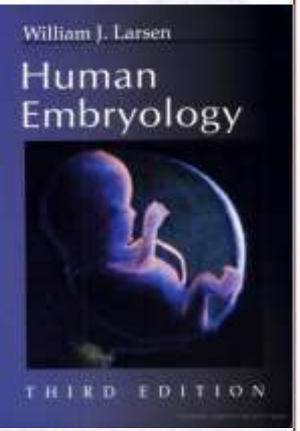
# Embryology. Male and Female Reproductive Cells

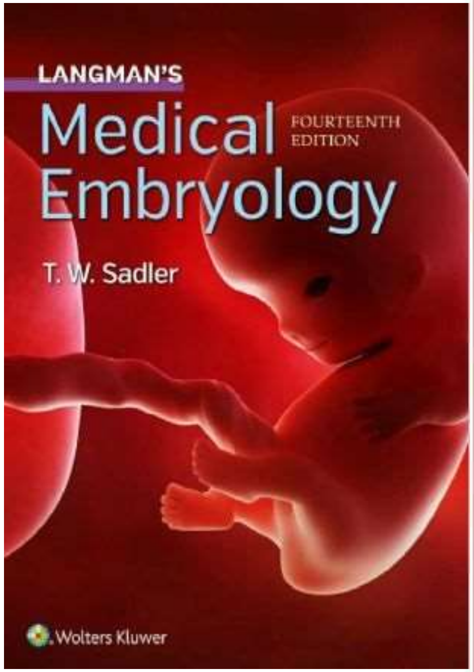
1. Introduction to Embryology
2. The molecular and genetic basis of embryonic development
3. Sex cells (gametes) and gametogenesis
4. Male and female gametes
5. Spermatogenesis and its regulation
6. Oogenesis and its regulation
7. Ovarial cycle and ovulation

# Embryology

- Embryology (Gr. *ἐμβρυον*, embryo + *logos*, study)
  - ✓ general embryology (embryogenesis)
  - ✓ special embryology (organogenesis)




- **Human Embryology**
- **Medical Embryology**
- prenatal development –
  280 days, 10 months:
  - ➢ embryonic period (embryo)
    – from fertilization to 8th week of development
    - preembryonic period (early development) – from the fertilization to 2nd week of gestation
    - embryonic period (late development) – from the 3rd week to the end of 2nd month
  - ➢ fetal period (fetus) – from the 9th developmental week to the birth
- postnatal development



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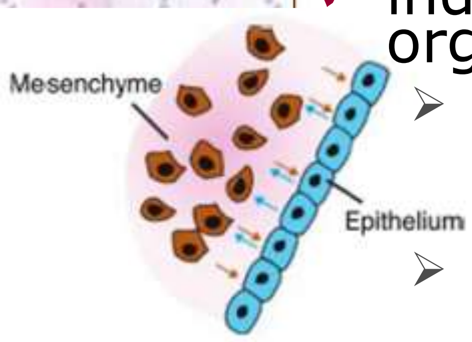
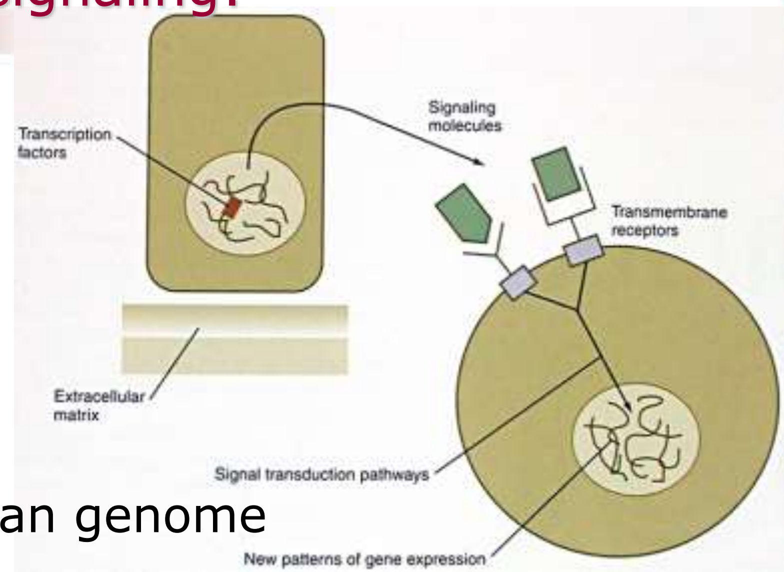
# Basis of embryonic development

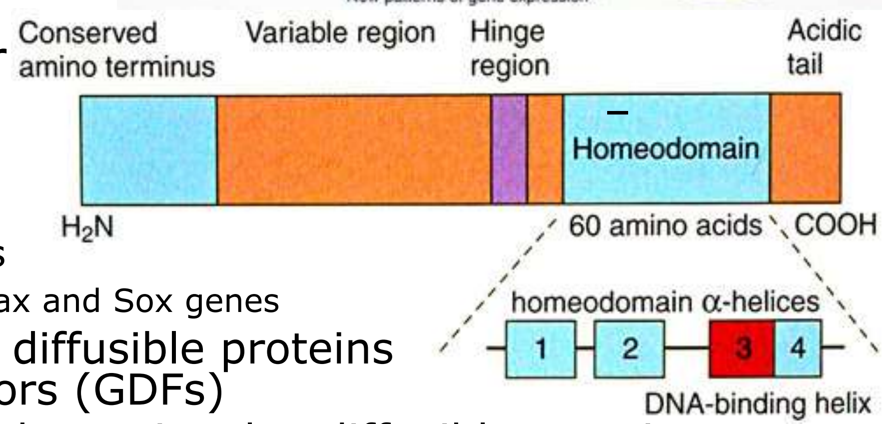
- Molecular regulation and signaling:

✓ induction and organ formation:
  - induction:
    - inducer – produces a signal
    - responder to that signal
  - epithelial-mesenchymal interactions
  - enhancers and silencers
  - 23 000 genes in the human genome




✓ cell-to-cell signaling
  - a ligand and receptor
  - transcription factors general and specific:
    - zinc ring proteins
    - homeodomain proteins
      - homeobox (HOX), Pax and Sox genes
  - paracrine signaling – diffusible proteins paracrine signaling factors (GDFs)
  - juxtacrine signaling: do not involve diffusible proteins
    - Notch pathway – a process analogous to paracrine signaling
    - ligands interact with receptors on neighboring cells
    - direct transmission by gap junctions



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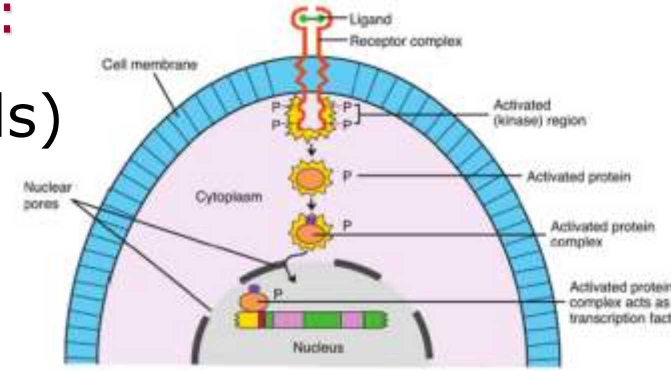
# Basis of embryonic development

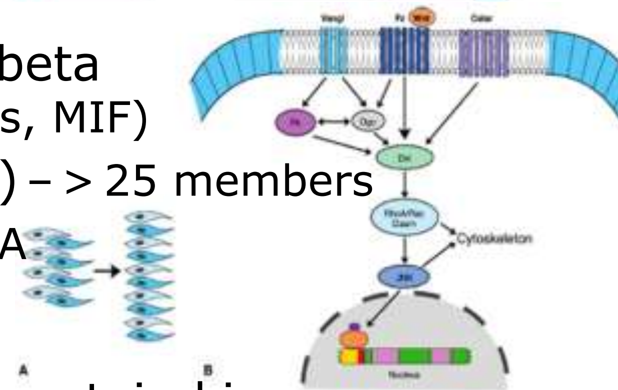
- Signal transduction pathways:

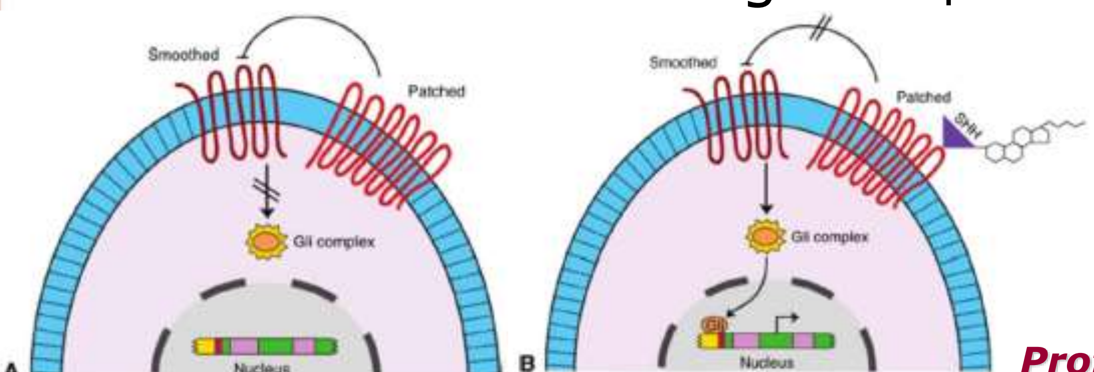
✓ signaling molecules (ligands)
  - four groups of GDFs
    - WNT proteins – at least 15 different WNT genes
    - hedgehog proteins (SHH)
    - transforming growth factor-beta (TGF-β)– > 30 members (BMPs, MIF)
    - fibroblast growth factor (FGF) – > 25 members
  - neurotransmitters – SER, GABA, NA

✓ transmembrane receptors

✓ first & second messengers – protein kinases



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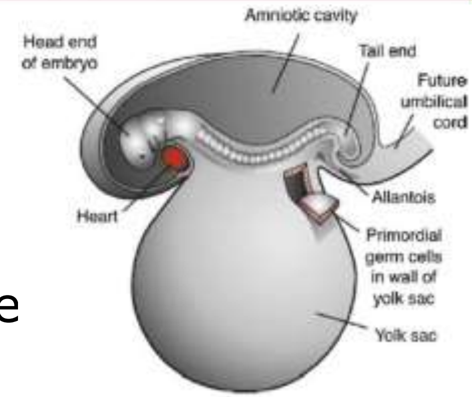
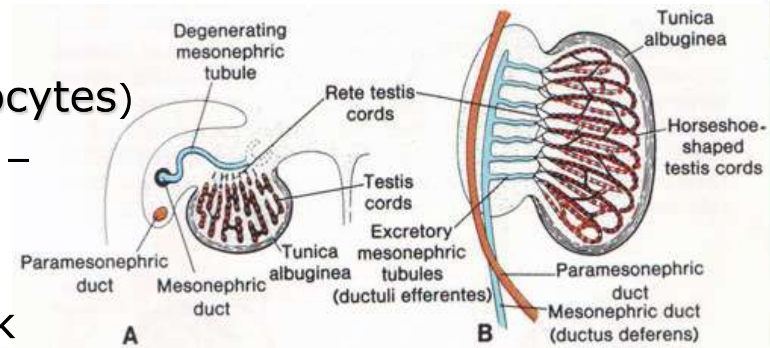
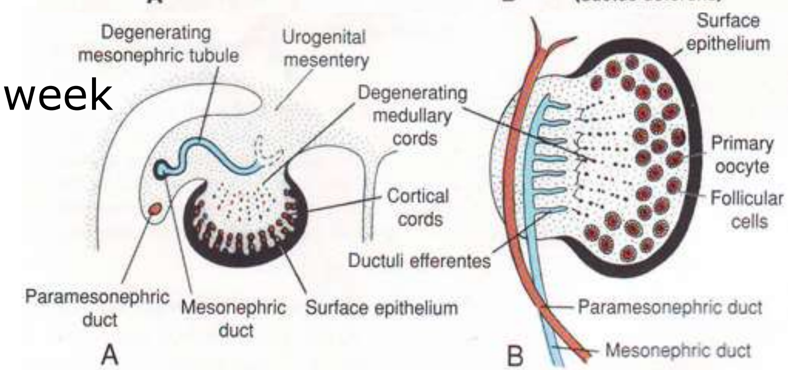
# Sex cells (gametes)

- Reproductive tissue:
  - ✓ a separate tissue – *A. Hadjiolov*, 1930
  - ✓ kind of epithelial tissue
- composition:
  - ✓ sex cells (gametes) – male and female
  - ✓ "somatic" cells
- embryonic origin:
  - ✓ primordial germ cells (gonocytes)
    - ➢ formation in the epiblast – 2nd week of gestation
    - ➢ movement to the wall of the yolk sac – 3rd week
    - ➢ migration toward the developing gonads – 5th week
    - ➢ formation of primary sex cords
    - ➢ sex differentiation – male and female
    - ➢ gametogenesis

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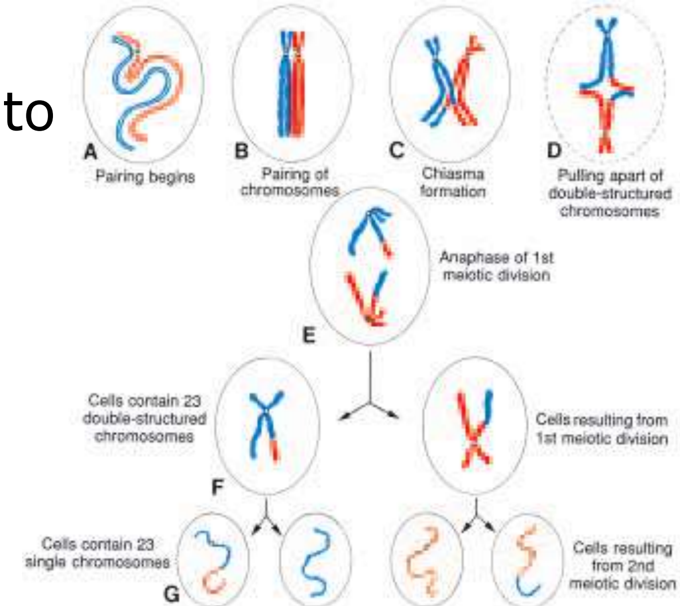
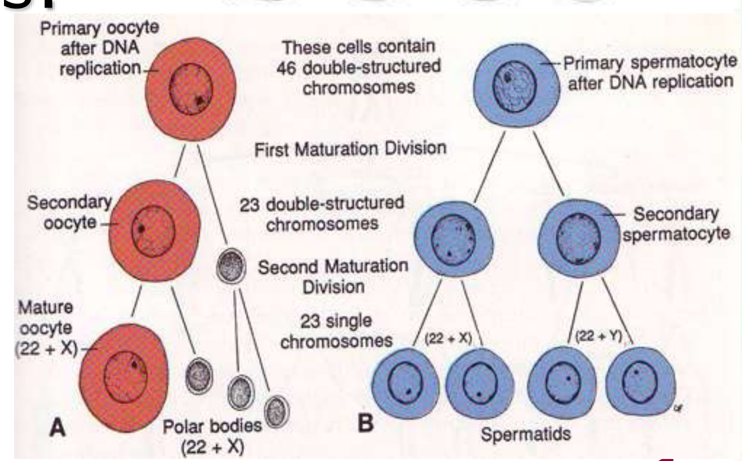
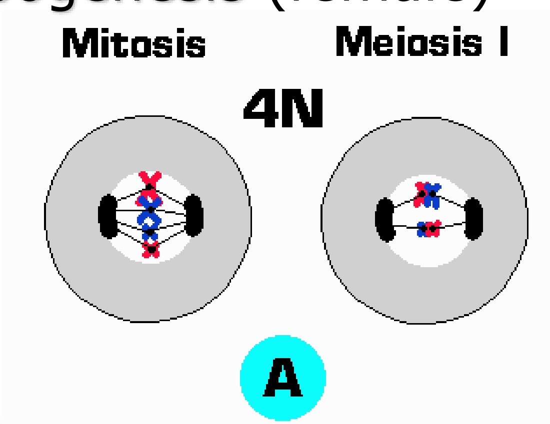
# Gametogenesis

- Gametogenesis:
  - ✓ conversion of germ cells into male and female gametes
  - ✓ the cell division that takes place in the germ cells to generate male and female gametes is meiosis
  - ✓ cytodifferentiation
- forms of gametogenesis:
  - ✓ spermatogenesis (male)
  - ✓ oogenesis (female)



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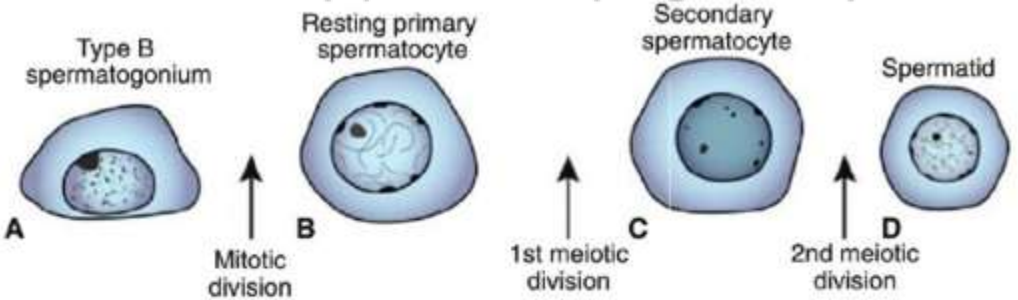
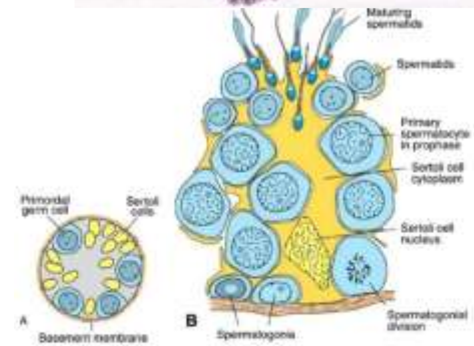
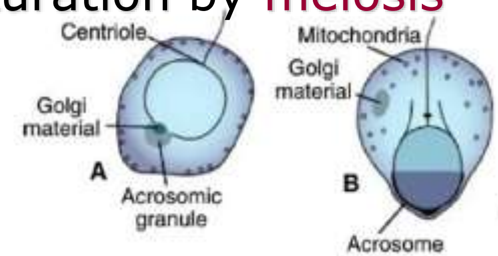
# Spermatogenesis

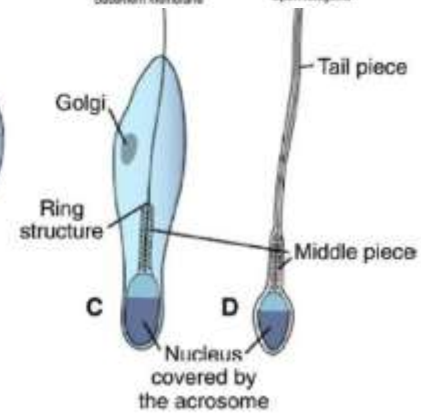
- spermatogonium ⇨ mature spermatozoon:
  - ✓ process duration – 74 days
  - ✓ ~300 million sperm cells/daily
- spermatocytopoiesis – in seminiferous tubules of the testis:
  - ✓ dividing of spermatogonia by mitosis (spermatocytogenesis)



  - ✓ growth and maturation by meiosis




  - ✓ cytodifferentiation of spermatids into spermatozoa (spermiogenesis)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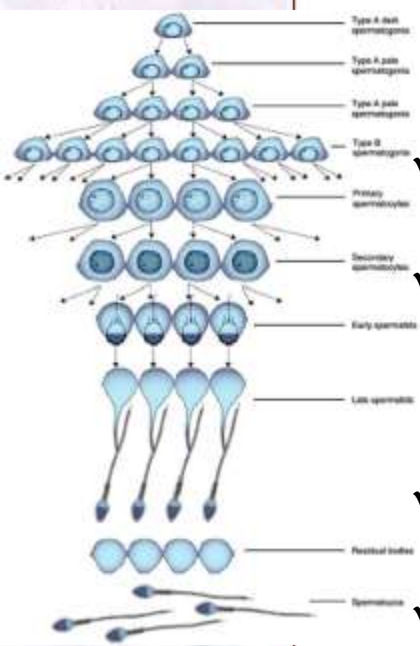
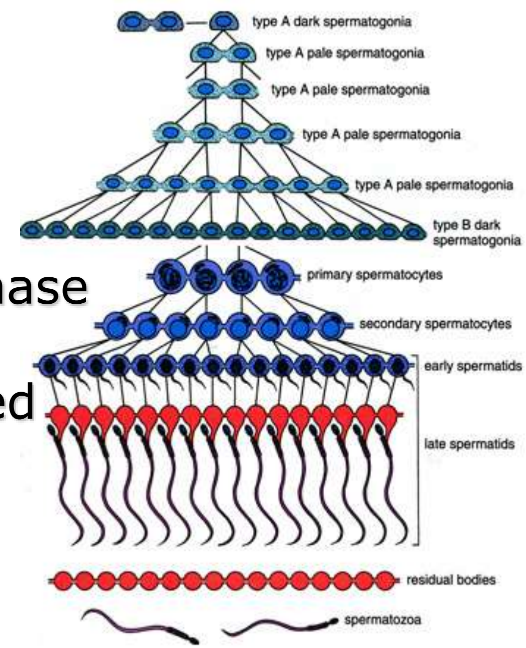
# Spermatogenesis

- peculiarities of spermatogenesis:



  - ✓ maturation of sperm begins at puberty
  - ✓ wavy and continuously course
    to a ripe old age
  - ✓ two meiotic divisions without interphase
  - ✓ four mature spermatozoa are formed
    from one spermatogonium
  - ✓ connected by cytoplasmic bridges
  - ✓ only after their separation from the residual bodies
    can the spermatozoa be considered isolated cells



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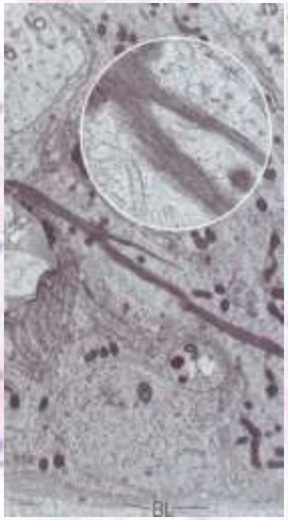
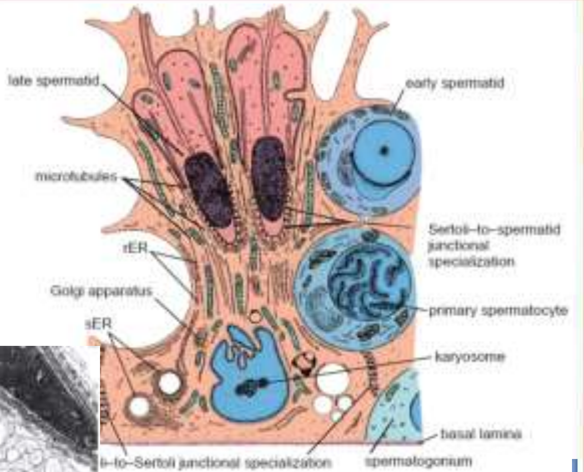
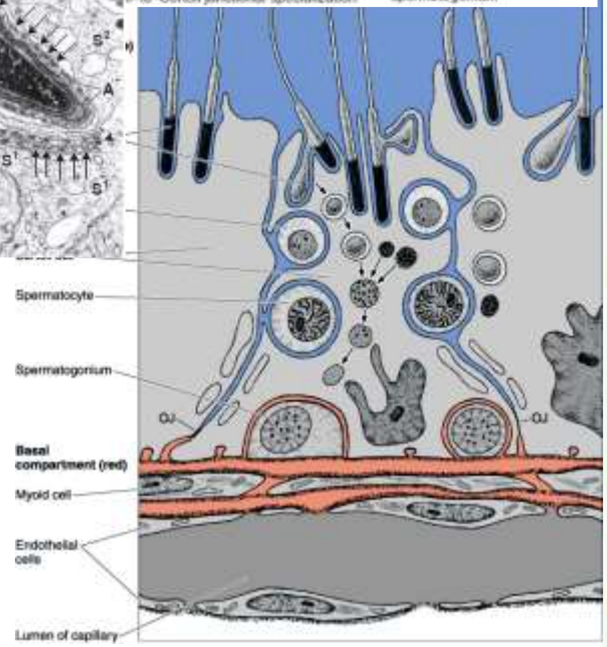
# Non-spermatogenic cells

***Enrico Sertoli* *(1842-1910)***



- *Sertoli* cells:
- sustentacular cells
  - ✓ derived from the surface epithelium
  - ✓ elongated pyramidal cells – 30-70 μm
  - ✓ *Charcot-Böttcher* crystals
  - ✓ basal and adluminal compartments
  - ✓ occluding junctions ⇨ blood-testis barrier
  - ✓ main functions:
    - ➢ support and protection of the developing spermatozoa
    - ➢ trophic (nutritional) role
    - ➢ phagocytosis – residual bodies
    - ➢ secretion (endocrine function) – inhibin, testis transferrin, androgen-binding protein (ABP), anti-Müllerian hormone (AMH)
    - ➢ bind vitamin E and A

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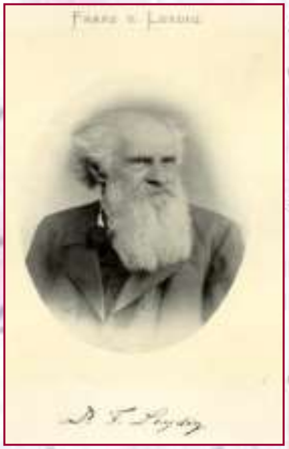
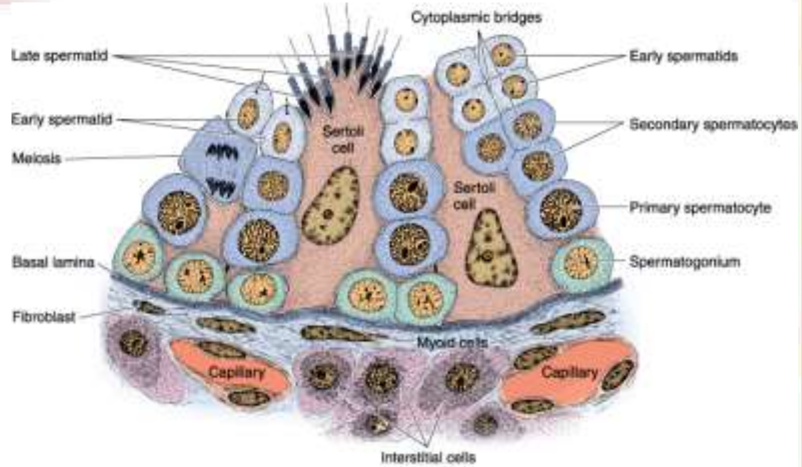
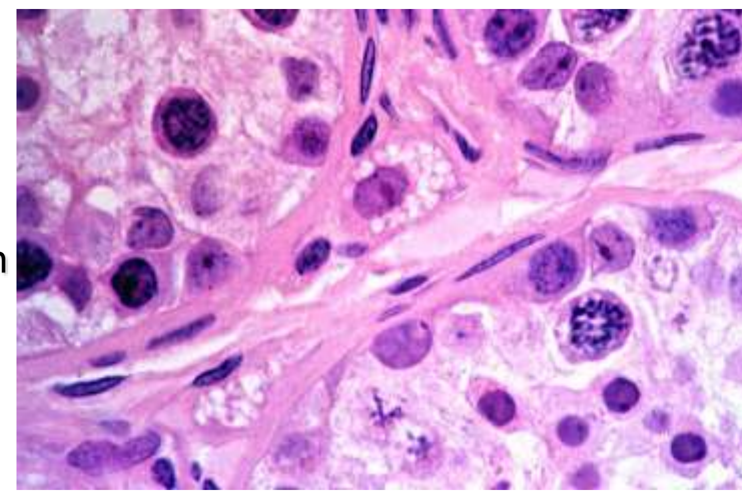
# Non-spermatogenic cells



**Franz von Leydig (1821-1908)**

- *Leydig* cells:
- interstitial cells – 1850
  - ✓ formation during the fetal period (during 16th and 20th week of gestation)
  - ✓ secondary interstitial cell – at puberty
  - ✓ located individually or in small groups around the capillaries
  - ✓ rounded or polygonal
  - ✓ well-developed sER
  - ✓ lipofuscin pigment
  - ✓ *Reinke*'s crystals – 3-20 µm
  - ✓ steroid-secreting cells – produce androgens (testosterone, androstenedione and dehydroepiandrosterone)
    ⇨ secondary male sex characteristics





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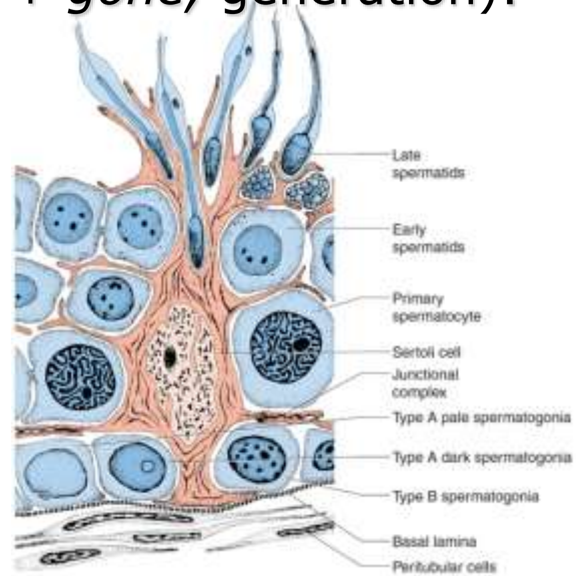
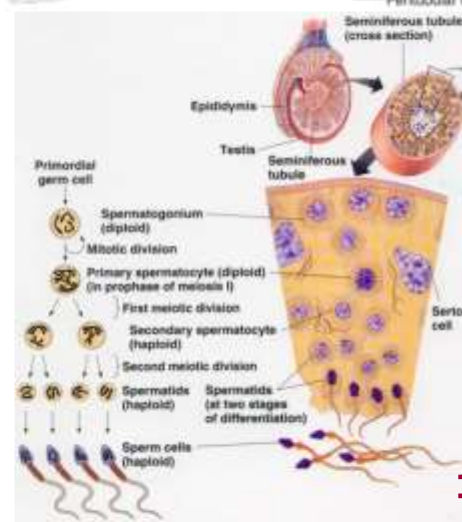
# Spermatogenic cells

- Spermatogonia (Gr. *sperma*, seed + *gone*, generation):
  - ✓ about 12 µm in diameter
  - ✓ situated next to the basal lamina of the epithelium
  - ✓ type A – stem cells
    - ➢ type *Ad* cells – divide rarely
    - ➢ type *Ap* cells – mitotic division
  - ✓ type B – progenitor cells (mitotic division – 16 days)

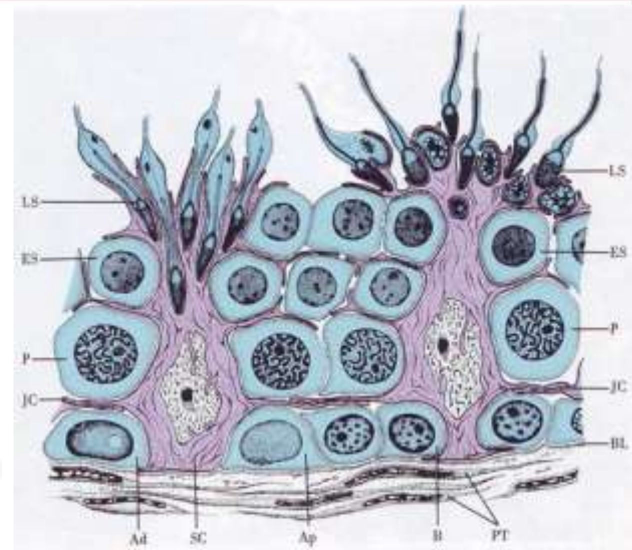


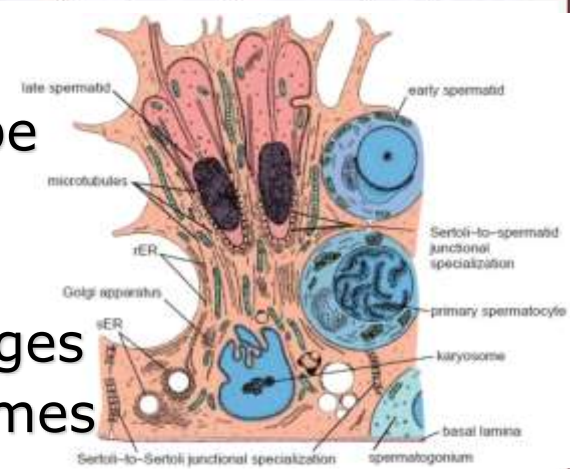

- Primary spermatocytes:
  - ✓ largest cells – 18-20 µm
  - ✓ enter a prolonged prophase of first meiotic division (22 days) – preleptotene spermatocytes
  - ✓ diploid – 46 (44, XY) chromosomes
  - ✓ 23 tetrades (2n DNA)



# Spermatogenic cells

- Secondary spermatocytes:
  - ✓ smaller cells – 12 μm
  - ✓ in meiosis II (16 days) – prespermatids
  - ✓ haploid – 23 chromosomes
  - ✓ normal amount of DNA (2n)
- Spermatids:
  - ✓ small cells – 7-8 μm
  - ✓ early spermatids – oval in shape
  - ✓ late spermatids – elongated
  - ✓ juxtaluminal location
  - ✓ connected by cytoplasmic bridges
  - ✓ haploid – contain 23 chromosomes
  - ✓ reduced amount of DNA – 1n
  - ✓ do not divide – undergo spermiogenesis





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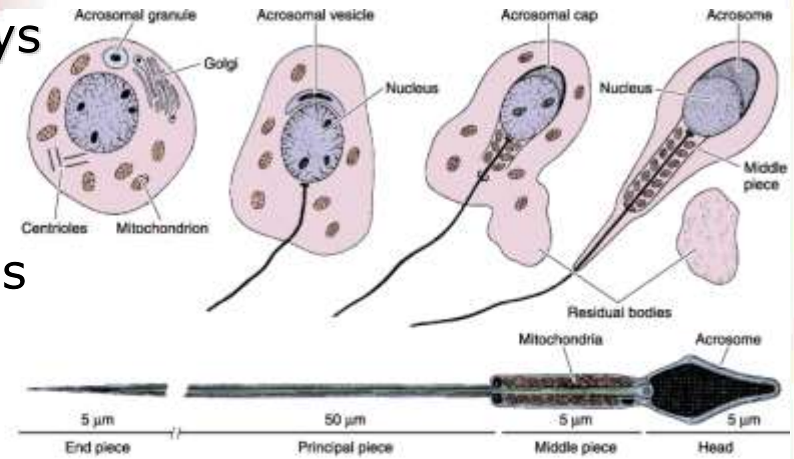
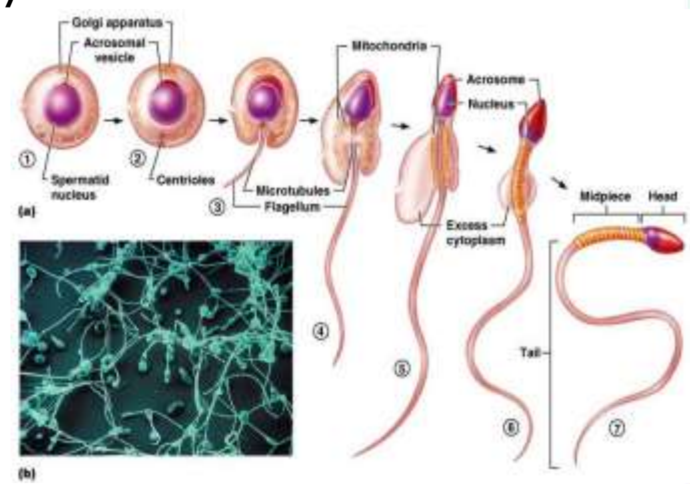
# Spermiogenesis

- spermatid ⇨ mature spermatozoon:
  - ✓ process duration – 24 days
- 3 phases:
  - ✓ *Golgi* phase
    - ➢ proacrosomal granules
  - ✓ acrosomal phase
    - ➢ acrosomal vesicle
    - ➢ acrosome – hydrolytic enzymes:
      - hyaluronidase
      - neuraminidase
      - acid phosphatase
      - acrosin (zonalysin)
  - ✓ maturation phase
    - ➢ residual bodies are shed
    - ➢ formation of spermatozoa
    - ➢ release of mature spermatozoa ⇨ spermiation



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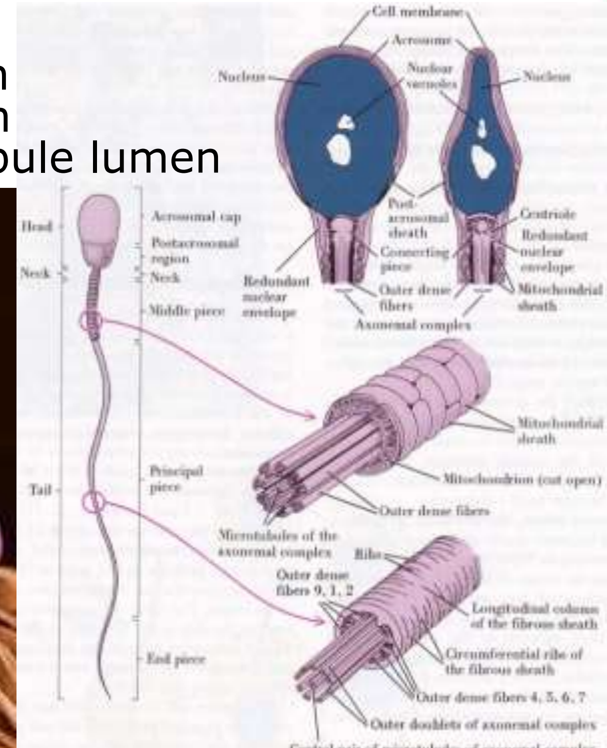
# Spermatozoon

***Anton van Leeuwenhoek (1632-1723)***

- Spermatozoon (Gr. *σπέρμα*, seed + *ζῷον*, living being):
  - ✓ mature male gamete
  - ✓ first observed in 1677
  - ✓ total length – 58-67 μm
  - ✓ spermiation – the process by which mature spermatids are released from Sertoli cells into the seminiferous tubule lumen



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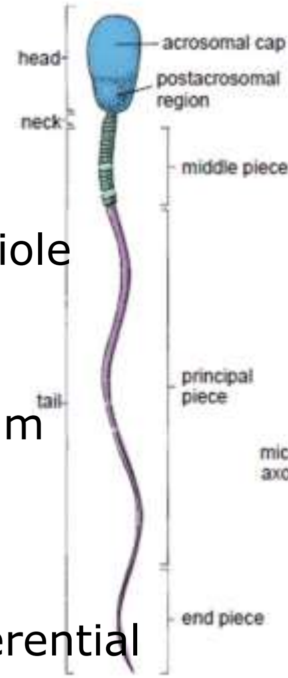
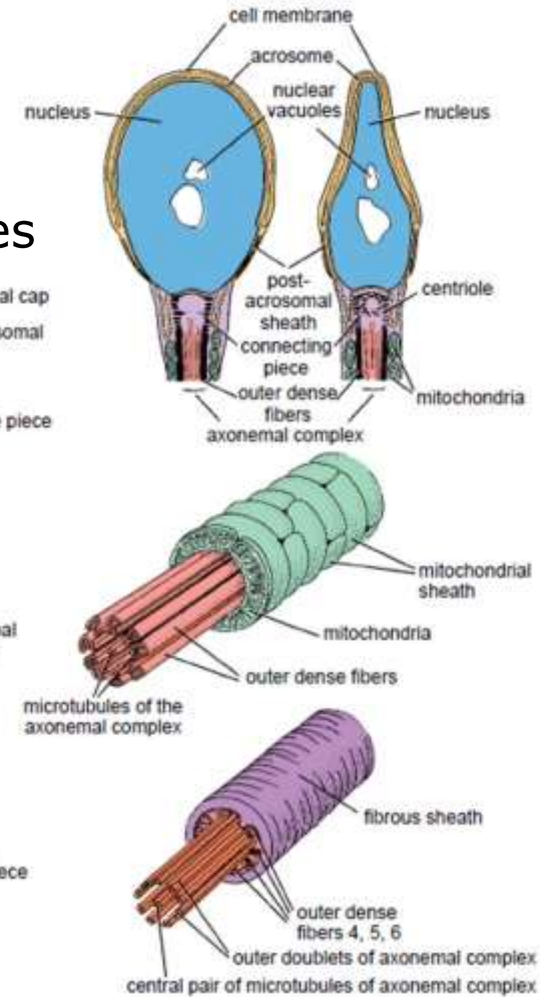
# Spermatozoon

- <u>Spermatozoon</u> (Gr. *σπέρμα*, seed + *ζῷον*, living being):
- structure:
  - ✓ head –
    length 5 µm; wide 3 µm; apex 1 µm
    - ➢ condensed nucleus, 1-2 vacuoles
    - ➢ acrosomal cap
- ✓ neck –
  length 0.3 µm; diameter 1 µm
  - ➢ covered by plasmalemma
  - ➢ basal body – proximal centriole
- ✓ tail – flagellum
  - ➢ middle piece –
    length 5-7 µm;diameter ~1 µm
    - axonemal complex
    - spiraled mitochondria
  - ➢ principal piece – 45-50 µm
    - longitudinal and circumferential fibrous sheaths
  - ➢ end piece – 5-7 µm
    - axoneme
    - surrounding plasmalemma



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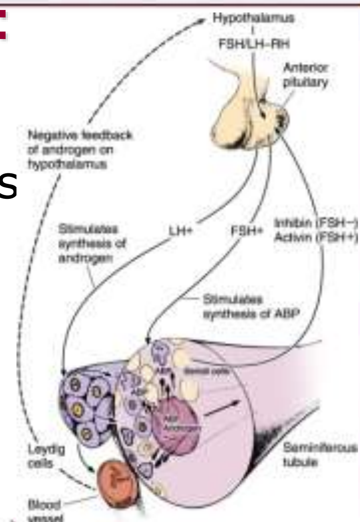
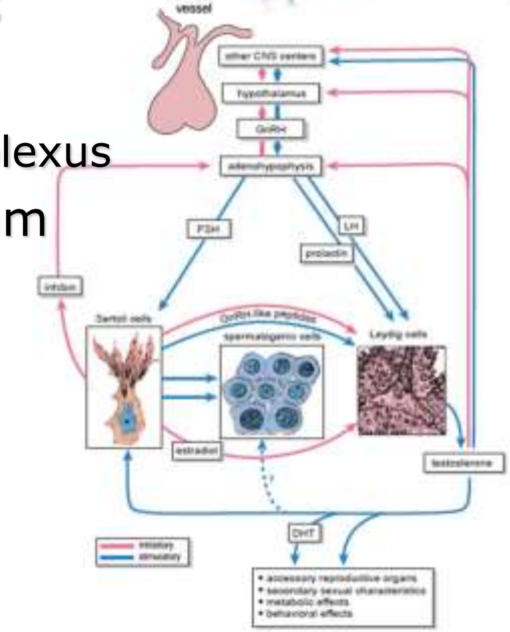
# Regulation of spermatogenesis

- Central regulation ⇨ stimulatory influence:
  - ✓ hypothalamus – GRH (hypophysis)
  - ✓ adenohypophysis – FSH (seminiferous tubules
    LH (*Leydig* cells)
- Local regulation – stimulatory and inhibitory:
  - ✓ *Sertoli* cells – ABP and inhibin
  - ✓ *Leydig* cells – testosterone
- Spermatogenesis regulating factors:
  - ✓ temperature ~35°C (cryptorchidism)
    - ➢ rich venous plexus, pampiniform plexus
    - ➢ evaporation of sweat from scrotum
    - ➢ contraction of cremaster muscles
  - ✓ malnutrition
  - ✓ alcoholism
  - ✓ action of certain toxic drugs
  - ✓ X-ray irradiation
  - ✓ disturbance in blood supply

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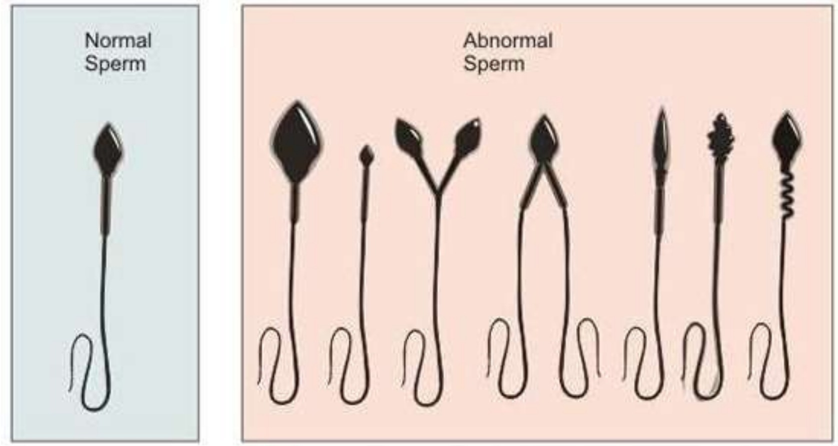
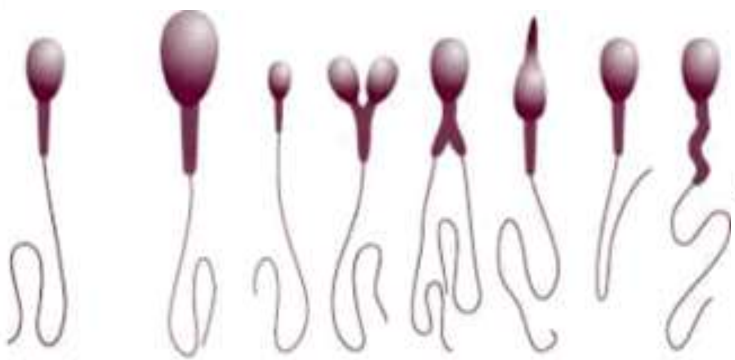
# Abnormal gametes

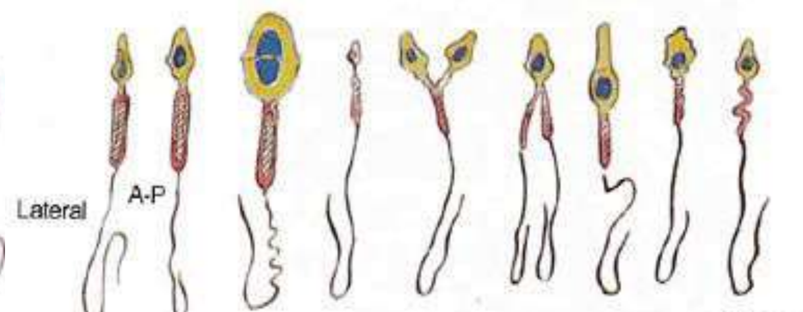
- in humans and in most mammals:

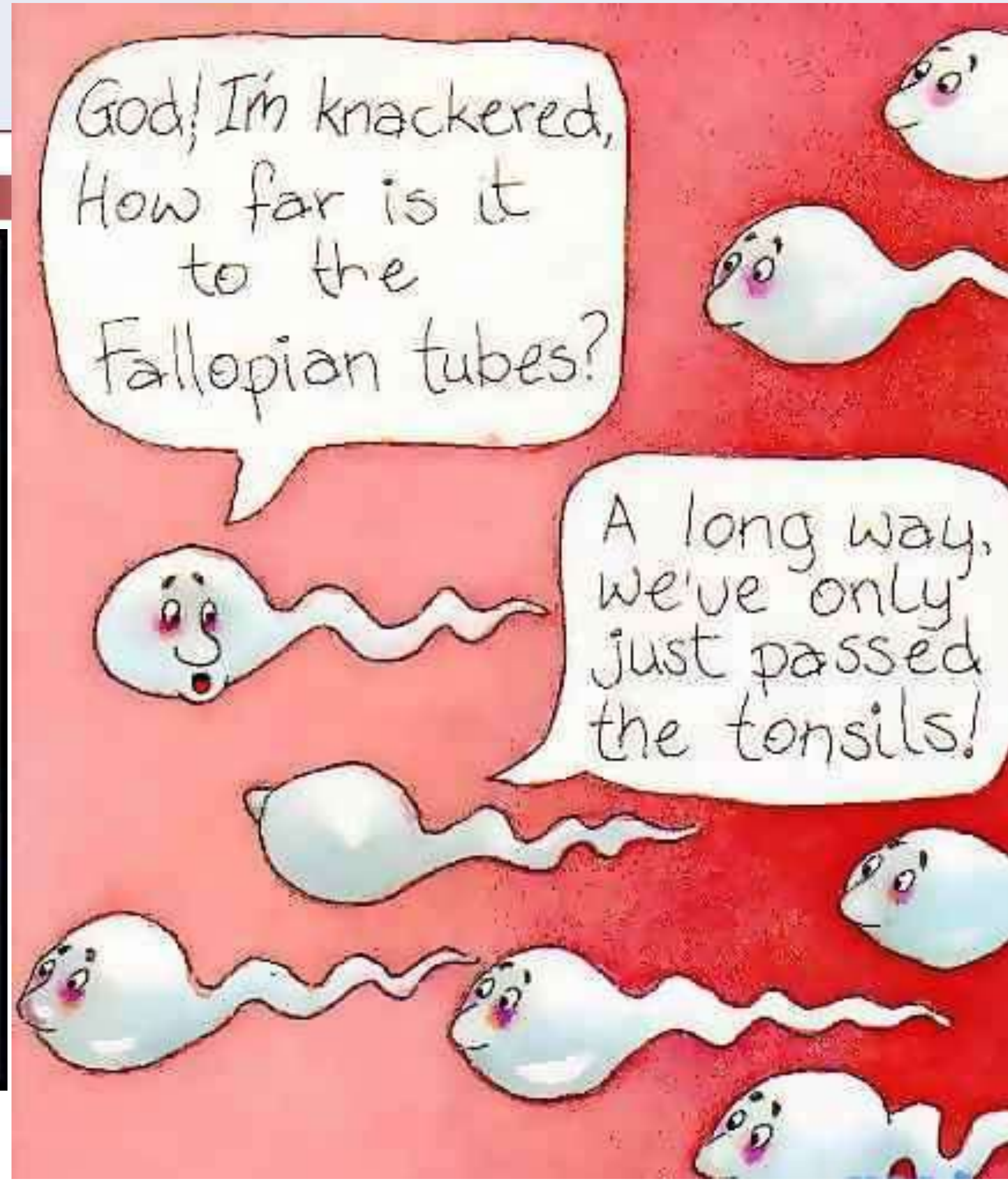
✓ abnormal spermatozoa – up to 10% of all spermatozoa

- ➢ abnormal head or tail
- ➢ giants or dwarfs
- ➢ sometimes are joined
- ➢ lack normal motility and probably do not fertilize oocytes



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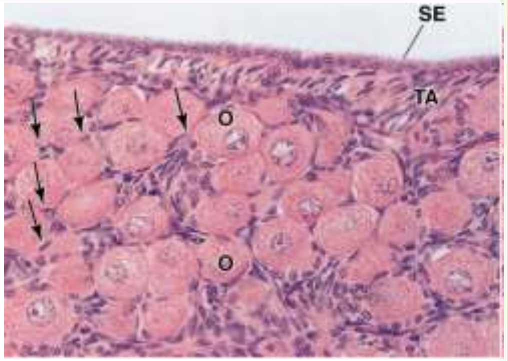
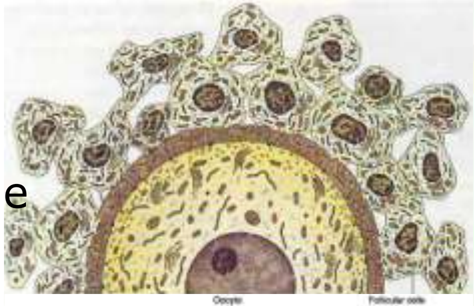
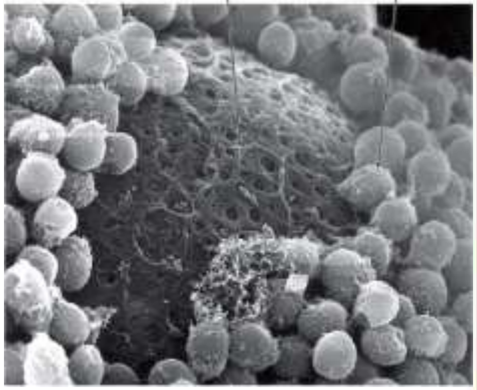
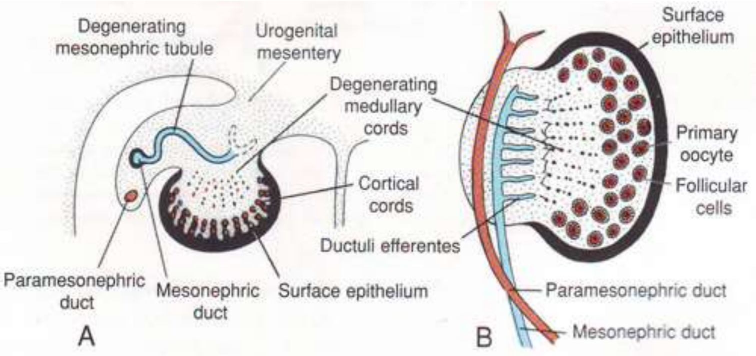
# Female gametes

- Female sex cells:
  - ✓ oocytes (Gr. *oon*, egg + *kytos*)
  - ✓ follicular (granulosa) cells
    - ➢ flat epithelial cells
    - ➢ defending function
    - ➢ secretory role – *liquor folliculi*
    - ➢ endocrine secretion – estrogens
    - ➢ ovulation ⇨ lutein cells – progesterone
  - ✓ thecal cells (thecocytes)
  - ✓ interstitial cells
  - ✓ hilus cells







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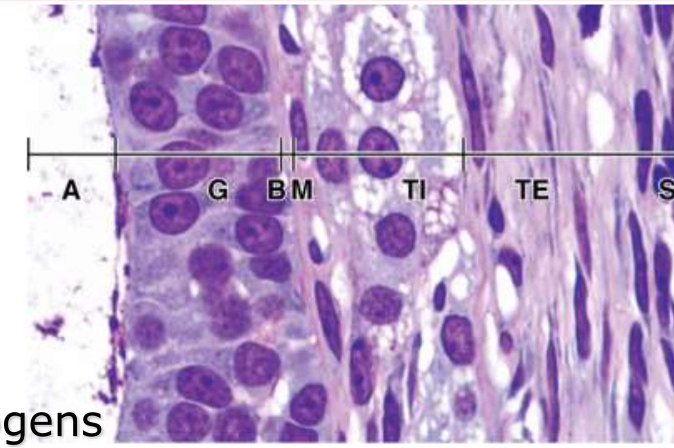
# Female gametes

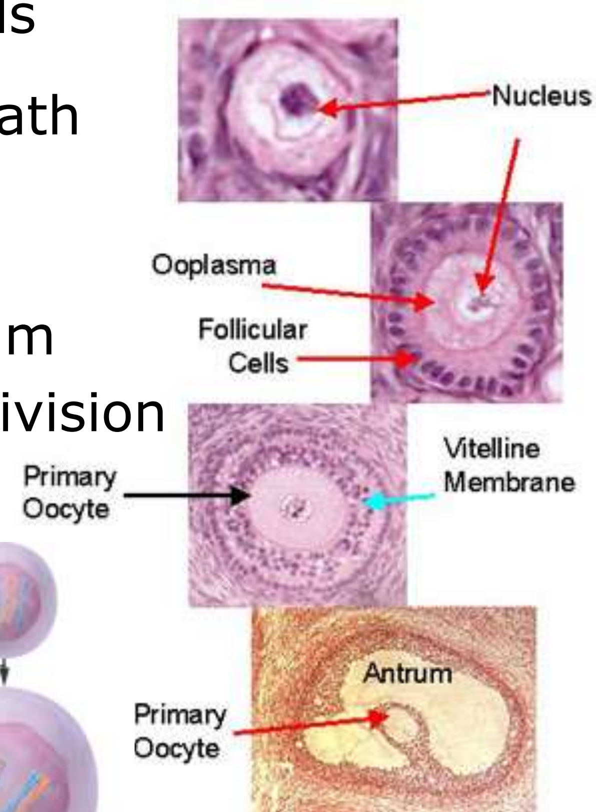
- Female sex cells:

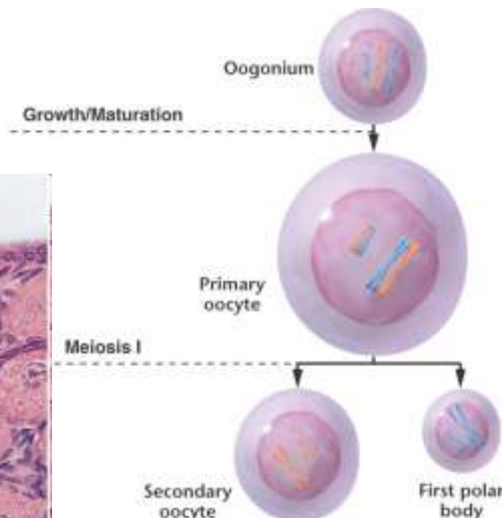



  - ✓ oocytes (Gr. *oon*, egg + *kytos*)
  - ✓ follicular cells
  - ✓ thecal cells (thecocytes)
    - ➢ build up the *theca interna*
    - ➢ steroid-secreting cells – estrogens
    - ➢ ovulation ⇨ lutein cells – progesterone
  - ✓ interstitial cells
    - ➢ active thecal cells
    - ➢ in small groups throughout the cortical stroma around vessels
    - ➢ source of ovarian androgens
  - ✓ hilus cells
    - ➢ in ovarian medulla
    - ➢ similar to *Leydig* cells in testis
    - ➢ produce testosterone

# Oocytes

- oogonia – mitotically active cells
  - ✓ reduction in number – cell death
  - ✓ primordial follicles
- primary oocytes:
  - ✓ medium-sized cells – 25-30 μm
  - ✓ prophase of the first meiotic division
  - ✓ diploid







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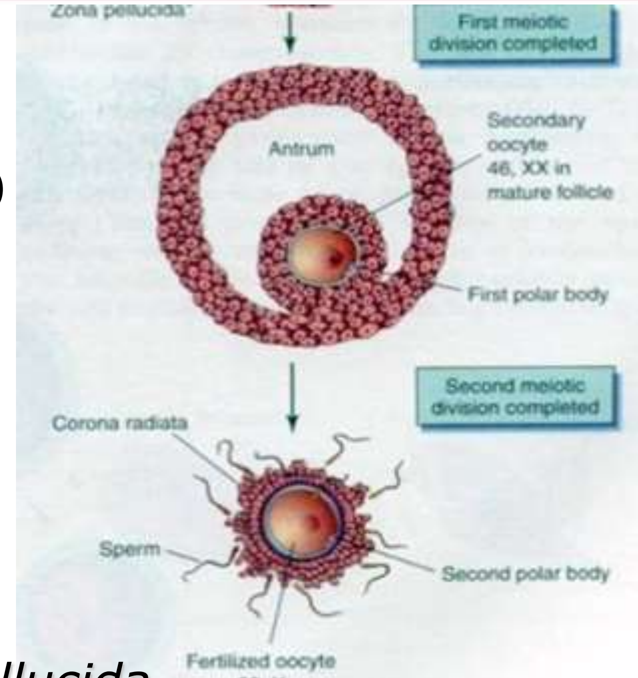
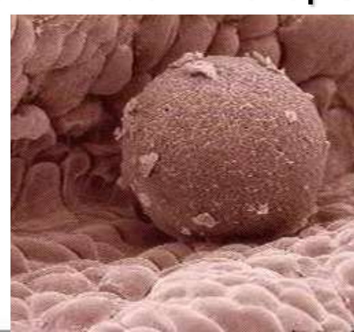
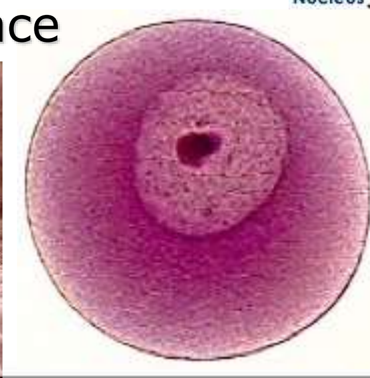
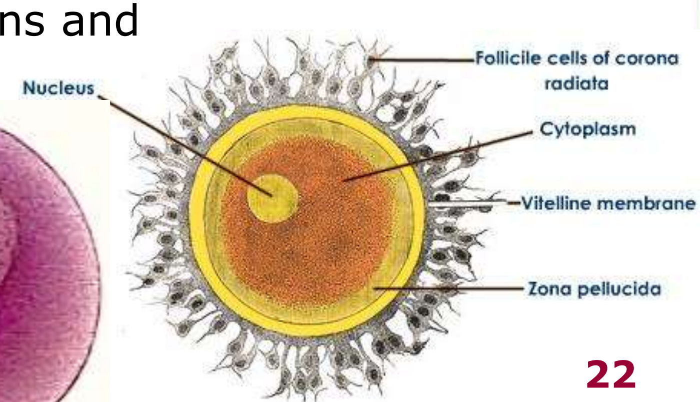
# Oocytes

- **secondary oocytes:**
  - ✓ larger in size – 40-50 μm
  - ✓ 2nd meiotic division (metaphase)
  - ✓ haploid – 23 chromosomes
  - ✓ normal amount of DNA (2n)
- **ovum (mature oocyte):**
  - ✓ large cell – 50-150 μm
  - ✓ large nucleus with haploid number of chromosomes
  - ✓ oolemma with microvilli
  - ✓ acidophilic PAS-positive *zona pellucida*, glycosaminoglycans, glycoproteins and sialic acid, source of fertilizine ⇨ perivitelline space



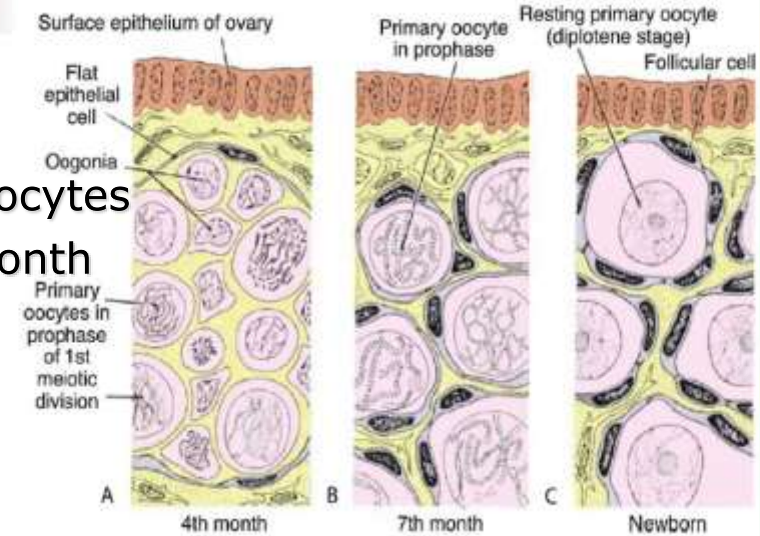



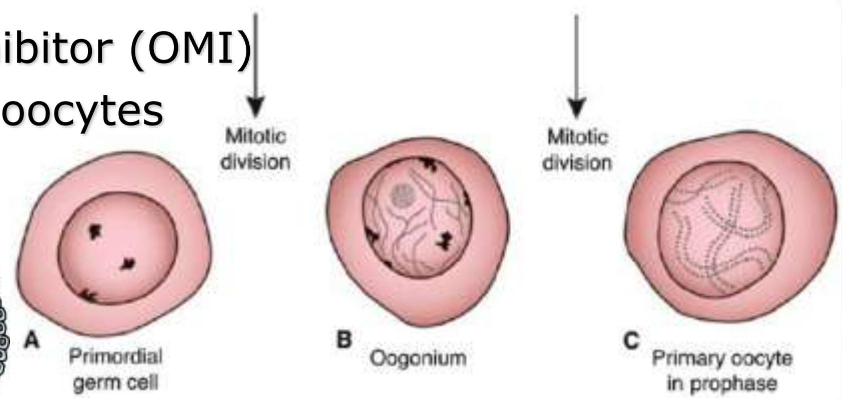


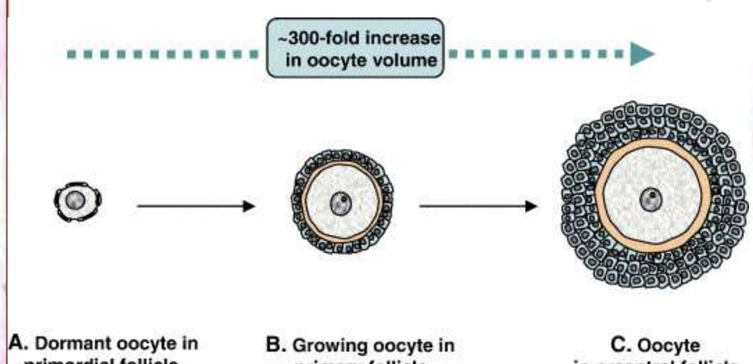


# Oogenesis

- oogonia ⇨ mature oocytes (ova):
  - ✓ in female gonads – ovaries
- prenatal stage:
  - ✓ period of proliferation – gonocytes
    - ✓ oogonia – 7 million/5$^{th}$ month
    - ✓ primary oocytes – 700000-2 million
- postnatal stage:
  - ✓ growth – primary oocytes
    - ➢ remain in prophase of meiosis I (diplotene stage)
    - ➢ oocyte maturation inhibitor (OMI)
  - ✓ maturation – secondary oocytes

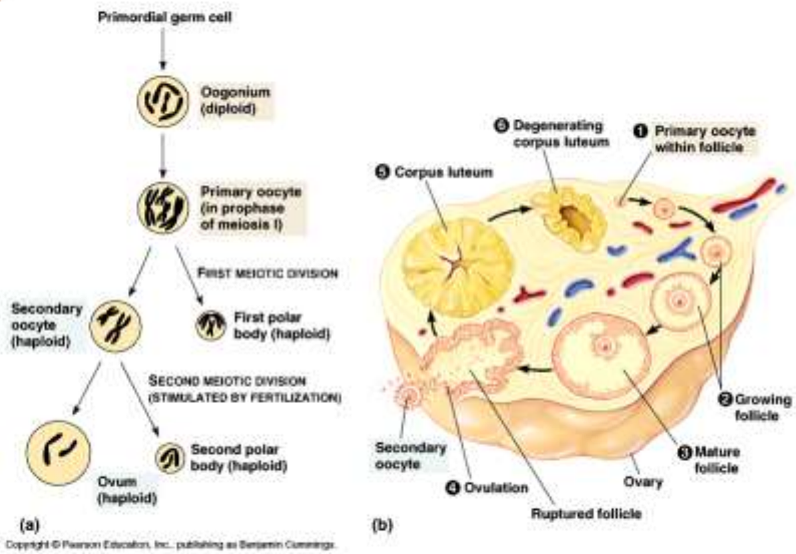


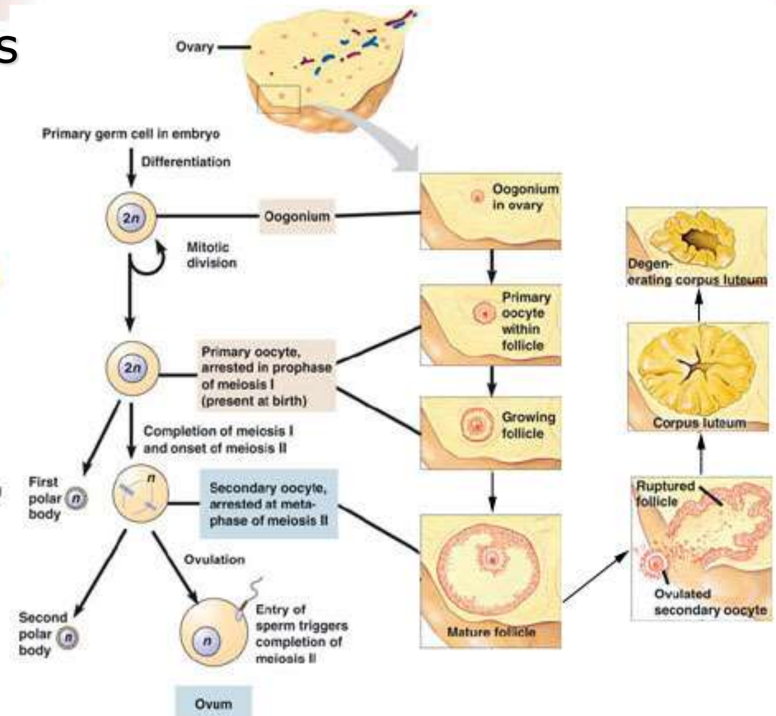



# Oogenesis

- oogonia ⇨ mature oocytes (ova):
  - ✓ in female gonads – ovaries

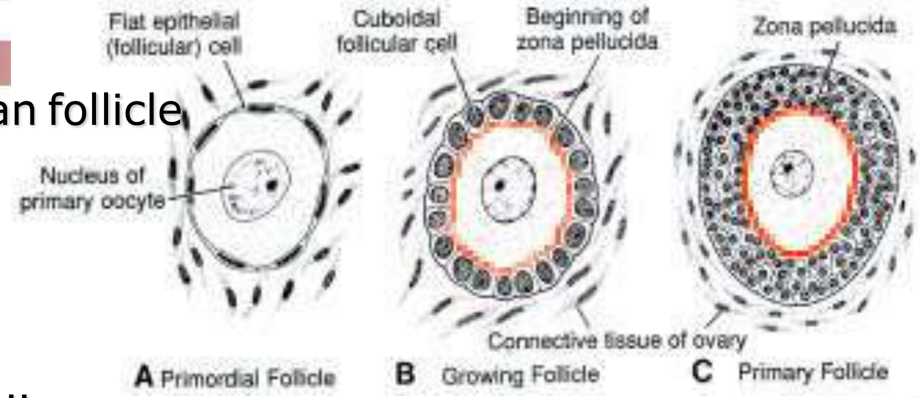


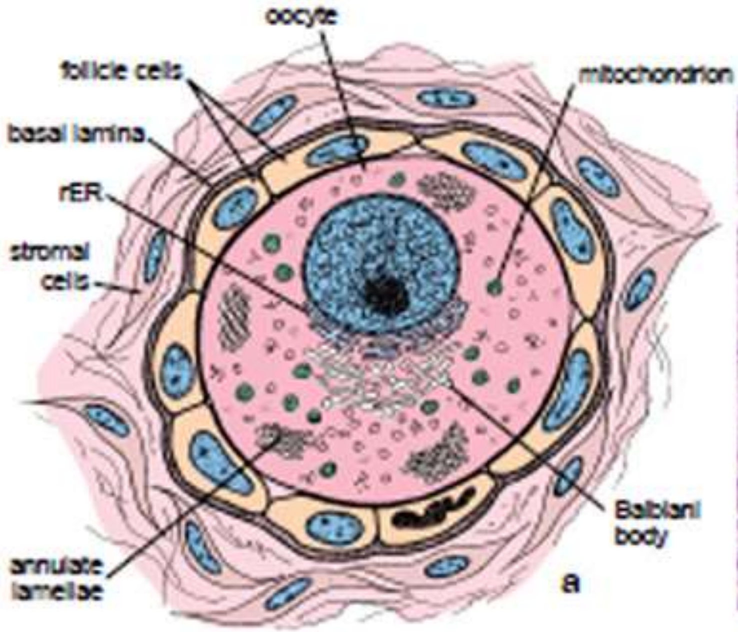

- peculiarities of oogenesis:
  - ✓ first meiotic division (meiosis I) begins during fetal life and is completed just before ovulation
  - ✓ meiosis II is completed only if the oocyte is fertilized
  - ✓ one mature oocyte (ovum) and three polar bodies are formed from one oogonium
  - ✓ different structural peculiarities in different animal species

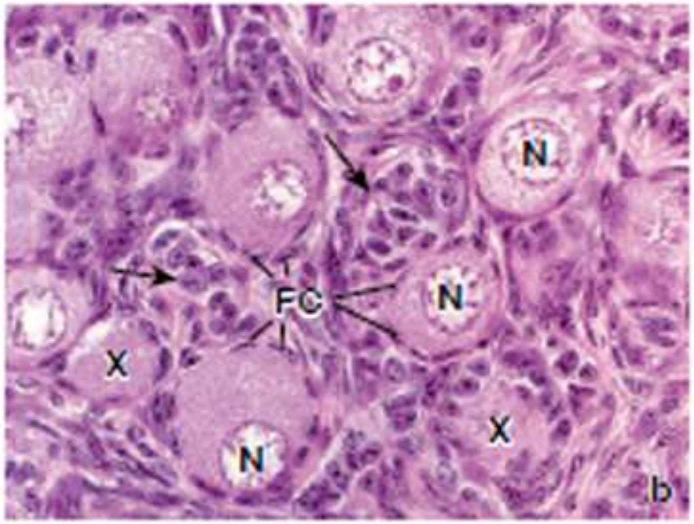
# Folliculogenesis

- Folliculogenesis:
  - maturation of the ovarian follicle
  - regulated by FSH




✓ primordial follicle
  - primary oocyte
  - flattened follicular cells (single layer)



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# Folliculogenesis

- Folliculogenesis:
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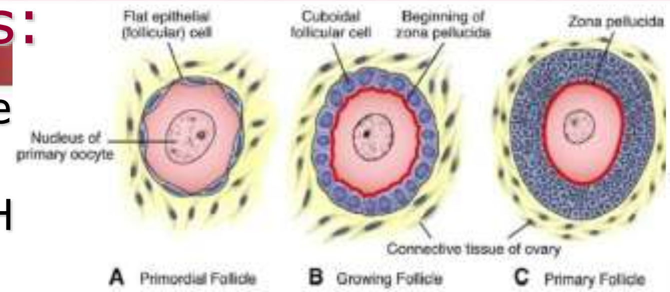
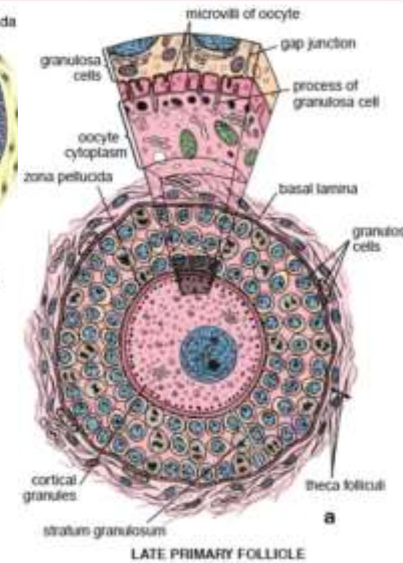
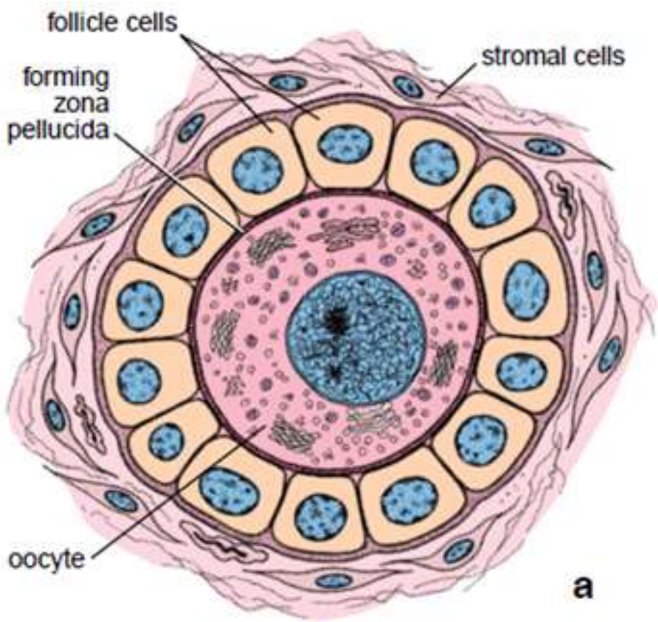
  - regulated by FSH

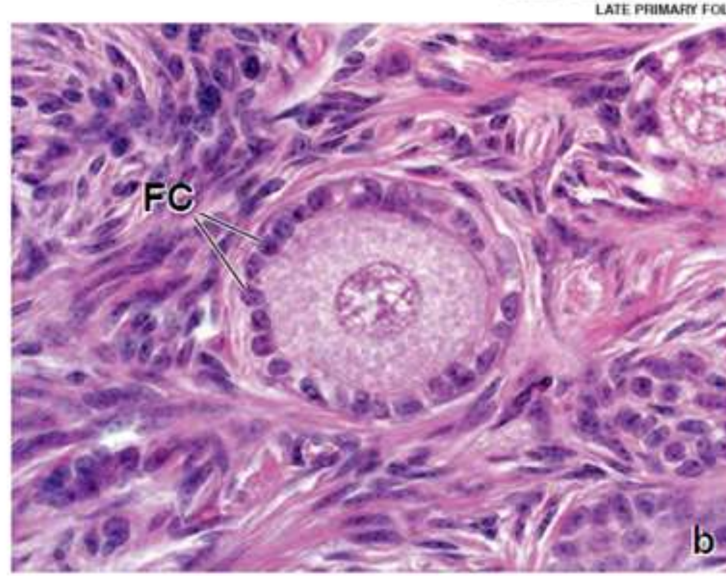



✓ primary follicle – 35-40 µm
- primary oocyte – 25-30 µm
- cuboidal follicular cells



PRIMARY FOLLICLE



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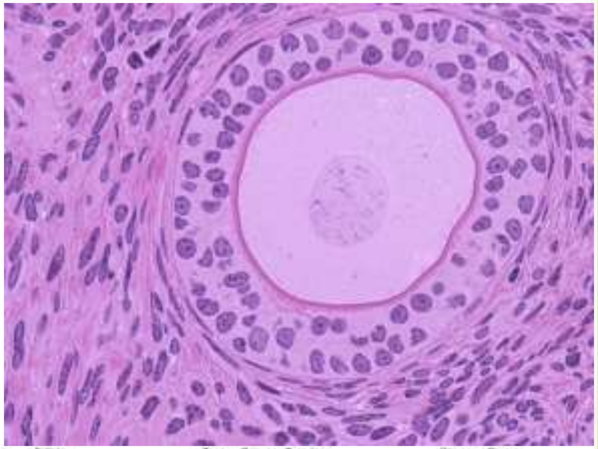
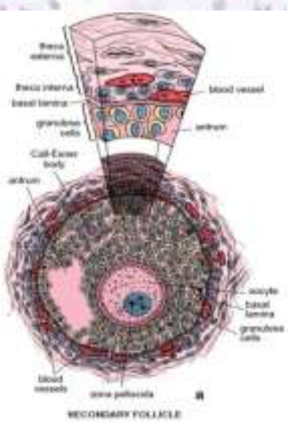
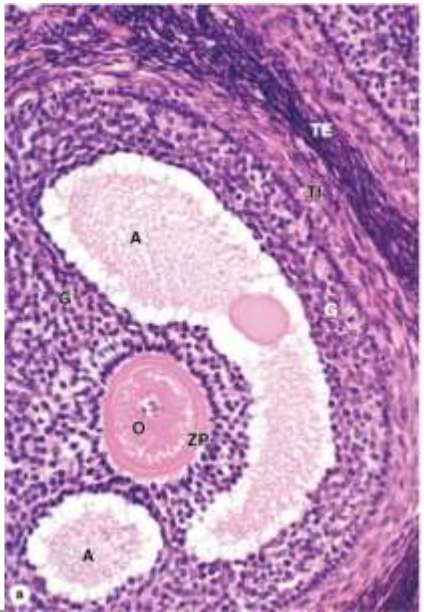
# Folliculogenesis

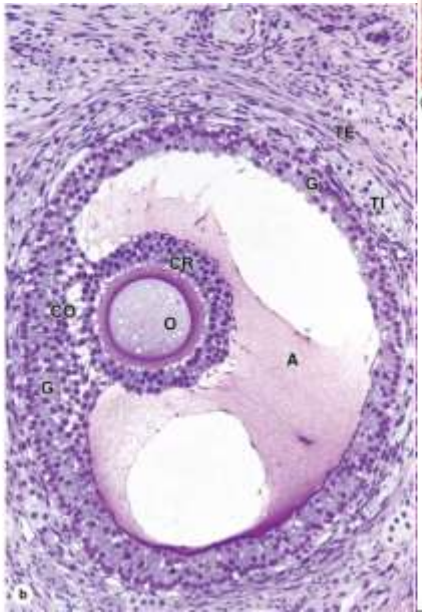
- Folliculogenesis:
  - maturation of the ovarian follicle
  - regulated by FSH

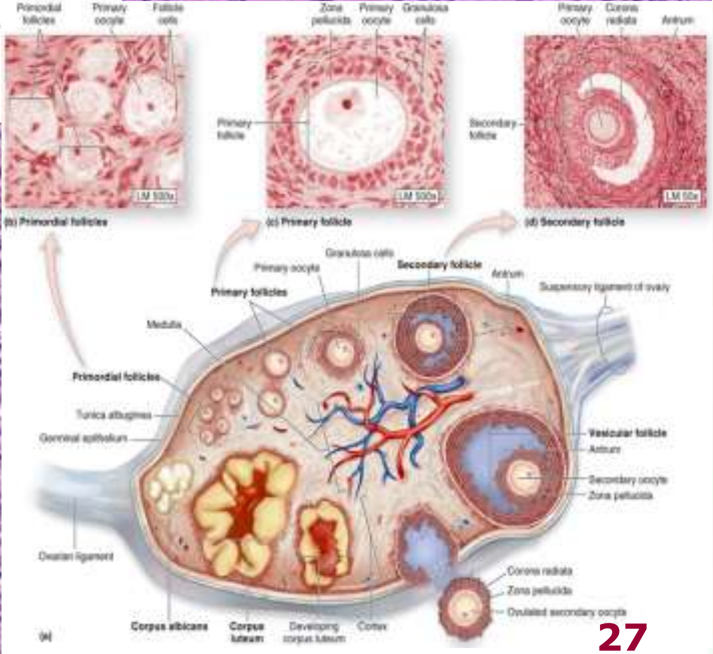
✓ secondary follicle (growing or antral) – 0.2 mm
  - primary oocyte (50-80 μm) with *zona pellucida*
  - follicular cells (several layers)
  - *antrum folliculi* and *liquor folliculi*
  - *theca folliculi interna et externa*







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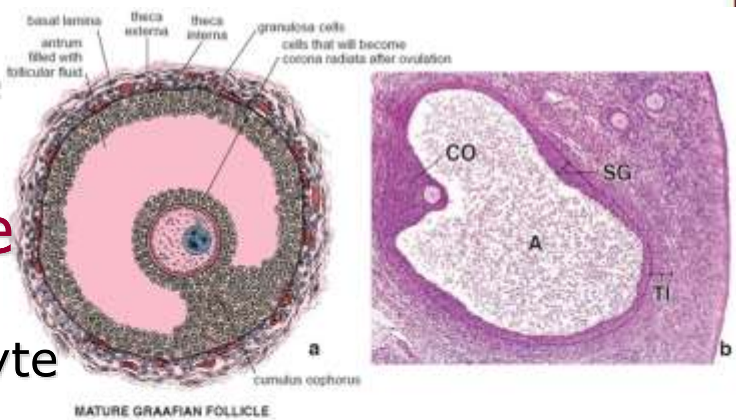
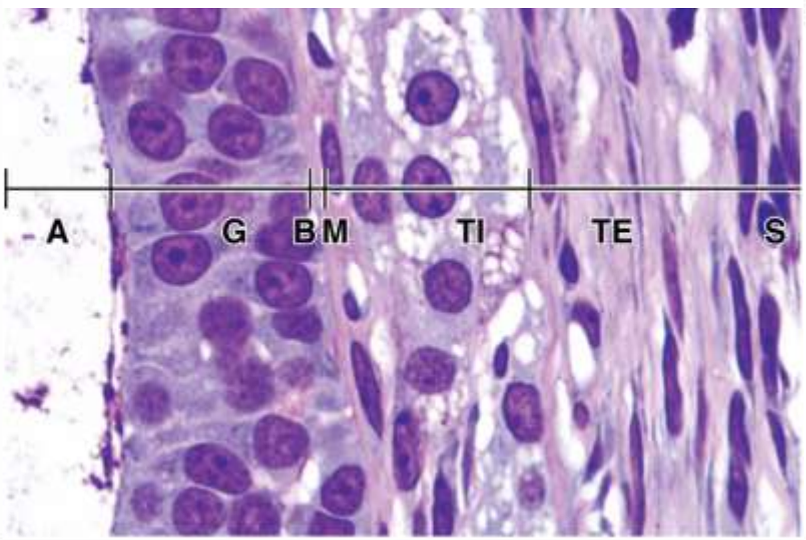
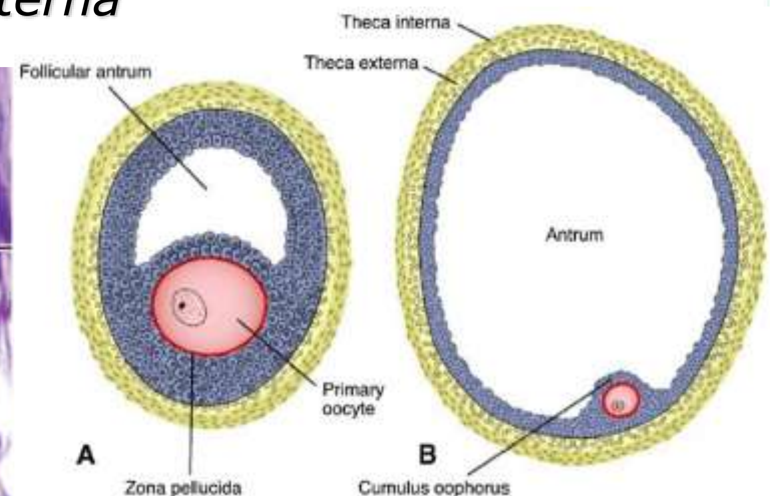
# Folliculogenesis

- Folliculogenesis:
  - maturation of the ovarian follicle
  - regulated by FSH

✓ mature (Graafian) follicle
– 10-25 mm
- large (125 µm) primary oocyte
  - ⇨ *cumulus oophorus (ovaricus)*
- *membrana (stratum) granulosa*
- *theca folliculi interna et externa*




***Regnier de Graaf (1641-1673)***



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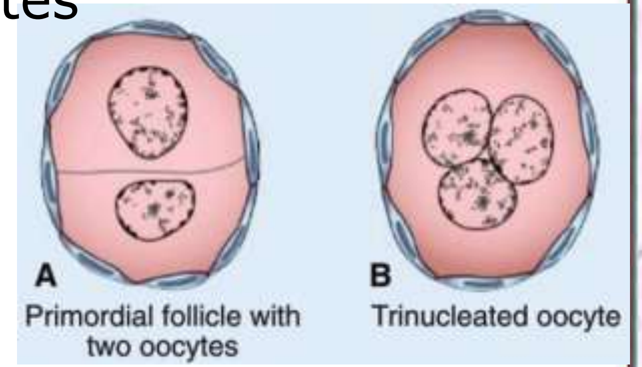
# Abnormal gametes

- in humans and in most mammals:
  - ✓ one ovarian follicle occasionally contains two or three clearly distinguishable primary oocytes
    - ➢ usually degenerate before reaching maturity
    - ➢ twins or triplets
  - ✓ one primary oocyte contains two or three nuclei
    - ➢ die before reaching maturity





Abnormal oocytes

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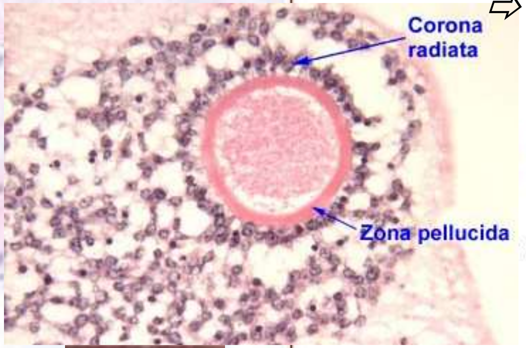
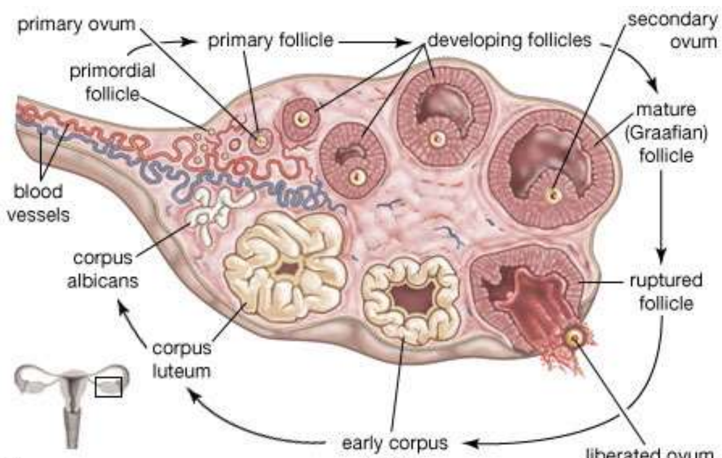
# Ovulation

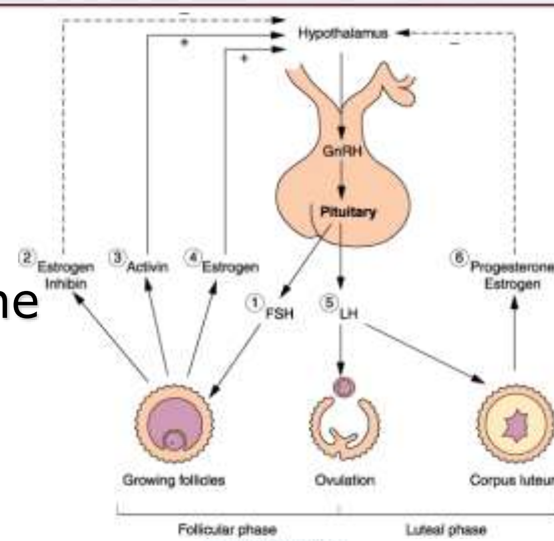
- ovulation:
  - ✓ the process by which an oocyte is released from the Graafian follicle
  - ✓ a cyclic process, blocked during pregnancy
  - ✓ takes place around 14-15 day
  - ✓ preovulatory Graafian follicle ⇨ *stigma*
  - ✓ liberation of the ovum ⇨ *corona radiata*

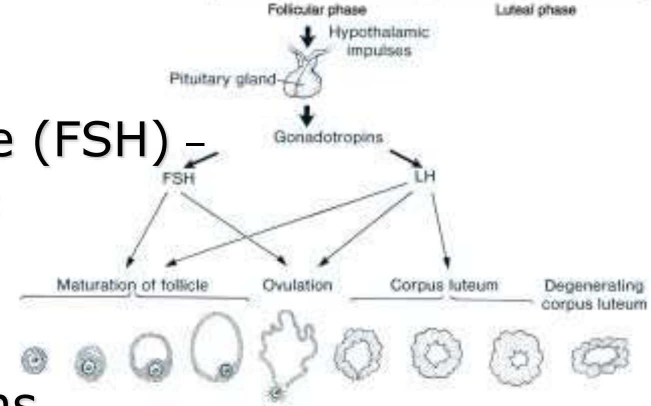


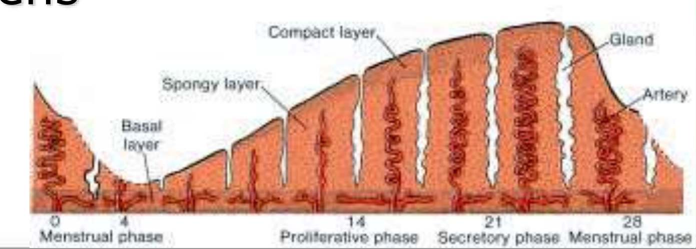



# Hormonal control of ovarian functions

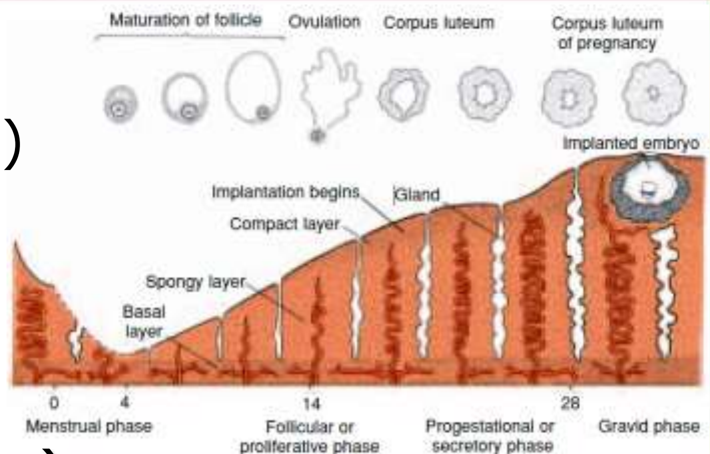
- regulation of the ovarian cycle:
  - ✓ hypothalamus
    - ➢ gonadotropin-releasing hormone (GnRH)
    - ➢ portal system
  - ✓ adenohypophysis – gonadotrope hormones:
    - ➢ follicle-stimulating hormone (FSH) – folliculogenesis ⇨ estrogens
    - ➢ luteinizing hormone (LH) – ovulation and development of corpus luteum ⇨ gestagens



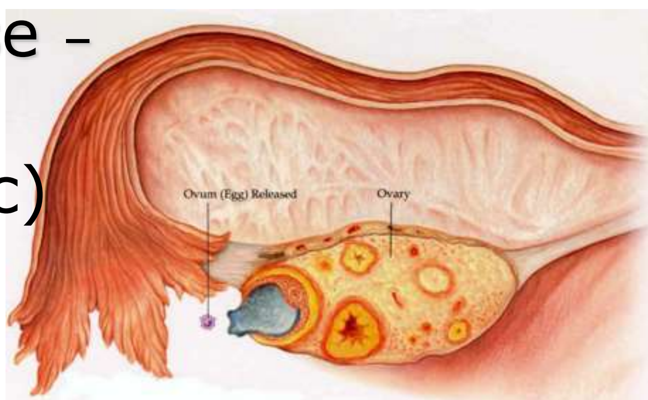



# Menstrual cycle




- menstrual cycle (28 days) – phases:
  - ✓ menstrual – 3-5 days (menstruation)
  - ✓ proliferative (follicular) phase – 5-14 day
  - ✓ secretory (luteal) phase – till 26-27 day
  - ✓ premenstrual (ischemic) phase – 1-2 days
- fertile window – ~7 days
  the time from 5 days before until 1–2 days after ovulation


**Menstrual cycle of 28 days**



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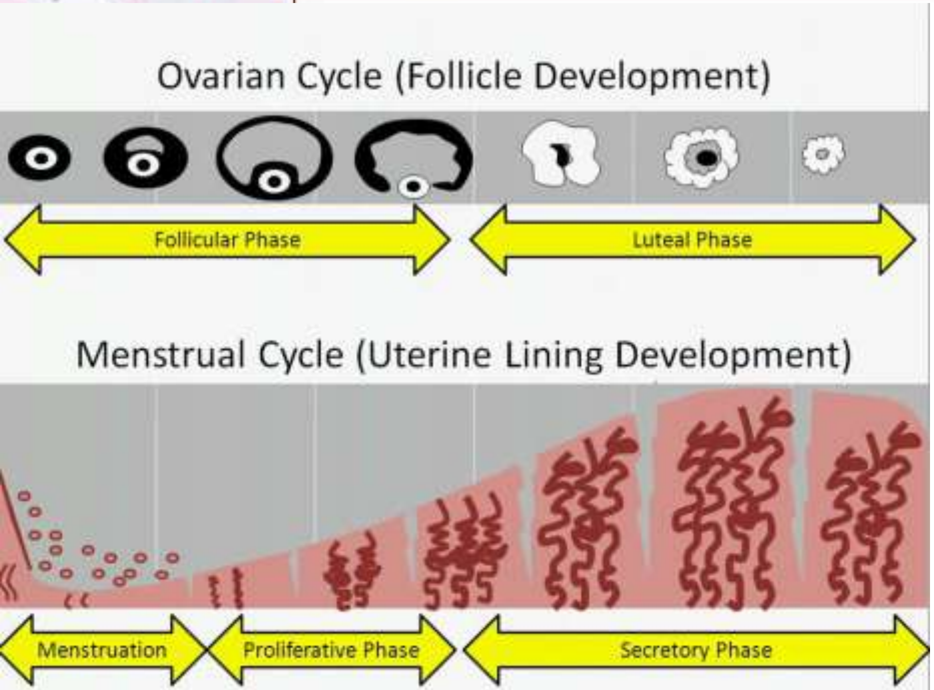
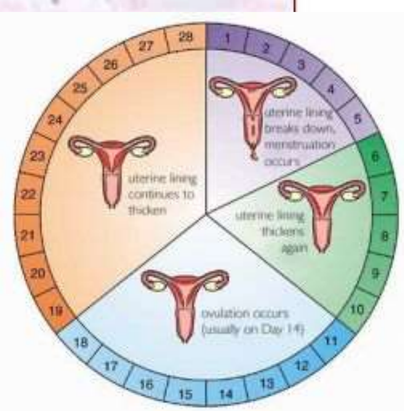
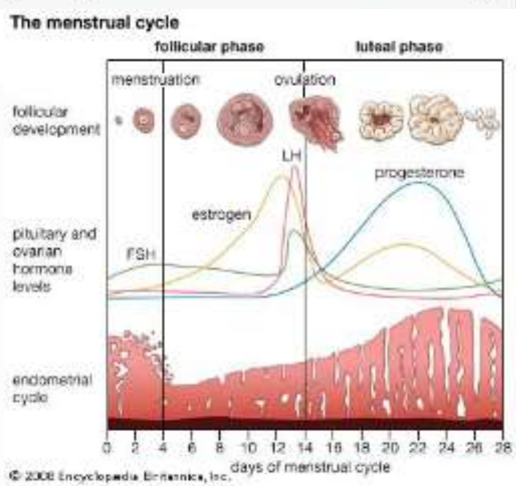
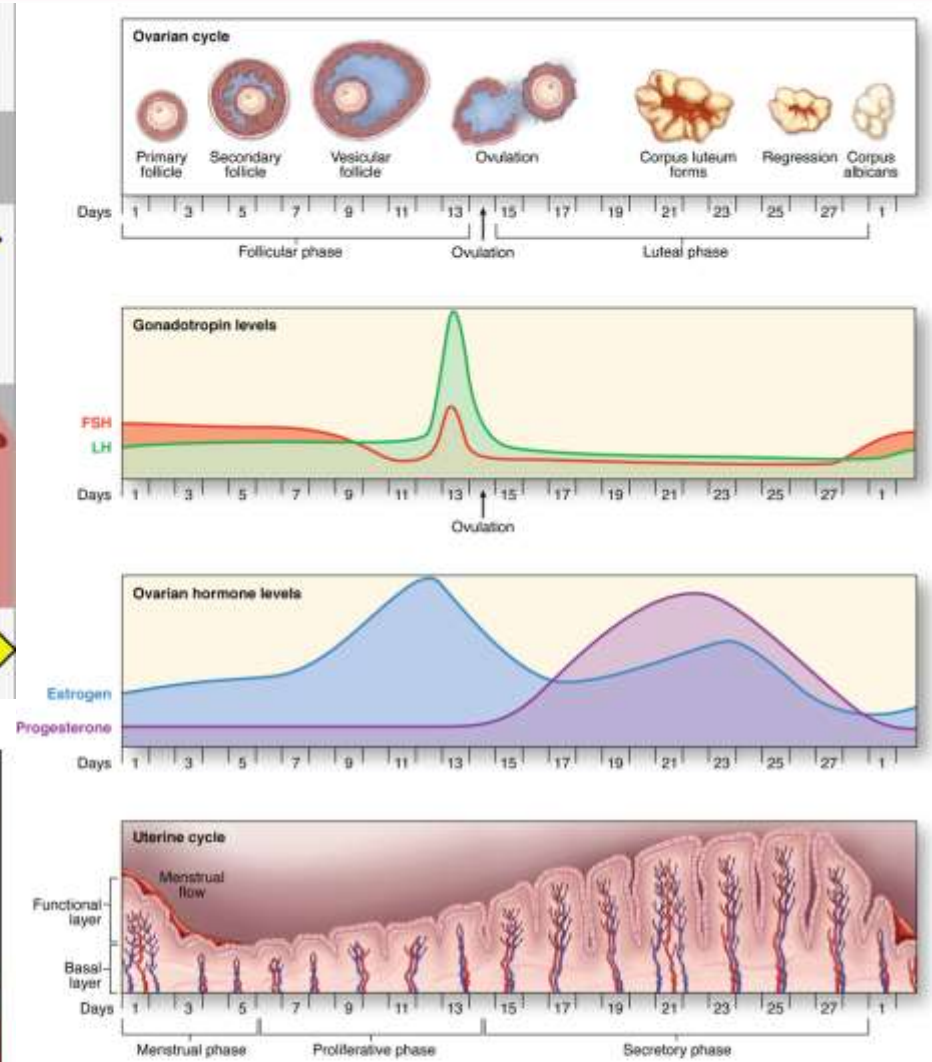
# Menstrual vs. ovarian cycle



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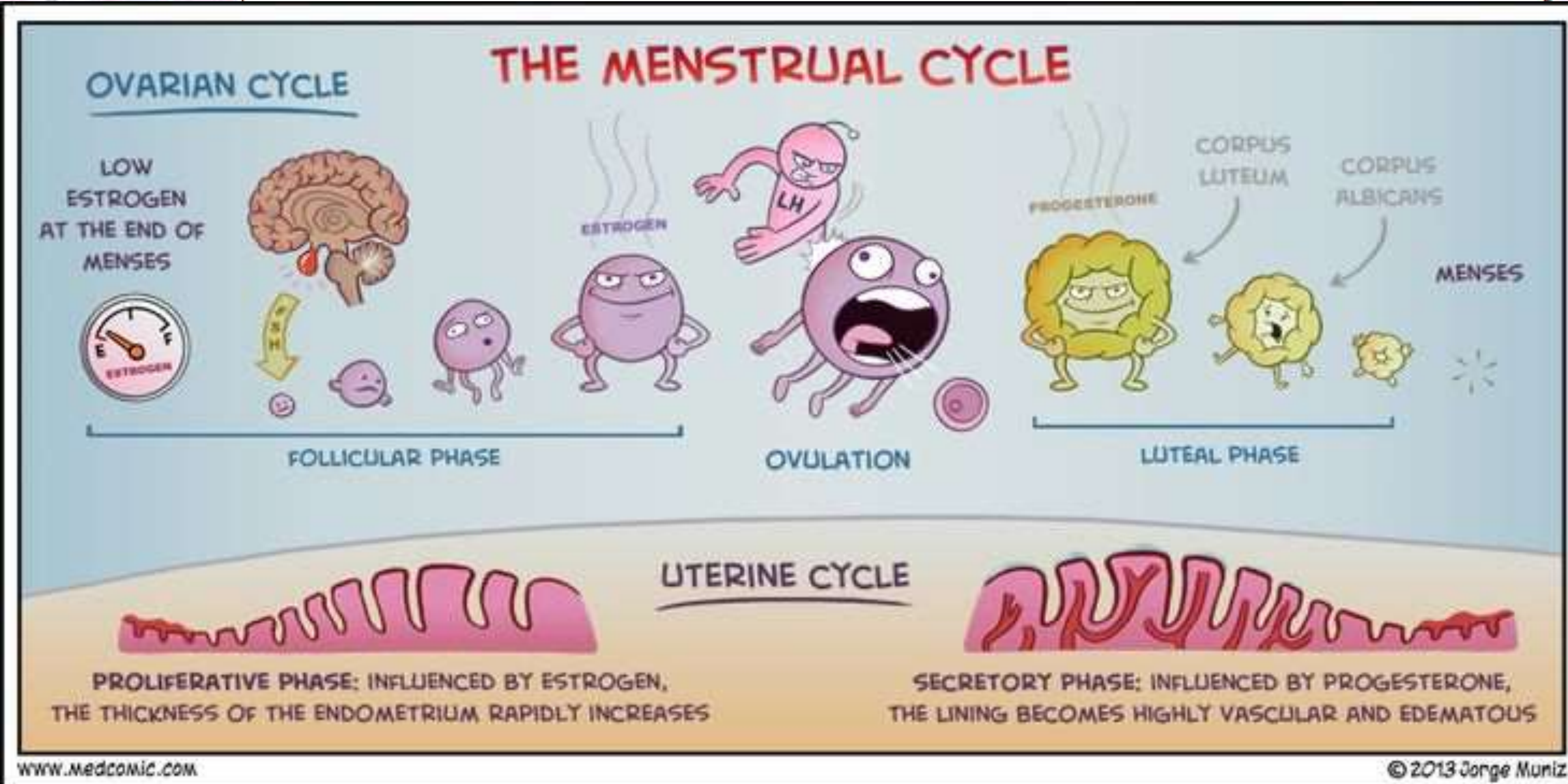
# Menstrual vs. ovarian cycle




*Thank you…*