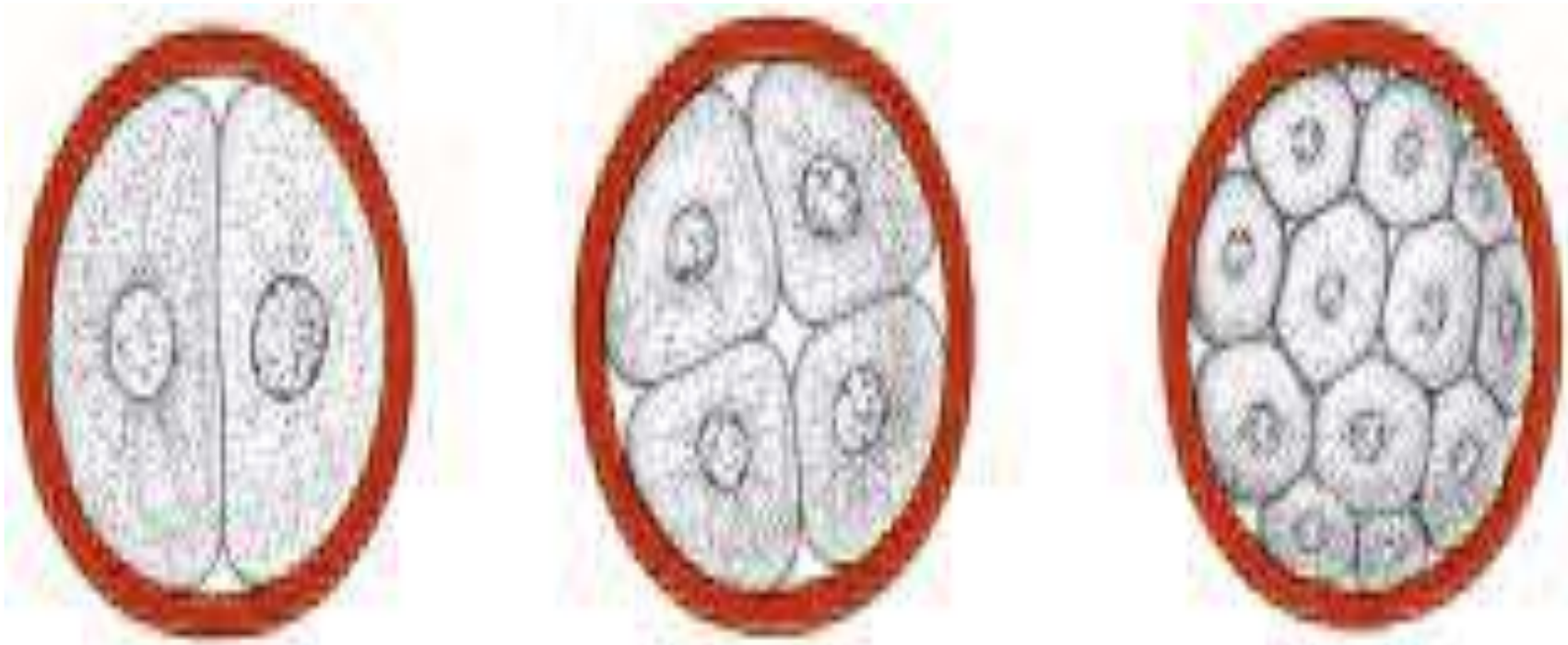
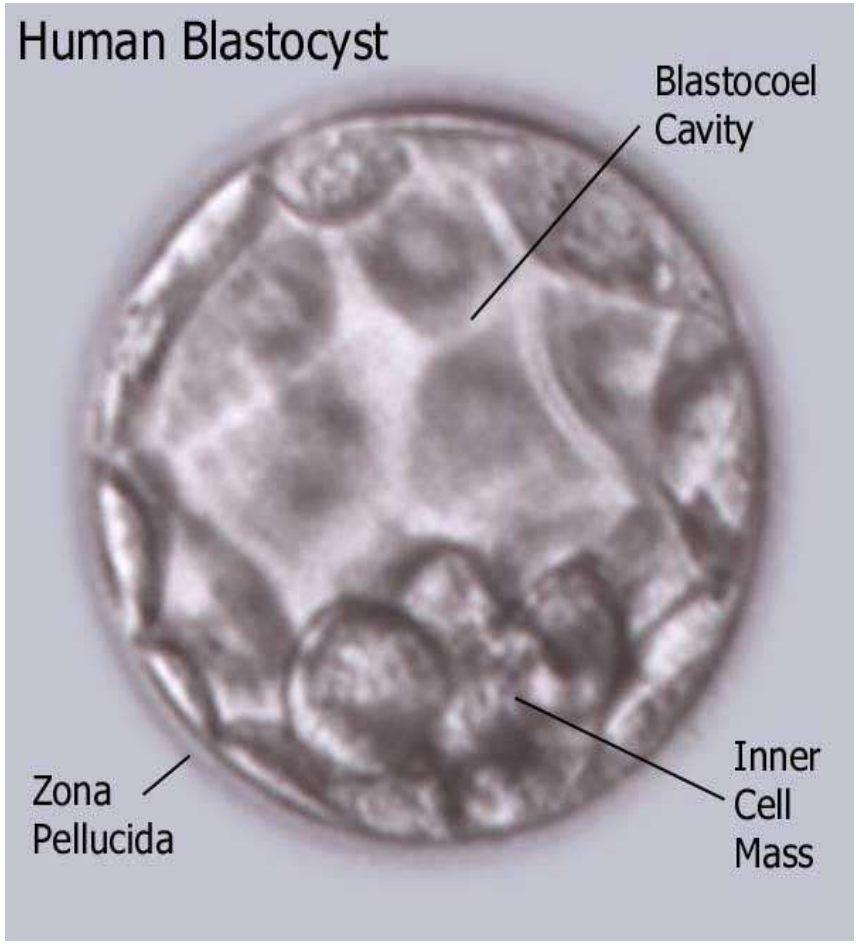
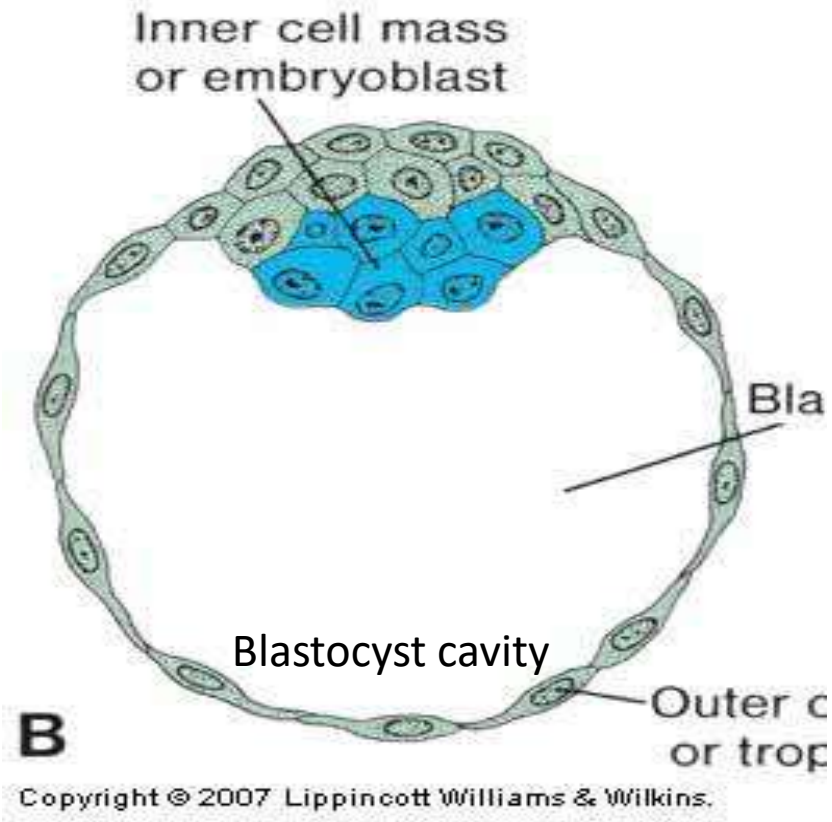
- After the third cleavage, blastomeres maximize their contact with each other, forming a compact ball of cells held together by tight junctions
- This process, **compaction,** segregates inner cells, which communicate extensively by gap junctions, from outer cells.
- Approximately 3 days after fertilization, cells of the compacted embryo divide again to form a 16-cell **morula** (mulberry
- Inner cells of the morula constitute the **inner cell mass,** and surrounding cells compose the **outer cell mass**


Two-cell stage
Four-cell stage
Morula

blastocyst


Human Blastocyst
Blastocoel Cavity
Zona Pellucida
Inner Cell Mass

- About the time the morula enters the uterine cavity, fluid begins to penetrate through the zona pellucida into the intercellular spaces of the inner cell mass.
- Gradually the intercellular spaces become confluent, and finally a single cavity, the **blastocele**
- At this time, the embryo is a **blastocyst.** Cells of the inner cell mass, now called the **embryoblast,** are at one pole, and those of the outer cell mass, or **trophoblast,** flatten and form the epithelial wall of the blastocyst. The zona pellucida has disappeared, allowing implantation to begin



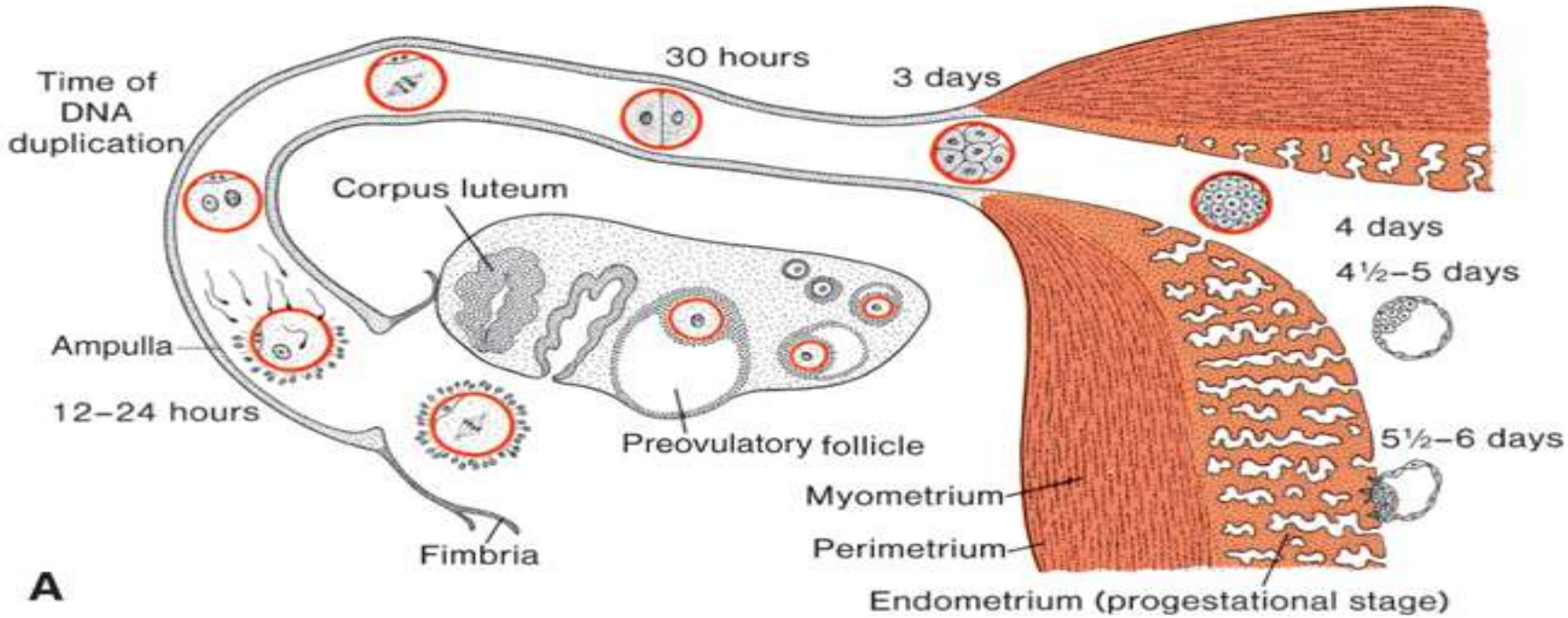
The inner cell mass and, the outer cell mass forming the outer wall of the blastocyst. Note the blastocyst cavity inside

Blastocyst formation

- At the time the morula enters the uterine cavity, a cavity called **blastocele** develops inside the intercellular space of the inner cell mass, and the embryo is now known as the **blastocyst**.
- The inner cell mass=**embryoblast**,
- the outer cell mass=**trophoblasts**
- **Embryoblast cells** give rise to tissues of the embryo proper
- while **trophoblast** forms the epithelial wall of the blastocyst and contributes to the placenta

implantation

- About the sixth day, the trophoblast starts penetrating the wall of the uterus.
- The penetration is aided by proteolytic enzymes produced by the trophoblast.
- By the end of 1st week of development, the zygote has passed the morula and blastocyst stage and has begun its implantation.
- By the end of 2nd week full implantation has taken place



Schematic diagram showing the movement of an oocyte until it is implanted in the uterine body

Uterus preparation for implantation

- The uterine wall is made up of 3 layers,
- the innermost= endometrium or mucosal lining,
- the middle smooth muscular=myometrium,
- the outer peritoneal covering lining= perimetrium
- Under the influence of progesterone, the uterine walls get thickened, become rich in blood supply, blood vessels become tortuous, and the secretory glands activity are increased, more glycogen is stored=decudious reaction

- Implantation normal occur in the **endometrium along the posterior or anterior wall** of the **body** of the uterus
- If fertilization occur, the glands shows increased secretory activities and then a dense capillary bed is formed
- If fertilization fails parts of the wall of the uterus (endomentrium) is shed off as menstrual flow.= Spongy and compact layers

C L I N I C A L C O R R E L A T E S

- **Abnormal Zygotes**
- The exact number of **abnormal zygotes** formed is unknown because they are usually lost within 2 to 3 weeks of fertilization, before the woman realizes she is pregnant, and therefore are not detected.
- Estimates are that as many as 50% of pregnancies end in spontaneous abortion and that half of these losses are a result of chromosomal abnormalities.
- These abortions are a natural means of screening embryos for defects, reducing the incidence of congenital malformations.
- Without this phenomenon, approximately 12% instead of 2% to 3% of infants would have birth defects

Uterine phases

- During this menstrual cycle, the uterine endometrium passes through three stages,

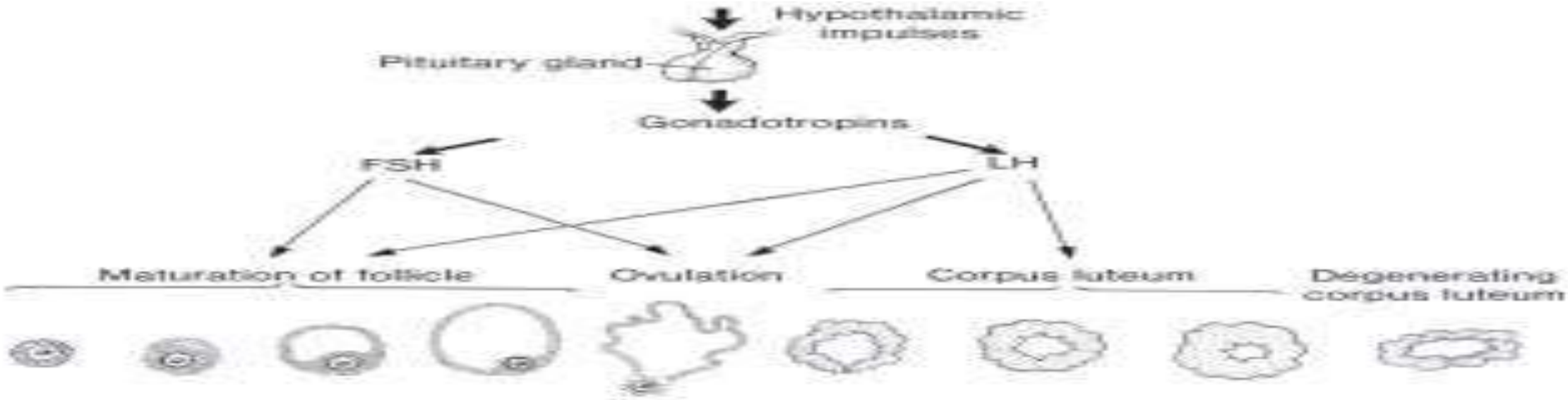
the **follicular** or **proliferative phase**

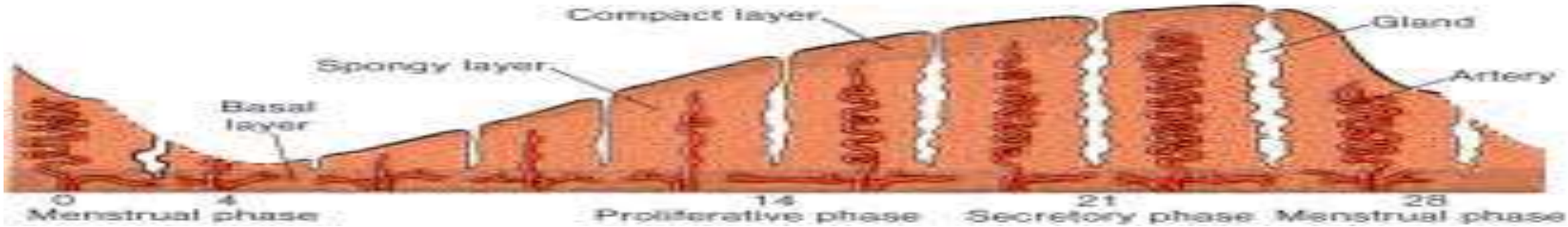
the **secretory** or **progestational phase,** and

the **menstrual phase**

- The proliferative phase begins at the end of the menstrual phase, is under the influence of estrogen, and parallels growth of the ovarian follicles.
- The secretory phase begins approximately 2 to 3 days after ovulation in response to progesterone produced by the corpus luteum.
- If fertilization does not occur, shedding of the endometrium (compact and spongy layers) marks the beginning of the menstrual phase.
- If fertilization does occur, the endometrium assists in implantation and contributes to formation of the placenta

- At the time of implantation, the mucosa of the uterus is in the secretory phase during which time uterine glands and arteries become coiled and the tissue becomes succulent.
- As a result, three distinct layers can be recognized in the endometrium: a superficial **compact layer,** an intermediate **spongy layer,** and a thin **basal layer**
- Normally, the human blastocyst implants in the endometrium along the anterior or posterior wall of the body of the uterus, where it becomes embedded


Hypothalamic impulses
Pituitary gland
Gonadotropins
FSH
LH
Maturation of follicle
Ovulation
Corpus luteum
Degenerating corpus luteum


Compact layer
Gland
Spongy layer
Artery
Basal layer
0
4
14
21
28
Menstrual phase
Proliferative phase
Secretory phase
Menstrual phase

- If the oocyte is not fertilized, venules and sinusoidal spaces gradually become packed with blood cells, and an extensive diapedesis of blood into the tissue is seen.
- When the **menstrual phase** begins, blood escapes from superficial arteries, and small pieces of stroma and glands break away. During the following 3 or 4 days, the compact and spongy layers are expelled from theuterus, and the basal layer is the only part of the endometrium that is retained
- This layer, which is supplied by its own arteries, the **basal arteries,** functions as the regenerative layer in the rebuilding of glands and arteries in the **proliferative phase**

Abnormal implantation

- Implantation may occur close to the internal os of uterus. Consequence is severe bleeding in the 2nd trimester and during delivery.
- Extrauterine (ectopic) pregnancies are common. It may occur in the abdominal cavity, ovary, or uterine tube.
- Ectopic pregnancy usually leads to death of the embryo and severe bleeding by the mother in the 2nd month.
- Majority ectopic occurs in uterine tube, most in the ampulla